

BOOK OF ABSTRACTS

XXXIV National Meeting of Biochemistry
October 20-25, 2024 | Mazatlán, Sinaloa



SMB  **SOCIEDAD
MEXICANA DE
BIOQUÍMICA**

BOOK OF ABSTRACTS

XXXIV National Meeting of Biochemistry

October 20-25, 2024 | Mazatlán, Sinaloa

SOCIEDAD MEXICANA DE BIOQUÍMICA

Mesa Directiva 2023-2025

Dr. Agustín Guerrero Hernández
Presidente

Dra. Libia Vega Loyo
Vicepresidente

Dra. . Lourdes Girard Cuesy
Secretaria Tesorera

Dra. Irma Romero Álvarez
Subsecretaria Tesorera

Comisión de Admisión 2023-2025

Dra. Libia Vega Loyo
Presidente

Dra. Mina Königsberg Fainstein
Dra. Lina Riego Ruiz
Dr. Jesús Alberto Olivares Reyes
Dr. Ricardo Espinosa Tanguma

D.A.R. 2024. MEMORIAS. XXXIV National Meeting of Biochemistry.

La reproducción o traducción de esta obra requiere el permiso escrito de la institución que lo edita.

Pueden reproducirse sin autorización pequeños fragmentos del texto y figuras aisladas, siempre que se den los créditos correspondientes. © Sociedad Mexicana de Bioquímica

Organizing Committee

Agustín Guerrero Hernández, Libia Vega Loyo, Lourdes Girard Cuesy, Irma Romero Álvarez

Editorial design: Paola Marfil Lara

Technical Support: María Teresa Castillo

Cover Image: Alejandro Maturano Mendoza

Published in Mexico, October 2024

ACKNOWLEDGEMENTS

We would like to express our gratitude to the following Institutions, Societies and Companies that provided financial and logistic support to this meeting:

Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional
Universidad Nacional Autónoma de México
Coordinación de la Investigación Científica, UNAM
Secretaría de Desarrollo Institucional, UNAM
Centro de Ciencias Genómicas, UNAM
Instituto de Fisiología Celular, UNAM
Instituto de Biotecnología, UNAM
Facultad de Química, UNAM
Revista de Educación Bioquímica (REB)
Panamerican Association for Biochemistry and Molecular Biology
Sociedad Española de Bioquímica y Biología Molecular
Secretaría de Turismo del Gobierno del Estado de Sinaloa
BIO-RAD Laboratories
CTR Scientific
Científica Senna
FALCON
GRUPO RAVER
QUÍMICA VALANER-MGI
UNIPARTS



Agradecemos al Consejo Nacional de Humanidades, Ciencias y Tecnologías (CONAHCYT) por el apoyo otorgado a la Sociedad Mexicana de Bioquímica para la organización de estos eventos, mediante el proyecto número 318661 "Apoyo para la organización de los Congresos Nacionales de la Sociedad Mexicana de Bioquímica, A.C." dentro de la convocatoria "Fortalecimiento de Actividades vinculadas con la Promoción, Difusión y Divulgación de las Humanidades, Ciencias, Tecnologías y la Innovación: Academias y Sociedades Científicas 2021".

The Organizing Committee thanks the following persons:

Drs. David Romero and Soledad Funes for their support on the achievement of CONACYT Project (318661).

The Academic Committee for their priceless support in the review of the oral abstracts, and the evaluation of "Premio Socios Fundadores".

Also the organizing committee is deeply grateful to all who helped in the preparation of the scientific program and to all the speakers for accepting our invitation unconditionally.

Teresa Castillo Martínez for her invaluable help in coordination and logistics of this event.

Biol. Andrea Ortiz Arcos for her dedication and support in the different stages of the organization of this meeting.

Lic. Angélica González Ceja for her help and support in the administrative issues with Cinvestav

Paola Marfil Lara for the assistance in preparation of this book.

Dear attendees:

On behalf of the organizing committee, Lourdes Girard, Libia Vega, and Irma Romero, I welcome you to this beautiful Mazatlán International Convention Center to hold the XXXIV Mexican Biochemistry Society Meeting.

We have put together a fantastic program to have a warm and fruitful interaction among all of us. We hope you enjoy the meeting, find new collaborations, and that the students get new opportunities to advance their work. This meeting will have plenary talks by internationally renowned scientists, in particular, we have the presence of Frances Arnold, the 2018 Nobel Prize in Chemistry laureate. Additionally, there will be plenary symposia, simultaneous symposia, oral presentations, meet and greet “having coffee with,” flash talks, and poster sessions. All these activities have been organized to facilitate the interaction among all participants.

The topics that will be covered in this meeting are state-of-the-art in biochemistry and include bioenergetics and biomembranes, signal transduction, molecular biology, protein structure, bioinformatics and systems biology, medical biochemistry, plant biochemistry, microbiology, toxicology, pharmacology, among others.

Mazatlán, the “Pearl of the Pacific,” isn’t just known for its stunning beaches and vibrant nightlife but also for its mouthwatering seafood that defines the city’s culinary scene. Sandy beaches line its 21km-long malecón (boardwalk), and it’s renowned for big-game fishing. In its Centro Histórico, or Old Mazatlán, one 19th-century landmark includes the performance hall Teatro Ángela Peralta. The Mazatlan aquarium has 34 saltwater tanks and 17 freshwater aquariums for visitors to admire a wealth of marine life. Interactive highlights include shows featuring sea lions, tropical birds, and live feedings of nurse sharks and surgeon fish.

TABLE OF CONTENTS

Scientific Program	9
Sunday, October 20	10
Monday, October 21	10
Tuesday, October 22	16
Wednesday, October 23	22
Thursday, October 24	28
Poster Session	33
Poster Session I	34
Poster Session II	59
Poster Session III	84
Poster Session IV	109
Plenary Lectures	134
Plenary Symposia	146
Simultaneous Symposia	159
Simultaneous Oral	190
Technical Talks	251
Poster Session	258
Basic Biochemistry	258
Biotechnology	377
Genetics, Epigenetics and Genetic Regulation	544
Immunology & Parasitology	653
Medicine, Health & Nutrition	691
Microbiology	803
Neurosciences & Neurobiology	928
Reactive Oxygen Species	983
Signal Transduction	1009
Systems Biology & Bioinformatics	1073
Toxicology & Pharmacology	1127
Others	1180
Author Index	1265



SCIENTIFIC PROGRAM

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

SUNDAY, OCTOBER 20

17:15 - 17:30	Welcome Ceremony (Mazatlán Room 2)
17:30 -18:15	Cultural Talk (Mazatlán Room 2) Baja California Rock Art Enah Montserrat Fonseca. Centro INAH Baja California. Chair: Agustín Guerrero. Cinvestav Zacatenco.
18:30-19:45	Opening Lecture I (Mazatlán Room 2) The genetic history of Mexico seen through a Paleogenomics lens María Ávila Arcos. Laboratorio Internacional de Investigación sobre el Genoma Humano, UNAM. Chair: Lourdes Girard. Centro de Ciencias Genómicas, UNAM.
20:00-22:00	Welcome Cocktail (<i>Explanada Mantarraya, MIC</i>)

MONDAY, OCTOBER 21

8:30-10:00	Plenary Symposia I (Mazatlán Room 2)
Impact of Nanotechnologies Applied to Human Health Chair: Libia Vega Loyo, Cinvestav Zacatenco.	
Bacterial growth inhibited by hydrophobic nano emulsions Jesús Carlos Ruiz Suárez, Cinvestav Unidad Monterrey.	
Challenges and opportunities of nanobiotechnology in the development of next-generation vaccines Francisca Villanueva Flores, CICATA IPN Unidad Morelos.	
Virus-based enzymatic nanoreactors for enzyme replacement therapy Rafael Vázquez Duhalt, Centro de Nanociencias y Nanotecnología, UNAM.	

<p>10:00-11:00</p>	<p>Plenary Lecture II (Mazatlán Room 2)</p> <p>The common bean (<i>Phaseolus vulgaris</i>)– <i>Rhizobium etli</i> N-fixing symbiosis: unraveling novel plant regulators through genetic/genomic approach</p> <p>Georgina Hernández Delgado, Centro de Ciencias Genómicas, UNAM. Chair: Lourdes Girard, Centro de Ciencias Genómicas, UNAM.</p>
<p>11:00-11:30</p>	<p>Coffee Break</p>
<p>11:30-12:45</p>	<p>Simultaneous oral Sessions 1, 2, 3</p>
<p style="text-align: center;">Session 1 (Mazatlán Room 1) Chair: Ruth Gutiérrez, Facultad de Medicina, UNAM.</p>	
	<p>Joint microbiome in a spondyloarthritis murine model: exploration of GUT-JOINT axis</p> <p>Susana Aideé González Chávez, Universidad Autónoma de Chihuahua.</p>
	<p>Energetic impairment in cardiometabolic heart failure: from transcriptomics and mitochondrial complexome profiling to cardiomyocyte respiration</p> <p>Abraham Méndez Fernández, Institute for Obesity Research, Tecnológico de Monterrey.</p>
	<p>Restorative effects of (+)-epicatechin in a rodent model of aging induced muscle atrophy: underlying mechanisms</p> <p>Carlos Palma Flores. Escuela Superior de Medicina, IPN.</p>
	<p>Targeting Ornithine Decarboxylase for Cancer Therapy: Assessing Inhibitor Efficacy on <i>in vitro</i> assays</p> <p>Beatriz Irene Arroyo Sánchez, Cinvestav Zacatenco.</p>
	<p>Overview of circulating microRNAs and genomics variants in prediabetes patients presenting different clinical courses: search for novel informative molecules</p> <p>Sandra Romero Córdoba, Instituto de Investigaciones Biomédicas, UNAM.</p>
<p style="text-align: center;">Session 2. Mazatlán Room 2 Chair: Gloria Soberón, Instituto de Investigaciones Biomédicas, UNAM.</p>	
	<p><i>Trichoderma atroviride</i> small RNA1 targets the <i>Arabidopsis</i> PRIM2 gene to establish a mutualistic relationship</p> <p>Sergio Casas Flores, Instituto Potosino de Investigación Científica y Tecnológica A.C.</p>

Cell division and morphology changes caused by *cenR* overexpression affect the symbiotic nitrogen fixation in *R. etli* CFN42

María M. Banda Hernández, Centro de Ciencias Genómicas, UNAM.

The role of the regulatory protein PhbF in the control of accumulation of the biodegradable plastic polyhydroxybutyrate in the bacterium *Azotobacter vinelandii*

Thalía Barrientos Millán, Instituto de Biotecnología, UNAM.

Linking Elevated Proteolysis with CpxR Regulatory Mechanisms in *Serratia marcescens* HU1848

Karla Lizeth De Anda Mora, Universidad Autónoma de Nuevo León.

Effect of Dengue virus serotype 2 (DENV2) on the Pregnane X Receptor (PXR) signaling pathway in mouse peritoneal macrophages

Carlos Daniel Bautista Olivier, Cinvestav Zacatenco.

Session 3. Mazatlán Room 3

Chair: Jesús Alberto Olivares Reyes. Cinvestav

Oligomeric regulation of cystathionine beta-synthase mediated hydrogen sulfide production modulates UPR induction

Eliás Nieto Zaragoza. Instituto de Fisiología Celular. UNAM

Multivalent Dynamic Colocalization of Avian Influenza Polymerase and Nucleoprotein by Intrinsically Disordered ANP32A Reveals the Molecular Basis of Human Adaptation

Aldo Román Camacho Zarco. Université Grenoble Alpes

Molecular Handcraft of Chimeric Proteins

José Arcadio Farías Rico. Centro de Ciencias Genómicas. UNAM

Decoding the mechanism governing the structural stability of wheat germ agglutinin

Enrique García Hernández. Instituto de Química. UNAM

Biochemical study of MDM2 and Rb mRNA interaction

Andrés Usiel Rodríguez Rodríguez. Universidad Autónoma de San Luis Potosí

11:30-12:45	Technical Talks (Carnaval Room 1, 2)
	Tools and Solutions for 3D Cell Culture Sebastián Hernández. UNIPARTS
	Quality Analysis Using the Agilent ProteoAnalyzer System and SDS-PAGE. A comparison of sizing and quantification performance Daniel Favato. Genomics Field Application Scientist. Latin America. Agilent Technologies (Patrocinado por Química Valaner)
12:45-13:45	Plenary Lecture III (Mazatlán Room 2) Channels of communication in sperm Alberto Darszon, Instituto de Biotecnología, UNAM Chair: Lourdes Girard. Centro de Ciencias Genómicas, UNAM
13:45-14:15	Flash Talks for poster advertising 1 (Mazatlán Room 2) Chair: Lourdes Girard. Centro de Ciencias Genómicas, UNAM
	FT1 Characterization and evaluation of the biological activity of the TA-PAsp polymer Ileana Castillo Tobías. Universidad Autónoma de Coahuila
	FT2 <i>In silico</i> analysis of the C8 clone and toxic activity of the ATP-dependent RNA helicase from the entomopathogenic bacterium <i>Serratia entomophila</i> Mor 4.1 in <i>Phyllophaga blanchardi</i> larvae (Coleoptera: Melolonthidae) Luz Areli Gorostieta Nava. Universidad Autónoma del Estado de Morelos
	FT3 Doing Bionanotechnology with dsDNA and CRISPR-dCas12a: Designing Linear and Branched Nanofibers Armando Hernández García. Instituto de Química. UNAM
	FT4 Mechanistic assessment of the biosynthesis of cyanobacterial secondary metabolites: Two cases José Alberto Martínez. Yereva Czech Academy of Sciences
	FT5 Characterization of recombinant modified trichocystatin TC-2 Jaime Ortega López. Cinvestav Zacatenco
	FT6 Effect of prolactin and 17β-estradiol on the adhesion and intracellular persistence of <i>Staphylococcus aureus</i> in bovine mammary epithelial cells Gladys Romero Corona. Universidad Michoacana de San Nicolás de Hidalgo

FT7 Immobilization of *thermomyces lanuginosus* lipase (TLL) via ionic-covalent interaction on heterofunctional glyoxyl-agarose supports for biodiesel production

Cristian David Salcedo Cuarán. Universidad del Valle

FT8 Characterization of DyP-type peroxidases and their use as biocatalysts in textile dyes decolorization

Jesús Alberto Segovia Cruz. Instituto de Biotecnología. UNAM

FT9 Genetic diversity and phenotypic variations in four native strains of *Bacillus* spp. biocontrol agents through pan-genome and BGC analysis

Hilda Mabel Sosa Esquivel. Universidad Autónoma de Zacatecas
"Francisco García Salinas"

FT10 Physiological evaluation of recombinant forms of granulocyte colony-stimulating factor

Miguel Ángel Valle Yañez. Cinvestav Zacatenco

FT11 Purification and Valorization of Urban Effluents Using Photobioreactors Operated with *Anabaena Inaequalis*

Wendy Zárate Hernández. Instituto Tecnológico de Veracruz

FT12 Dynamics of Bacterial Communities in Response to Disturbances

Roberto Carlos Alvarez Martínez. Universidad Autónoma de Querétaro

FT13 Key Proteins for Regeneration in *A. mexicanum*: Transcriptomic Insights from Aged and Juvenile Limbs

Aylin Del Moral Morales. Universidad Autónoma Metropolitana

FT14 Functional and structural studies of a novel endolysin from ICPI phage active against multidrug-resistant bacteria

Laura Angélica Espinosa Barrera. Universidad de Colima

FT15 Unraveling the distinct biases of the genomic landscape of lung adenocarcinoma from Mexican patients

Bertha Rueda Zarazúa. Instituto Nacional de Medicina Genómica. UNAM

14:15-15:30

Lunch

15:30-17:00

Simultaneous Symposia 1, 2, 3
(Mazatlán 1, 2, 3)

1. Symposium on neurobiology

Mazatlán Room 1

Chair: Susana Castro Obregón, Instituto de Fisiología Celular, UNAM

Role of autophagy in neurodevelopment and brain aging

Susana Castro Obregón, Instituto de Fisiología Celular, UNAM

Circadian rhythms as regulators of energy homeostasis

Lucía Mendoza Viveros, Instituto Potosino de Investigación Científica y Tecnológica, A. C.

Social neuroscience and culture of peace

Roberto Mercadillo Caballero, Universidad Autónoma Metropolitana-Iztapalapa

**2. Growth control and metabolism in fungi
in memoriam Luis Caspeta Guadarrama**

Mazatlán Room 2

Chair: Alicia González, Instituto de Fisiología Celular, UNAM

Branched-chain amino acid pathway regulation and its connection to carbon metabolism during yeast diauxic shift

Ximena Escalera Fanjul, Theoretical Biophysics, Humboldt University, Berlin

The convergence of molecular and metabolic responses to thermo-acidic stress in yeast

Alfredo Martínez Jiménez, Instituto de Biotecnología, UNAM

The ergosterol biosynthesis pathway in *Candida* spp: antifungal compounds and their metabolic implications

Lourdes Villa Tanaca, Escuela Nacional de Ciencias Biológicas, IPN

3. Advances in protein structure, function and evolution

Mazatlán Room 3

Chairs: Georgina Garza Ramos & Daniel Alejandro Fernández. Facultad de Medicina, UNAM

Redox biocatalysis at the IBt-UNAM: molecular basis and applications of oxidoreductases

Marcela Ayala Aceves. Departamento de Bioingeniería. Instituto de Biotecnología. UNAM

Metal-crystallin interactions: The bioinorganic facet of cataract disease

Liliana Quintanar Vera. Departamento de Química y Centro de Investigación sobre el Envejecimiento. Cinvestav

	<p>Recent progress in predicting the structure and reactivity of peptides and proteins Joel Ireta Moreno. Departamento de Química. UAM Iztapalapa</p>
	<p>Search for new bacterial enzymes with potential clinical or biotechnological applications Rosario A. Muñoz Clares. Departamento de Bioquímica. Facultad de Química. UNAM</p>
17:00-18:00	<p>Having Coffee with ... (Carnaval Room 3)</p> <p>Georgina Hernández Delgado, Alberto Darszon, Susana Castro Obregón, Alicia González Manjarrez, Rafael Vázquez Duhalt</p>
17:00-19:00	<p>Poster Session 1</p> <p>BT – BIOTECHNOLOGY I BT1 – BT53 G – GENETICS, EPIGENETICS AND GENETIC REGULATION I G1 – G36 MH – MEDICINE, HEALTH & NUTRITION I MH1 – MH31 NN – NEUROSCIENCES & NEUROBIOLOGY I NN1 – NN27 SB – SYSTEMS BIOLOGY & BIOINFORMATICS I SB1 – SB26 ST – SIGNAL TRANSDUCTION AND CELL DIFFERENTIATION I ST1 – ST31 O – OTHERS I O1 – O40</p>

TUESDAY, OCTOBER 22

8:30-10:00	<p>Plenary Symposia II Hispano – Mexicano (Mazatlán Room 2)</p> <p>New Aspects on Cellular Regulation of Senescence and Differentiation Chair: Libia Vega Loyo, Cinvestav Zacatenco</p>
	<p>Neonatal T-Cells María Angélica Santana Calderón, Centro de Investigación en Dinámica Celular, UAEM</p>
	<p>From Single-Exon to Multiomics: Mapping the Impact of Intronless Genes on Cancer Biology Katia Aviña Padilla, Cinvestav Irapuato</p>
	<p>Reaching new horizons in cellular senescence research by leveraging a brand new multi-senescent cell type catalog Carlos Anerillas Aljama, Centro de Biología Molecular Severo Ochoa. CSIC-UAM. España</p>

10:00-11:00	<p>Plenary Lecture IV (Mazatlán Room 2)</p> <p>Mitochondria in global and local intracellular calcium signaling Gyorgy Hajnoczky. Thomas Jefferson University. USA Chair: Agustín Guerrero. Cinvestav Zacatenco</p>
11:00-11:30	Coffee break
11:30-12:45	Simultaneous oral Sessions 4, 5, 6

Session 4
Mazatlán Room 1
Chair: Dario Olicón. Escuela Nacional de Ciencias Biológicas, IPN.

<p>Design and production of a H7N3 highly pathogenic avian influenza recombinant vaccine Miguel Ángel Castillo Martínez. Cinvestav Zacatenco</p>
<p>Search for leading molecules targeting the replicative core of the SARS-CoV-2 polymerase for the development of specific antiviral drugs Armando Cruz Rangel. Instituto Nacional de Medicina Genómica</p>
<p>Structural design of a genetic circuit to obtain a biosensor América Selene Gaona Mendoza. Universidad de Guanajuato</p>
<p>Design and implementation of a cell surface display system on <i>Neurospora crassa</i> using native cell wall protein, ACW-1, as anchor Ana Sofía Ramírez Pelayo. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.</p>
<p>Structural Insights into the Glutathione Transferase Sigma Class from <i>Taenia solium</i>: Crystallographic Analysis and Functional Implications Ricardo Miranda Blancas. Instituto de Biotecnología. UNAM</p>

Session 5
Mazatlán Room 2
Chair: Gloria Saab. Instituto de Biotecnología, UNAM

<p>Aim11: a novel yeast assembly factor for complex IV and supercomplexes Ulrik Hiram Pedroza Dávila. Instituto de Fisiología Celular, UNAM</p>
<p>Benzopyrene degradation induces changes in antioxidant and detoxifying metabolism in <i>Debaryomyces hansenii</i> Francisco Javier Padilla Garfías. Instituto de Fisiología Celular, UNAM</p>

β -hairpin of *Thermus thermophilus* laccase: A regulator of enzymatic activity?

Leticia León Luna. Instituto de Biotecnología. UNAM

Exploring the quaternary assembly of a homomultimeric Catechol 1,2-dioxygenase: An Integrative study

Arisbeth Guadalupe Almeida Juárez. Instituto de Biotecnología. UNAM

At physiological pH and temperature, L-tyrosine can inhibit the formation of amyloid fibers of human lysozyme

Santos Arturo López Guzmán. Universidad Autónoma Metropolitana

Session 6

Mazatlán Room 3

Chair: Luis D. Alcaráz. Facultad de Ciencias, UNAM

OAEVOB: Online-Adjusted EVOLutionary Biclustering algorithm to analyze gene expression data

Edgardo Galán Vásquez. Instituto de Investigaciones en Matemáticas Aplicadas y en Sistemas, UNAM

Regulatory relationships between genes are not linked to direct transcription factor-target interactions in gene regulatory networks

Diego Arturo Velázquez Trejo. Instituto Nacional de Medicina Genómica. UNAM

A Modular Toolbox to Study Biological Systems: Real-Time Volatilomics and Mass Spectrometry Imaging under Ambient Conditions

Robert Winkler. Cinvestav Irapuato

Effect of co-culture of *A. flavus* and *P. ostreatus* on the expression of genes associated with the biosynthesis of aflatoxins and detoxifying enzymes

Luis Jesús Martínez Tozcano. Centro de Investigación en Biotecnología Aplicada. IPN

Design of de novo proteins and enzymes using generative language models

Sergio Romero Romero. Instituto de Fisiología Celular. UNAM

11:30-12:45

**Technical Talks
(Carnaval Room 1, 2)**

MACS Technology: Cell Separation

Laura López. UNIPARTS

Let's optimize your purification process

Carlos Bravo. BIO-RAD

12:45-14:15

Simultaneous Symposia 4, 5, 6
(Mazatlán Room 1, 2, 3)

4. Edges in mitochondrial knowledge

Mazatlán Room 1

Chair: Héctor Miranda Astudillo. Instituto de Investigaciones Biomédicas, UNAM

Mitochondrial Complement 1q Binding Protein (C1qbp) interacts with Cyclophilin D and regulates mouse heart Oxidative Phosphorylation and Permeability Transition

Manuel Gutiérrez Aguilar. Departamento de Bioquímica, Facultad de Química, UNAM

LRPPRC and SLIRP synergise to maintain sufficient and orderly mammalian mitochondrial translation

Diana Rubalcava Gracia Medrano. Department of Medical Biochemistry and Biophysics, Karolinska Institutet

The Divergent Biogenesis of the Respiratory Complex III in Malaria Parasites

Aldo Garcia Guerrero. Department of Biochemistry, University of Utah School of Medicine

Mitochondrial respirasome: diversity among conserved function

Héctor Vicente Miranda Astudillo. Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas, UNAM

5. Insights into plant adaptation: from growth to resilience

Mazatlán Room 2

Chair: Patricia Coello Coutiño. Facultad de Química, UNAM

Deciphering the epigenetic network of plant stem cell niche maintenance and plasticity

María de la Paz Sánchez. Instituto de Ecología, UNAM

Visualizing reactive oxygen species in living plant cells under biotic and abiotic responses

Luis Cárdenas. Instituto de Biotecnología, UNAM

Role of beneficial rhizobacteria in plant adaptation to environmental stress

José López Bucio. IIQB- Universidad Michoacana de San Nicolás de Hidalgo.

Plant signaling mechanisms under energy stress

Patricia Coello. Facultad de Química, UNAM

6. Symposium on biochemistry and molecular biology of bacteria

Mazatlán Room 3

Chair: Guadalupe Espín Ocampo. Instituto de Biotecnología, UNAM

	<p>Phenazines, biotechnological compounds produced by <i>Pseudomonas aeruginosa</i>: gene regulation and overproduction. Miguel Cocotl Yañez. Departamento de Microbiología y Parasitología. Facultad de Medicina, UNAM</p>
	<p>Genes required for the formation of virulence-provoking bacterial sphingolipids Otto Geiger. Centro de Ciencias Genómicas, UNAM</p>
	<p>Towards Enhanced Production of Biodegradable Plastics: Strategies to Improve Yield and Quality in <i>Azotobacter vinelandii</i> Daniel Segura González. Instituto de Biotecnología, UNAM</p>
14:15-14:30	Group photo
14:15-15:30	Lunch
15:30-16:30	<p>Plenary Lecture V (Mazatlán Room 2)</p> <p>Innovation by Evolution: Bringing New Chemistry to Life Frances H. Arnold. California Institute of Technology. USA Chair: Agustín Guerrero. Cinvestav Zacatenco</p>
16:30-17:00	<p>Flash Talks for poster advertising (Mazatlán Room 2)</p> <p>Chair: Agustín Guerrero. Cinvestav Zacatenco</p>
	<p>FT16 Study of the ABCG transporter family of <i>Metarhizium guizhouense</i> HA11-2 during its mycorrhizal association Víctor Manuel García Vera. Universidad de Guanajuato</p>
	<p>FT17 Relevance of protein O-glycosylation during the interaction of <i>Sporothrix schenckii</i> with the host Manuela Gómez Gaviria. Universidad de Guanajuato</p>
	<p>FT18 Killer yeast and glucose mediated synthesis of nanoparticles with biological effect Carlos Alberto Molina Vera. Universidad Autónoma de Querétaro</p>

FT19 Study of the endophytic capacity of entomopathogenic fungi

Jesús Ernesto Ramírez Nieto. Universidad de Guanajuato

FT20 Whole genome sequencing of candida auris strain isolated in Monterrey, Mexico

Alí Fernando Ruiz Higareda. Universidad Autónoma de Nuevo León

FT21 The wild plant *Conopholis alpina*, and their visiting honey bees (*Apis mellifera*) carry identical bacterial strains under natural conditions

Mary José Salas Limón. Universidad Autónoma de Puebla

FT22 Clinical value of gene expression profiling by digital PCR for the detection of leukemic cells in pediatric population

Adriana del Carmen Aguilar Lemarroy. Instituto Mexicano del Seguro Social

FT23 Interaction between genetic variants in vitamin D metabolism genes: impact on rheumatoid arthritis susceptibility and hypovitaminosis D

Bertha Campos López. Universidad de Guadalajara

FT24 Insulin pathway changes on the gene expression in adipose tissue modulated by ellagic acid in obese Wistar rat induced with high fat diet

Ramón Castillo Correa. Escuela Superior de Medicina. IPN

FT25 Genetic variants related with neoadjuvant chemotherapy response in breast cancer patients

Luis Felipe Jave Suárez. Universidad de Guadalajara

FT26 The increase in methylation of the ABCG2 gene is related to the presence of its Q141K allele and lower expression of the gene in peripheral blood of patients with gout and controls

Ámbar López Macay. Instituto Nacional de Rehabilitación

FT27 Comparative genomic analysis of *Candida glabrata* clinical isolates to understand antifungal resistance

Ana Lizbeth López Marmolejo. Instituto Potosino de Investigación Científica y Tecnológica A.C.

FT28 The forgotten side of the GPN-loop GTPase Npa3: the carboxy-terminal domain is a critical regulator of the GTPase core function

Manuel de Jesús Ochoa Valdez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

	<p>FT29 Unveiling molecular differences in 3D cell Culture of Triple Negative Breast Cancer cells and their resemblance to Tumor Tissue Mercedes Olvera Valencia. Escuela Nacional de Medicina y Homeopatía. IPN</p> <p>FT30 Determination of Antioxidant Activity and Phenolic Compounds in Fermented Cocoa Bean Shell Infusion Sebastián Cervera Pereyra. Universidad Juárez Autónoma de Tabasco</p>
17:00-18:00	<p>Having Coffee with ... (Carnaval Room 3)</p> <p>Gyorgy Hajnoczky, Frances Arnold, Carlos Anerillas, Angélica Santana, Otto Geiger</p>
17:00-19:00	<p>Poster Session 2</p> <p>BB – BASIC BIOCHEMISTRY I BB1 – BB37 BT – BIOTECHNOLOGY II BT54 – BT100 G – GENETICS, EPIGENETICS AND GENETIC REGULATION II G37 – G71 MH – MEDICINE, HEALTH & NUTRITION II MH32 – MH66 M – MICROBIOLOGY I M1 – M40 ROS – REACTIVE OXYGEN SPECIES ROS1 – ROS25 SB – SYSTEMS BIOLOGY & BIOINFORMATICS II SB27 – SB52</p>
19:00-20:00	<p>Business Session (Carnaval Room 1)</p>

WEDNESDAY, OCTOBER 23

8:30-10:00	<p>Plenary Symposia III (Mazatlán Room 2)</p> <p>Antimicrobial drug resistance and genomic epidemiology of superbugs Chair: Santiago Castillo Ramírez. Centro de Ciencias Genómicas, UNAM</p> <p>Evolutionary dynamics of carbapenem-resistant <i>Pseudomonas aeruginosa</i> in Chile: the impact of the ST654 'high risk' clone Andrés Opazo. Universidad de Concepción. Santiago de Chile</p> <p>Genomic diversity and antimicrobial resistance genes of <i>Salmonella enterica</i> in Mexico Adrián Gómez Baltazar. Universidad Autónoma de Querétaro</p> <p>One Health and pan genomic epidemiology of a superbug Santiago Castillo Ramírez. Centro de Ciencias Genómicas, UNAM</p>
-------------------	--

10:00 – 11:00	<p>Plenary Lecture VI (Mazatlán Room 2)</p> <p>Histamine: the (relatively) new kid in town José Antonio Arias Montaña. Department of Physiology, Biophysics and Neurosciences. Cinvestav Chair: Irma Romero Álvarez. Facultad de Medicina, UNAM</p>
11:00 – 11:30	Coffee break
11:30-12:45	<p>Simultaneous Oral sessions 7, 8, 9 (Mazatlán 1,2,3)</p>

SESSION 7
(Mazatlán Room 1)
Chair: Claudia González Espinosa Cinvestav Sede Sur

His-tagged proapoptotic KLAK peptide: A dual purpose strategy and their anticancer activity *in vitro*
Aldo Omar González Cruz. Universidad Autónoma de Nuevo León

Exposure to superparamagnetic iron oxide nanoparticles induced hepatorenal toxicity in BALB/C mice
Babatunde Oluwafemi Adetuyi. Cinvestav Zacatenco

New small-molecule CD36 antagonists
Marco Velasco Velázquez. Facultad de Medicina, UNAM

Effects of pollutant phenanthrene on glutathione-dependent enzymes and glutathione content in the shrimp *Penaeus vannamei*
Laura Camacho Jiménez. Centro de Investigación en Alimentación y Desarrollo, A.C.

Effect of pH on the GSSG-dependent substrate inhibition of Thioredoxin glutathione reductase. An initial velocity and progress curve-based study
Juan Luis Rendón Gómez. Facultad de Medicina. UNAM

SESSION 8
(Mazatlán Room 2)
Chair: Carlos Ricardo González Ruiz. IPICYT

Wnt signaling in Cancer Stem Cell: Towards the Non-canonical pathway
Miguel Ángel Sarabia Sánchez. Facultad de Medicina. UNAM

Individual miRNAs regulate sets of transcripts that encode for physically interacting specific proteins with each other
Alma Lilia Hernández Olvera. Instituto Nacional de Medicina Genómica. UNAM

Transcriptomic Profile of Seq-1 Peptide in Steatotic Hepatocytes: Implications in liver fibrosis

Sandra Daniela Calixto Tlacomulco. Facultad de Medicina. UNAM

Exploring chromatin dynamics during *in vitro* decidualization of human endometrial stromal cells

Alejandra Monserrat Retis Reséndiz. Facultad de Química. UNAM

The acetogenin laherradurin and the alkaloid liriodenine promote apoptosis and autophagy induction through mitochondrial dynamics dysregulation in colorectal cancer *in vitro*

Izamary Delgado Waldo. Facultad de Medicina. UNAM

SESSION 9

(Mazatlán Room 3)

Chair: Víctor Manuel Ayala García – Universidad Juárez del Estado de Durango

Characterization of the GlyOx from *Azotobacter vinelandii* and its Application for Glycine Biosensing

Andrés Zárate Romero. Centro de Nanociencias y Nanotecnología. UNAM

Consortium of microorganisms for polystyrene biodegradation

María Marcela Robles Machuca. Universidad Autónoma de Nayarit

Plant cell wall degrading enzymes in the secretome of *Colletotrichum lindemuthianum* pathotypes

Karla Morelia Díaz Tapia. Universidad Michoacana de San Nicolás de Hidalgo

Unveiling the uptake and the early genetic responses of *Arabidopsis* to growth promoting microbial volatiles

Zulia Fernandina Nieves López. Cinvestav Irapuato

Functional characterization of a non-specific phospholipase C (*PvNPC4*) in bean-rhizobia symbiosis and root development

Ronald Pacheco Sánchez. Instituto de Biotecnología. UNAM

11:30-12:45

**Technical Talks
(Carnaval Room 1, 2)**

Espectrometría de masas con tecnología Orbitrap y sus principales aplicaciones en ciencias de la vida

Filippo Bedani, ISASA

CIENTIFICA SENNA

<p>12:45-13:45</p>	<p>Plenary Lecture VII (Mazatlán Room 2)</p> <p>Rotary Engines of Life: Exploring ATP Synthase and Cristae Formation in <i>Polytomella parva</i> Diego González Halphen. Instituto de Fisiología Celular. UNAM Chair: Irma Romero Álvarez. Facultad de Medicina. UNAM</p>
<p>13:45-14:15</p>	<p>Flash Talks for poster advertising 3 (Mazatlán Room 2)</p> <p>Chair: Irma Romero Álvarez. Facultad de Medicina. UNAM</p>
	<p>FT31 Characterization of sphenotoxin from <i>Ophryacus sphenophrys</i> and its comparison with classic crotoxin Tania Corkidi Zajur. Instituto de Biotecnología. UNAM</p>
	<p>FT32 The methionine synthase in plant development Sara Margarita Garza Aguilar. Escuela de Ingeniería y Ciencias. Tecnológico de Monterrey</p>
	<p>FT33 Tips to increase the thermal stability of a recombinant protein: the practical case of CGI-58 Miriam Livier Llamas García. Instituto Potosino de Investigación Científica y Tecnológica A.C.</p>
	<p>FT34 Biochemical characterization of orotate phosphoribosyltransferase from the phytopathogen <i>Pseudomonas cichorii</i> Amelia López Albores. Instituto Tecnológico de Tuxtla Gutiérrez</p>
	<p>FT35 Aldosterone effects on the expression of ryanodine receptors and their participation in the calcium dynamics of rat mesenteric arteries Hiram Lozano Ruiz. Cinvestav Zacatenco</p>
	<p>FT36 Subcellular localization plays a determinant role on the functional diversification of the paralogous proteins BAT1 and BAT2 Yamile Paredes Chiquini. Instituto de Fisiología Celular. UNAM</p>
	<p>FT37 Hinges, springs, and sticky surfaces determine the preferred conformations of CRE in the absence of LOXP and prime it for binding Nina Pastor Colón. Universidad Autónoma del Estado de Morelos</p>
	<p>FT38 Characterization of Human Epidermal Growth Factor (hEGF)-induced Ca²⁺ release in HeLa Cells Amanda Renté Alpizar. Cinvestav Zacatenco</p>
	<p>FT39 Obesity leads to transcriptomic alterations in one-carbon metabolism-related genes: in silico analysis of GTEx study Jesús Daniel Cantú Ruiz. Institute for Obesity Research. Tecnológico de Monterrey</p>

FT40 Exploring the Connection Between Vitamin B12 Deficiency and Obesity

Erika Castaño Moreno. Institute for Obesity Research. Tecnológico de Monterrey

FT41 “Alaches”, quelites that besides being nutritious, have effect on *Helicobacter pylori*

Erika Gómez Chang. Facultad de Medicina. UNAM

FT42 Estrogen modulation of CaMKII and calcium handling proteins in H9c2 hypertrophied myotubes

Silvia Araceli López Morán. Centro de Investigación Biomédica. Tecnológico de Monterrey

FT43 Ca²⁺ Overload-Induced Mitochondrial Dysfunction is an Early Risk Factor for Lethal Ventricular Arrhythmias

Felipe de Jesús Salazar Ramírez. Escuela Nacional de Medicina. Tecnológico de Monterrey

FT44 Physiological response and organic interactions of berrycactus in wistar rats with metabolic syndrome (MS)

Daniela Joyce Trujillo Silva. CONAHCyT. Instituto Potosino de Investigación Científica y Tecnológica A.C.

14:15-14:30	Group photo
14:15-15:30	Lunch
15:30-17:00	Simultaneous Symposia (Mazatlán Room 1, 2, 3)

7. ROS in fungi, turtles and rats: janus favorite molecules

(Mazatlán Room 1)

Chair: Alejandro de las Peñas. Instituto Potosino de Investigación Científica y Tecnológica

Evidence of oxidative stress responses of green turtles (*Chelonia mydas*) to differential habitat conditions in the Mexican Caribbean

Vanessa Labrada Martagón. Facultad de Ciencias. Universidad Autónoma de San Luis Potosí

***Candida glabrata* (*Nakaseomyces glabratus*) requires the GSH/ Glutathione reductase and Thioredoxin/Thioredoxin reductase systems for mitochondrial function and oxidative stress response**

Guadalupe Gutiérrez Escobedo. División de Biología Molecular. Instituto Potosino de Investigación Científica y Tecnológica

Targeting senescent cells to decrease oxidative stress and neuroinflammation and prevent cognitive impairment due to chronic obesity in middle-aged female rats.

Mina Königsberg Fainstein. Departamento de Ciencias de la Salud. Universidad Autónoma Metropolitana - Iztapalapa

8. Recent progress in signal transduction research

(Mazatlán Room 2)

Chair: Angélica Rueda y Sánchez de la Vega. Cinvestav Zacatenco

Regulation of insulin signaling: multiple factors and molecular mechanisms involved

Jesús Alberto Olivares Reyes. Departamento de Bioquímica, Cinvestav

Epithelial to mesenchymal transition in breast cancer: EGF signaling

Eduardo Monjaraz Guzmán. Instituto de Fisiología. Benemérita Universidad Autónoma de Puebla

SERCA pumps as key regulators of calcium signaling in blood vessels

Angélica Rueda y Sánchez de la Vega. Departamento de Bioquímica. Cinvestav

9. Mitochondria in health and disease

(Mazatlán Room 3)

Chair: Gerardo García Rivas. Institute for Obesity Research. TecSalud

Role of mitochondrial fusion proteins in cell homeostasis

Antonio Zorzano. University of Barcelona

Exercise regulation of mitochondrial function in metabolic dysfunction-associated steatotic liver disease

Rodrigo Troncoso. INTA, Chile

Depression-like behaviour is comorbidity in a model of heart failure with preserved ejection fraction induced by high-fat diet and L-NAME

Gerardo García Rivas. IOR, TecSalud

Avocado oil: a promising alternative against chronic degenerative diseases through its effects on mitochondria

Christian Cortes Rojo. Universidad Michoacana de San Nicolás de Hidalgo

17:00-18:00

Having Coffee with ... (Carnaval Room 3)

Diego González Halphen, José Antonio Arias Montaña, Sergio Grinstein, Mina Königsberg, Andrés Opazo

17:00-19:00

Poster Session 3

BB – BASIC BIOCHEMISTRY II BB38 – BB78
 BT – BIOTECHNOLOGY III BT101 – BT158
 G – GENETICS, EPIGENETICS AND GENETIC REGULATION III G72 – G106
 MH – MEDICINE, HEALTH & NUTRITION III MH67 – MH109
 M – MICROBIOLOGY II M41 – M83
 TP – TOXICOLOGY & PHARMACOLOGY I TP1 – TP25

8:30-10:00	Plenary Symposia IV (Mazatlán Room 2) Wanderings in Biochemistry Chair: Agustín Guerrero. Cinvestav Zacatenco
	Tales of a pathogenic yeast Irene Castaño Navarro. Instituto Potosino de Investigación Científica y Tecnológica
	Wanderlust: from genome rearrangements to cell division cycle in Rhizobium David Romero Camarena. Centro de Ciencias Genómicas, UNAM

THURSDAY, OCTOBER 24

	The road of cellular communication in different models Teresa Hernández Sotomayor. Sociedad Mexicana de Bioquímica
10:00-11:00	Plenary Lecture VIII (Mazatlán Room 2) The journey of glucose in the body: from absorption to glucose uptake by the muscle Amira Klip. The Hospital for Sick Children. University of Toronto. Canada Chair: Irma Romero Álvarez. Facultad de Medicina, UNAM
11:00-11:30	Coffee break
11:30-12:45	Simultaneous oral Sessions 10, 11, 12
SESSION 10 (Mazatlán Room 1) Chair: Alejandro Zentella Dehesa. Instituto de Investigaciones Biomédicas, UNAM	
	Cytotoxic properties and gene expression modulating effects of cinnamon essential oil (<i>Cinnamomum zeylanicum</i>) in cervical cancer cells Rubén Piña Cruz. Universidad de Guadalajara
	BORIS: A Novel Oncogene and Molecular Player in Glioblastoma Pathogenesis Ernesto Soto Reyes Solís. Universidad Autónoma Metropolitana Cuajimalpa

Crosstalk between Wnt/ β -catenin and PI3K/AKT pathways mediated by HOTAIR/HIF1 α axis in cervical cancer

Samuel Trujano Camacho. Universidad Autónoma Metropolitana Iztapalapa

Participation of Exon 18 Variants of the SCN8A Gene in the Proliferation and Metastatic Behavior of Cervical Cancer Cell Lines

Tonantzin Guadalupe Anguheven Ledezma. Instituto de Fisiología Celular. UNAM

LINC01087/miR-130a-3p/FOXA1 axis regulates the cell proliferation, migration and vasculogenic mimicry in breast cancer cells in a three-dimensional (3D) microenvironment

Sandra Lizeth Alvarado Ortigoza. Universidad Autónoma de la Ciudad de México

SESSION 11

(Mazatlán Room 2)

Chair: Ángel Andrade Torres. Universidad Autónoma de Nuevo León

Identification of differential proteins produced in hypo- and hyper-virulent mutants grown in wheat coleoptiles compared to the wild type strain of *Fusarium graminearum*

Ana Lilia Martínez Rocha. Universidad de Guanajuato

Bacterial microbiota during embryonic development: a maternal inheritance

Yendi Navarro Noya. Universidad Autónoma de Tlaxcala

HpGroEL protein is secreted by *Helicobacter pylori* and binds other secreted proteins, its chaperonin activity is maintained at extreme pH

José de Jesús Olivares Trejo. Universidad Autónoma de la Ciudad de México

Testosterone and estradiol induce the expression of *Actinobacillus seminis* secreted proteases

Gerardo Antonio Ramírez Paz y Puente. Facultad de Estudios Superiores Iztacala. UNAM

Adaptive Stress Response to Nitrogen Limitation in the Antarctic Yeast *Rhodotorula mucilaginosa* M94C9

Miguel Ángel Rosas Paz. Facultad de Ciencias. UNAM

SESSION 12

(Mazatlán Room 3)

Chair: Rodolfo Pastelin Palacios, Facultad de Química, UNAM

Spontaneous calcium transients in striatal astrocytes: evidence from a preclinical model of autism

Daniel Reyes Haro. Instituto de Neurobiología. UNAM

	<p>Effect of protein overexpression and silencing EiMyb23 on the encystment of <i>Entamoeba invadens</i> Jatziry Daniela Ocampo Ulloa. Cinvestav Zacatenco</p>
	<p>Resveratrol inhibits the PI3K/Akt insulin pathway by activating classical PKCs Karla Daniela Hernández González. Cinvestav Zacatenco</p>
	<p>Physiological effect of the interaction between the mitochondrial maize hexokinase 4 and the beta-glucosidase aggregating factor 1 Andrés Burgos Palacios. Facultad de Química. UNAM</p>
12:45-13:45	<p>Plenary Lecture IX (Mazatlán Room 2)</p> <p>AI Approaches in Protein Bioinformatics Daisuke Kihara. Department of Biological Sciences, Department of Computer Science. Purdue University Chair: Libia Vega Cinvestav Zacatenco</p>
13:45-14:15	<p>Flash Talks for poster advertising 4 (Mazatlán Room 2)</p> <p>Chair: Libia Vega. Cinvestav Zacatenco</p>
	<p>FT45 Anti-<i>Helicobacter pylori</i> activity, cytotoxicity and <i>in vivo</i> gastroprotective effect of diacetylcurcumin and its metal derivatives Almanelly Agabo Martínez. Facultad de Medicina UNAM</p>
	<p>FT46 The combined therapy of diclofenac-itraconazole reduces the size of <i>Madurella mycetomatis</i> grains and stimulates cytokines of the Th1 and Th17 profile Iván Alejandro Banda Flores. Universidad Autónoma de San Luis Potosí</p>
	<p>FT47 Transcriptional characterization of the antitumor activity of laherradurin on an <i>in vitro</i> model of colorectal cancer Eduardo Pérez Arteaga. Facultad de Medicina. UNAM</p>
	<p>FT48 Cannabidiol increases adipogenesis on 3T3-L1 pre-adipocytes via PPARγ Helen Yarimet Lorenzo Anota. Institute for Obesity Research. Tecnológico de Monterrey</p>
	<p>FT49 BUB 1 is a nodular kinase that predicts overall survival in osteosarcoma, liposarcoma, synovial. sarcoma, and leiomyosarcoma Frida Citlali Rodríguez Izquierdo. UAM Iztapalapa. Instituto Nacional de Cancerología</p>

FT50 The IL-6 induces metastatic properties in luminal breast cancer cells through GPR30

Ana Carolina Tirado Garibay. Universidad Michoacana de San Nicolás de Hidalgo

FT51 Identification of Aim24 as a yeast complex II membrane domain assembly factor

Yolanda Margarita Camacho Villasana. Instituto de Fisiología Celular. UNAM

FT52 Agronomic and molecular evaluation of polyploid tomato

Fátima Clarita Cota Ruiz. Cinvestav Zacatenco

FT53 p53 Mutants Induce an Aggressive Phenotype through Overexpression of miR-27b-5p in Cancer

José Edwin Dolores García. Universidad Autónoma Metropolitana Iztapalapa

FT54 The inhibitory effect of rTBL-1 on colorectal cancers *in vitro* is related to the presence of the epidermal growth factor receptor

Teresa García Gasca. Universidad Autónoma de Querétaro

FT55 Modulation of glutamate uptake by the subacute activation of the histamine H₃ receptor in rat cerebro-cortical astrocytes in primary culture

Yrving Daniel Díaz De Lucio. Cinvestav Zacatenco

FT56 Impact of Intestinal Low-Grade Inflammation on Purinergic-Calcium Signaling in Enteric Glial Cells

Irving Israel Vega Juárez. Universidad de Colima

FT57 *In vivo* role of T γ δ cells in a lupus mouse model induced by NPA stabilization

Edgar Iván Galarce Sosa. Escuela Nacional de Ciencias Biológicas. IPN

FT58 *Plasmodium vivax* mitochondrial DNA polymorphism, and its relationship to genes encoding sexual proteins and the infection pattern in mosquitoes *Nyssorhynchus albimanus* and *Anopheles pseudopunctipennis*, Chiapas, Mexico

Lilia González Cerón. Instituto Nacional de Salud Pública

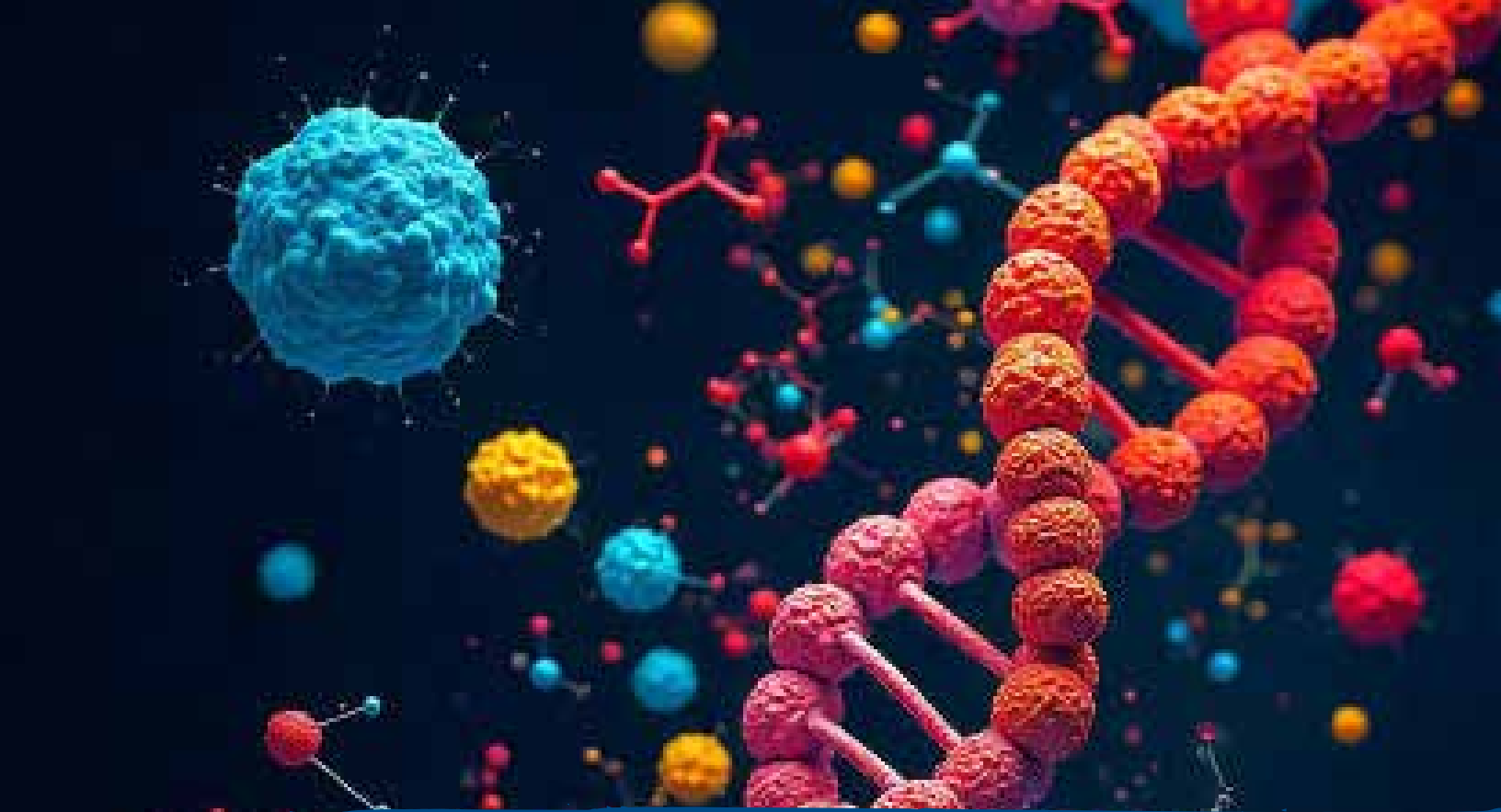
FT59 Impact of the form of birth and effect of Flagellin on the activation of neonatal CD4⁺ and CD8⁺ T cells

Carlos Jesús Ventura Martínez. Universidad Autónoma del Estado de Morelos

14:15-15:15

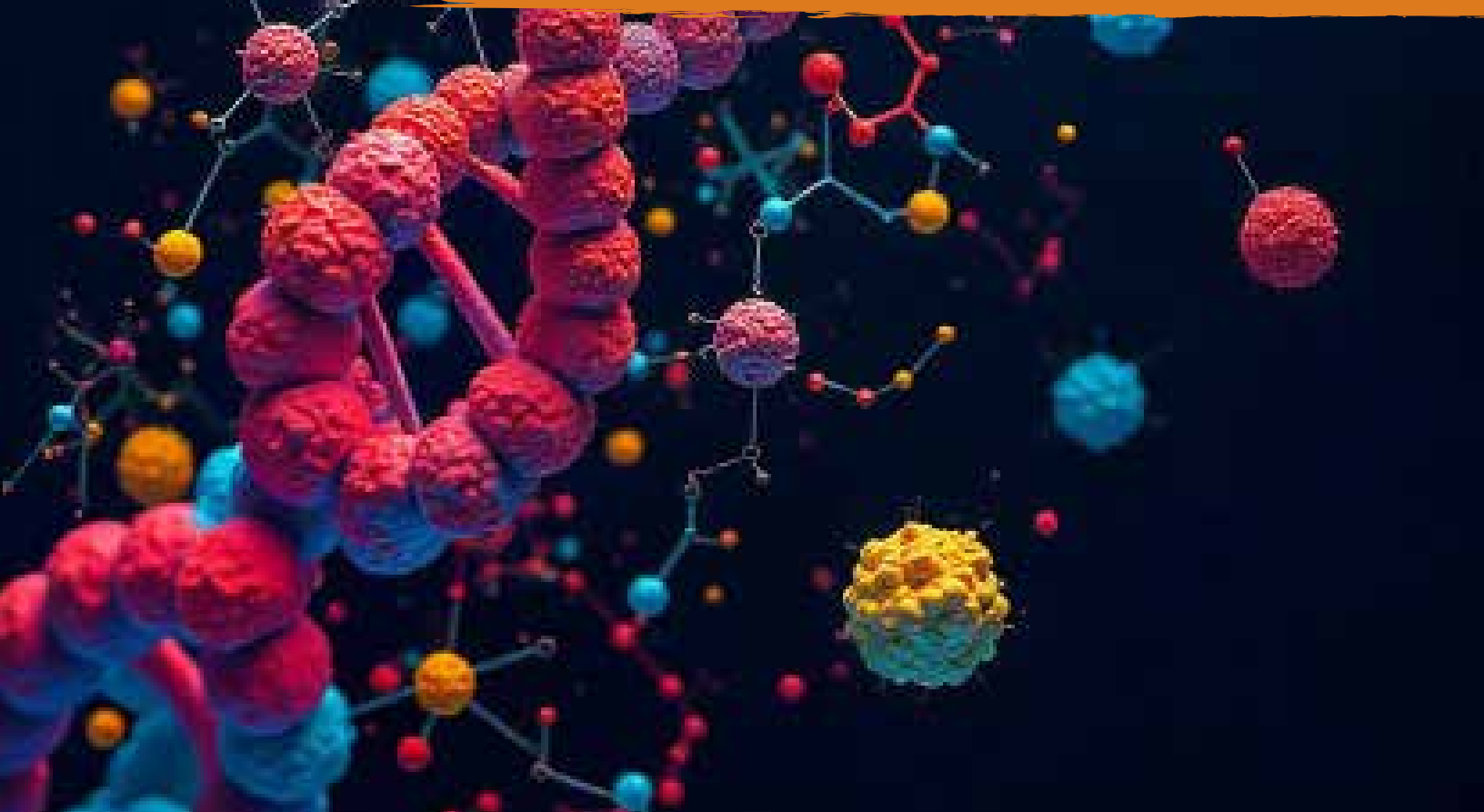
Lunch

15:15-17:15	Poster Session 4 BB – BASIC BIOCHEMISTRY III BB79 – BB118 IP – IMMUNOLOGY & PARASITOLOGY IP1 – IP38 M – MICROBIOLOGY III M84 – M124 NN – NEUROSCIENCES & NEUROBIOLOGY II NN28 – NN54 TP – TOXICOLOGY & PHARMACOLOGY II TP26 – TP52 ST – SIGNAL TRANSDUCTION & CELL DIFFERENTIATION II ST32 – ST63 O – OTHERS II O41 – O83
15:15-16:15	Having Coffee with ... (Carnaval Room 3) Amira Klip, Daisuke Kihara, David Romero
17:15-18:15	Closing Lecture (Mazatlán Room 2) Membrane remodelling in endocytosis and phagocytosis Sergio Grinstein. The Hospital for Sick Children. University of Toronto. Canada Chair: Agustín Guerrero. Cinvestav Zacatenco
18:15 - 18:45	Final announcements and closing ceremony
20:00-1:00	Farewell dinner (Explanada Mantarraya, MIC)



POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY



POSTER SESSION I
MONDAY, OCTOBER 21
17:00 - 19:00

BIOTECHNOLOGY I

BT1

Characterization and evaluation of the biological activity of the TA-PAsp polymer

Ileana Castillo Tobías. Universidad Autónoma de Coahuila

BT2

In silico analysis of the C8 clone and toxic activity of the ATP-dependent RNA helicase from the entomopathogenic bacterium Serratia entomophila Mor 4.1 in Phyllophaga blanchardi larvae (Coleoptera: Melolonthidae)

Luz Areli Gorostieta Nava. Universidad Autónoma del Estado de Morelos

BT3

Doing Bionanotechnology with dsDNA and CRISPR-dCas12a: Designing Linear and Branched Nanofibers

Armando Hernández García. Universidad Nacional Autónoma de México

BT4

Mechanistic assessment of the biosynthesis of cyanobacterial secondary metabolites: Two cases

Jose Alberto Martinez Yerena. Czech Academy of Sciences

BT5

Characterization of recombinant modified trichocystatin TC-2

Jaime Ortega López. Cinvestav Zacatenco

BT6

Effect of prolactin and 17 β -estradiol on the adhesion and intracellular persistence of Staphylococcus aureus in bovine mammary epithelial cells

Gladys Romero Corona. Universidad Michoacana de San Nicolás de Hidalgo

BT7

Immobilization of thermomyces lanuginosus lipase (tll) via ionic-covalent interaction on heterofunctional glyoxyl-agarose supports for biodiesel production

Cristian David Salcedo Cuarán. Universidad del Valle

BT8

Characterization of DyP-type peroxidases and their use as biocatalysts in textile dyes decolorization

Jesús Alberto Segovia Cruz. Universidad Nacional Autónoma de México

BT9

Genetic diversity and phenotypic variations in four native strains of Bacillus spp. biocontrol agents through pan-genome and BGC analysis

Hilda Mabel Sosa Esquivel. Universidad Autónoma de Zacatecas "Francisco García Salinas"

BT10

Physiological evaluation of recombinant forms of granulocyte colony-stimulating factor

Miguel Ángel Valle Yañez. Cinvestav Zacatenco

BT11

Purification and Valorization of Urban Effluents Using Photobioreactors Operated with Anabaena Inaequalis

Wendy Zárate Hernández. Instituto Tecnológico de Veracruz

BT12

Bacteria isolated from an asphaltene rock from the Gulf of Mexico are capable of degrading oil at low temperatures

Libertad Alejandra Adaya García. Universidad Nacional Autónoma de México

BT13

Co-culture of Daldinia eschscholzii and Humphreya coffeata

Alma Rosa Agapito Ocampo. Universidad Autónoma del Estado de Morelos

BT14

Optimization of paramylon production in Euglena gracilis using different light spectra

Zhaida Itzel Aguilar González. Universidad Nacional Autónoma de México

BT15

Expression of chagasin chimeras with 4 epitopes from trypanosoma cruzi antigen, TSA-1, in Pichia pastoris

Paola Eugenia Agustín Vélez. Cinvestav Zacatenco

BT16

Heterologous expression of scorpion toxin from Chihuahua

Carolina Alvarado González. Universidad Autónoma de Chihuahua

BT17

The national laboratory for analysis of biotechnological molecules and drugs (LAMMB) challenges and perspectives

Elianeth Amaro Encarnación. Universidad Nacional Autónoma de México

BT18

Polyuronide synthases in filamentous fungi

Mariandrea Victoria Aranda Barba. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BT19

Functional and Structural Analysis of Basidiomycete Fungal Lignocellulolytic Enzymes for Biotechnological Applications

Silvia Armenta Jaime. Universidad Autónoma del Estado de Hidalgo

BT20

Intracellular oxidative stress at low temperatures in the phytopathogen bacterium Pseudomonas savastanoi pv. phaseolicola NPS3121

Jackeline Lizzeta Arvizu Gómez. Universidad Autónoma de Nayarit

BT21

CLE14 peptide inhibits regeneration and callogenesis in Arabidopsis

Adrián Ávalos Rangel. Universidad Michoacana de San Nicolás de Hidalgo

BT22

Functional drink from wine industry waste

José Ramón Barraza Amaya. Universidad Autónoma de Coahuila

BT23

Improvement of pigment production by filamentous fungi

Iliana Geraldine Barreto Flores. Instituto Tecnológico Superior de Irapuato

BT24

Optimization of the fermentation process to produce whiskey

John Carlos Botia Becerra. Universidad de Guanajuato

BT25

Enhancing denitrification with electric fields in Paracoccus denitrificans

Arturo Cadena Ramírez. Universidad Politécnica de Pachuca

BT26

Green technologies for the treatment of pesticides in water basins

David Alfonso Camarena Pozos. Centro de Innovación Aplicada en Tecnologías Competitivas

BT27

Production and optimization of induced Serratia marcescens carboxylesterases for the degradation of polyolefins

Amador Roberto Campos Valdez. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BT28

Solutions based on copper ions as antifungal control in soil against the phytopathogenic fungus Neopestalotiopsis spp.

Jesús Emiliano Cantador Campos. Instituto Politécnico Nacional

BT29

Immobilization of Bacillus subtilis on magnetite nanoparticles for arsenic removal in water

César Abdiel Cárdenas Villanueva. Instituto Tecnológico de Durango

BT30

New biodegradable plastics (Poly-3-Hydroxyalkanoates) produced by Azotobacter vinelandii OP recombinant strains

Alma Luz Carmona Brito. Universidad Nacional Autónoma de México

BT31

Auxin symplastic transport modulates root hairs development of Arabidopsis induced by Azospirillum baldaniorum Sp245

Elizabeth Carrillo Flores. Universidad Michoacana de San Nicolás de Hidalgo

BT32

Identification of antigens in the immunoproteome of listeria monocytogenes

Kimberly Arisbeth Casal Rodríguez. Universidad Autónoma de Sinaloa

BT33

Comprehensive strategies for the discovery of plastic-degrading microorganisms and enzymes

Leticia Casas Godoy. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BT34

Enzymatic saccharification of Agave durangensis bagasse for the production of biohydrogen

Gerardo Javier Chaidez Gurrola. Universidad Juárez del Estado de Durango

BT35

Regeneration in vitro of jipi palm (Carludovica palmata Ruiz & Pavón) mediated by thidiazuron

Samuel Alejandro Chan Poot. Centro de Investigación Científica de Yucatán A.C.

BT36

Hsa-miR-206 effect on tumorigenesis and chemoresistance of HCT116 colorectal cancer cells

Lizbeth Guadalupe Chávez Ramos. Universidad Autónoma de Sinaloa

BT37

Molecular and physiological characterization of yqcK locus of Bacillus subtilis and its metabolic role within the ars arsenic response operon

Elizabeth Cisneros Lozano. Universidad Juárez del Estado de Durango

BT38

Removal of pharmaceuticals compounds (PCs) used in COVID-19 treatment present in wastewater by constructed wetlands

Jessica Fernanda Contreras Mojica. Centro de Innovación Aplicada en Tecnologías Competitivas

BT39

Host-microbiota interactions drive the probiotics enrichment in the microbiota of L. vannamei: A hologenome perspective

María Fernanda Cornejo Granados. Universidad Nacional Autónoma de México

BT40

Comparative analysis of the osteogenic effect of the mixture of chitosan hydrogel with bone or bladder ECM hydrogels and bone mineral

Juan Luis Cota Quintero. Universidad Autónoma de Sinaloa

BT41

Evaluation of the thermostability of the mutant PET hydrolase

Danna Marely Cruz Rico. Universidad Autónoma del Estado de Hidalgo

BT42

Generation and propagation of improved varieties of malt and forage barley: An alternative crop for vulnerable areas in the state of Jalisco

Dianey Celeste Cruz Muñoz. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BT43

Identification of Signaling Molecules Involved in Bacterial Communication Isolated from Lichens

Miriam Joceline Cruz Ramos. Instituto Tecnológico Superior de Irapuato

BT44

Characterization of wild blueberry (*Vaccinium leucanthum*) by conventional and FT-IR spectroscopic techniques

Acela Cuamatzi de Gante. Centro de Investigación en Biotecnología Aplicada-IPN

BT45

Exploring esterase/lipase production in *Aspergillus fumigatus* for plastic degradation

Roberto Javier De la Rosa Mora. Universidad Autónoma de Nayarit

BT46

Generation of mutant strain disrupted in *aseR* gene for studying the arsenic response of *ase* operon in *Bacillus subtilis*

Miguel Ángel de la Torre Arellano. Universidad Juárez del Estado de Durango

BT47

The genotoxic effect of fluoride presents in endemic soil on onion (*Allium cepa*) bulb cells

Ángel Ricardo Díaz Duarte. Universidad Juárez del Estado de Durango

BT48

Identification of bacterial proteases isolated from a mangrove in Nayarit

Fernando Díaz Partida. Universidad Autónoma de Nayarit

BT49

Development of an Inoculant to Accelerate the Composting Process of Bovine Manure

Frida Sofía Doroteo Platas. Universidad Autónoma de Puebla

BT50

Functional characterization of HUB proteins in the phytohormone signaling network in Arabidopsis thaliana

Andrea Duarte Elías. Instituto Tecnológico Superior de Irapuato

BT51

Obtaining Starch from Pachyrhizus erosus as an Alternative to Conventional Starches

Jimena Carolina Enríquez Fonseca. Instituto Politécnico Nacional

BT52

Antioxidant activity and phenol content of liquid-fermentation of Morchella esculenta

Martha Elena Espin Reza. Universidad Autónoma del Estado de Morelos

BT53

Induction of the tannase system in marine fungi for PET (polyethylene terephthalate) degradation

Jossie Fernanda Espinoza Ávila. Universidad de Guadalajara

GENETICS, EPIGENETICS AND GENETIC REGULATION I

G1

Expression of the CCD4-3 gene in different tissues of the N4 and P12 morphotypes of Bixa orellana L. by qRT-PCR real time

Margarita Aguilar Espinosa. Centro de Investigación Científica de Yucatán A.C.

G2

Agave as a model to understand the molecular genetic circuits controlling development and cell wall metabolism of hard fibers

Fulgencio Alatorre Cobos. Centro de Investigación Científica de Yucatán A.C.

G3

Determination of NR3C1 gene methylation and expression levels in peripheral blood and post-mortem brain tissue and their association with suicide

Yazmín Mercedes Amador Segovia. Universidad Juárez del Estado de Durango

G4

Association of the SNP HLA-DRB1*07:01, HLA-B*55:01 and IgE levels with hypersensitivity reactions to betalactamic antibiotics in COVID-19 patients in the Mexican population

Paola Nataly Andrade Valdez. Instituto Politécnico Nacional

G5

Frequency of Kdr mutations in the VGSC of Aedes aegypti vector

Annete Itzel Apodaca Medina. Universidad Autónoma de Occidente

G6

MEOX2 as an epigenetic regulator of genes involved in lung cancer carcinogenesis

Leonel Armas López. Universidad Nacional Autónoma de México

G7

Determining the length of the MAOA-UVNTR polymorphism in a sample of suicidal subjects and control subjects

Samuel Arroyo Carranza. Universidad Juárez del Estado de Durango

G8

Chromatin state changes between primary cortex astrocytes induced to senescence and gliosis with palmitate

Karla Estephania Ávila Galicia. Universidad Autónoma Metropolitana

G9

Ellagic acid modulates the mRNA expression of TNF pathway, NLRP3 inflammasome, NFkB1 and FTO in adipose tissue of diet-induced obese Wistar rat

Antonio Ávila Guerrero. Instituto Politécnico Nacional

G10

Generation of the libR/flcA mutant of Azospirillum brasilense Sp7

Ana Margarita Balderas Peña. Universidad Autónoma de Puebla

G11

MEDIATOR 18 mediator complex subunit association in phosphate scarcity response

Maria Fernanda Ballesteros Barrera. Universidad Michoacana de San Nicolás de Hidalgo

G12

Determination of TPH1 gene expression levels in post-mortem brain tissue and its association with suicide

Norma Gabriela Barraza Méndez. Universidad Juárez del Estado de Durango

G13

Effect of the expression of the long NON-CODING RNA XIST regarding the oncogen MYC, in the treatment of patients with acute myeloid leukemia

Susana Bernardo Hernández. Hospital Regional de Alta Especialidad de Ixtapaluca

G14

MIR124-3 polymorphisms and neurodevelopment in children Vfrom Durango, México.

Katzumy Lizbeth Blancas Olvera. Universidad Juárez del Estado de Durango

G15

Ligand-dependent XRE binding patterns of aryl hydrocarbon receptor (AHR) on CYP1A1 gene promoter

Itzel Adriana Cabildo Delgado. Cinvestav Zacatenco

G16

Genotyping of the rs737865 polymorphism of the comt gene in suicidal subjects and control subjects using real-time PCR

Jesus Eduardo Castañeda Betancourt. Universidad Juárez del Estado de Durango

G17

Silencing of ABCG2 gene expression in HCT15 colon cells by transfection of a specific sgRNA using the CRISPR-Cas9 technique

Jesús Fabián Cervantes Meneses. Instituto Nacional de Rehabilitación

G18

Role of Hypoxia Response Factors from Phaseolus vulgaris during a nodule development

Ana I. Chávez Martínez. Universidad Nacional Autónoma de México

G19

Effect of the fliW-gene deletion on the extracellular electron transfer and biofilm production in Geobacter sulfurreducens

Jessica Cholula Calixto. Universidad Nacional Autónoma de México

G20

Msn4 is an important regulator of the Environmental Stress Response in Candida glabrata

Yazmín Contreras Bravo. Instituto Potosino de Investigación Científica y Tecnológica A.C.

G21

Transcription factors and DNA motifs in NaStEP promoter regulate gene expression in self-incompatible Nicotiana glauca

Yuridia Cruz González Zamora. Universidad Nacional Autónoma de México

G22

Association of the rs4986791 polymorphism of the tlr4 gene in outpatients with the severity grade of COVID-19

Ingrid Cuevas Cárdenas. Universidad Juárez del Estado de Durango

G23

Global DNA methylation levels are associated with cardiometabolic risk and clinical disease activity in systemic lupus erythematosus patients

Ulises De la Cruz Mosso. Universidad de Guadalajara

G24

Microevolution of Candida glabrata in the presence of fluconazole

Rosa Lilian Díaz Chávez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

G25

Loss of the tumor suppressor miR-122 promotes cell migration and up-regulation of BORIS/CTCF in triple-negative breast cancer

Mauricio Flores Fortis. Universidad Autónoma Metropolitana Cuajimalpa

G26

Exploring the Transcriptome of MCF-7 Breast Cancer Cells Treated with Capsicum annuum L. var. fascinato

Roberto Jorge García Mendoza. Universidad Autónoma de Querétaro

G27

"La" protein in Leishmania major: analysis of its interactions with other proteins and RNA molecules

Sergio García De la Cruz. Universidad Nacional Autónoma de México

G28

Association of the DNMT1 protein with long non-coding RNAs: MalaT1, UCA1 and HOTAIR in cervical cancer

Janis Aislinn García Flores. Instituto Nacional de Cancerología

G29

rs 2278163 Variant of the DLX3 gene and its relationship to the severity of dental fluorosis in women in Durango city

Oscar Martin Garcia Rosales. Universidad Juárez del Estado de Durango

G30

Interplay between chaperones and the sorting platform for hierarchical secretion of proteins through the injectisome

Ricardo Gaspar Lino. Universidad Nacional Autónoma de México

G31

Identification of genetic variants associated with treatment toxicity in pediatric patients with acute lymphoblastic leukemia

Francisco Javier Gaytán Cervantes. IMSS

G32

Epigenetic analysis in adipocytes differentiated from mesenchymal stem cells of the human umbilical cord of newborns from healthy, obese, and diabetic mothers under high glucose and cholesterol conditions

Emily Jocelyn Gómez Flores. Instituto Nacional de Perinatología Isidro Espinosa de los Reyes

G33

Analysis of small non-coding RNAs expression profiles and their impact on Malignant Pleural Mesothelioma Development

Carolina González Torres. Centro Médico Nacional. IMSS Siglo XXI

G34

Gene regulation of CTA1 in C. glabrata is controlled by different cis and trans-acting elements after being exposed to different stimuli

Carlos Ricardo González Ruiz. Instituto Potosino de Investigación Científica y Tecnológica A.C.

G35

Targeting CTCFL oncogene using CRISPR-Cas13 technology

Paula González Hernández. Universidad Autónoma Metropolitana

G36

Chromatin remodeling protein Abf1 regulates expression of adhesin encoding genes in Candida glabrata

Grecia Hernández Hernández. Instituto Potosino de Investigación Científica y Tecnológica A.C.

MEDICINE, HEALTH & NUTRITION I

MH1

Genomic analysis of mutations in the bruton tyrosine kinase gene in patient with X-LINKED agammaglobulinemia and gastric cancer

Dulce Jazmin Alvarez Olvera. Universidad Autónoma de la Ciudad de México

MH2

Boron derivatives as ACE/AChE inhibitors for the control of hypertension: Design, synthesis and in silico evaluation

Erik Andrade Jorge. Instituto Politécnico Nacional

MH3

Differential effect of MSCs on the immunomodulation capacity of breast cancer cells

Víctor Manuel Arenas Luna. Universidad Panamericana campus Ciudad de México

MH4

Antiobesogenic effect of D-Limonene, Ellagic acid, Gallic acid, and P-Coumaric acid in the liver of rats fed with high-fat diet

Luis Alberto Ayala Ruiz. Universidad Michoacana de San Nicolás de Hidalgo

MH5

Search for genetic biomarkers of risk for secondary failure to sulfonylureas

Mónica Vianney Ayala Martínez. Universidad Nacional Autónoma de México

MH6

Estimate of the consumption of NON-Caloric sweeteners in adults and their association with the risk of metabolic diseases

Edgar Daniel Barreras Duran. Universidad Estatal de Sonora

MH7

Analysis of the expression of enzymatic oncological clinical biomarkers in human saliva

Silvia Barrios Aguilar. Universidad Autónoma de Sinaloa

MH8

Development of an Enzyme-Linked Immunosorbent Assay (ELISA) for the rapid detection of the non-structural protein 1 (NS1) of the Dengue virus

Maidoly Batista Fernández. Cinvestav Zacatenco

MH9

Wood smoke extract induces alterations in mitochondrial biogenesis and increased production of reactive oxygen species in normal human fibroblasts

Carina Becerril Berrocal. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas

MH10

Effect of glycine betaine on the proliferation of human colon adenocarcinoma cells HT-29

Eiby Grisell Benitez Ibarra. Universidad de Sonora

MH11

The Importance of Nutrition in Alzheimer´s Disease

Oswaldo Bivian Paredes. Universidad Autónoma del Estado de Hidalgo

MH12

Sequestosome1 nuclear translocation after DNA damage in lung epithelial cells

Sandra Cabrera Benítez. Universidad Nacional Autónoma de México

MH13

Effect of empaglifozin treatment on magnesemia in metabolic síndrome

Juan Carlos Cabrera Ángeles. Instituto Nacional de Cardiología "Ignacio Chávez"

MH14

Analysis of Pulmonary Surfactant Protein C during Aging

Uriel Camacho Silverio. Universidad Nacional Autónoma de México

MH15

Effect of compounds from the aqueous extract of Erythraea tetramera Schiede on pancreatic lipase activity

Abelardo Camacho Luis. Universidad Juárez del Estado de Durango

MH16

Polar Fraction of Lophocereus schotti decreases TGFB1 Gene Expression in the Progress of Chemically Induced Hepatocarcinogenesis

Marina Campos Valdez. Universidad de Guadalajara

MH17

Effect of Egregia menziesii Algae Extracts on Viability and Migration of Rat Intestinal IEC-6 Cell Line

Christian Gidonni Carrión Hernández. Universidad Veracruzana

MH18

Evaluation of the effect of the GgXN polymer on the behavior of induced hemiparkinsonism in rat

Joshua Imanol Cervantes Andraca. Universidad Nacional Autónoma de México

MH19

Transcriptomic Analysis of Bone Neof ormation in the DBA/1 Mouse Model of Spondyloarthritis: Role of Multiple Osteogenic Mechanisms

Eduardo Chaparro Barrera. Universidad Autónoma de Chihuahua

MH20

One-carbon metabolism and fetal growth in the princesa cohort

Carolina Chetirquen Silva. Instituto Nacional de Medicina Genómica

MH21

Intermittent fasting reduces mitochondrial function in the colon to revert obesity-driven oxidative stress by modulation of the gut microbiota and the metabolome profile

Mary Carmen Citlally Condado Huerta. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán

MH22

Fungal Metabolites as Inhibitors of Shikimate Kinase from Methicillin-Resistant Staphylococcus aureus

Azhiel Contreras Chávez. Universidad Juárez del Estado de Durango

MH23

Changes in ovarian morphology and ultrastructure in offspring of mice with polycystic ovary syndrome

Paola Estefania Del Valle Ruvalcaba. Universidad Nacional Autónoma de México

MH24

Human sperm dysfunction is associated with overweight, obesity and oxidative stress

Aida Guadalupe Díaz Martel. Universidad Autónoma de Querétaro

MH25

Tumor-Educated Platelets in Hepatocellular Carcinoma: a Multicentric Study

Mariela Judith Domínguez Domínguez. Universidad Veracruzana

MH26

Curcuminoids effects on antioxidant system in plasma and liver of rats

María Teresa Espinosa García. Universidad Nacional Autónoma de México

MH27

Antioxidant Charge Reverts High-Density Lipoprotein (HDL)-Induced Endothelial Dysfunction in Women with Acute Coronary Syndrome

Diego Estrada Luna. Universidad Autónoma del Estado de Hidalgo

MH28

Residual glycosidase activity disguise the effect of commercial digestive enzymes on in vitro digestion of phenolic glycosides

César Femat Castañeda. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

MH29

Evaluation of HBB protein expression in extracellular vesicles isolated from plasmas of women with BI-RADS 4 classification

Juan Carlos Fernández Hernández. Escuela Superior de Medicina

MH30

Anosmia, odynophagia, and ageusia in COVID-19 positive patients

Karen Elizabeth Franco Reyes. Universidad Juárez del Estado de Durango

MH31

Formulation of a cookie with the addition of flour from wine industry residues

Carlos Emilio Galindo Corona. Universidad Autónoma de Coahuila

NEUROSCIENCES & NEUROBIOLOGY I

NN1

Cognitive stimulation increases microglial phagocytosis in an Alzheimer's disease model

David Francisco Aguilar Ávila. Universidad Nacional Autónoma de México

NN2

Age-related increases in skeletal muscle alpha-synuclein monomeric and oligomeric forms

Ana Leticia Arriaga Guerrero. Universidad Nacional Autónoma de México

NN3

Long and very-long chain ceramides promotes an anxiety-like behavior in female mice through microglia activation in cortex

Sofía Bernal Vega. Universidad Autónoma de Nuevo León

NN4

Impaired hippocampal neurogenesis and spatial learning deficit in adult Wistar rats with acquired hypothyroidism

Pablo Edson Bustamante Nieves. Instituto Politécnico Nacional

NN5

Effects of Acetyl-L-Carnitine and L-Carnitine on the Differentiation Efficiency and the Morphology of Motor Neurons Derived from mouse Embryonic Stem Cells

Maricela Cabrera Castro. Universidad Nacional Autónoma de México

NN6

Possible inflammation mediated by NF- κ B p65 in the retina on short-term induced-diabetes in a rat model

Alejandro Canizales Ontiveros. Universidad Nacional Autónoma de México

NN7

Effect of high-carbohydrate diet on mitophagy and mitochondrial dynamics of the frontal cortex of male Wistar rats

Karen Carreto Meneses. Universidad Autónoma de Puebla

NN8

LC noradrenergic neurons are involved in the lack Hypothalamus-Pituitary-Thyroid axis response to cold in stressed animals

Andrea Castillo Campos. Universidad Nacional Autónoma de México

NN9

Co-infection model of cytomegalovirus and ZIKA virus in human neural progenitor cells

Claudia Castillo Martín del Campo. Universidad Autónoma de San Luis Potosí

NN10

Study of Tibolone administration on NADPH oxidase expression in a murine model of traumatic spinal cord injury

Nadia Tzayaka Castillo Mendieta. Centro Médico Nacional Siglo XXI. IMSS

NN11

Induction, phosphorylation and protein interactions of PEBP1 under early focal cerebral ischemia/reperfusion in the rat hippocampus

Febe Elena Cázares Raga. Cinvestav Zacatenco

NN12

Effects of 2-APB on dopaminergic pathways

Rebeca Chávez Campos. Instituto Politécnico Nacional

NN13

Tibolone improves motor recovery, regulates neuroinflammation and gliosis in a model of traumatic spinal cord injury

Angélica Coyoy-Salgado CONAHCYT. CMN Siglo XXI. IMSS

NN14

Cephalic alterations produced by conditional inactivation of Plpp3 in the Wnt1expression domains

Sheyla Sabel Cruz Cruz. Universidad Nacional Autónoma de México

NN15

Maternal diabetes modifies the transcription of genes important for the division of cortical neural stem cells: Role of FOXP2

Diana Sarahi De la Merced García. Instituto Nacional de Perinatología Isidro Espinosa de los Reyes

NN16

New antiepileptic targets modified by levetiracetam treatment in dentate gyrus of rats with temporal lobe epilepsy

Verónica Díaz Villegas. Instituto Politécnico Nacional

NN17

Cognitive stimulation reduces neuroinflammation and increases Klotho levels in the hippocampus of mouse Alzheimer's Disease model

Gustavo de Jesús Fabián Rodríguez. Universidad Nacional Autónoma de México

NN18

Antioxidant effect of Psacalium decompositum in an Alzheimer's disease mouse model

Paulina Flores Galicia. Instituto Nacional de Geriatria

NN19

Effect of Sulforaphane or Dasatinib + Quercetin on cognitive impairment and neuroinflammation in an experimental model of chronic obesity in middle-age female Wistar rats

Rosa Pamela Flores Torres. Universidad Autónoma Metropolitana Iztapalapa

NN20

Study of morphological alterations caused by Plpp3 deficiency in the cephalic neural crest of mouse embryos

Alejandro Elliot Flores Oliva. Universidad Nacional Autónoma de México

NN21

Maqui berry decreases oxidative stress and inflammation in the hippocampus of ozone-exposed rats

Christian Guerra Araiza. Centro Médico Nacional "Siglo XXI". IMSS

NN22

Hypothalamic transcriptomics under different high-fat diets

Ruth Gutiérrez Aguilar. Universidad Nacional Autónoma de México

NN23

Evaluation of the effect of metformin on learning and long-term memory in Type 2 diabetic mice

Joan Aldae Hernández Poblano. Universidad Autónoma de Coahuila

NN24

Effect of a High-carbohydrate Diet on Inflammation and Microglial Phenotype in the Adenohypophysis of Postpubertal Wistar Rats

Hugo Hernández Frago. Universidad Autónoma de Puebla

NN25

GH treatment decrease astrogliosis in the post stroke

Martín Hernández Lucas. Universidad Nacional Autónoma de México

NN26

Leptomerin reduces locomotor and spatial memory deficits caused by intrahippocampal injection of A β 1-42

Dulce Hernández Sacramento. Universidad Autónoma de Puebla

NN27

Role of sodium phenylbutyrate on hippocampal morphological changes of hyperammonemic rats induced with CCl $_4$

Brenda Hernández Juárez. Universidad Autónoma de Puebla

SIGNAL TRANSDUCTION AND CELL DIFFERENTIATION I

ST1

Identification of novel c-di-AMP binding proteins that regulate mutagenesis and DNA repair in *Bacillus subtilis*

Karen Abundiz Yáñez. Universidad de Guanajuato

ST2

Regulation of the activity of insulin-like growth 1 receptor by estradiol in breast cancer cell MCF-7

Esteban Acosta Ramos. Cinvestav Zacatenco

ST3

α 1A-Adrenergic receptor function: roles of IL3 and Cterm phosphosites

Rocío Alcántara Hernández. Universidad Nacional Autónoma de México

ST4

Thrombin-induced NF κ B activation in retinal pigmented epithelium cells (RPE)

Daniela Alejandra Alvarado Fernández. Universidad Nacional Autónoma de México

ST5

Effect of proinflammatory cytokines on the expression of molecular markers of the cancer stem cell phenotype in PC3 cells

Osiris Evelyn Aparicio Carrillo. Universidad Autónoma de Puebla

ST6

Unlocking the potential of approved drugs for the inhibition of PTP1B in cancer therapy

Luis Enrique Arias Romero. Universidad Nacional Autónoma de México

ST7

Leptin modifies the metastatic potential in castration-resistant prostate cancer cells (PC3) through the TRPM7 channel

Rubén Ávalos López. Universidad Autónoma de Puebla

ST8

Evaluation the role of Ser176 phosphorylation in the activity and localization of SnRK1

Beatriz Alejandra Ávila Castañeda. Universidad Nacional Autónoma de México

ST9

Initiation of intracellular trafficking of the LPA3 receptor after its stimulation

Elias Uriel Avila García. Universidad Nacional Autónoma de México

ST10

Adrenoceptor α 1A (ADRA1A) integrates a signaling signature statistically linked to longer survival of liver hepatocellular carcinoma patients

Yarely Mabel Beltrán Navarro. Universidad Nacional Autónoma de México

ST11

Role of HPV16 E1 protein in the activation NF- κ B signaling pathway

Alicia Beltrán Soto. Instituto Nacional de Cancerología

ST12

Ouabain promotes claudin-1, -2, and -4 autophagic degradation through oxidative stress and AMPK activation in MDCK cells

Jessica Paulina Campos Blázquez. Cinvestav Zacatenco

ST13

Role of AhR in Cell Morphology during Neuronal Differentiation of SH-SY5Y Cells

Jimena Cris Campos Arce. Cinvestav Zacatenco

ST14

Testosterone increases the expression of KV channels and enhances the airway smooth muscle relaxation induced by P2Y4 and adenylyl cyclase signaling

Abril Carbajal-García. Universidad Nacional Autónoma de México

ST15

Thyroid Hormone T3 Exerts Distinct Regulatory Effects on Metabolism in White, Brown, and Brite Adipocytes

Lidia Itzel Castro Rodríguez. Cinvestav Zacatenco

ST16

Participation of E6 and E7 HPV16 oncoproteins in cell invasion and migration mediated by the down regulation of RhoE/Rnd3 GTPase

Yesenia Cid Cruz. UNAM/Instituto Nacional de Cancerología

ST17

[Ca²⁺]_i] distribution in spermatozoa from two populations of Sceloporus grammicus along an altitudinal gradient

Gabriela Contreras González. Universidad Autónoma Metropolitana Iztapalapa

ST18

Effect of Corticotropin-Releasing Factor (CRF) on the activation of MAPK and PI3K/Akt pathways induced by Insulin-like Growth Factor-1 (IGF-1) in CHO-K1 cells

Carlos De Jesús Quiroz. Cinvestav Zacatenco

ST19

PTP1B modulates endothelial-to-mesenchymal transition during TNF-induced endothelial dysfunction

José Esparza López. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán

ST20

Effect of VPH16 E6 and E7 on mast cell migration

Zyanya Patricia Espinosa Riquer. Instituto Nacional de Cancerología

ST21

Role of the Ire1-mediated endoplasmic reticulum stress pathway during the development of the tracheal system in Drosophila melanogaster

María Fernanda Frutis Osorio. Universidad Nacional Autónoma de México

ST22

Role of STAT5 in energy mitochondrial metabolism genes regulation in cervical cancer cells stimulated with Interleukin 2

Rubén Alejandro Fuentes Pascacio. Universidad Nacional Autónoma de México

ST23

Analysis of the relationship between the JAK/STAT pathway and the death receptor CD95 to induce proliferation and survival of cervical cancer cells

Armando García Velasco. Universidad Nacional Autónoma de México

ST24

Aberrant Epithelial Reprogramming in the Lung from Mmp8-Mmp13 double knockout mouse impairs fibrosis resolution

Ángeles García-Vicente. Universidad Nacional Autónoma de México

ST25

Effect of Insulin on Vascular Endothelial Growth Factor-Induced Actions in Endothelial Cells EA.hy926

Maikel González Matos. Cinvestav Zacatenco

ST26

Toll-like receptor (TLR)-4 activation induces mast cell migration and participates on their incorporation to solid tumors

Daniel Guerrero Morán. Cinvestav Sede Sur

ST27

STAT3 role in the autophagy activation in cervical cancer cells treated with high doses of IL-2 and DDP

Adriana Gutiérrez Hoya. Universidad Nacional Autónoma de México

ST28

Role of MEOX2 in modulating the expression and/or activation of MEKK1/2 promoting cell proliferation in lung cancer

Brenda Naomi Hernández Ramos. Universidad Nacional Autónoma de México

ST29

Effect of caffeine on metastatic potential in non-small cell lung cancer (A549)

Yarely Rocío Herrera Sánchez. Universidad Autónoma de Puebla

ST30

Jasmonic acid regulates cell death and regeneration in the Arabidopsis roots of a mutant with compromised cell viability

Pedro Iván Huerta Venegas. UMSNH/Instituto de Ecología A.C.

ST31

Cell senescence modifies phenotype and function of Mast Cells

Alfredo Ibarra Sánchez. Cinvestav Sede Sur

SYSTEMS BIOLOGY & BIOINFORMATICS I

SB1

Dynamics of Bacterial Communities in Response to Disturbances

Roberto Carlos Álvarez Martínez. Universidad Autónoma de Querétaro

SB2

Key Proteins for Regeneration in *A. mexicanum*: Transcriptomic Insights from Aged and Juvenile Limbs

Aylin Del Moral Morales. Universidad Autónoma Metropolitana

SB3

Functional and structural studies of a novel endolysin from ICP1 phage active against multidrug-resistant bacteria

Laura Angélica Espinosa Barrera. Universidad de Colima

SB4

Unraveling the distinct biases of the genomic landscape of lung adenocarcinoma from Mexican patients

Bertha Rueda Zarazua. Universidad Nacional Autónoma de México

SB5

Effect of the Lys62Ala Mutation on the Thermal Stability of BstHPr Protein by Molecular Dynamics

Salomón de Jesús Alas Guardado. Universidad Autónoma Metropolitana

SB6

Experimental and computational study of benzazepiniones as potential inhibitors of *Mycobacterium tuberculosis* HadAB dehydratase

Armando Alberto Ambrosio Huerta. Universidad Nacional Autónoma de México

SB7

Analysis of gene variations in tumour and healthy tissues of acral lentiginous melanoma patients

Orlando Ángeles Martínez. Universidad Mexiquense del Bicentenario

SB8

Identification and Functional Characterization of the Genetic Circuits Controlling Cell Wall Metabolism in *Jipi Palm* (*Carludovica* spp.)

Jorge Luis Araujo Sánchez. Centro de Investigación Científica de Yucatán A.C.

SB9

Computational screening to predict microrna targets in the flavivirus 3' untranslated region for antiviral development

Rodolfo Gamaliel Avila Bonilla. Cinvestav Zacatenco

SB10

Viral metagenomics in hypersaline environments

María Guadalupe Ayala Rodríguez. Universidad Autónoma del Estado de Morelos

SB11

Clustering Bacterial Promoter Sequences Using Supervised Machine Learning

Paulo Cambranis Boldo. Universidad Nacional Autónoma de México

SB12

Microalgal peptides against the main protease (Mpro) of SARS-CoV-2: In silico evaluation

David Mauricio Cañedo Figueroa. Universidad Autónoma de Sinaloa

SB13

Evolutionary history and distribution analysis of rhamnosyltransferases in the fungal kingdom

Joaquín Omar Chávez Santiago. Universidad de Guanajuato

SB14

Bioinformatic Analysis of Whole Genomes of Staphylococcus aureus Isolated from the Skin of Atopic Dermatitis Patients and Healthy Subjects

Sergio Alland Colorado Cortés. Universidad Nacional Autónoma de México

SB15

Transcriptomic analysis of the infective process of the mistletoe Psittacanthus calyculatus in the host tree mesquite Prosopis laevigata

Marco Ramsés Cota Pineda. Centro de Investigación en Alimentación y Desarrollo, A.C.

SB16

Early and late transcriptional responses to auxins and cytokinins during the induction of somatic embryogenesis in Coffea canephora

Marcos David Couoh Cauch. Centro de Investigación Científica de Yucatán A.C.

SB17

Microbial community dynamics through metagenomics in two experimental biodigesters for rural community use

Erika Viridiana Cruz Bonilla. Cinvestav Irapuato

SB18

Study of perivascular fiber formation in Agave fourcroydes through a transcriptomic approach

José Roberto Cruz Balam. Centro de Investigación Científica de Yucatán A.C.

SB19

In Silico Inhibitory Potential of Peptides HRA and PK-2 Against the Fusion Protein F of Respiratory Syncytial Virus

Deyanira Judith De Anda Alejandrez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

SB20

How to model the structure-function relationship of proteins?

Gabriel Del Río Guerra. Universidad Nacional Autónoma de México

SB21

Analysing the co-expression and protein-protein interaction networks related to thermotolerance in Arabidopsis thaliana

Ana Karen Diaz Martinez. Instituto Tecnológico Superior de Irapuato

SB22

Theoretical study of the NamH hydroxylase of Mycobacterium tuberculosis as a potential target in the development of new antibiotics

Roberto Sealtiel Farías Gaytán. Universidad Nacional Autónoma de México

SB23

Phenotypic and genotypic evaluation of antibiotic resistance in Pseudomonas aeruginosa isolated from hydrothermal water samples from Chignahuapan, Puebla

Jennifer Vianey Fernández Doderó. Universidad Autónoma de Puebla

SB24

Developing a Generative AI Assistant for RegulonDB: Enhancing Data on E. coli K-12 Transcriptional Regulation

Ma del Socorro Gama Castro. Universidad Autónoma de la Ciudad de México

SB25

Evaluation of two taxonomic assignment methods for the study of lizard gut microbiome using metagenomic data

Elizabeth Selene Gomez Acata. Universidad Autónoma de Tlaxcala

SB26

Pipeline implementations using the Galaxy platform for reference assembly of chloroplast genome from transcriptome data

Ana Carolina González Trillo. Instituto Politécnico Nacional

OTHERS I

O1

Effect evaluation of Curcumin and Resveratrol compounds on cancer stem cell-enriched cultures derived from cancer cell lines

Jorge Aguilar Meza. Instituto Nacional de Cancerología

O2

Mesenchymal-Amoeboid Transition in Triple-Negative Breast Cancer line, MDA-MB-231

Arturo Aguilar Rojas. Instituto Mexicano del Seguro Social

O3

Towards big data analysis of a superbug's molecular epidemiology

Omar Alejandro Aguilar Vera. Universidad Nacional Autónoma de México

O4

Study of the stability of the Rb protein and its modulation by MDM2

Gabriel Alonso Pérez. Universidad Autónoma de San Luis Potosí

O5

Manifestaciones bucales presentes en pacientes con enfermedad de Parkinson de la ciudad de Durango y su relación con la progresión de esta enfermedad

Leslie Mariana Apodaca Ayala. Universidad Juárez del Estado de Durango

O6

Molecular Insights into Folate Dynamic Metabolism in Common Bean (*Phaseolus vulgaris*) Seeds During Storage

Itzel Astrid Aviña Ávalos. Tecnológico de Monterrey

O7

Common bean transcription factors from the AGL/MADS and SPL families drive the expression of the Mir172c gene, a key regulator of common bean N-fixing symbiosis

Litzy Ayra Pardo. Universidad Nacional Autónoma de México

O8

Rapid identification of bioactive compounds from agave by MS-DART as potential biomarkers for archaeological studies

Emanuel Bojórquez Quintal. El Colegio de Michoacán, A.C.

O9

Studying nucleophagy in the longest-lived rodent, the Naked Mole-Rat

Luis Antonio Buendía Sánchez. Universidad Nacional Autónoma de México

O10

Laccase proton relay mechanism from *Thermus thermophilus* HB27

María Cristina Cardona Echavarría. Universidad Nacional Autónoma de México

O11

Resistin and adiponectin modulate ABCA1 and ABCG2 transporters in an in vitro placenta model

Amanda Bexidiu Castillo Escoto. Instituto Nacional de Perinatología Isidro Espinosa de los Reyes

O12

Rapid characterization of commercial essential oils major components by direct methods (ATR-IR and MS-DART)

Ana V. Coria Téllez. El Colegio de Michoacán, A.C

O13

SERCA pumps counteract tetanic contractions while maintaining the force amplitude of single twitch contractions

Adan Dagnino Acosta. Universidad de Colima

O14

Purification and characterization of the esterase TA0887 from Thermoplasma acidophilum

Alejandro Delgado Rey. Instituto Potosino de Investigación Científica y Tecnológica A.C.

O15

Characterization of the Inhibitory Effects of Coagulase-Negative Staphylococci from Healthy Subjects on the Growth of Staphylococcus aureus Isolated from Atopic Dermatitis Patients

Leticia Itzel Diaz Ramos. Universidad Nacional Autónoma de México

O16

Molecular docking of glycerol kinase driven monophosphorylations of small alcohols

Wendy Itzel Escobedo Hinojosa. Universidad Nacional Autónoma de México

O17

Generation of Human Antibodies in NSG Mice Against Specific Targets of Alzheimer's Disease

Jeniffer Espinoza Hernández. Instituto Politécnico Nacional

O18

Studies of the effect of the interaction of the E1B- 55K protein with RNA on transcription and splicing of adenovirus late viral mRNA

Diana Laura Flores Garcia. Universidad Autónoma del Estado de Morelos

O19

Gender leadership in bacteriology and Archaea research in Latin America

Luis Ernesto Fuentes Ramírez. Universidad Autónoma de Puebla

O20

Cyclic Nucleotide-Gated Ion Channels: A Potential New Player in Plant Antiviral Defense Against Begomovirus Infection

Ana María García Montelongo. Universidad de Colima

O21

HGF modulates the MAPK/ERK 1/2 pathway to exert its hepatoprotective effect in a murine model of experimental cholestasis

Maricruz García Barrera. Universidad Autónoma Metropolitana Iztapalapa

O22

Relationship between external pH, Ca²⁺ oscillations and intracellular pH in human spermatozooids

Ángela Raquel García Martínez. Universidad Nacional Autónoma de México

O23

Study of calcium oscillations induced by membrane depolarizing pulses in human sperm

Mariam Gasga Tehuintle. Universidad Nacional Autónoma de México

O24

Sequence space analysis of homologous proteins with (β/α)₈ fold

Estefani Gaytan Nuñez. Universidad Nacional Autónoma de México

O25

Surface engineering of the encapsulin nanocompartment of Myxococcus xanthus for the generation of protein delivery vehicles

Sac Nicté Gómez Barrera. Universidad Nacional Autónoma de México

O26

Phylogeography of Zamia loddigesii Miq in the Gulf of Mexico

Marcel Arturo Gómez García. Universidad Veracruzana

O27

The role of the ubiquitination in the replication of the neurotropic astrovirus VA1

Jaqueline Gómez Reyes. Universidad Nacional Autónoma de México

O28

Role of SERCA pumps in the shape of the contraction-relaxation kinetics of soleus muscle and its effect on force production and fatigue

Ana Marcela González Bedoy. Universidad de Colima

O29

Description of SYCP3 accumulations in primary spermatocytes of the first spermatogenic wave of prepubertal mice

Luis Pablo Guzmán Vargas. Universidad Nacional Autónoma de México

O30

Effects of melatonin on human sperm in vitro capacitation

Gabriela Hernández Silva. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán

O31

Nanovaccines design against Vibrio cholerae

Alejandro Huerta Saquero. Universidad Nacional Autónoma de México

O32

Expression of MICAL2 protein in oral squamous cell carcinoma. Bibliographic review pilot study

José Manuel Huizar Reyes. Universidad Juárez del Estado de Durango

O33

Participation of estrogen and progesterone receptors, and PKA signaling in the expression regulation of unfolded protein response genes during decidualization in immortalized human endometrial stromal cells

Jesús Iván Jiménez Rivera. Universidad Nacional Autónoma de México

O34

Uso del examen escrito para evaluar algunas competencias en la asignatura de Bioquímica y Biología Molecular: Una nueva perspectiva para un viejo instrumento

Kevin David Laguna Maldonado. Universidad Nacional Autónoma de México

O35

Bioprospecting of endophytic fungi from macroalgae of the mexican caribbean sea

Ixchel Adriana Loa Ramírez. Universidad Nacional Autónoma de México

O36

Vigor characterization of wild and cultivable amaranth seeds

Alberto Antonio Lomelí Bernal. Instituto Potosino de Investigación Científica y Tecnológica A.C.

O37

Fluoracil's Senolytic Potential: Targeting Selective Cell Death in Senescent Cells

Norma Edith López Díaz Guerrero. Universidad Autónoma Metropolitana Iztapalapa

O38

Antitumour and antimigratory activity of glaucolid E in breast cancer cells

Hugo Aramís López Meléndez. Universidad Nacional Autónoma de México

O39

Subcellular localization of phosphomimetic forms of Rb

Leonardo Lozoya Lozoya. Universidad Autónoma de San Luis Potosí

O40

In silico determination of pKa AND logD of novel voltage-gated potassium ion channel blockers based on 4-aminopyridine

Jair Alejandro Madera Hurtado. Universidad de Guadalajara

POSTER SESSION II
TUESDAY, OCTOBER 22
17:00 - 19:00

BASIC BIOCHEMISTRY I

BB1

Evaluation of specific L-glutaminase activity in the unconventional yeast

Rhodotorula mucilaginosa

Paola Itzel Acosta Valdelamar. Universidad Nacional Autónoma de México

BB2

Are Zea mays HXK7-8 glucose sensors?

David Acosta Muro. Universidad Nacional Autónoma de México

BB3

Overexpression of TIMP3 decreases migration in A549 cell line

Arantxa Melissa Aguilar López. Instituto Nacional de Enfermedades Respiratorias "Ismael Cosío Villegas"

BB4

Inhibition of p62 reduces mobility and intracellular calcium in mouse spermatozoa

Charmina Aguirre Alvarado. IMSS

BB5

Effect of chitosan on the permeability of different yeasts strains

Minerva Araiza Villanueva. Universidad Nacional Autónoma de México

BB6

The C-terminal fragment of one of the asparaginyl endopeptidases (AEPs) of

Trichomonas vaginalis works as an AEP proteolytic activity inhibitor

Rossana Arroyo Verastegui. Cinvestav Zacatenco

BB7

Phenotypical responses of seedlings from Arabidopsis thaliana mpk3 and mpk6 mutants to high temperatures

David Fernando Barrera Gómez. Universidad Nacional Autónoma de México

BB8

Chemical Remotion of Glycan Moieties to Enhance Protein Identification by LC-MS/MS

Esaú Bojórquez Velázquez. Instituto de Ecología

BB9

Development of a Tool for the Assessment of Critical Thinking in Medical Students

Oliva Briz Tena. Universidad Nacional Autónoma de México

BB10

Relationship between the enzyme glucose-6-phosphate dehydrogenase (ZWF1) and hydrogen sulfide production in the yeast *Saccharomyces cerevisiae*

María Jatziry Camacho Campos. Universidad Nacional Autónoma de México

BB11

Role of the *KINrg1* and *KIRtg3* proteins from *K. lactis* in glucose repression, retrograde response and mitochondrial DNA integrity

Carlos Campero Basaldua. Universidad Nacional Autónoma de México

BB12

The moonlight protein *Alt1*

Eduardo Cardona Guerrero. Universidad Nacional Autónoma de México

BB13

Hepatoprotective effect of bacterial cyclodipeptides in obesity rats model

Ángela Regina Castillo Jiménez. Universidad Michoacana de San Nicolás de Hidalgo

BB14

Phloem exudate proteins of grafted and non-grafted beans (*Phaseolus sp.*) cultivated under irrigation and water restriction during grain filling

Martha Itsel Cayetano Marcial. Colegio de Postgraduados

BB15

A biocomputational and site-directed mutagenesis approach to recover the acid phosphatase/phytase activity of *EhHAPP49*, an atypical amoebic enzyme

Guadalupe Alejandra Chávez Melendrez. Universidad Autónoma de Baja California

BB16

Fumarate reductase in *Rhodotorula mucilaginosa* and its possible physiological roles

Natalia Chiquete Félix. Universidad Nacional Autónoma de México

BB17

Introgression of drought tolerance of *Phaseolus acutifolius* A. Gray to *Phaseolus vulgaris* L

Marcela Cilia García. Colegio de Postgraduados

BB18

pBr-2-APB is an inefficient endoplasmic reticulum calcium releasing agent

Rodrigo Contreras Gaytán. Cinvestav Zacatenco

BB19

DPE2 activity in bean fruit pericarp is post-translationally regulated

Gerardo Contreras Ruiz. Universidad Nacional Autónoma de México

BB20

Lactate deshydrogenasa and Its Relevance in Cancer Biology: Regulation, Isoenzymes, and Inhibition Mechanisms

Carla Isabela Cuevas Bañuelos. Instituto Politécnico Nacional

BB21

Chlorotracker: a photoactivable 1,8-naphthalimide derivative as a probe for chloroplast imaging

Karen Melissa Dahlman Cabrera. Universidad Nacional Autónoma de México

BB22

Engineering of nanocompartments based on encapsulin a from myxococcus xanthus for use as a protein release system

Willy Ángel Delgado Tapia. Universidad Nacional Autónoma de México

BB23

Evaluation of the antifungal effect of the extract of Moringa oleifera on Aspergillus parasiticus

Edmar de Jesús Díaz García. Universidad Autónoma Benito Juárez de Oaxaca

BB24

MCT11 Transporter overexpression in vitro leads to lipid accumulation

Zuleima Natali Domínguez. Velázquez. Cinvestav Zacatenco

BB25

Evaluation of the changes in carbon fixation and carbohydrate content between diverse bread wheat genotypes tolerant and sensitive to heat stress

Ana Laura Encinas Montes. Centro de Investigación en Alimentación y Desarrollo, A.C.

BB26

Identification and Evaluation of Tannins and Flavonoids of Nephelium lappaceum of industrial interest

Adelma Escobar Ramírez. Universidad Juárez Autónoma de Tabasco

BB27

Impact of the W165F Mutation in pkBADH on the Formation of the pkBADH-NAD⁺ Complex

Anabel Félix Arredondo. Universidad de Sonora

BB28

Defining essential and non-essential chloroplast ribosomal proteins in Nicotiana tabacum and Arabidopsis thaliana

Emanuel Osmar Flores Camargo. Instituto Politécnico Nacional

BB29

The combination of three drugs modifies the metabolism in colorectal cancer cells

Laura Cecilia Flores García. UNAM. Instituto Nacional de Cancerología

BB30

The translation factor eIF4E is a key mediator of Doxorubicin resistance: Insights from a triple-negative breast cancer model

Héctor Frayde Gómez. Universidad Autónoma de Baja California

BB31

Determination of the activity of the enzymes in the primary assimilation of nitrogen in the embryogenic system of Coffea canephora

Laura Chanel Fuentes Vázquez. Centro de Investigación Científica de Yucatán A.C.

BB32

Isolation of intact *P. parva* and *C. reinhardtii* plastids for the standardization of an in vitro protein import model

Sergio Fuentes Hernández. Universidad Nacional Autónoma de México

BB33

Fluoride concentration in the breast milk of lactating women in the city of Durango

Sergio Alberto Galindo Najera. Universidad Juárez del Estado de Durango

BB34

Characterization of mitochondrial Ca^{2+} regulation in HeLa cells

Martín Leonardo Gallegos Gómez. Cinvestav Zacatenco

BB35

Challenges in the Solubility and Folding of Recombinant Lipases: The Case of LipGOM6

Elena Lizbeth García Villegas. Universidad Autónoma del Estado de Morelos

BB36

Specificity and Mechanisms of c-type Cytochrome Biogenesis in Malaria Parasites

Aldo E. García Guerrero. University of Utah

BB37

Characterization of the VDAC family in maize and the physiological role of the isoforms ZmVDAC1b and ZmVDAC4b in drought stress

Donají Azucena García Ortiz. Universidad Nacional Autónoma de México

BIOTECHNOLOGY II

BT54

Insights in the antibacterial structure-function relationship of the pepper defensin J1-1

Georgina Estrada Tapia. Centro de Investigación Científica de Yucatán A.C.

BT55

Overproduction of medically and biotechnologically relevant phenazines using a mutant in *rsma* of *Pseudomonas aeruginosa* ID4365 with attenuated virulence

Misael Josafat Fabían Del Olmo. Universidad Nacional Autónoma de México

BT56

Identification of an antigenic protein from *Helicobacter pylori* through prediction of T and B epitopes and production of the recombinant protein in *Escherichia coli*

Brenda Yarely Fernández Madrid. Instituto Tecnológico de Durango / TecNM

BT57

Colorimetric evaluation and pigment content in leaves of different varieties of *A. hypochondriacus*, *A. hybridus* and *A. caudatus* in vegetative stage

Guadalupe Abigail Flores Pérez. Instituto Politécnico Nacional

BT58

Finding novel alkyl glucoside-producing and methanol-tolerant variants from an *AmyA* alpha-amylase library

Kimberly Flores Rivera. Universidad Nacional Autónoma de México

BT59

Cellulolytic activity optimization of enzymatic extracts from a native strain of *Bacillus subtilis* isolated from *Agave durangensis* bagasse

Miguel Antonio Franco Vazquez. Universidad Juárez del Estado de Durango

BT60

Synthesis of gold nanoparticles using a lipid-rich extract from mexican avocado seed and evaluation of its cytotoxicity on murine melanoma cells

Minerva Frutis Murillo. Universidad Michoacana de San Nicolás de Hidalgo

BT61

Construction of an expression vector for the *SPSK_04019* gene encoding an *Hsp70* from *Sporothrix schenckii* and expression of the *rHSP70* Protein

Connie Gallegos Rivera. Universidad Juárez del Estado de Durango

BT62

Identification and isolation of strains of agronomic interest in agricultural soil with clay texture

Jesus Garcia Pereyra. Instituto Tecnológico del Valle del Guadiana

BT63

Evaluation of the effect of zinc oxide nanoparticles on jalapeño pepper (*Capsicum annuum*) crop. Bioaccumulation, metabolic profile and bacteriome

Luis Alberto García Casillas. Universidad de Guadalajara

BT64

Green synthesis of silver nanoparticles and its effects on plants

Cristina Garcidueñas Piña. Universidad Autónoma de Aguascalientes

BT65

Effect of dissolved oxygen in the growth of a recombinant glycoprotein-producing *Pichia pastoris*

Rogelio Diego Gaytan Castro. Universidad Nacional Autónoma de México

BT66

Expression of membrane receptors in cell-free systems from *Escherichia coli* and their use in the generation of biosensors

Karen Stephania González Ponce. Universidad de Guanajuato

BT67

Metzal as a texturizing agent in the development of barley (*Hordeum vulgare*)

Neftali Esteban Guzmán Mendoza. Universidad Autónoma del Estado de Hidalgo

BT68

Plant Growth-Promoting Bacteria enhances the biomass of *Agastache mexicana* subsp *mexicana* and improves the production of high-value secondary metabolites in greenhouse conditions

Héctor Daniel Hermenegildo Rosas. Universidad Autónoma del Estado de Morelos

BT69

Expression of the recombinant BoDef1: a Class I Defensin from *Bixa orellana*

Raquel Fabiola Hernández Díaz. Centro de Investigación Científica de Yucatán A.C.

BT70

Identification of virulence gen *fimh* in clinic isolates of *Klebsiella pneumoniae*

Nubia Francelica Hernández Sánchez. Universidad Autónoma de Sinaloa

BT71

In silico analysis of hotspots in replicative polymerases and their relationship with gynecological cancer

Kayley Aileen Hernández Ramírez. Universidad Autónoma del Estado de Hidalgo

BT72

Biotechnological synthesis of precursors of interest in the production of SSRIs

Olga Livier Jasso Durón Instituto Politécnico Nacional

BT73

Molecular Identification of a Highly Polyhydroxybutyrate-Producing Strain

Alfonso Jiménez Adón. Instituto Politécnico Nacional

BT74

Ex situ fermentation biomass monitoring by impedance spectroscopy

José Raúl Junco Carmona. Instituto Politécnico Nacional

BT75

Amylase production from solid state fermentation and submerged liquid fermentation by thermotolerant filamentous fungi obtained from the Tolantongo caves

Adriana Jazmín Legorreta Castañeda. Instituto Politécnico Nacional

BT76

Analysis of the effect of vascular and mesophyll expression of a common bean aquaporin PvAQP1 on Arabidopsis thaliana growth

Valeria Alexandra López Toledo. Cinvestav Zacatenco

BT77

Identification of antigens in the immunoproteome of porphyromonas gingivalis: progress

Héctor Samuel López Moreno. Universidad Autónoma de Sinaloa

BT78

R-LuxHR: Quantitative homologous recombination reporter system

Eduardo López Urrutia. Universidad Nacional Autónoma de México

BT79

O/W emulsions stabilized with modified chipilin protein (Crotalaria longirostrata)

Diana Itzel López Monterrubio. Universidad Autónoma de Chapingo

BT80

Extracellular Fragmented Self DNA acts a damage signal in Neochloris oleoabundans

Nancy Edith Lozoya Pérez. Centro de Innovación Aplicada en Tecnologías Competitivas, A.C.

BT81

Phenological evaluation in Capsicum annum plant by the application of chitosan nanoparticles

Eva Mariela Lunar Mata. Universidad de Guanajuato

BT82

Cloning of flippase IA of Giardia intestinalis

Carmen Alejandra Luque Camacho. Universidad Autónoma de Sinaloa

BT83

Evaluation of microalgae growth Porphyridium cruentum in relation to light intensity

Mildred Yael Manriquez Zamora. Instituto Politécnico Nacional

BT84

Systematic evaluation of selective antimicrobial peptides

Gabriel Marcelino Pérez. Universidad Nacional Autónoma de México

BT85

Phylogenetic analysis of *Aspergillus flavus* enzymes with high potential for PET (polyethylene terephthalate) degradation

Noemí Marchena Echeverría. Universidad de Guadalajara

BT86

Advancements in barley variety development: technologies and applications

Julio Armando Massange Sánchez. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BT87

Effect of different C:N and magnesium ratios on PHB accumulation in *Bacillus thuringiensis*

Yazmin Melendez Xicohtencatl. Instituto Politécnico Nacional

BT88

Relation between environmental conditions and organic fertilizers in corn seeds quality

Lucila Méndez Morán. Universidad de Guadalajara

BT89

Characterization of a recombinant protease inhibitor from broccoli (*Brassica oleracea* var. *italica*)

Maria Fernanda Mendoza Acosta. Universidad de Guanajuato

BT90

Formulation and characterization of raspberry-based products produced in Huejotzingo, Puebla

Maricarmen Meneses Juárez. Instituto Politécnico Nacional

BT91

Encapsulation of the *Bacillus subtilis* PAD123-Pars::gfpmut3a biosensor in alginate beads for the detection of arsenic in water

Noé de Jesús Miranda Breceda. Universidad Juárez del Estado de Durango

BT92

Antimicrobial and growth-promoting properties of silver nanoparticles from *Beta vulgaris* L. leaves on *M. bombycina* and *S. undatus* in vitro

José Francisco Morales Domínguez. Universidad Autónoma de Aguascalientes

BT93

Genetic transformation of *Mimosa tenuiflora* for the establishment of hairy root cultures

Jorge Humberto Mundo Ariza. Universidad Autónoma del Estado de Morelos

BT94

Unveiling Remote Homologs of Fungal Cell Wall Proteins Using AI-Based Structural Predictions

Pablo Valentín Navarro Enguilo. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BT95

Formulation of an organo-mineral fertilizer inoculated with plant growth-promoting bacteria

Melissa Nuñez Valdés. Universidad Autónoma de Puebla

BT96

Agavin promotes beneficial microbes in the shrimp microbiota

Adrián Ochoa Leyva. Universidad Nacional Autónoma de México

BT97

Separation and characterization of the charge variants of the monoclonal antibody bevacizumab

Maricela Olvera Rodríguez. Instituto de Biotecnología

BT98

Synthesis of xanthan gum derivatives and its biological properties

Karen Onofre Rentería. Universidad Autónoma de Coahuila

BT99

Bioinformatic analysis of the relationship between the environment and microorganisms for maize yield simulation

Jimena Ortiz Laris. Instituto Tecnológico Superior de Irapuato

BT100

Effect of SO_x from simulated flue gas on growth and gene expression of S-compounds in the microalgae *Desmodesmus abundans* RSM

Adriana Pacheco Moscoa. Tecnológico de Monterrey

GENETICS, EPIGENETICS AND GENETIC REGULATION II

G37

Clinical value of gene expression profiling by digital PCR for the detection of leukemic cells in pediatric population

Adriana del Carmen Aguilar Lemarroy. Instituto Mexicano del Seguro Social

G38

Interaction between genetic variants in vitamin D metabolism genes: impact on rheumatoid arthritis susceptibility and hypovitaminosis D

Bertha Campos López. Universidad de Guadalajara

G39

Insulin pathway changes on the gene expression in adipose tissue modulated by ellagic acid in obese Wistar rat induced with high fat diet

Ramón Castillo Correa. Instituto Politécnico Nacional

G40

Genetic variants related with neoadjuvant chemotherapy response in breast cancer patients

Luis Felipe Jave Suárez. Universidad de Guadalajara

G41

The increase in methylation of the ABCG2 gene is related to the presence of its Q141K allele and lower expression of the gene in peripheral blood of patients with gout and controls

Ámbar López Macay. Instituto Nacional de Rehabilitación

G42

Comparative genomic analysis of Candida glabrata clinical isolates to understand antifungal resistance

Ana Lizbeth López Marmolejo. Instituto Potosino de Investigación Científica y Tecnológica A.C.

G43

The forgotten side of the GPN-loop GTPase Npa3: the carboxy-terminal domain is a critical regulator of the GTPase core function

Manuel de Jesús Ochoa Valdez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

G44

Unveiling molecular differences in 3D cell Culture of Triple Negative Breast Cancer cells and their resemblance to Tumor Tissue

Mercedes Olvera Valencia. Instituto Politécnico Nacional

G45

Epigenetic regulation of ATP2A3 and CACNA1H genes in HepG2 cells

Guadalupe Hernández Martínez. Universidad Veracruzana

G46

Functional study of Maf1 in the protozoan parasite Leishmania major

Aldo Rodrigo Hernández Zamarripa. Universidad Nacional Autónoma de México

G47

Metabolic processes associated with chemotaxis responses in Kosakonia cowanii

María Inés Hernández Castellanos. Universidad Autónoma de Querétaro

G48

Detection of single-nucleotide polymorphisms in BLK, STAT4 and IRF5 genes associated with pediatric antiphospholipid syndrome by real-time PCR genotyping

Tanya Paola Hidalgo Romero. IMSS

G49

MCF-7 cells modify capacitive calcium entry (CCE) by exposure to methanolic extracts of Capsicum annuum L. var. Fascinatum (FAS)

Joel Hurtado Patiño. Universidad Autónoma de Querétaro

G50

Evaluation of the expression of miRNAs contained in extracellular vesicles of patients diagnosed with COVID-19

Nancy Vanessa Iglesias Vázquez. Hospital Regional de Alta Especialidad de Ixtapaluca

G51

Role of flotillin from Phaseolus vulgaris during a mutualistic interaction with Rhizobium tropici

Pamela Jiménez Chávez. Universidad Nacional Autónoma de México

G52

Estradiol, medroxyprogesterone, and camp differentially regulate unfolded protein response genes during decidualization in human immortalized endometrial stromal cells

Miguel Ángel Jiménez Beltrán. Universidad Nacional Autónoma de México

G53

Identification of SNORNAS that interact with lipids using the LIPID-RNA SEQ technique in Saccharomyces cerevisiae

Irma Angélica Jiménez Ramírez. Centro de Investigación Científica de Yucatán A.C.

G54

Unveiling the landscape of LINC00052 molecular mechanisms in breast cancer cells by bioinformatic and experimental analyses

Miguel Ángel Juárez Mancera. Instituto Nacional de Medicina Genómica

G55

Analysis of the interaction network between non-coding RNAs and the transcription factors c-Jun and c-Fos and its effect on the expression of genes that mediate renal hypertrophy

Isaac Virgilio Martínez Cárdenas. Universidad Autónoma de la Ciudad de México

G56

Post-translational regulation of tissue-specific basic Helix-Loop-Helix transcription factors through heterodimerization

Kenya Lizbeth Mena Garcés. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas

G57

In silico and cellular differences associated to the cell division process between the A and B races of the colonial microalga Botryococcus braunii

Xóchitl Morales de la Cruz. Cinvestav Irapuato

G58

Characterization of ABC-type transporters in Candida glabrata

Mariana Montserrat Morales Avila. Instituto Potosino de Investigación Científica y Tecnológica A.C.

G59

Study of promoter activity, subcellular localization, and overexpression of ERULUS during mutualistic interactions

Rogelio Morales Sotelo. Universidad Nacional Autónoma de México

G60

Genomic sequencing of four pathotypes of Colletotrichum lindemuthianum and analysis of virulence genes

Ma. Irene Morelos Martínez. Universidad Michoacana de San Nicolás de Hidalgo

G61

An interplay between histone exchange and tRNA expression impacts cellular proteostasis and aging

Erica Moreno Méndez. Universidad Nacional Autónoma de México

G62

Detection of Human Papillomavirus using PCR in situ in Prostatic Cancer

Guadalupe Morquecho Ramos. Instituto Politécnico Nacional

G63

Regulation of the antifungal alkylresorcinol lipid production by phosphate in Azotobacter vinelandii

Andrea Viridiana Moyao Mejía. Universidad Nacional Autónoma de México

G64

Molecular study of Tau131, subunit of transcription factor TFIIIC, in the human pathogen Trypanosoma brucei

Leonardo Munguía Aguirre. Universidad Nacional Autónoma de México

G65

PEROXIDASE 35 gene is involved in lateral root primordium morphogenesis and is regulated at the epigenetic level

Selene Napsucialy Mendivil. Universidad Nacional Autónoma de México

G66

Differential display of expressed genes of Trichoderma asperellum during growth on PET (Polyethylene terephthalate)

Adriana Guadalupe Orozco García. Universidad de Guadalajara

G67

Study of SYCP3 and its perinuclear accumulations in primary spermatocytes during the rat first spermatogenic wave

Sebastián Pacheco Gutiérrez. Universidad Nacional Autónoma de México

G68

GST, NQO1, and CC16 polymorphisms as potential disease severity biomarkers in influenza-related pneumonia

Margarita Isabel Palacios Arreola. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas

G69

Tripartite Motif Containing 25 is upregulated in glioblastoma

Eva Guadalupe Palacios Serrato. Universidad Autónoma de la Ciudad de México

G70

A comparative study in the regulome of Gallibacterium anatis strain ESV200 against Pasteurellaceae family and E. coli bacteria

Nain Gabriela Pedroza Viveros. Universidad Autónoma de Puebla

G71

Exploring the genetic bases of retrotransposon expression and its impacts on cellular aging

Cesia Daniela Pérez Aguilar. Cinvestav Irapuato

MEDICINE, HEALTH & NUTRITION II

MH32

Inhibition of the early development of fibrosis in chronic kidney disease

Juan Carlos Gallardo Pérez. Instituto Nacional de Cardiología "Ignacio Chávez"

MH33

Etiology, treatment and survival of patients with hepatocellular carcinoma in Veracruz, México

Mariela Judith Domínguez Domínguez. Universidad Veracruzana

MH34

Evaluation of the effect of a guar gum derivative in a murine model of obesity

Ehekatzin García Valdés. Instituto Politécnico Nacional

MH35

The effect of oxLDL on a cellular model of HER2+ breast cancer

Jeshua Johayra Garcia Soberanes. Universidad Autónoma de Baja California

MH36

Effect of two O-GLCNAc transferase inhibitor drugs on VPH-positive cervical cancer cells

Ulises González González. Universidad Autónoma Benito Juárez de Oaxaca

MH37

The microbiota and the production of secondary and tertiary bile acids depend on the type of protein consumed

Omar Granados Portillo. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán

MH38

Mitochondrial expression of Kir5.1 and Kir6.2 channels in the progression of triple-negative breast carcinoma

María del Carmen Gutiérrez Galván. Universidad de Colima

MH39

Nopal and Pirfenidone Improve Body and Liver Weights, and Insulin Resistance in a Mouse Obesity Model with Diethylnitrosamine

Jorge Gutiérrez Cuevas. Universidad de Guadalajara

MH40

Kinetic and structural characterization of small molecules capable of inhibiting Src homology-2-containing protein tyrosine phosphatase 2

Brenda Paola Gutiérrez Soto. Universidad Juárez del Estado de Durango

MH41

Tear proteins as potential biomarkers in retinoblastoma

Jesús Hernández Monge. Universidad Autónoma de San Luis Potosí

MH42

Alterations in the functioning of the heart induced by changes in energy metabolism due to tumoral growth

María de la Luz Hernández Esquivel. Instituto Nacional de Cardiología "Ignacio Chávez"

MH43

Effect of *Ibervillea sonorae* (Wereke) on oxidative stress in the kidney during diabetes

Ana Lilia Hernández Alba. Instituto Nacional de Cardiología "Ignacio Chávez"

MH44

Effect of cancer cells-derived conditioned medium on cardiomyocyte energy metabolism

Fernando Emiliano Jiménez Mondragón. Instituto Nacional de Cardiología "Ignacio Chávez"

MH45

Blackberry juice fermented with two consortiums of lactic acid bacteria: physicochemical and antioxidant properties during storage

Liliana Lugo Zarate. Universidad Autónoma del Estado de Hidalgo

MH46

Effect of Glycine Betaine on P53 and Cas3 Expression in Human Colon Adenocarcinoma Cells HT-29

Lizeth Anahí López Castro Universidad de Sonora

MH47

Evaluation electroacupuncture effect in patients with neuropathy diabetic by genetic markers of CD4 polarization

Carina López Leyva. IMSS

MH48

Maternal polycystic ovary syndrome changes the architecture and cell function of pancreatic islets in mouse descendants

Rosa Isela López Castillero. Universidad Nacional Autónoma de México

MH49

Metabolic and ultrastructural modifications in liver of offspring born of polycystic ovary syndrome mouse model

Erick Gabriel López Cruz. Universidad Nacional Autónoma de México

MH50

Anti-fibrogenic role of IFC-305 in fibroblasts from patients with Idiopathic Pulmonary Fibrosis and its effects on mitochondrial function

Erika Rubí Luis García. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas

MH51

Effect of Callistemon citrinus phytosomes on brain and gut microbiota of High Fat Diet rat model

Oliver Rafid Magaña Rodríguez. Universidad Michoacana de San Nicolás de Hidalgo

MH52

Identification of Endometrial Stem Cell Markers in Menstrual Blood

Gloria Erika Mejia Carmona. Universidad Autónoma de Ciudad Juárez

MH53

Resveratrol and Quercetin Improve Sarcopenic Obesity by Regulating the Angiotensin II/AT1 Pathway and Myostatin Concentrations in a Metabolic Syndrome Rat Model

Jimena Alejandra Méndez Castro. Instituto Nacional de Cardiología "Ignacio Chávez"

MH54

Participation of calreticulin in human fetal membranes

Montserrat Mijares Rodríguez. Instituto Nacional de Medicina Genómica

MH55

Characterization of cardiovascular risk phenotypes and their association with clinical activity and metabolic endotoxemia in systemic lupus erythematosus patients

Paulina Esmeralda Mora Garcia. Universidad de Guadalajara

MH56

Evaluation of a potential signaling pathway involved in the antihypertrophic effect of cannabidiol(CBD) in cardiac myoblasts

Carolina Morales Ochoa. Instituto Tecnológico de Estudios Superiores de Monterrey

MH57

Proteomic analysis of targets deregulated by the miRNAs miR-221, miR-145 and Let-7c involved in prostate cancer

Gabriela Carolina Morales Sandoval. Instituto Politécnico Nacional

MH58

Efectividad del óxido de aluminio sobre la adhesión de bracket en esmalte fluorótico

Anareli Mota Morales. Universidad Juárez del Estado de Durango

MH59

Development of a biopolymer as a system delivery for antidiabetic secondary metabolites from purple sweet potato

Alethia Muñoz Ramírez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

MH60

Characterization of Nrf2 role and modulation effect in a chemoresistant Breast Cancer model

Andrea Muñoz Ayala. Universidad Autónoma de Baja California

MH61

Antilipemic effect of the ethanol fraction of justicia spicigera on the liver of rats fed with a high-fat diet

Marina Murillo Villicaña. Universidad Michoacana de San Nicolás de Hidalgo

MH62

Effect of consuming standardized meals with different macronutrient proportions of metabolic flexibility

Charlotte Olivares Yañez. Universidad Nacional Autónoma de México

MH63

Resistina induce migración e invasión en células de cáncer prostático PC3: Rol de vesículas extracelulares

Mario Israel Oregel Cortez. Universidad Autónoma de Baja California

MH64

Effect of diazoxide and moderate-intensity exercise on the function of mitochondria isolated from skeletal muscle during obesity

Jessica Ortega Pérez. Universidad Michoacana de San Nicolás de Hidalgo

MH65

Antioxidant capacity of pigmented maize from Mexico

Estela Eiko Osawa Martínez. Cinvestav Zacatenco

MH66

Genetic Detection of Dengue Virus Serotypes using the CRISPR-Cas12a System

Marco Antonio Piñón Chávez. Universidad Nacional Autónoma de México

MICROBIOLOGY I

M1

Study of the ABCG transporter family of *Metarhizium guizhouense* HA11-2 during its mycorrhizal association

Víctor Manuel García Vera. Universidad de Guanajuato

M2

Relevance of protein O-glycosylation during the interaction of *Sporothrix schenckii* with the host

Manuela Gómez Gaviria. Universidad de Guanajuato

M3

Killer yeast and glucose mediated synthesis of nanoparticles with biological effect

Carlos Alberto Molina Vera. Universidad Autónoma de Querétaro

M4

Study of the endophytic capacity of entomopathogenic fungi

Jesús Ernesto Ramírez Nieto. Universidad de Guanajuato

M5

Whole genome sequencing of *Candida auris* strain isolated in Monterrey, Mexico

Alí Fernando Ruiz Higareda. Universidad Autónoma de Nuevo León

M6

The wild plant *Conopholis alpina*, and their visiting honey bees (*Apis mellifera*) carry identical bacterial strains under natural conditions

Mary Jose Salas Limón. Universidad Autónoma de Puebla

M7

Effect of Epinephrine and Norepinephrine in the *Gallibacterium anatis* biofilm composition and structure

Alicia Noemi Aguilar Fuentes. Universidad Nacional Autónoma de México

M8

Reversion of antibiotic-resistant phenotype in escape bacteria through actinobacteria secondary metabolites

Vanessa N. Alcántara Garduño. Instituto Politécnico Nacional

M9

Bridging Nutritional Gaps: The Role of Microbiomes in host productivity

Luis D. Alcaráz. Universidad Nacional Autónoma de México

M10

Plant response to the plant growth promoting rhizobacterium *Achromobacter* sp. 5B1 under phosphate and nitrate deprivation

Joseline Suhail Alejo Guerra. Universidad Michoacana de San Nicolás de Hidalgo

M11

Identification through transcriptomics, of genes involved in the swarming of *Pectobacterium brasiliense* BF20 overexpressing *exl1* gene, and its relation to expansin *Exl1*

Gloria Alejandra Altamirano Cruz. Universidad Nacional Autónoma de México

M12

The sweet competition: the bacterial lectin *PirB* vp from *Vibrio parahaemolyticus* and its implication in the colonization of ecological niches

Cassandra Adilenne Alvarado Verdín. Universidad Autónoma de Nayarit

M13

Analysis of diversity in the CRISPR-Cas system of *Cronobacter sakazakii* isolated from food samples

Ana Karen Álvarez Contreras. Instituto Politécnico Nacional

M14

Identification and characterization of contemporaneous clinical isolates of *Candida glabrata*

Oscar Daniel Aranda Moreno. Instituto Potosino de Investigación Científica y Tecnológica A.C.

M15

Identification of molecules with antibacterial activity from different fungal pathogens

Jesús Antonio Arroyo García. Instituto Potosino de Investigación Científica y Tecnológica A.C.

M16

Exploring the impact of glyphosate on orange tree microbiomes: a meta-taxonomic approach

Diana Montserrat Baeza Magaña. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

M17

Phenotypic and Bioinformatic Study of gene *cdgE* of *Azospirillum baldaniorum* Sp245

Nora Georgina Barrera Olvera. Universidad Autónoma de Puebla

M18

Study of the mechanism of control of C-5 alginate epimerases by the second messenger c-di-GMP

Víctor V Barrios Rafael. Universidad Nacional Autónoma de México

M19

Structural models of the phosphatase enzyme and mutants in *Escherichia coli* involved in the synthesis of trehalose and bacterial desiccation

Alejandra Bernabé Allende. Universidad Autónoma de Puebla

M20

Distribution of Huanglongbing in orange fruit producer zone of San Luis Potosí and neighboring states

Karla Grisel Calderón González. Universidad Autónoma de San Luis Potosí

M21

Effect of bacterial cyclodipeptides on gut microbiota composition and nutrient absorption in an obesity model

Marlene Estefania Campos Morales. Universidad Michoacana de San Nicolás de Hidalgo

M22

Gene of no orthodoxy multisensor histidine kinase is involved in motility type swarming and swimming in the rhizobacterium *Azospirillum baldaniorum* Sp245

Uriel Cardona Baltazar. Universidad Autónoma de Puebla

M23

Study of the histidine-kinase *RetS* as part of the multi-kinase system *GacS* (MKN-*GacS*) in *Azotobacter vinelandii*

Miguel Castañeda Lucio. Universidad Autónoma de Puebla

M24

Isolation and biochemical identification of *Lactobacillus* spp. extracted from products of animal origin

Azul Castrillo Sanchun. Universidad Autónoma Metropolitana Xochimilco

M25

Genetic study of melanin synthesis in *Azotobacter vinelandii*

María Fernanda Chávez Jacobo. Universidad Autónoma de Puebla

M26

Phenotypic and genomic characterization of *Streptomyces pakalii* sp. nov., a novel species of Mexican actinomycete with anti-biofilm and anti-quorum sensing activity in ESKAPE bacteria

Michelle A. Chávez Hernández. Instituto Politécnico Nacional

M27

Expression of Specialized Metabolites by an OSMAC Approach Applied to *Streptomyces*

Uriel Colin Camacho. Universidad Nacional Autónoma de México

M28

Bacteria from the skin microbiota of the axolotl *Ambystoma altamirani* show growth-inhibitory activity against antibiotic-resistant strains of *Staphylococcus aureus*

Cynthia Margarita Concepción Acosta. Universidad Nacional Autónoma de México

M29

Cloning and overexpression of *fliC* gene from *Azospirillum argentinense* REC3 in the expression vector pGEX-4T1

Ana Gabriela Cordova Mejía. Universidad Autónoma de Puebla

M30

Impact of the over expression of a gene codifying for a hybrid C-DI-GMP regulating protein of *Azospirillum baldaniorum* SP245

David Ricardo Cortés Sotres. Universidad Autónoma de Puebla

M31

Study of the small RNA regulator (sRNA) *ErsA* involved in the synthesis of alginates in *Azotobacter vinelandii*

Juan Carlos De Lima Mar. Universidad Autónoma de Puebla

M32

The loss-of-function of the methyltransferase *KMT6* on *Trichoderma atroviride* modifies the expression of secondary metabolite gene clusters that compromise the root gravitropism of *Arabidopsis*

Saraí Esparza Reynoso. UGA. LANGEBIO. Cinvestav Irapuato

M33

The complex regulation of sigma factor *RpoS* proteolysis in *Azotobacter vinelandii*

Elda Guadalupe Espín Ocampo. Universidad Nacional Autónoma de México

M34

Symbiosis induction between *Rhizobium etli* and *Saccharomyces cerevisiae*

Ilse Berenice Espinoza Hernández. Universidad Nacional Autónoma de México

M35

The administration of a recombinant Tepary bean (*Phaseolus acutifolius*) lectin affects rat bacteriome

Juan Brandon Araujo Mendoza. Universidad Autónoma de Querétaro

M36

Isolation of pathogens from food and assessment of their antimicrobial resistance

Maricela Esteban Méndez. Instituto Politécnico Nacional

M37

Isolation and Characterization of Spoilage Molds of Strawberry (*Fragaria x ananassa* Duchense) marketed in Durango

Cinthia Andrea Estrada Rodríguez. TecNM/Instituto Tecnológico de Durango

M38

Effects of chitosan on growth of fungal species involved in building biodeterioration

Joshua Fernandez Jasso. Instituto Politécnico Nacional

M39

Analysis of blaOXA-72 gene regulatory region in *Acinetobacter baumannii*

Diana Flores Percino. Universidad Autónoma de Puebla

M40

Effect of Iron on the Growth and Virulence of *Pseudomonas syringae* pv. *phaseolicola* NPS3121

Kenia Franquez Gómez. Universidad Autónoma de Nayarit

REACTIVE OXYGEN SPECIES

ROS1

Determination of Antioxidant Activity and Phenolic Compounds in Fermented Cocoa Bean Shell Infusion

Sebastián Cervera Pereyra. Universidad Juárez Autónoma de Tabasco

ROS2

YwqN Encodes a NADP(H)/FMN-Dependent Quinone Reductase That Protects *Bacillus subtilis* from Oxygen Radical Genotoxicity

Karen Abundiz Yáñez. Universidad de Guanajuato

ROS3

Enzymatic antioxidant response during physiological cardiac hypertrophy induced by pregnancy

José Bruno Apodaca Fuentes. Universidad de Sonora

ROS4

Effect of vitamin C on renal markers of oxidative stress induced by methotrexate in psoriasis

Elodia Nataly Diaz De la Cruz. Universidad Michoacana de San Nicolás de Hidalgo

ROS5

Effect of potential hydrogen on hydrogen sulfide production in *Saccharomyces cerevisiae*

Mariana Michell Garcia Reyes. Universidad Nacional Autónoma de México

ROS6

Effect of Oxidative Stress on the mRNA Levels of Il-1 β , Il-6, and Il-10 in the Intestine of Rats Exposed to Ozone

Denisse García Sánchez. Universidad Nacional Autónoma de México

ROS7

Hydrogen peroxide detoxification through of the thioredoxin system in the cysticerci of Taenia

Alberto Guevara Flores. Universidad Nacional Autónoma de México

ROS8

Partial purification and biochemical characterization of thioredoxin reductase (TrxR) from the insect Shelfordella tartara

Iván Emilio Ibarra Estrada. Universidad Nacional Autónoma de México

ROS9

Antioxidant effect of the water: methanol fraction of the ethyl acetate extract of Potentilla indica on renal mitochondria of rats with type 2 diabetes mellitus

Cinthia Itzel Landa Moreno. Universidad Michoacana de San Nicolás de Hidalgo

ROS10

Assessment of oxidation markers, MAPK activation and their potential association in cervical cancer

Karen Andrea Larrauri Rodríguez. Universidad Autónoma de Puebla

ROS11

Cyanidin effects on oxidative stress and mitochondrial biogenesis in PAE cells, during hypoxia-reoxygenation

Melani León Martínez. Instituto Nacional de Cardiología "Ignacio Chávez"

ROS12

Characterization of the cytosolic and mitochondrial antioxidant system of Ustilago maydis under oxidative conditions

Aranzazú López Jaime. Facultad de Ciencias Biológicas. BUAP

ROS13

Effect of selenium supplementation on asexual reproduction of Taenia crassiceps cisticerci

José de Jesús Martínez González. Universidad Nacional Autónoma de México

ROS14

Exploratory analysis of antioxidant and detoxification enzymatic systems in Dendroctonus genus (Coleoptera Curculionidae)

José de Jesús Martínez González. Universidad Nacional Autónoma de México

ROS15

8-OxoG Leads to Stress Survival and Evolution in Bacillus subtilis

Lissett Esther Martínez Magaña. Universidad de Guanajuato

ROS16

Evaluation of the antioxidant activity of the damage suppression protein (Dsup) of the tardigrade *Ramazzottius varieornatus*

Mariel Alexis Quiven Feria. Cinvestav Zacatenco

ROS17

Resveratrol as an antioxidant potential in the regulation of oxidative stress in the prefrontal cortex during aging in Wistar rats

Raúl Vladimir Ramírez Olmos. Universidad Autónoma de Puebla

ROS18

Elucidation of a *DisA*-independent checkpoint mechanism during germination/outgrowth of *Bacillus subtilis* spores

Alejandra Rangel Mendoza. Universidad de Guanajuato

ROS19

In vitro* studies of iron chelating capacity and antioxidant activity of the ethanolic extract of *Eryngium carlinae

María de los Ángeles Rangel Rosales. Universidad Michoacana de San Nicolás de Hidalgo

ROS20

Effect of PCL/F-68 nanoparticles loaded with curcumin on the viability of *Taenia crassiceps Cysticerci* (CESTODA)

Lucero Reyes García. Universidad Nacional Autónoma de México

ROS21

Evaluation of antioxidant capacity of various *Moringa oleifera* based herbal products using cytochrome C

Carlos Alejandro Rocha Martínez. Universidad Juárez del Estado de Durango

ROS22

Differential changes in redox state induced by senolytic and senomorphic treatments in the serum of middle-aged female rats during chronic obesity

Verónica Salas Venegas. Universidad Autónoma Metropolitana Iztapalapa

ROS23

Evaluation of the photo-protection and repair effects exerted by the carotenoids (lutein, zeaxanthin and β -carotene) against photodamage caused by UV radiation in human keratinocytes

Judith Guadalupe Santos Ceseña. Universidad Autónoma de Querétaro

ROS24

Xanthine Oxidase-Regulation of 8-OxoG-Promoted *Bacillus subtilis* Fitness

María José Toledo Ramírez. Universidad de Guanajuato

ROS25

Partial purification of thioredoxin reductase (TrxR) from *Dendroctonus valens*

César Vásquez Lima. Universidad Nacional Autónoma de México

SYSTEMS BIOLOGY & BIOINFORMATICS II

SB27

Comparative transcriptomics of the pharmacological response to the compound AZD5363 in two human ductal adenocarcinoma cell lines

Rebeca González Ortiz. Universidad Nacional Autónoma de México

SB28

Plant thermomorphogenesis: protein-protein interaction and gene coexpression networks in *Arabidopsis thaliana*

Fernanda Lizeth Gutiérrez Palomeque. Instituto Tecnológico Superior de Irapuato

SB29

Searching for structural transitions in a parallel protein network

Juan Martín Hernández Castillo. Universidad Nacional Autónoma de México

SB30

In silico study of galanin receptors as potential targets for the treatment of depression

Beatriz Hernández Estrada. Universidad Autónoma de Querétaro

SB31

Detection of transcriptional determinants of cellular longevity in human tissues using a machine learning-based approach

Jorge Adrián Islas Ortiz. INMEGEN. Universidad Nacional Autónoma de México

SB32

Search for selective inhibitors on PTP1B of peptide nature

Rodolfo Aarón Lizárraga Valadez. Universidad Nacional Autónoma de México

SB33

Identification of molecular markers, SNPs, associated with morphological characteristics of *Bixa orellana* L.

Ana Lucía López Gurgua. Centro de Investigación Científica de Yucatán A.C.

SB34

Molecular Dynamics and Docking Analysis of Quercetin and Patuletin of Evaluating Antitumor Activity

Aarón Saúl López Carrasco. Universidad Nacional Autónoma de México

SB35

Modeling Krebs cycle from rat liver, heart, and hepatoma mitochondria

Álvaro Marín Hernández. Instituto Nacional de Cardiología "Ignacio Chávez"

SB36

Prediction of the structural effect of variants of uncertain significance detected by NGS in Mexican children with B-cell acute lymphoblastic leukemia

Daniel Alejandro Martínez Anaya. Instituto Nacional de Pediatría

SB37

Vector capacity of Aedes aegypti through the horizontal infection mechanism of the dengue larva-larva virus

Mayra Mejía Sánchez. Universidad Autónoma de Sinaloa

SB38

Unveiling the Plastic-Degrading Potential of Eukaryotes: A Computational Approach

Andrés Méndez Zamora. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

SB39

Role of the PAS Domain in the activity of Diguanylate Cyclase or Phosphodiesterase of the CdgD Protein in Azospirillum baldaniorum Sp245

Dulce Andrea Montes Pérez. Universidad Autónoma de Puebla

SB40

Exploring the therapeutic potential of berry cactus in rat microbiota composition with Metabolic Syndrome

Cesaré Ovando Vázquez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

SB41

Evolution of hypoxia response factors in the Plantae kingdom

Janet Palacios Martínez. Universidad Nacional Autónoma de México

SB42

Prediction of the three-dimensional structure of the Nrg1-Rtg3 chimeric complex

Edgar Adrián Ramírez González. Universidad Nacional Autónoma de México

SB43

PhaZ as a possible PET degrader?

Graciela Jesed Reyes Trujillo. Universidad Autónoma de Puebla

SB44

Analysis of the interaction between TMPRSS2, ACE2, and furin against spike of SARS-COV-2

Víctor Rodríguez Escobar. Universidad Autónoma Benito Juárez de Oaxaca

SB45

Machine learning prediction of bacterial-phage specificity using genomic fingerprints obtained by virtual hybridization

Juan Antonio Rodríguez de la Cruz. Instituto Politécnico Nacional

SB46

Structural analysis of SARS-CoV-2 Mpro protease variants for the discovery of broad-spectrum inhibitory drugs using in silico tools

Carlos Alfonso Sánchez López. Instituto Tecnológico Superior de Irapuato

SB47

In silico analysis of the binding of the OppA protein of Yersinia pseudotuberculosis with different lipases

Andrea Sánchez Ríos. Universidad Autónoma de Puebla

SB48

AI Revolutionizing Water Quality: Harnessing Microalgae and Bacteria for Sustainable Remediation

Mishael Sánchez Pérez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

SB49

Differential molecular interactions between iberiotoxin and human SLO3 and SLO1 potassium channels

Jorge Arturo Torres Juárez. Universidad Autónoma de Querétaro

SB50

Microbial Diversity Analysis via Metagenomics in 'El Iztete' Public Landfill, Tepic Nayarit

Jesús Alberto Urbar Ulloa. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

SB51

Comparative genomic analysis of cuticular biosynthesis in fleshy fruits

José Alberto Valenzuela Avilés. Centro de Investigación en Alimentación y Desarrollo, A.C.

SB52

Unlocking nitrogen's role during somatic embryogenesis induction of C. canephora: A transcriptomic view

Brigitte Valeria Vargas Morales. Centro de Investigación Científica de Yucatán A.C.

POSTER SESSION III
WEDNESDAY, OCTOBER 23
17:00 - 19:00

BASIC BIOCHEMISTRY II

BB38

Characterization of sphenotoxin from ophryacus sphenophrys and its comparison with classic crotoxin

Tania Corkidi Zajur. Universidad Nacional Autónoma de México

BB39

The methionine synthase in plant development

Sara Margarita Garza Aguilar. Tecnológico de Monterrey

BB40

Tips to increase the thermal stability of a recombinant protein: the practical case of CGI-58

Miriam Livier Llamas García. Instituto Potosino de Investigación Científica y Tecnológica A.C.

BB41

Biochemical characterization of orotate phosphoribosyltransferase from the phytopathogen *Pseudomonas cichorii*

Amelia López Albores. Instituto Tecnológico de Tuxtla Gutiérrez

BB42

Aldosterone effects on the expression of ryanodine receptors and their participation in the calcium dynamics of rat mesenteric arteries

Hiram Lozano Ruiz. Cinvestav Zacatenco

BB43

Subcellular localization plays a determinant role on the functional diversification of the paralogous proteins *BAT1* and *BAT2*

Yamile Paredes Chiquini. Universidad Nacional Autónoma de México

BB44

Hinges, springs, and sticky surfaces determine the preferred conformations of Cre in the absence of loxP and prime it for binding

Nina Pastor Colón. Universidad Autónoma del Estado de Morelos

BB45

Characterization of Human Epidermal Growth Factor (hEGF)-induced Ca²⁺ release in HeLa Cells

Amanda Renté Alpizar. Cinvestav Zacatenco

BB46

Impact of drought stress and recovery irrigation on amino acid accumulation and antioxidant activity in Mexican soybean

Julio César García Rodríguez. Cinvestav Irapuato

BB47

Molecular interaction between P53, P21 and biomolecules present at Manilkara zapota seeds

Vanessa García Alcudia. Universidad Juárez Autónoma de Tabasco

BB48

Cloning, Expression and Purification of Recombinant Hemagglutinins from Avian Influenza Virus Type A

Margarita Fernanda Gómez Meza. Instituto Nacional de Pediatría

BB49

Relationship between maize cell cycle-related regulators and the repair protein RAD51A

Estefany Damaris Guerrero Molina. Universidad Nacional Autónoma de México

BB50

Exploring the structural and functional changes of Vibrio cholerae pyruvate kinase

Gloria Hernández Alcántara. Universidad Nacional Autónoma de México

BB51

In vitro study of physicochemical properties of novel voltage-gated potassium ion channel blockers based on 4-Aminopyridine

Karen Sofía Hernández Medina. Universidad de Guadalajara

BB52

Structure and phylogeny of crotoxin subunits A and B

David Hernández Herrera. Instituto Politécnico Nacional

BB53

Evaluation of the DNA gyrase inhibitory effect of natural products and derivatives with known anti-Helicobacter pylori activity

Erick Hernández Hipólito. Universidad Nacional Autónoma de México

BB54

Red and Infrared light as an enhancer for mitochondrial respiration of keratinocytes cells

Manuel Alejandro Herrera López. Universidade De São Paulo

BB55

Study of peripheral domains in structure-function of isocitrate lyase (ICL) from Pseudomonas aeruginosa

Mildred Jiménez Báez. Universidad Michoacana de San Nicolás de Hidalgo

BB56

Purification and characterization of chimeric recombinant variants of human deubiquitinase USP2 in relation to its specificity for ubiquitin K63 chains

Josahandy Jiménez Peralta. Universidad Nacional Autónoma de México

BB57

Effect of growth temperature on catalase activity of *Rhodococcus equi*

Francisco Javier Juárez Castañeda. Universidad Juárez del Estado de Durango

BB58

The glucose sensor HXK1 is involved in the cold acclimation and freezing survival of *Arabidopsis thaliana* plants

Beatriz King Díaz. Universidad Nacional Autónoma de México

BB59

A novel central metabolism control by cell cycle

Aurora Lara Núñez. Universidad Nacional Autónoma de México

BB60

Study of the cell death induced by the Iztli peptide 1 in *Saccharomyces cerevisiae* reveals an unknown genetic connection between arrest, metabolism and mating

Ma. Teresa Lara Ortiz. Universidad Nacional Autónoma de México

BB61

Studies on the physiological role of the putative gentisaldehyde dehydrogenase from *Pseudomonas aeruginosa*

Alexey Llopiz. Universidad Nacional Autónoma de México

BB62

Characterization of the Cdk subunit regulators TvCKS1 and TvCKS2 in *Trichomonas vaginalis*

Karla Concepción López Pacheco. Universidad Nacional Autónoma de México

BB63

A Seasonal Proteomic Study of *Centruroides exilicauda* venom from Baja California, México

Joaquín López Carrillo. Centro de Investigación Científica y de Educación Superior de Ensenada

BB64

Mitochondrial iron metabolism is disrupted in a murine model of liver steatosis

Luis Alberto Luévano Martínez. Tecnológico de Monterrey

BB65

Peptide VSAK derived from the C-terminal region of CETPI, attenuates cellular responses associated with LPS

Ismael Luna Reyes. Universidad Nacional Autónoma de México

BB66

A computational and experimental approach to develop a FRET-based substrate for assessing the proteolytic activity of Mycobacterium tuberculosis MarP

Pablo Alfonso Madero Ayala. Universidad Autónoma de Baja California

BB67

Effect of a triazaspiran-type molecule on migration and invasion of MDA-MB-231 tumor cells

Jesús Omar Medina Cruz. Universidad Autónoma de Baja California

BB68

Wolbachia spp. infection in Drosophila melanogaster

Ofelia Mendez Romero. Universidad Nacional Autónoma de México

BB69

Phenolic Profiling of Native Chromatic Corn Varieties

Liliana Hortencia Méndez Barredo. Instituto de Ecología, A.C.

BB70

Pitavastatin: Its Potential Adverse Effect Evaluate in Hypercholesterolemic CD-1 Male Mice

Rodrigo Miranda Zamora. Universidad Nacional Autónoma de México

BB71

Analysis of fatty acid transport mediated by the MCT11 transporter

Nicole Justine Moreno Licona. Cinvestav Zacatenco

BB72

Characterization of the roles PRIMPOL in DNA integrity maintaining in Arabidopsis thaliana

Johan Olalde Hernández. Universidad Nacional Autónoma de México

BB73

Metabolic factors associated with metabolically healthy obesity

Marisol Olivares Arévalo. Universidad Nacional Autónoma de México

BB74

A new ER Ca²⁺ release agent synthesized from 2-APB tested in HeLa cells

David Olvera Rangel. Cinvestav Zacatenco

BB75

Induction of systemic resistance in Arabidopsis against phytopathogens by the beneficial fungus Trichoderma atroviride through modulation of serine biosynthesis

Daniel David Pacheco Rodríguez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

BB76

Alpha amylase inhibition by silver nanoparticles biosynthesized with Cnidocolus aconitifolius

Eduardo Padilla Camberos. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BB77

Proteomic analysis of the response to gibberellic acid in Capsicum annuum

Diego de Jesús Pantoja Gutiérrez. Instituto Tecnológico de Celaya

BB78

Modulation of the Hexosamine Biosynthetic Pathway and O-GlcNAcylation Influences the Localization of the CD36 Receptor in Macrophages

Yobana Pérez Cervera. Universidad Autónoma Benito Juárez de Oaxaca

BIOTECHNOLOGY III

BT101

In vitro assisted refolding of the recombinant Trypanosoma cruzi antigen TSA-1

Esdras Enoc Pacindo Cabrales. Cinvestav Zacatenco

BT102

Antifungal Effect of Clove Ethanol Extract (Syzygium aromaticum) on the in vitro and biochemical inhibition of Phytophthora sp

Adolfo Padilla Mendiola. Universidad Juárez del Estado de Durango

BT103

B. licheniformis α -amylase engineering guided by machine learning-coupled directed evolution to introduce transglucosidic activity

José Alberto Parra Espinoza. Universidad Nacional Autónoma de México

BT104

Purification and characterization of cytotoxic molecules from a LIPID-RICH extract from mexican avocado seed (Persea americana var. drymifolia)

María Guadalupe Pérez Esquivel. Universidad Michoacana de San Nicolás de Hidalgo

BT105

Study of an isolated Penicillium sp that uses plastic polymers as carbon source

Martha Lizeth Pérez Méndez. Universidad de Guanajuato

BT106

Evaluation of the interaction between bacteria and photobionts isolated from lichens

Britany Irlanda Pérez Barrón. Instituto Tecnológico Superior de Irapuato

BT107

Impact of phosphite on gene expression in the filamentous fungus *Trichoderma atroviride*

Camilo Pérez Salazar. UGA-LANGEBIO. Cinvestav

BT108

Humanization of Chimeric Anti-CD20 Antibody Rituximab via CDR-Grafting

Daniel Jesús Pérez Vega. Instituto Politécnico Nacional

BT109

Design and generation of a genetic construction for ectopic expression of the *ArsR C104S* mutant repressor in *Bacillus subtilis* for organic arsenic detection

Miguel de Jesús Piña Mena. Universidad Juárez del Estado de Durango

BT110

Metabolic changes in HTB 177 lung cancer cells in response to the anticancer extract of *Coleus hadiensis*

Karla Ramírez Estrada. Universidad Autónoma de Nuevo León

BT111

Muscle growth promotion by a chimeric myostatin immunogen delivery by an antigen display and gene delivery baculovirus vector

Vianey Ramírez Andoney. Universidad Nacional Autónoma de México

BT112

One way to evolve C family DNA polymerases

Andrea Ramón García. Universidad Autónoma del Estado de Hidalgo

BT113

Isolation and characterization of native strains from Lithium-Containing mining tailings

Kevin Samuel Renteria Ortiz. Instituto Tecnológico de Durango

BT114

Modifying the product specificity by protein engineering of an archaeal cyclomaltodextrin glucanotransferase from a deep-sea hydrothermal vent

Ana María Rincón Murillo. Instituto Tecnológico de Colima

BT115

Production of a β -1,4-Endoglucanase enzyme from *Bacillus subtilis* in *Saccharomyces cerevisiae*

Joel Rios Alvarado. Instituto Tecnológico de Durango

BT116

Facile Synthesis of PEGylated Fe₀ Au heteronanoparticles, as high potential biomedical use

Maria Ana Rivera Soto. Universidad Autónoma de la Ciudad de México

BT117

Isolation and functional characterization of collagen from fish scales

Crisalejandra Rivera Pérez. Centro de Investigaciones Biológicas Del Noroeste S.C.

BT118

Newest insights into bixin biosynthesis in achiote, *Bixa orellana* L.

Renata Rivera Madrid. Centro de Investigación Científica de Yucatán A.C.

BT119

Methodological strategies for biosynthesis and characterization of iron nanoparticles using *Trichoderma harzianum*

Ángeles Alitzel Rivera Román. Universidad Autónoma del Estado de Hidalgo

BT120

Treatment of dairy wastewater using natural coagulants derived from agro-industrial wastes

Ana Rosa Rocha Vallejo. Centro de Innovación Aplicada en Tecnologías Competitivas

BT121

Soluble expression of chagasin chimeras harbouring four TSA-1 epitopes in *Escherichia coli*

Marco Ibrim Rodríguez Sánchez. Cinvestav Zacatenco

BT122

Modification of nopal pectin with quaternary ammonium salts to give it antibacterial, anticancer and biocompatibility properties

Tania Milena Rodríguez Amaya. Universidad Autónoma de Coahuila

BT123

Study of interaction mechanisms between *Pleurotus ostreatus* and heavy metals (chromium and lead) in cultivation media primarily composed of lignocellulosic residues

Pamela Romo Rodríguez. TecNM Campus Pabellón de Arteaga

BT124

Humanization of an IgE antibody: recombinant expression and strategy to recover affinity towards its allergen

Edgard David Rosas Ramírez. Instituto Politécnico Nacional

BT125

Improving xylose metabolism in *Saccharomyces cerevisiae*

Jaime Rosas Diaz. Universidad Nacional Autónoma de México

BT126

Evaluation of the interaction between lichenic bacteria and non-lichenic fungi

Julieta Rosas Vallejo. Instituto Tecnológico Superior de Irapuato

BT127

Antifungal activity of *Erigeron canadensis* and its establishment of callus in vitro culture

Andrea Ruiz Betancourt. Universidad Autónoma Metropolitana Iztapalapa

BT128

In vitro antifungal activity of ethanolic extracts from leaf and stem residues of Mexican oregano (*Lippia graveolens* Kunth) on *Fusarium* spp.

Estela Ruiz Baca. Universidad Juárez del Estado de Durango

BT129

SHINE3 and MYB31 transcription factors regulate cuticle biosynthesis and cell wall remodeling during soursop (*Annona muricata*) fruits ripening

Héctor Adán Ruiz Ortega. Centro de Investigación en Alimentación y Desarrollo, A.C.

BT130

Immobilization of α -amylase for alkyl-glucosides production

Gloria Saab Rincón. Universidad Nacional Autónoma de México

BT131

Identifying bacteria with cutinase activity for the degradation of plastic from the Tepetitlic Lagoon

Magdiel Salas Montenegro. Universidad Autónoma de Nayarit

BT132

Analysis of cell migration and proliferation in exposure to mefloquine on the MDA-MB-231 cell line

Andrey Salazar Mendoza. Universidad Autónoma de Baja California

BT133

Neutralization of the neurotoxin activity of Alpha-latrotoxin from the black widow spider using monoclonal antibodies

Fátima Sánchez Ramírez. Universidad Nacional Autónoma de México

BT134

Phytotoxicity and genomic instability of the interaction using nanoparticles with nitrogenated fertilizers in the In vivo Allium Cepa and Capsicum Model

Alejandro Sánchez González. Universidad Autónoma de Baja California

BT135

Development of an edible coating and film from carboxymethylcellulose, mesquite GUM, and a dairy industry waste product

Maria Cristina Sánchez Aguirre. Universidad Autónoma de Coahuila

BT136

Characterization of protease enzymes from the ZH2 strain of the genus *Geobacillus* spp.

Eric Alejandro Sánchez Rosas. Instituto Politécnico Nacional

BT137

GMO Study Landscape: Content, Citation, and Geography

Cinthia Valentina Soberanes Gutiérrez. Consejo Nacional de Humanidades, Ciencias y Tecnologías

BT138

Search for a DNA aptamer to identify Acinetobacter baumannii using the cell-SELEX technique

Emili kenya Solís López. Universidad Autónoma de la Ciudad de México

BT139

Production In silico of new cancer immunotherapeutic chimeric protein designed with PD-1 IgV-domain-Fc

Eva Cristina Sosa Sillas. Universidad Autónoma de Sinaloa

BT140

Antioxidant and antimicrobial evaluation of in vitro cultures of Ludwigia octovalvis (Jacq.)

Raven Stephany Abigail Tadeo. Cuenca Universidad Autónoma Metropolitana

BT141

Evaluation of the implementation of the CRISPR-Cas9 system for the modification of the ATP6 gene of Paracoccus denitrificans

Sergio Aarón Tinajero Vargas. Cinvestav Zacatenco

BT142

Screening method for the directed evolution of a fungal peroxygenase able to catalyze the oxidation of emerging pollutants

Alina Elizabeth Torres Aguirre. Universidad Nacional Autónoma de México

BT143

Design of a PCR-RFLP for malaria genomic surveillance in Mexico

Ramón Gabriel Trejo. Sánchez. Instituto Politécnico Nacional

BT144

Proteomic an metabolite profile during the germination of amaranth seeds (Amaranthus spp.)

Fernando Daniel Urbieto Villalobos. Instituto Potosino de Investigación Científica y Tecnológica A.C.

BT145

Generation and characterization of albumin-cystatin microparticles for the control of plant pathogens

Silvia Edith Valdés Rodríguez. Cinvestav Irapuato

BT146

Exploring Fluorescent Protein iLOV as Versatile Intragenic Reporter in Capsicum annum L

Lilian Gabriela Valencia Turcotte. Universidad Nacional Autónoma de México

BT147

Estimation of arsenic concentration in drinking water wells using the As BIOSENSOR BsWCBgfpmut3a

Luz Idalia Valenzuela García. Centro de Investigación en Materiales Avanzados

BT148

Design and generation of a genetic construction for the disruption of the Bacillus subtilis arsR gene

Evelin Aylin Valenzuela Villalobos. Universidad Juárez del Estado de Durango

BT149

Importance of understanding the correlation between chronic inflammation, the immune response and membrane remodeling, as a restorative treatment strategy

Alba Adriana Vallejo Cardona. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BT150

Elucidation of lactose metabolism in Actinobacillus succinogenes 130Z

Nadia Mariela Varela Pérez. Universidad Nacional Autónoma de México

BT151

Revaluation of metzal residue as a texturizing agent in the vegetative development of lettuce (Lactuca sativa L.)

Carlos Getzael Vazquez Osorno. Universidad Autónoma del Estado de Hidalgo

BT152

Lipid and carotenoid producing yeast recovered from urban wastewater: an evaluation of their biotechnological potential

Dana Lorena Vázquez de los Reyes. Universidad Nacional Autónoma de México

BT153

Immunoproteomic Identification of a Type 1 Fimbriae in Klebsiella pneumoniae

Ulises Vega Castillo. Universidad Autónoma de Sinaloa

BT154

Evaluation of the effect of RNAi silencing on the putative Hyaluronic Acid Synthase (HAS) gene of Mucor lusitanicus

Claudia Cecilia Vega García. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C.

BT155

Evaluation of the microbiological and enzymatic susceptibility of acrylic hydrogels crosslinked with lignin-modified

Gilberto Velázquez Juárez. Universidad de Guadalajara

BT156

Determination of parameters for low-frequency electromagnetic stimulation of MDA-MB-231 cells in vitro

Fernando Verduzco Valdez. Universidad Autónoma de Baja California

BT157

Effect of vaccine HB-ATV-8 on cell signalling in a 3D in vitro model of atherosclerosis

Gloria Stephanie Villa Jaimes. Universidad Nacional Autónoma de México

BT158

A Novel cis-Element as a Potential Regulator in Begomovirus Complementary Strand Synthesis

Daniel A. Mendoza Magaña. Universidad de Colima

GENETICS, EPIGENETICS AND GENETIC REGULATION III

G72

The role of Arabidopsis sRNA2 and sRNA3 and their potential target At_IncRNA1 in establishing a mutualistic relationship with Trichoderma

Kumari Rashmi. Instituto Potosino de Investigación Científica y Tecnológica A.C.

G73

Evolving the theory of Hypermobility Ehlers-Danlos Syndrome from connective tissue disease to a mechanosensing pathology

Ishar Arad Retana Arellano. Universidad Nacional Autónoma de México

G74

Identification of differential expressed microRNAs in gastric carcinogenesis

Diego Reyna Colín. Universidad Autónoma Metropolitana Cuajimalpa

G75

Long Non-coding RNA LINC00115 in response to 5-FU pharmacological treatment in colorectal cancer

Lil Rodríguez Aboy. Universidad Nacional Autónoma de México

G76

TFIIIC subunit Tau131 in Trypanosoma brucei: identification of its protein interactors

Mariani Guadalupe Rodríguez López. Universidad Nacional Autónoma de México

G77 Evolution of the early recombinosome in plants

Arnaud Ronceret. Universidad Nacional Autónoma de México

G78

Epigenetic changes in the GAPDH gene during hypoxia and reoxygenation stress in Penaeus vannamei

Jesús Adelmo Rosas Anaya. Centro de Investigación en Alimentación y Desarrollo, A.C.

G79

rs2070935, rs2289671 and rs17027 variants of GFAP and their association with neurological sequelae of COVID-19

Alma Cristina Salas Leal. Universidad Juárez del Estado de Durango

G80

Modulation of the Killer effect by replacing K⁺ with Na⁺ in the culture medium

Jorge Salinas Celestino. Universidad Autónoma de Querétaro

G81

Role of the oncogene BORIS (CTCF1) in transcriptional regulation of the VEGFA gene promoter

Ana Sofía Salmerón Oms. Universidad Autónoma Metropolitana Cuajimalpa

G82

Transcriptional evaluation of key genes involved in the tissue regeneration process of Ambystoma mexicanum

Cynthia G. Sámano Salazar. Universidad Autónoma Metropolitana Cuajimalpa

G83

LncRNAs landscape during the hypoxia-induced vasculogenic mimicry in breast cancer cells

Gricelda Sánchez Sánchez. Universidad Autónoma de la Ciudad de México

G84

CXCR4 splicing isoform expression in response to typical Hypersensitivity Pneumonitis signaling

Daniela Fernanda Sánchez Sánchez. Instituto Nacional de Enfermedades Respiratorias
Ismael Cosío Villegas

G85

miR-193b and its possible influence over DDR genes through ANRIL regulation in triple-negative breast cancer-derived cell lines

Julio Alejandro Sánchez Pérez. Universidad Nacional Autónoma de México

G86

Role of CREBA and CREMT in the regulation of the murine CATSPER3 promoter

Diego Eduardo Sánchez Jasso. Cinvestav Zacatenco

G87

Effect of Panobinostat (LBH589) on gene acetylation regulation of Ca²⁺ signaling during epithelial-mesenchymal transition in the breast MCF10A cell line

Ana Cecilia Sánchez Trujillo. Universidad Nacional Autónoma de México

G88

Identification of Post-transcriptional Regulatory Sequences ERE-Type in Cancer-Related Genes

César Augusto Sánchez Bocanegra. Universidad Autónoma de Querétaro

G89

Telomere and Subtelomere Organization in Ustilago maydis Chromosomes

Adriana Inclán Popoca. Universidad Autónoma de Puebla

G90

Study of mitochondrial DNA variants associated to breast cancer development

Jessica Jazmin Saucedo Rivas. Instituto Nacional de Medicina Genómica

G91

Is there a conserved machinery of executive function evolution and development across mammalian species? A comparative transcriptomics perspective

María Fernanda Shehin Mina. INMEGEN. Universidad Nacional Autónoma de México

G92

Solanum lycopersicum cv. Micro-Tom as a tool to characterize carotenoid cleavage dioxygenases from Bixa orellana L.

Diana Laura Sierra Ulín. Centro de Investigación Científica de Yucatán A.C.

G93

Epigenetic characterization of ADAMTS17 gene promoter during tissue regeneration in Ambystoma mexicanum

Violeta Guadalupe Silva Díaz. Universidad Autónoma Metropolitana

G94

Effect of pharmacological inhibition of Crml on cardiac dysfunction in a progeria mouse model

Angélica Soberano Nieto. Cinvestav Zacatenco

G95

Study of the signaling pathways that maintain the DNA integrity in mitochondria and chloroplast in plants

Diana Solano Arguello. Universidad Nacional Autónoma de México

G96

Transcriptional contribution of a human heterodimeric basic Helix-Loop-Helix Transcription Factor when isolated from the human cell context

Ana Lilia Torres Machorro. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas

G97

Detection of fusion genes in patients diagnosed with acute lymphoblastic leukemia

Ruth Trejo Martínez. Universidad Autónoma de San Luis Potosí

G98

Autophagy characterization during aging in human dermal fibroblasts

Juan José Unzueta Mendoza. Universidad Autónoma Metropolitana Iztapalapa

G99

Mechanosensing and inflammation in Premature Rupture of Membranes

Lourdes Vadillo Pérez. INMEGEN. Universidad Nacional Autónoma de México

G100

Analysis of the DNA methylation pattern of the SFN gene promoter in early stages of the K14E7 murine model

Luz del Carmen Valerio Jácome. Cinvestav Zacatenco

G101

Use of Neural Networks for the Analysis and Classification of Gliomas through their Epigenetic Patterns

David Valle García. Instituto Nacional de Neurología y Neurocirugía Manuel Velasco Suárez

G102

Genetic variant rs2074192 of the ACE2 gene and its association with COVID-19 severity

Jason Enrique Vázquez Navarro. Universidad Juárez del Estado de Durango

G103

Can the biochemical interaction between Killer strains and non-toxinproducing strains of Saccharomyces cerevisiae result in resistance?

Azul Vázquez López de la Fuente. Universidad Autónoma de Querétaro

G104

Abf1 is a DNA binding protein involved in different DNA processes in Candida glabrata

Laura Angélica Vera Salazar. Instituto Potosino de Investigación Científica y Tecnológica A.C.

G105

IRF4 regulates TNFAIP3 expression in CD4+ T cells of cutaneous T cell lymphoma and T cell acute lymphoblastic leukemia

Georgina Victoria Acosta. Hospital Juárez de México

G106

Epigenetic mechanisms associated with the expression of Protocadherin 18 in a model of Epithelial-Mesenchymal Transition in MCF10A, MCF7, and MDA-MB-231 cells

Pablo Sinuhé Gómez González. Universidad Nacional Autónoma de México

MEDICINE, HEALTH & NUTRITION III

MH67

Obesity leads to transcriptomic alterations in one-carbon metabolism-related genes: in silico analysis of GTEx study

Daniel Cantú Ruiz. Tecnológico de Monterrey

MH68

Exploring the Connection Between Vitamin B12 Deficiency and Obesity

Erika Castaño Moreno. Tecnológico de Monterrey

MH69

"Alaches", quelites that besides being nutritious, have effect on Helicobacter pylori

Erika Gómez Chang. Universidad Nacional Autónoma de México

MH70

Estrogen modulation of CaMKII and calcium handling proteins in H9c2 hypertrophied myotubes

Silvia Araceli López Morán. Instituto Tecnológico de Estudios Superiores de Monterrey

MH71

Ca²⁺ Overload-Induced Mitochondrial Dysfunction is an Early Risk Factor for Lethal Ventricular Arrhythmias

Felipe de Jesús Salazar Ramírez. Tecnológico de Monterrey

MH72

Physiological response and organic interactions of berrycactus in wistar rats with metabolic syndrome (MS)

Daniela Joyce Trujillo Silva. Instituto Potosino de Investigación Científica y Tecnológica A.C.

MH73

Hyperglycemia and oxidant stress in an adult population from Mexico City

Irais Poblete Naredo. Cinvestav Zacatenco

MH74

Mitochondrial Dysfunction in the Model of Spondyloarthritis in DBA/1 Mice

Rodrigo Prieto Carrasco. Universidad Autónoma de Chihuahua

MH75

Comparison of chemo cytotoxicity in a 2D vs 3D model by LDH in primary cell culture from patients with pancreatic cancer

Valeria Punzo Mora. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán

MH76

Unlocking the lithium potential: a review of its anticancer properties

Laura Itzel Quintas Granados. Universidad Autónoma de la Ciudad de México

MH77

Evaluation of the apoptotic effect of the OIP4BAD peptide in jurkat cells of acute lymphoblastic leukemia and PBMC

Ana Mayra Reséndiz Corona. Universidad Autónoma de Querétaro

MH78

Identification of biomarkers for diagnosis and prognosis of pediatric medulloblastoma

Alicia María Reveles Espinoza. Instituto Politécnico Nacional

MH79

Analysis of DAPK1 Methylation in Apoptosis Resistance in Idiopathic Pulmonary Fibrosis derived Fibroblasts

Mariana Río de la Loza Rodríguez. Universidad Nacional Autónoma de México

MH80

Impact of periodontal disease on mental health

Andrea Yamile Rivas Escobedo. Universidad Juárez del Estado de Durango

MH81

Expression of SERCA2 and SERCA3 genes in a model of Epithelial-Mesenchymal Transition in MCF10A cells and breast cancer cell lines in response to Resveratrol

Gabriela Rodríguez Rodríguez. Universidad Nacional Autónoma de México

MH82

Evaluation of first- and second-generation platforms for messenger RNA synthesis as a potential technology in vaccine development

Griselda Rodríguez Martínez. Hospital Infantil de México Federico Gómez

MH83

Correlation of Gonadotropin-Releasing Hormone Receptor Expression in Breast Cancer Cell Lines and Its Implication as a Therapeutic Target

Daniela Rodríguez Cruz. IMSS. Cinvestav

MH84

Prevalence and classification of C-shaped Canal and Radix in Mandibular Molars using Cone-Beam Computed Tomography in Mexican Population

Ángel Gustavo Romo Mireles. Universidad Juárez del Estado de Durango

MH85

Looking for molecular biomarker's childhood acute lymphoblastic leukemia

Eliel Ruiz May. Instituto de Ecología

MH86

Novel anti-giardial compounds in Larrea tridentata and their in vitro effects on Giardia lamblia trophozoites

Ángela Daniela Saldaña Alvarado. Universidad Autónoma de Coahuila

MH87

Mexican Ganoderma lucidum mushroom extracts induce "BRCAness" phenotype and metabolic vulnerabilities in triple-negative breast cancer: A complementary medicine study

Ivan Salido Guadarrama. Instituto Nacional de Perinatología Isidro Espinosa de los Reyes

MH88

Autophagy induction after lung exposure to Saccharopolyspora rectivirgula in C57BL6 mice

Andrea Sánchez Barajas. Universidad Nacional Autónoma de México

MH89

A skinomic approach to the study of health, pathologies, aging and anti-aging of the skin

Juan Manuel Sánchez Contreras. Universidad Nacional Autónoma de México

MH90

Damage heterogeneity during induction of Hepatocarcinogenesis by chronic administration of DEN and 2-AAF through 13- and 18- weeks in Wistar Rats

Jaime Sánchez Meza. Universidad de Guadalajara

MH91

Exploring SARS-CoV-2 Gene Expression in Mexican Patients: Insights from Molecular Analysis

Joana Sánchez Medina. Universidad Nacional Autónoma de México

MH92

Gli1 expression associated with overall survival in locally advanced cervical cancer

Gabriela Carolina Morales Sandoval. Universidad Nacional Autónoma de México

MH93

β3 subunit expression in normal breast epithelial cells and highly metastatic triple-negative breast cancer cells

Juan Manuel Sandoval Valverde. Universidad de Colima

MH94

Discovery of Potential Inhibitors of Dehydroquinase Dehydratase from Methicillin-Resistant Staphylococcus aureus through Computer-Aided Drug Design

Ana Karina Segovia Parra. Universidad Juárez del Estado de Durango

MH95

Detection of EMT markers in the renal carcinogenic process of an in vivo experimental model

José Dolores Solano Becerra. Universidad Nacional Autónoma de México

MH96

O-GlcNAcylation decreases the expression of alpha2-6 sialic acid by activating the PI3-kinase/Akt pathway in oral cavity squamous cell cancer

Carlos Josué Solórzano Mata. Universidad Autónoma Benito Juárez de Oaxaca

MH97

Ibervillea sonorae (wereke) enhances glucose signaling in the skeletal muscle of diabetic rats

Vanessa Wendi Suarez Barrios. Instituto Nacional de Cardiología "Ignacio Chávez"

MH98

Antioxidant effect of the unsaponifiable fraction of avocado oil on oxidative stress and glucose metabolism in animal model of non-alcoholic fatty liver disease

Olin Torres Isidro. Universidad Michoacana de San Nicolás de Hidalgo

MH99

Influence of ECM Stiffness and Geometric Constraints on the Morphology and Mechanical Response of Lung Fibroblasts

Maria Fernanda Toscano Marquez. Instituto Nacional de Enfermedades Respiratorias
Ismael Cosío Villegas

MH100

PIEZO1-RHO/PMLC/IGF1 axis in skeletal muscle under micronutrients effects in an aging model

Anya Ayelen Valdez Hernández. Instituto Politécnico Nacional

MH101

Design of a novel multiplex qPCR system to detect pathogenic Mucoralean fungi from clinical human samples

Marco Iván Valle Maldonado. Universidad Michoacana de San Nicolás de Hidalgo

MH102

Modulatory effect of avocado oil on mitochondrial permeability transition pore in wistar rats fed a high-fat and high-fructose diet

Manuel Alejandro Vargas Vargas. Universidad Michoacana de San Nicolás de Hidalgo

MH103

The unsaponifiable fraction of avocado oil improves mitochondrial function in rats with non-alcoholic fatty liver disease

Manuel Alejandro Vargas Vargas. Universidad Michoacana de San Nicolás de Hidalgo

MH104

Evaluation of platelet function in women post-chemotherapy for breast cancer: a pilot study

Itzel Patricia Vásquez Martínez. Universidad Autónoma Benito Juárez de Oaxaca

MH105

Fisetin and regular aerobic exercise enhance glutathione redox status in the brain of rats with diabetes

Gabriel Vazquez Arredondo. Universidad de Guanajuato

MH106

Characterization of benzimidazole derivatives as inhibitors of Src homology 2 domain-containing protein tyrosine phosphatase 1

Héctor Abdel Vázquez Lechuga. Universidad Juárez del Estado de Durango

MH107

Analysis of potential drugs with leptin-binding capacity

Brandon Jorge Romero Rodríguez. Instituto Politécnico Nacional

MH108

Nicotinamide decreases protein oxidation in the retina, RPE, and LIVER in a hyperglycemic model

Juan David Villeda González. Universidad Nacional Autónoma de México

MH109

Hepatoprotective effect of Empagliflozin/Metformin co-treatment in Metabolic Syndrome

Oscar René Zambrano Vásquez. Instituto Nacional de Cardiología "Ignacio Chávez"

MICROBIOLOGY II

M41

Genetic variability of tuberculosis complex in livestock farmers in Oaxaca

Heidi Paola Galván Antonio. Universidad Autónoma Benito Juárez de Oaxaca

M42

Effect of flavonoid compounds on the multidrug-resistant yeast *Candida auris*

Jonathan García Hernández. Instituto Politécnico Nacional

M43

Point mutations in *Salmonella Typhimurium InvF* transcriptional regulator affect its function

Laura Elena García Cortés. Instituto Politécnico Nacional

M44

Interaction of *Wickerhamomyces anomalus* with fungi isolated from crops of economic interest

Karina García Gutiérrez. Instituto Politécnico Nacional

M45

Metagenomic sequencing of respiratory syncytial virus from isolates in northeastern Mexico

Julio César Garza Cabello. Universidad Autónoma de Nuevo León

M46

Identification of SR proteins in *Ustilago maydis*

Mayra Patricia Gaspariano Cholula. Benemerita Universidad Autónoma de Puebla

M47

Obtaining bioethanol from brewer's malt bagasse

Venus Melissa Gómez Lagunas. Universidad Autónoma de San Luis Potosí

M48

Phenotypic, genotypic and metabolomic characterization of Streptomyces albidoflavus J29 ori2 with antifungal activity on Candida spp.

Adilene González Silva. Instituto Politécnico Nacional

M49

Identification of virulence factors (efaA, asa1, acel and esp) in biofilm-forming Enterococcus faecium and E. faecalis

Christian González Reyes. Universidad Autónoma de Nayarit

M50

Role of the S. cerevisiae orthologous proteins KINrg1 and KIRtg3 from the aerobic yeast K. lactis in respiratory metabolism

Yael González Tinoco. Universidad Nacional Autónoma de México

M51

Structural study of TLA-1 β -lactamase in complex with tazobactam

César Alberto González Guzmán. Universidad Nacional Autónoma de México

M52

Uncovering the functional role of the RND pumps in R. etli CFN42

Adrián González Martínez. Universidad Nacional Autónoma de México

M53

The mitochondrial fusion protein Fzo1 is essential in the fungus Podospora anserina

Mariana Aline González Manríquez. Universidad Nacional Autónoma de México

M54

Participation of the ORF PA2305/ambB in the virulence of Pseudomonas aeruginosa PAO1

Ximena Hernandez Ramos. Universidad Michoacana de San Nicolás de Hidalgo

M55

Identification of proteins associated with isoforms of the eukaryotic Translation Initiation Factor 5A (eIF5A) in Fusarium graminearum

Jorge Eduardo Hernández Espinosa. Universidad de Guanajuato

M56

Antimicrobial resistance in meat samples

Hugo Antonio Hernández Pérez. Universidad Nacional Autónoma de México

M57

Characterization of the mycelial growth of fungi from an arid environment

Miguel Emiliano Hernández Fernández. Instituto Politécnico Nacional

M58

Microbiological and physicochemical characterization of kombucha

Liliana Alejandra Hernández Espinosa. Instituto Politécnico Nacional

M59

Microbiological characterization of Tenate cheese through a microbiological-genomic analysis

Mariana Hernández Suárez. Universidad Autónoma del Estado de Hidalgo

M60

Risk of the prevalence of Staphylococcus aureus in artisanal cheeses sold in Mexico city

Marcos Francisco Hernández Robles. Instituto Politécnico Nacional

M61

Variability of longitudinal clinical isolates of Candida glabrata

Marco Josué Hernández Chávez. Instituto Potosino de Investigación Científica y Tecnológica A.C.

M62

The pqsA mutation in Pseudomonas aeruginosa PAO1 modifies the production of virulence factors depending on the growth stages

Jaime Adrian Hurtado Solorzano. Universidad Michoacana de San Nicolás de Hidalgo

M63

Transcription factors involved in root cap development drive root growth direction upon inoculation with the beneficial rhizobacterium Achromobacter sp. 5B1

Kirán Rubí Jiménez Vázquez. Universidad Michoacana de San Nicolás de Hidalgo

M64

The qscR mutation in Pseudomonas aeruginosa PAO1 modifies the biosynthesis of acyl homoserine lactones dependent on the growth phase

Kevin Ledesma Muñiz. Universidad Michoacana de San Nicolás de Hidalgo

M65

Study of Bacillus cereus from food samples: Prevalence, Toxigenic Profile and Antibiotic resistance response

Brenda Sofía Loaeza Cruz. Instituto Politécnico Nacional

M66

Isolation and genome sequencing of Vibrio parahaemolyticus strains in Litopenaeus vannamei specimens from an aquaculture farm in Sonora

Jesús Daniel López Loera. Universidad Autónoma de Sinaloa

M67

The plant growth promoting rhizobacterium Achromobacter sp. 5B1, rescues Arabidopsis seedlings from alkaline stress by enhancing root organogenesis and hormonal responses

José López Hernández. Universidad Michoacana de San Nicolás de Hidalgo

M68

Role of cell-end protein TEA-5 in the growth and development of Neurospora crassa

Pedro Alejandro López García. Centro de Investigación Científica y de Educación Superior de Ensenada

M69

Search for pathogenic bacteria and antibiotic resistance in the water of Xochimilco canals

Daniel López Pastrana. Universidad Nacional Autónoma de México

M70

Determination of isoniazid tolerance of BCG pasteur ATCC 35734 and its isogenic mutant derivative BCGDBC1419c as planktonic and biofilm cultures

Luis Manuel Lorenzo Rodríguez. Universidad de Guadalajara

M71

Antimicrobial activity from Magnolol and Honokiol from Magnolia spp on periimplantitis causing pathogen

Jesús Agustín Loyola Alonso. Universidad de Guanajuato

M72

Anti-quorum sensing effect of the cyclodipeptide cyclo (Pro-Tyr), cyclo (Pro-Val), and cyclo (Pro-Phe) on the Rhl system in Pseudomonas aeruginosa PAO1

Valeria Madrigal Pineda. Universidad Michoacana de San Nicolás de Hidalgo

M73

Design of a multiplex PCR for the detection of Mycoplasma and Ureaplasma species associated with reproductive disorders in cattle

Anabelle Manzo Sandoval. Instituto Nacional de Investigaciones Forestales Agrícolas y Pecuarias

M74

Antifungal activity of immobilized cystatin microparticles of albumin

María Karina Manzo Valencia. Cinvestav Irapuato

M75

Microbiological analysis of indoor air quality in the Universidad Iberoamericana Torreón

José Antonio Martínez Villalba. Universidad Iberoamericana Torreón

M76

Effect of epinephrine and norepinephrine on the expression of virulence factors in Pasteurella multocida

Mayra Martínez Alfonzo. Universidad Nacional Autónoma de México

M77

Actinomycete metabolites isolated from Mexican jungle soils with antimicrobial and anti-virulence activity against antimicrobial-resistant pathogenic bacteria

Araceli Martínez Larios. Instituto Politécnico Nacional

M78

Assessing Antibiotic Resistance in Bacteria Isolated from Street Food Stalls

Rubén Martínez Rojas. Instituto Politécnico Nacional

M79

Analysis of NAD⁺ self-sufficiency in Avibacterium paragallinarum

Sergio Alberto Matias Vera. Universidad Autónoma de Puebla

M80

Identification and implications of the N501Y mutation in SARS-CoV-2 isolates from a Mexican population

Ángel Medina Reyes. Universidad de Guadalajara

M81

Bacterial adhesins implicated in root and gut attachment

Gerardo Mejía Vázquez. Universidad Nacional Autónoma de México

M82

Microbial Examination of Food: Comparative study of Faecal Coliforms Presence in Cooked vs. Uncooked Foods

Ángel Alejandro Meneses Sánchez. Instituto Politécnico Nacional

M83

Production and Characterization of Chitinase Activity by Bacillus spp. Strains with Antagonistic Activity Against Fusarium verticillioides

Ma. Fernanda Meneses Hernández. Universidad Nacional Autónoma de México

TOXICOLOGY & PHARMACOLOGY I

TP1

VIAN-c4551 in eye drops reaches the retina in supratherapeutic concentrations and reverses diabetes-induced excessive vasopermeability in the retina of rodents

Elva Adán Castro. Universidad Nacional Autónoma de México

TP2

Effect of pioglitazone on renal function after exposure to cadmium in Wistar rat

Daniel Isaac Alcántara Jara. Universidad Autónoma de Puebla

TP3

Chlorophyll quantification in aquatic plant (Elodea canadensis) exposed to heavy metal stibnite (Sb₂S₃) in bioassay

Ana Paola Rivera Torres. Universidad Juárez del Estado de Durango

TP4

Genotoxic effect in onion bulbs (Allium cepa) due to exposure to sphalerite (ZnS)

Gerardo Alfonso Anguiano Vega. Universidad Juárez del Estado de Durango

TP5

Antitumor activity of incomptine a in a murine breast cancer model by downregulation of hexokinase II

Ángel Giovanni Arietta García. Universidad Autónoma Metropolitana Iztapalapa

TP6

Effect of chronic ozone pollution on IL-6 and CD4 levels in the hippocampus of rats

Pamela Barragán Reséndiz. Universidad Nacional Autónoma de México

TP7

Oleic Acid Induce Neuroprotection in Brain Regions of Rats treated with Dexrazoxane

David Calderón Guzmán. Instituto Nacional de Pediatría

TP8

Exploring Lithium Salts for Autophagy Induction and Cell Viability: Novel Therapeutic Strategies for Cervical Cancer

Alejandro Israel Castillo Montoya. Universidad Nacional Autónoma de México

TP9

Evaluation of ecotoxic effects of estibanite (Sb_2S_3) in aquatic plant model (*Elodea canadensis*)

Luis Adolfo Cenicerros Morales. Universidad Juárez del Estado de Durango

TP10

Identification of Potential Agonists from Natural Sources for PPAR- γ in the Era of Machine Learning

Flavio F. Contreras Torres. Tecnológico de Monterrey

TP11

Differential Expression of Piezo1 in Prostate Cancer Cell Lines

Jesús Emiliano Covarrubias Lobatón. Universidad Nacional Autónoma de México

TP12

Molecular coupling of Diels-Alder adduct derived from anthranilic acid on COX 1 and 2

Eric Suriel Cruz Ruíz. Instituto Politécnico Nacional

TP13

Changes in liver function in rats chronically exposed to fluoride prenatally and postnatally

Fernanda Marlen Enríquez Sánchez. Universidad de Guadalajara

TP14

Efficacy of the cytotoxic effect of 6-pentadecyl salicylic anacardic acid in olive oil nanodrops in different cell lines

Elizabet Estrada Muñiz. Cinvestav Zacatenco

TP15

Evaluation of the administration of a phytochemical to a murine model of acute gastritis

Verónica Edith Gallegos Hernández. Universidad Nacional Autónoma de México

TP16

Impact of Lithium Chloride and Lithium Carbonate on DNA Repair and Apoptosis in Cervical Cancer Cells

Juan Carlos García Acosta. Universidad Nacional Autónoma de México

TP17

Bactericidal and bacteriostatic activity of secondary metabolites from *Magnolia vovidesii* against oral bacteria

Alma Rosa González Pérez. Universidad de Guanajuato

TP18

Evaluation of the antiviral activity against *Chikungunya* of the dichloromethane extract of the stem of *Ocimum basilicum* in vitro

José Carlos González Arce. Instituto Politécnico Nacional

TP19

VIAN-c4551 in eye drops inhibits the retinal vasopermeability induced by the intravitreal injection of VEGF in rats and mice

Daniela Granados Carrasco. Universidad Nacional Autónoma de México

TP20

Anacardic 6-Pentadecyl Salicylic Acid Modifies the Expression of Cell Cycle-Related Proteins in Jurkat and Peripheral Blood Mononuclear Cells

Gabriela Guerrero Palomo. Cinvestav Zacatenco

TP21

Repositioning imipramine for in vitro antiparasitic effects in *Giardia lamblia*

Xareni Zinereth Herrera Valero. Universidad Autónoma de Coahuila

TP22

Electrophysiological Analysis of *Centruroides exilicauda* Venom on Ion Channel Activity in Mouse Dorsal Root Ganglion Neurons

Darién Huerta González. Centro de Investigación Científica y de Educación Superior de Ensenada

TP23

Effect of chronic β -carotene treatment on stem markers of breast cancer cell lines

Juan Carlos Juárez Cruz. Universidad Autónoma de Guerrero

TP24

Discovering Potential Analgesics from Baja California Bark Scorpion Venom (*Centruroides exilicauda*) Using a Metabolomic Approach

Eugenio Leyva Figueroa. Centro de Investigación Científica y de Educación Superior de Ensenada

TP25

Study of the effect of glutathione peroxidase in organotypical cultures of rat cartilage exposed to nanoparticles with glutathione

Laura Denise López Barrera. Universidad Nacional Autónoma de México

POSTER SESSION IV
THURSDAY, OCTOBER 24
15:15 - 17:15

BASIC BIOCHEMISTRY III

BB79

Redesign of shikimate dehydrogenase toward cofactor specificity exchange using artificial intelligence

Raúl Iván Pérez Bermúdez. Universidad Nacional Autónoma de México

BB80

Screening of a chemical fragment library for the search of inhibitors against the acyl

homoserine lactone synthase of *Acinetobacter baumannii*

Jesús Ricardo Pérez Velázquez. Instituto Nacional de Pediatría

BB81

Choosing sides in LOXP and the importance of the initial colonization by CRE

Marco Antonio Pérez Castillo. Universidad Autónoma del Estado de Morelos

BB82

Changes in lipid and proteome composition of the inner and outer liver's mitochondrial membranes and its correlation with mitochondrial respiration in experimental type 1 diabetes

Ramiro Perusquía García. Universidad Nacional Autónoma de México

BB83

Identification of *Paramecium multimicronucleatum* mitochondrial ATP synthase

Maria Guadalupe Quintanar Solís. Universidad Nacional Autónoma de México

BB84

Generation of a quantification system for *Galleria mellonella* apolipoprotein-III

Uriel Ramírez Sotelo. Universidad de Guanajuato

BB85

Partial purification and characterization of the NADH dehydrogenase of the respiratory chain of *Bacillus subtilis*

Alexia Ramírez Ávila. Universidad Nacional Autónoma de México

BB86

The pyruvate kinase from *Pyrobaculum aerophilum* shares with other *Thermoproteales* unique features of the catalytic lid.

Leticia Ramírez Silva. Universidad Nacional Autónoma de México

BB87

Evaluation of the Intensity of the Killer Effect in *Saccharomyces cerevisiae* using different carbon sources and its relationship with Mitochondrial Biogenesis

Eric Ramos Ojeda. Universidad Autónoma de Querétaro

BB88

Kill me if you can, the long-lived yeast *Rhodotorula mucilaginosa*

Carolina Ricardez García. Universidad Nacional Autónoma de México

BB89

Kinetic characterization of eukaryotic mitochondrial ATP synthases

Anaiza Rico Luna. Universidad Nacional Autónoma de México

BB90

Biochemical response of four bread wheat genotypes subjected to heat stress

Edgar Robles Rodríguez. Centro de Investigación en Alimentación y Desarrollo, A.C.

BB91

Kinetics of Two Recombinant *Capsicum Annuum* L. Soluble Inorganic Pyrophosphatases with Different Phylogenetic Origin

Rogelio Rodríguez Sotres. Universidad Nacional Autónoma de México

BB92

Effect of Metabolic Syndrome on the expression of the ryanodine receptor and the SERCA pump in rat cerebral arteries

Daniela Rodríguez Benito. Cinvestav Zacatenco

BB93

Identifying a hydrophobin class I from *Agaricus bisporus*: production of different amyloid-like fibrils

Jesús Rojas Osnaya. Universidad Autónoma Metropolitana Cuajimalpa

BB94

Metabolic Changes and Antioxidant Response in *Ustilago maydis* Grown in Acetate

Lucero Romero Aguilar. Universidad Nacional Autónoma de México

BB95

Antioxidant capacity of asparagus under different growing methods

Adela Guadalupe Rosas Rosas. Universidad Autónoma de Chapingo

BB96

Phylogenetic distribution and reconstruction of ancestral phytases

Rogelio Iván Rosas López. Universidad Nacional Autónoma de México

BB97

Cannabidiol attenuates palmitic acid-induced inflammatory signaling in human macrophages by cannabidiol: insights into metabolic stress mitigation

Mayte Rueda Munguía. Instituto Tecnológico de Estudios Superiores de Monterrey

BB98

The roles of transcription factor SOG1 of Arabidopsis thaliana beyond of its nuclear function in root

Karem Sánchez Martínez Universidad Nacional Autónoma de México

BB99

The use of Thioflavin T to estimate the Plasma Membrane Potential (PMP) in different yeast strains and the effect of pH

Norma Silvia Sánchez Sánchez. Universidad Nacional Autónoma de México

BB100

Akt and PKC inhibitors activate an ER Ca²⁺ leak involving Sec61 translocon in HeLa cells

Lizeth Sandoval Vázquez. Cinvestav Zacatenco

BB101

Zika virus regulates cell survival through modulation of miR-125a expression and mitochondrial function

María E Santana Román. Universidad Nacional Autónoma de México

BB102

In silico, In vitro and In vivo analyzes demonstrate that arsenic affects cellular respiration by interacting with cytochrome C and ubiquinone

Erick Sierra Campos. Universidad Juárez del Estado de Durango

BB103

Effect of the Ethanolic Leaves Extract of Callistemon citrinus on the Antioxidant Activity in the Liver of Rats Administered with Indomethacin

Uriel Noe Solano Candia. Universidad Michoacana de San Nicolás de Hidalgo

BB104

Morphological and genetic characterization of fruit dehiscence in Bixa orellana L.

Rocío Tamayo García. Centro de Investigación Científica de Yucatán A.C.

BB105

Expression, Purification, and Characterization of 3-dehydroquinase synthase (DHQS) from Acinetobacter baumannii

Atl Emiliano Tarbuck Valle. Universidad Nacional Autónoma de México

BB106

Fermentation of cacao to modulate its content of active compounds

Tatiana Torres Loera. Cinvestav Zacatenco

BB107

Crystallization of the catechol 1,2-dioxygenase enzyme from *Stutzerimonas stutzeri* GOM2

Dalia Isabel Torres Aguirre. Universidad Autónoma del Estado de Morelos

BB108

Functional analysis of the catalase-peroxidase of phytopathogenic fungus *Ustilago maydis*

Marisol Ubaldo Aguilar. Universidad Nacional Autónoma de México

BB109

¿Why *Bacillus licheniformis* can make an aerobic respiratory whit cyanide in the media?

Daniel Uribe-Ramírez. Instituto Politécnico Nacional

BB110

Characterization of mitochondrial complex I from the diatom *Phaeodactylum tricornutum*

Gloria Vargas Romero. Universidad Nacional Autónoma de México

BB111

Determination of LCM and β -glucosidase in Naltel Maize sprouts from the Sierra Norte, Oaxaca

Pedro Ronel Vásquez Díaz. Instituto Tecnológico de Oaxaca

BB112

Physicochemical properties of the B domain (catalytic lid) of the pyruvate kinase from *Thermofilum pendens*

Alicia Vega Segura. Universidad Nacional Autónoma de México

BB113

The substrate of glucose-6-phosphate dehydrogenase of *Pseudomonas aeruginosa* induces a change in its quaternary structure

Roberto Velasco García. Universidad Nacional Autónoma de México

BB114

Structural keys behind unusual product diversification of a non-classical CGTase

Beatriz Velazquez Cruz Universidad de Colima

BB115

On the role of Pro16 in the stability, folding kinetics and function of the LAO binding protein from *Salmonella typhimurium*

Alejandro Fernández Velasco. Universidad Nacional Autónoma de México

BB116

Effect of *Pleurotus djamor* on mitochondrial function and expression of GRP-75 and uncoupling proteins in an insulin-resistant model

Erica Karime Ventura García. Universidad Autónoma de Coahuila

BB117

EFL1 interdomain communication is disrupted as a consequence of the R1086Q mutation

Jonathan Alejandro Zúñiga Domínguez. Universidad Nacional Autónoma de México

BB118

Characterization of sucrose synthase gene family in common bean (*Phaseolus vulgaris* L.) subjected to moisture restriction during pod filling

Norma Cecilia Morales Elias. Colegio de Postgraduados.

IMMUNOLOGY & PARASITOLOGY

IP1

In vivo role of T γ δ cells in a lupus mouse model induced by NPA stabilization

Edgar Iván Galarce Sosa. Instituto Politécnico Nacional

IP2

Plasmodium vivax mitochondrial DNA polymorphism, and its relationship to genes encoding sexual proteins and the infection pattern in mosquitoes Nyssorhynchus albimanus and Anopheles pseudopunctipennis, Chiapas, México

Lilia González Cerón. Instituto Nacional de Salud Pública

IP3

Impact of the form of birth and effect of Flagellin on the activation of neonatal CD4⁺ and CD8⁺ T cells

Carlos Jesús Ventura Martínez. Universidad Autónoma del Estado de Morelos

IP4

Role of metalloproteinases in the midgut of Aedes aegypti mosquitoes infected with dengue virus

Sofía Alanís Muñoz. Instituto Nacional de la Salud Pública

IP5

Involvement of the α SMasa6 from E. histolytica in the repair of plasma membrane damage induced by pore-forming proteins

Sairy Yarely Andrade Guillen. Universidad de Guanajuato

IP6

Macrophage migration inhibitory factor (MIF) contributes to macrophage-M1 polarization by regulating mitochondrial function

Antonio Andrade Meza. Universidad Nacional Autónoma de México

IP7

Exosomes secreted by PBMCs infected with Helicobacter pylori transport TNF- α modulates EMT, migration and invasion in AGS cells

Josefina Atrisco Morales. Universidad Autónoma de Guerrero

IP8

Induction of trained immunity by bovine respiratory disease vaccine in cattle

Marisol Báez Magaña. Universidad Michoacana de San Nicolás de Hidalgo

IP9

Mitochondrial dynamics in B cell response against lipidic antigens

Giovanna Berenice Barrera Aveleida. Instituto Politécnico Nacional

IP10

Analysis of entomopathogenic properties of *P. syringae* CDBB-B1293 against *Aedes aegypti*, the main vector of the dengue virus

Valeria Calderón Frontana. Cinvestav Zacatenco

IP11

Effect of supplementation with L-arginine and L-serine on the cytokine profile of neonates born by cesarean section and vaginal delivery

Ingrid Yaritzit Carreón Cortés. Universidad Autónoma del Estado de Morelos

IP12

Association of the metabolic state and activation response of neonatal and adult CD4⁺ and CD8⁺ T lymphocytes

Francisco Castañeda Gómez. Universidad Autónoma del Estado de Morelos

IP13

Stimulating the immune function of neonatal CD4⁺ T cells with cytokines

Alejandra Cedillo Baños. Universidad Autónoma del Estado de Morelos

IP14

Nucleolar factor Pescadillo is required for cell proliferation in the human parasite *Trypanosoma brucei*

Laila Espinosa Renteria. Universidad Nacional Autónoma de México

IP15

Molecular study of the Pseudouridine synthase Cbf5 in the human pathogen *Leishmania major*

Luis Enrique Florencio Martínez. Universidad Nacional Autónoma de México

IP16

Characterization of Tau138 and its role in RNA Pol III transcription and chromatin organization in trypanosomatid parasites

Eduardo García Huerta. Universidad Nacional Autónoma de México

IP17

The deletion of Smad7 enhances the inhibitory effect of TGF- β on the effector functions of CD8⁺ T lymphocytes

Verónica Yutsil García Rasilla. Universidad Nacional Autónoma de México

IP18

Obtention of human neutralizing antibodies against dengue virus through the selection of B-cells

Dulce Desireé Izar Marín. Instituto Politécnico Nacional

IP19

Study of NFAT activation in porcine alveolar macrophages via hrGFP reporter gene expression

Eunice López Ahumada. Universidad Nacional Autónoma de México

IP20

B55 β as a pivotal molecule in arthritis development and anti-TNF- α therapy

Dulce Malvaez Luis. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán

IP21

Effect of two conditionings of Citrus aurantium on the proliferation of Entamoeba histolytica trophozoites cultured in vitro

Ivette Andrea Martínez Santiago. Universidad Nacional Autónoma de México

IP22

Identification of protein factors involved in the atypical biogenesis of the 60S ribosome subunit in the human pathogen Leishmania major

Aldo Yael Miranda Muñoz. Universidad Nacional Autónoma de México

IP23

Population regulation of Schinus molle on trophozoites of Entamoeba histolytica in axenic culture

Arely Morales Vaca. Universidad Nacional Autónoma de México

IP24

Variante rs10109853C/T de IDO2 y su relación con artritis reumatoide

Betsabe Guadalupe Najera Ruiz. Universidad Autónoma de Guerrero

IP25

Prevalence of Virulence Genes in Staphylococcus aureus Isolated from Patients with Atopic Dermatitis and Their Effect on TSLP Production by Human Keratinocytes

Claudia Leticia Vanegas Flores. Universidad Nacional Autónoma de México

IP26

The sialomucin CD43 induces autophagy-dependent cell death in lymphoid tumors during starvation

Erick Israel Pérez García. Universidad Nacional Autónoma de México

IP27

The NS1 protein of the dengue virus promotes early secondary tissue infection of Aedes aegypti mosquitoes

Edgar Quezada Ruiz. Cinvestav Zacatenco

IP28

Activation of NKT cells and Tg δ lymphocytes in response to mycobacterial lipids

Claudia Albany Reséndiz Mora. Instituto Politécnico Nacional

IP29

Study on the anti-inflammatory potential of DL-3-hydroxy-3-ethyl-3-phenylpropionamide, an anticonvulsant drug, in microglia derived from mice with epilepsy

Claudia Albany Reséndiz Mora. Instituto Politécnico Nacional

IP30

Immunometabolic control of neonatal CD4⁺ T cell responses

Jonathan Ricardo Robles Reyes. Universidad Autónoma del Estado de Morelos

IP31

6-Pentadecyl Salicylic Acid Modifies the Secretion of Cytokines of Human Natural Killer Cells in Vitro

Christyna de los Ángeles Rosales González. Cinvestav Zacatenco

IP32

New liposomal amphotericin B derivative: A potential chagas disease treatment

Lisset Torres Martínez. Universidad Nacional Autónoma de México

IP33

Optimizing Human Serum Pretreatment Blood Serum Pretreatment for Accurate Total Endotoxin Quantification Using LAL Assay

Keren Tovar Presas. Universidad Autónoma de Ciudad Juárez

IP34

Identification of perchloric acid-soluble proteins from *Trichomonas vaginalis*

Laura Isabel Vázquez Carrillo. Universidad Autónoma de la Ciudad de México

IP35

Evaluation of the immune response in rats during physiological cardiac hypertrophy induced by pregnancy

José Alberto Verduzco Palafox. Universidad de Sonora

IP36

Macrophage polarization analysis in a mouse model of Systemic Lupus Erythematosus triggered by lipid antigens

Carlos Wong Baeza. Instituto Politécnico Nacional

IP37

Immunological Endotypes of Atopic Dermatitis in the Mexican Population: Differential Expression of CLA⁺ T Cells and CCR4/CCR10 Receptors

Luis Carlos Zumaya Pérez. Universidad Nacional Autónoma de México

IP38

KLF10: A Crucial Transcription Factor in Macrophage Defense Against *Mycobacterium tuberculosis*

Hilda C. Leyva Sánchez. Universidad Nacional Autónoma de México

MICROBIOLOGY III

M84

Microbiological characterization of Mother of Vinegar and its inhibitory effect

Gloria Beatriz Molina Valdés. Instituto Politécnico Nacional

M85

The post-transcriptional regulator RsmA mediates the protective response against pyocyanin overproduction in Pseudomonas aeruginosa ID4365

Luis Fernando Montelongo Martínez. Universidad Nacional Autónoma de México

M86

The chorionic gonadotropin hormone decreases the expression of possible virulence factors of Klebsiella pneumoniae in vitro

Juan Fernando Montes García. Universidad Nacional Autónoma de México

M87

Identification of a pesticidal crystal protein (Cry8Ba) in the genome of the Metarhizium brunneum

Aida Gabriela Mora Acebedo. Universidad de Guanajuato

M88

Role of Sargassum fluitans as a Biological Reducer of Gold in the Synthesis of Nanoparticles and its Bactericidal Impact

Bryan Leonardo Naredo Rojas. Universidad Autónoma de Puebla

M89

Study of the role of the histidine-kinase RPF2 in alginate biosynthesis in Azotobacter vinelandii

Mariana Ochoa López. Universidad Autónoma de Puebla

M90

Development of a biosensor for the detection of Streptococcus pneumoniae

José de Jesús Olivares Trejo. Universidad Autónoma de la Ciudad de México

M91

Biotechnological Potential of Thermotolerant Wastewater Yeasts for High-Temperature Biofuel Production

Miguel Ángel Orozco González. Universidad Nacional Autónoma de México

M92

Modulation of the Virulence of Multidrug-Resistant E. coli O104:H4 by Subinhibitory Concentrations of Ampicillin

Yaraymi Ortiz Reyes. Universidad Autónoma de Nuevo León

M93

Visualization of the Neurospora crassa developmental cycle in mutant strains lacking hydrophobins

Omar Páez Gómez. Universidad Nacional Autónoma de México

M94

Mitochondria proteome of mosquito cells infected with dengue virus

Victoria Pando Robles. Instituto Nacional de Salud Pública

M95

Interaction of the MibR protein with the regulatory region of the ipdC gene of Azospirillum brasilense Sp7

José Cristian Pérez Gómez. Universidad Autónoma de Puebla

M96

Characterization of the anti-quorum sensing and anti-biofilm activity of diverse actinomycetes isolated from soils of the Mexican jungle against pathogenic bacteria resistant to antibiotics

Itzel Plascencia Ortiz. Instituto Politécnico Nacional

M97

Evaluation of the immunogenic properties of surface proteins from Gallibacterium anatis 12656-12

Ana Karen Ponce Quiroz. Universidad Autónoma de Puebla

M98

The unexplored diversity of yeasts from open fermentations in Mexico

Nancy Judit Quintana Rodríguez. UGA. Cinvestav Irapuato

M99

Genome analysis of Gluconacetobacter sp. UAPS01-405, isolated from the non-photosynthetic parasitic plant Conopholis alpina

Rigel Quintero Luis. Universidad Autónoma de Puebla

M100

Aldehyde Dehydrogenase Diversity and 6-OL-ALDH overexpression in Azospirillum genomes

Albverto Ramírez Mata. Universidad Autónoma de Puebla

M101

Biochemical and microbiological characterization of the intestinal microbiome of the Tenebrio molitor larva

Axel Reyes Coatl. Universidad Autónoma de Puebla

M102

Phenotypic characterization of the libR gene of Azospirillum brasilense sp7

Sandra R. Reyes Carmona. Universidad Autónoma de Puebla

M103

Physiological and symbiotic differences of pyruvate carboxylase and phosphoenolpyruvate carboxylase in Rhizobium phaseoli

Alma Ruth Reyes González. Universidad Nacional Autónoma de México

M104

Avin 41190: A Novel Regulatory Element in the Multikinase Network GacS (MKN-GacS) in Azotobacter vinelandii

Itzel Rodríguez Guerra Universidad Autónoma de Puebla

M105

The BARA/SIRA and CSR systems regulates the expression of a putative carbonic anhydrase enzyme encoded in the PSLT virulence plasmid of salmonella enterica serovar typhimurium

Yoselin Xiomara Rodríguez Muñoz. Universidad Nacional Autónoma de México

M106

The potential of Periconia macrospinoso HAGJ2 isolated from agave tequilana to control plant pathogens

Fernando Uriel Rojas Rojas. Universidad Nacional Autónoma de México

M107

The role of Epinephrine and Norepinephrine in promoting the growth and virulence factor expression of Mannheimia haemolytica is significant.

Verónica Rosales Islas. Universidad Nacional Autónoma de México

M108

Study of splicing and Nonsense Mechanism Decay (NMD) factors in Ustilago maydis

Kate Ariadna Rossano Gutiérrez. Universidad Autónoma de Puebla

M109

Elucidating the function of Azotobacter vinelandii phasins proteins: PhbP2 and PhbP3 are required for the degradation of the biodegradable plastic polyhydroxybutyrate

Jessica Ruiz Escobedo. Universidad Nacional Autónoma de México

M110

Characterization of gut microbiota in cafeteria diet-fed mice model of obesity

Jessica Ruvalcaba Galindo. Universidad de Colima

M111

Identification of antibiotic resistance in oral bacteria from dogs with periodontal disease

Yosahandy Palacios Castañón. Universidad La Salle Bajío

M112

Search of maize seed endophytes with antagonistic activity against Fusarium verticillioides

Stefhany Segoviano Correa. Universidad Nacional Autónoma de México

M113

Study of bacteriophages to control of multi-drug resistant bacteria

Omar Alejandro Sepúlveda Robles. CMN. Siglo XXI. IMSS

M114

Study and characterization of new compounds with antivirulence and antifungal activity produced by Streptomyces albidoflavus J25 isolated from Mexican jungle soil

Daniela Elysa Solorio Escamilla. Instituto Politécnico Nacional

M115

Genomic insights into three clinical Serratia marcescens strains of environmental origin

Faviola Tavares Carreón. Universidad Autónoma de Nuevo León

M116

Detection of Bacillus cereus as a strategy to assess food safety in Mexico City

María Denisse Tejada Ramírez. Instituto Politécnico Nacional

M117

Nernst Equation versus Killer effect in Saccharomyces cerevisiae

Brenda Tellez de la Garza. Universidad Autónoma de Querétaro

M118

Antimicrobial potential of heliotropium angiospermum against Salmonella SP., isolated from chicken

David Tirado Torres. Universidad de Guanajuato

M119

Evaluation of the antimicrobial activity of buddleja scordioides against Salmonella SP., isolated from chicken

Guadalupe Vázquez Rodríguez. Universidad de Guanajuato

M120

Risk of the presence of Escherichia coli in vegetables marketed in Mexico city

Carlos Ramón Vázquez Quiñones. Instituto Politécnico Nacional

M121

Bioinformatic analysis of glycine derivative of 22-oxocolestenyl as potential anti-cancer agent

Ana Karen Villa Merlán. Universidad Autónoma de Puebla

M122

Mitogen-Activated Protein Kinase Hog1: Key Regulator for Enhancing Riboflavin Overproduction in the Yeast Debaryomyces hansenii

Diana Villarreal Huerta. Universidad Nacional Autónoma de México

M123

Analysis of genes cdgE and hkhB in Azospirillum baldanorium Sp245 involved in biofilm formation and motility

María Luisa Xiqui Vázquez. Universidad Autónoma de Puebla

M124

Characterization of the antibiotic effect and metal resistance in the C4 strain of Providencia stuartii

Gokabeth Zarco Hernández. Instituto Politécnico Nacional

NEUROSCIENCES & NEUROBIOLOGY II

NN28

Modulation of glutamate uptake by the subacute activation of the histamine H3 receptor in rat cerebro-cortical astrocytes in primary culture

Yrving Daniel Díaz De Lucio. Cinvestav Zacatenco

NN29

Impact of Intestinal Low-Grade Inflammation on Purinergic-Calcium Signaling in Enteric Glial Cells

Irving Israel Vega Juárez. Universidad de Colima

NN30

Resveratrol as a modulator of the glutathione system and its neuroprotective effect on the prefrontal cortex

Brenda Hernández Pérez. Universidad Autónoma de Puebla

NN31

Sex-dependent effects on neurodevelopment of perinatal exposure to glyphosate herbicides in the rat

Isela Hernández Plata. Universidad Nacional Autónoma de México

NN32

Effect of agomelatine on the CPF 5-HT_{2c} receptor in a rat model of major depression

Daniel Juárez Serrano. Universidad Autónoma de Puebla

NN33

Catalase as a biomarker of oxidative stress related to depressive disorders

Francisco Javier Lievanos Ruiz. Universidad Michoacana de San Nicolás de Hidalgo

NN34

RNA-seq and cytokine secretion comparison in primary rat cerebral cortex astrocytes induced to cellular senescence or gliosis with palmitate

Michel López Teros. Universidad Autónoma Metropolitana Iztapalapa

NN35

Depression-like behaviour is comorbidity in a model of heart failure with preserved ejection fraction induced by high-fat diet and L-NAME

Roger Alexis Maldonado Ruiz. Tecnológico de Monterrey

NN36

Mutation analysis in the RCBTB1 gene causing retinal dystrophy: a model for mapping related genes in Mexico

Claudia Mancilla Simbro. Universidad Autónoma de Puebla

NN37

Functional characterization of human embryonic stem cell derived-chromaffin cells

Hilda Angélica Martínez Becerril. Universidad Nacional Autónoma de México

NN38

Obesity induced by a diet rich in fats and sugars affects spatial memory in ovariectomized female CD1 mice

Heidy Martínez Pacheco. Universidad Veracruzana

NN39

Alterations in spatial memory of CD-1 mice strain feed with Mexican cafeteria diet

César Amador Mendoza Calles. Universidad Veracruzana

NN40

Sensitivity of TUBB4A to microtubule stabilization and depolymerization

Aurora Citlalli Montes Centeno. Universidad de Guanajuato

NN41

Mitochondrial Dynamics in the Retina at the Early Onset of Diabetes in a Rat Model of Type I Diabetes

Frida Paulina Muñiz Ruvalcaba. Universidad Nacional Autónoma de México

NN42

Unraveling the Cortical Origins and Dopaminergic Modulation of Beta Oscillations in Parkinson's Disease Models

Aidán Alejandro Ortega García. Universidad Nacional Autónoma de México

NN43

Characterization and differentiation of human induced pluripotent stem cells into midbrain dopaminergic neurons

Andrés Paredes Rocha. Universidad Nacional Autónoma de México

NN44

Muscarinic modulation of Calcium currents in striatal ChAT's interneurons: the impact of M1 receptor

Yohana Parrado Quintana. Universidad Nacional Autónoma de México

NN45

Role of testosterone in the gain of stemness characteristics in cells derived from human glioblastoma

Juan Carlos Quintero Gallegos. Instituto Nacional de Perinatología. UNAM

NN46

The calcium hypothesis in Alzheimer's disease

Luis Ángel Ramírez Cipriano. Universidad Autónoma del Estado de Hidalgo

NN47

Metabolic and epigenetic effects of β -hydroxybutyrate during the differentiation of human pluripotent cells to dopaminergic neurons

Margareht Reyes Aldana. Universidad Nacional Autónoma de México

NN48

Inactivation of Plpp3 in neural crest cells leads to neonatal cardiac failure

Erick Antonio Rojas Pacheco. Universidad Nacional Autónoma de México

NN49

Impact of Dual Inhibition of Pak1 and CaMK2 Kinases on Glioblastoma Oncogenesis

Héctor Iván Saldívar Cerón. Universidad Nacional Autónoma de México

NN50

Enhancement of locomotor function and attenuation of apoptosis by the use of tibolone as a treatment for traumatic spinal cord injury

Stephanie M. Sánchez Torres. CONAHCYT. Centro Médico Nacional Siglo XXI. IMSS

NN51

Activity modulation of striatal cholinergic interneurons mediated by activation of dopamine D1-like receptors

Maricela Lizbeth Saucedo Bello. Universidad Nacional Autónoma de México

NN52

Molecular cloning, functional characterization, and differential expression of two novel GABA_AR-like subunits from red swamp crayfish *Procambarus clarkii*

Iván Uriel Valladares Hernández. Cinvestav Zacatenco

NN53

Progesterone treatment restores normal blood-brain barrier function in a rat model of chronic cerebral hypoperfusion

Amairani Maydelin Vázquez Espadin. Universidad Autónoma Metropolitana Iztapalapa

NN54

Epigenetic control of neuron activation and memory in aging

G. Aleph Prieto. Universidad Nacional Autónoma de México

SIGNAL TRANSDUCTION AND CELL DIFFERENTIATION II

ST32

Cannabidiol increases adipogenesis on 3T3-L1 pre-adipocytes via PPAR γ

Helen Yarimet Lorenzo Anota. Tecnológico de Monterrey

ST33

BUB 1 is a nodular kinase that predicts overall survival in osteosarcoma, liposarcoma, synovial. sarcoma, and leiomyosarcoma

Frida Citlali Rodríguez Izquierdo. UAM I / Instituto Nacional de Cancerología

ST34

The IL-6 induces metastatic properties in luminal breast cancer cells through GPR30

Ana Carolina Tirado Garibay. Universidad Michoacana de San Nicolás de Hidalgo

ST35

Spermatozoa adhesion to an immobilized fibronectin matrix alters their physiology and increases their survival

Coral Yamilet Jorge Cruz. Cinvestav Zacatenco

ST36

Possible Involvement of Pak1 and CaMKII in Insulin Secretion by Pancreatic Beta Cells

Nely Gisela López Desiderio. UNAM/Instituto Politécnico Nacional

ST37

Insights into the regulation of SnRK1 through phosphorylation by upstream kinases

Carmen Aitana López Zequeira. Universidad Nacional Autónoma de México

ST38

Lipid droplets in plant cells: interaction with nuclei and the endoplasmic reticulum

Alejandro López Hernández. Universidad Michoacana de San Nicolás de Hidalgo

ST39

Thrombin-induced activation of focal adhesion proteins in the Retinal Pigment Epithelium (RPE)

Katya Manzanita Quintero. Universidad Nacional Autónoma de México

ST40

The role of apoptosis in the development of the Drosophila tracheal system and its relationship with protruding cells of the tracheal dorsal trunks

Pedro Alberto Marmolejo Cruz. Universidad Nacional Autónoma de México

ST41

Specific Temporal Requirement of Prox1 Activity During Pancreatic Acinar Cell Development

Angélica Sofía Martínez Ramírez. Northwestern University

ST42

Activation of the IGF-1/IGF-1R increases chemoresistance to paclitaxel of non-small cell lung cancer cell line (A549)

Max Alejandro Maximino Rojas. Universidad Autónoma de Puebla

ST43

Interferon-stimulated gene 15 and ISGylation levels are modulated by IFN-gamma in medulloblastoma cells

Karen Haidee Medina Abreu. Universidad Autónoma de la Ciudad de México

ST44

Mechanisms of Action and Secretion of the TGF- β Cytokine in Colorectal Cancer

Christian Daniel Meza Juárez. Universidad Nacional Autónoma de México

ST45

Evaluation of the effect of amiodarone and desethylamiodarone on cell proliferation and migration in lung cancer cells, A549

Zurisadai Morales Herrera. Universidad Autónoma de Puebla

ST46

RANKL differentially regulates breast cancer stem cells according to RANK or LGR4 activation

Alejandro Ordaz Ramos. Instituto Nacional de Medicina Genómica

ST47

Diastolic dysfunction, excitation-contraction coupling defects and arrhythmias in absence of mitochondria Ca^{2+} uptake

Julietta Palomeque. Tecnológico de Monterrey

ST48

Role of indoleamine serotonin in modulating cell death in the root meristem of *Arabidopsis thaliana*

Ramón Pelagio Flores. Universidad Michoacana de San Nicolás de Hidalgo

ST49

Participation of the Rac1 GTPase in progressive motility in guinea pig and mouse sperm

Danelia Ramirez Ramirez. Cinvestav Zacatenco

ST50

Pxd1 and organelle trafficking: Implications for Sexual Development in *Podospora anserina*

Matías Ramírez Noguez. Universidad Nacional Autónoma de México

ST51

Loss-of-function of MEDIATOR 12 or 13 subunits causes the swelling of root hairs in response to sucrose and abscisic acid in *Arabidopsis*

Javier Raya González. Universidad Michoacana de San Nicolás de Hidalgo

ST52

Effect of 17 β -estradiol on salbutamol-induced airway smooth muscle relaxation

Jorge Eduardo Reyes García. Universidad Nacional Autónoma de México

ST53

Evaluation of a triazaspirane-type derivative on cell viability, proliferation and migration in MDA-MB-231 breast cancer cells

Bryan Alexis Rivera Suárez. Universidad Autónoma de Baja California

ST54

TMEM16A regulates sperm cell volume through RhoA and the actin cytoskeleton

Ana Lilia Roa Espitia. Cinvestav Zacatenco

ST55

Role of STAT3 on the regulation of critical genes for metabolic rearrangement in cervical cancer cells after IL-2 treatment

Rodrigo Rojas Mercado. Universidad Nacional Autónoma de México

ST56

Sperm viability is regulated by intracellular calcium levels through the CaSR/PI3K/Akt pathway, which probably regulates TRPV channels in guinea pig sperm

Mónica Lizbeth Salgado Lucio. Cinvestav Zacatenco

ST57

Expression and associated epigenetic mechanisms of DSC3 and DSG3 during Epithelial-Mesenchymal Transition induced by TGF- β and EGF in breast cancer cell lines

Ángel Salmerón Hernández. Universidad Nacional Autónoma de México

ST58

Trachea-epidermis interactions as a model of coordinated morphogenesis in Drosophila development

Eduardo Sánchez Cisneros. Universidad Nacional Autónoma de México

ST59

Discovery of PTP1B ligands through drug repurposing for type 2 diabetes therapy: in silico and in vitro studies

Ximena Valeria Sánchez Nava. Universidad Nacional Autónoma de México

ST60

The expression of TAZ transcriptional co-regulator is induced by TGF- β /SMAD pathway in hepatic cancer cells

Marcela Sosa Garrocho. Universidad Nacional Autónoma de México

ST61

Study of the role played by the non canonical Wnt/Ca²⁺ pathway in the chemoresistance of colon cancer cells

Andrea Terán Ramos. Universidad Nacional Autónoma de México

ST62

Thrombin-induced cytokine secretion from retinal Müller glia

Naomi Vargas Martínez. Universidad Nacional Autónoma de México

ST63

The Protein Tyrosine Phosphatase 1B (PTP1B) Promotes the Migration of Colon Cancer Cells Through the Activation of a VAV1-Rac1 Signaling Pathway

Olga Villamar Cruz. Universidad Nacional Autónoma de México

TOXICOLOGY & PHARMACOLOGY II

TP26

Anti-Helicobacter pylori activity, cytotoxicity and in vivo gastroprotective effect of diacetylcurcumin and its metal derivatives

Almanelly Agabo Martínez. Universidad Nacional Autónoma de México

TP27

The combined therapy of diclofenac-itraconazole reduces the size of Madurella mycetomatis grains and stimulates cytokines of the Th1 and Th17 profile

Iván Alejandro Banda Flores. Universidad Autónoma de San Luis Potosí

TP28

Transcriptional characterization of the antitumor activity of laherradurin on an in vitro model of colorectal cancer

Eduardo Pérez Arteaga. Universidad Nacional Autónoma de México

TP29

Kidney and liver damage from a combined NSAID-antifungal therapy for treatment of eumycetoma

Manuel Martínez Hernández. Universidad Autónoma de San Luis Potosí

TP30

Evaluation of Antineoplastic Compounds in Breast Cancer Cell Lines

Natalia Mata de los Rios. Universidad Autónoma de Chihuahua

TP31

Behavioral and GABAergic alterations after chronic atrazine exposure in the female albino rat

María Soledad Mendoza Trejo. Universidad Nacional Autónoma de México

TP32

Evaluation of artichoke (Cynara scolymus) extract and weight control in a model of induced menopause

Nazarett Tabita Mendoza Centeno. Universidad Nacional Autónoma de México

TP33

Curcumin and Metformin enhances cytotoxicity and cell death on cervical cancer cell lines

Marisol Mir García. Instituto Nacional de la Salud Pública

TP34

Antimicrobial activity of synthetic peptides derived from scorpion Superstitionia donensis

Rodolfo Miranda Espino. Universidad Nacional Autónoma de México

TP35

Antifungal miconazole inhibits Kv10.1 and Nav1.7 ion channels

Enikar Manuel Morales Patlán. Universidad Nacional Autónoma de México

TP36

Machine Learning Innovates Drug Discovery Processes: Potential Ligands for Cannabinoids

Ana C. Murrieta. Tecnológico de Monterrey

TP37

Protective effect of an aqueous extract of Maqui Berry in an acute model of carbon tetrachloride-induced liver injury in Wistar rats

Carlos Enrique Orozco Barrios. Centro Médico Nacional Siglo XXI. IMSS

TP38

Characterization of the Venom of the Scorpion *Nullibrotheas allenii*

Nancy Sarai Ortiz Ventura. Universidad Nacional Autónoma de México

TP39

p,p'-DDE alters monocyte-to-macrophage differentiation markers of human peripheral blood mononuclear cells

José Ricardo Palacios Valladares. Cinvestav Zacatenco

TP40

Antimicrobial and antifungal properties of methanolic extracts from three species of *Magnoliopsida*

Angélica Eulalia Pérez Cid. Instituto Politécnico Nacional

TP41

Urinary 1-hydroxypyrene as a biomarker of exposure to polycyclic aromatic hydrocarbons in pregnant women in the Puebla City

Jesús Azahel Piña López. Universidad Autónoma de Puebla

TP42

Preparation and characterization of matrix microspheres of acetylsalicylic acid and acetaminophen, using sodium alginate by the ionic gelation technique

Edith Cecilia Rivera Meza. Instituto Politécnico Nacional

TP43

Biophysical and physicochemical characterization of novel potassium channel blocker 3-Fluoro-5-Methylpyridin-4-Amine

Marina Sofía Rodríguez Rangel. Universidad de Guadalajara

TP44

Establishment of a Doxorubicin-Resistant SW620 Cell Line for Multidrug Resistance Studies

Yanet Eunice Rodríguez López. Universidad Nacional Autónoma de México

TP45

Synergical effect of *spc13*-dic whit commonly used antibiotics in gram negative, positive and multi-resistant bacteria

Carmen Itzamatul Rodríguez Alejandro. Universidad Autónoma del Estado de Morelos

TP46

Pharmacological potential of the sap of *Sedum rubrotinctum* R.T. Clausen for the treatment of eye infections

Anuar Salazar Gómez. Universidad Nacional Autónoma de México

TP47

Anticancer effects of morin hydrate targeting cytochromes P450 in pediatric rhabdomyosarcoma cells

Rebeca Santes Palacios. Instituto Nacional de Pediatría

TP48

In vitro gene therapy model to assess the anticancer activity of the proapoptotic peptide CTMP-4

María Teresa Silva Guzmán. Universidad Autónoma de Nuevo León

TP49

Does SPC13, an antimicrobial activity found in the venom of the *Scolopendra vpolymorpha*, also have histone H3 properties?

Lucero Valladares Cisneros. Universidad Autónoma del Estado de Morelos

TP50

Identification of anti-CD36 antibodies and characterization of their biological activity

Marco Velasco Velázquez. Universidad Nacional Autónoma de México

TP51

Lithium-Induced Regulation of Cellular Processes in Colorectal Cancer: An In Vitro Analysis

Edgar Yebran Villegas Vázquez. Universidad Nacional Autónoma de México

TP52

Immune characterization effect of scorpion venom from municipality of Coronado, Chihuahua

Adrian Rivas Muñoz. Universidad Autónoma de Chihuahua

OTHERS II

O41

Identification of Aim24 as a yeast complex II membrane domain assembly factor

Yolanda Margarita Camacho Villasana. Universidad Nacional Autónoma de México

O42

Agronomic and molecular evaluation of polyploid tomato

Fatima Clarita Cota Ruiz. Cinvestav Zacatenco

O43

p53 Mutants Induce an Aggressive Phenotype through Overexpression of miR-27b-5p in Cancer

José Edwin Dolores García. Universidad Autónoma Metropolitana Iztapalapa

O44

The inhibitory effect of rTBL-1 on colorectal cancers in vitro is related to the presence of the epidermal growth factor receptor

Teresa García Gasca. Universidad Autónoma de Querétaro

O45

Analysis of the binding of Mdm2 protein with Rb mRNA

Alondra Mariana Martínez Partida. Universidad Autónoma de San Luis Potosí

O46

Surveillance of Natural Killer Cells in Patients with Tick-Borne Diseases

Mariana Ivonne Martínez Rivera. Universidad Autónoma de Chihuahua

O47

Characterization of the functional relationship between the retrograde pathway and cell longevity in *Saccharomyces cerevisiae*

Paola Abril Medina Flores. Universidad Nacional Autónoma de México

O48

Dinamic of phosphatidylserine exposure during capacitation in human sperm

Ivonne Medina Ruiz. Universidad Nacional Autónoma de México

O49

The E1B-55KDa oncoprotein binds to the adenoviral genome and regulates gene transcription

Eduardo Mundo Nájera. Universidad Autónoma del Estado de Morelos

O50

Life after death?

Christopher Navarro Martínez. Universidad Nacional Autónoma de México

O51

Labeling of recombinant lectin with nanoparticles and its analysis in microscopy

Lizet Angélica Núñez Corona. Universidad Autónoma de Querétaro

O52

MDM2 controls the expression levels of p53 and RB tumour suppressors

Vanesa Olivares Illana. Universidad Autónoma de San Luis Potosí

O53

IFC-305 treatment induces apoptosis mediated by miR92a-3p in liver cancer cells

Bibiana Ortega Domínguez. Universidad Nacional Autónoma de México

O54

GM1 dimerization induces an increase in the intracellular Ca²⁺ concentration and the acrosome reaction in capacitated human sperm

Iván Oseguera López. Universidad Nacional Autónoma de México

O55

Post-embryonic activation of the primary root apical meristem cells in some Cactaceae species

Juan Manuel Palacios Corona. Universidad Nacional Autónoma de México

O56

Carvacrol improves the contractile properties of skeletal muscle under low-intensity electrical stimulation

Daniel Perea Ruiz. Universidad de Colima

O57

Charge distribution contributes to in vitro protein desiccation protection of a group of highly charged anhydrobiotic intrinsically disordered proteins

Diego Pérez Villanueva. Universidad Nacional Autónoma de México

O58

miR-107 Expression as a Potential Predictor of Castration Resistance in Prostate Cancer liquid biopses

Jonathan Puente Rivera. UACM. Hospital Juárez de México

O59

Mitigation of Emerging Diseases in Wild Animals through Pharmacimicrobiomics

Jimena Ramírez Villarreal. Universidad Autónoma de Querétaro

O60

The common bean mutant (*Phaseolus vulgaris*) non-nodulating mutant: *nnod(2114)* is affected in the infection thread development during symbiosis with rhizobia

Rocio Reyero Saavedra. Universidad Nacional Autónoma de México

O61

Unveiling the Proteomic Impact of Calcium Carbide Ripening on Maradol Papaya Seeds

Francisco Antonio Reyes Soria. Instituto de Ecología, A.C.

O62

Comparative Insights into Glutathione Transferases in *Taenia solium*: *Ts24GST*, *Ts25GST* and *Ts26GST*

Cinthyia Alejandra Rocha Sánchez. Universidad Nacional Autónoma de México

O63

Measuring Macromolecular Crowding in *Arabidopsis* Root Cells from Different Developmental Zones

Delia L. Rodríguez Bustos. Universidad Nacional Autónoma de México

O64

Comparative analysis of the septin 2 interactome in mosquito cells (Aag2) infected and not infected with dengue virus

Jose Ángel Rubio Miranda. Cinvestav Zacatenco

O65

Protein conformational dynamics and thermal adaptation

Jorge Emiliano Salinas López. Universidad Nacional Autónoma de México

O66

The common bean (*Phaseolus vulgaris*) non-nodulating mutant nnod(1895) is affected in the initial step of rhizobial infection during the N-fixing symbiosis

Karina San Vicente Trujillo. Universidad Nacional Autónoma de México

O67

Development of a Single-Cycle Non-Classical HAstV Replicon System for Studying Viral Replication and Inhibitors

Guadalupe Sánchez Flores. Universidad Autónoma del estado de Morelos

O68

Effect of Zinc Oxide Nanoparticles Against Rotavirus Infectivity

Juan Diego Siary Leyva. Universidad de Sonora

O69

The follicle stimulating hormone modulates protein tyrosine phosphorylation, hyperactivation and acrosome reaction during human sperm capacitation

Viridiana Estefany Solis Ayala. Universidad Nacional Autónoma de México

O70

Molecular modeling of the effect of non-synonymous ABCC4 mutations over MRP4 conformation and substrate binding

Valeria Jacqueline Soto Ontiveros. Universidad Autónoma de Querétaro

O71

In Silico Analysis of Structural Motifs in Antibodies with Biotherapeutic Potential Against SARS-CoV-2

Wanda Yael Tlalmis Guzmán. Instituto Politécnico Nacional

O72

Expression of the CCD4-1, CCD4-2, CCD4-3, CCD4-4 genes in different tissues of the N4 morphotype of *Bixa orellana* L. by PCR

Diego Torres Pech. Centro de Investigación Científica de Yucatán A.C.

O73

Phytochemical profile of mexican myrtle (*Psidium sartorianum*) leaves extracts and infusion by traditional method

José Guadalupe Torres Salazar. Universidad Autónoma de Occidente

O74

A kalanchoe flammea extract in combination with drugs can help reduce the tumor growth of cancer cell lines in an in vivo model

Heriberto Abraham Valencia González. Instituto Nacional de Cancerología/UNAM

O75

Beneficial element or not? Exploring the effects of TiO₂ nanoparticles on common bean (Phaseolus vulgaris L.)

Jessica Denisse Valle García. Cinvestav Zacatenco

O76

Molecular characterization of binding site of β -CCB in oligodendroglial GABA_A receptor $\alpha 3\beta 2\gamma 1$

María Berenice Varela Correa. Universidad Nacional Autónoma de México

O77

Targeting Conserved Epitopes of Influenza Virus Proteins to Dendritic Cells for Generating a Heterotypic Immune Response

Felipe de Jesús Vázquez Alba. Universidad Autónoma del Estado de Morelos

O78

Functional Role Assessment of Deltasatellites in the Accumulation Dynamics of Begomoviral Titles in Host Plants

José Manuel Vázquez Rodríguez. Universidad de Colima

O79

Towards the Allotopic Expression of a Chimeric COX3 Gene in the Yeast Saccharomyces cerevisiae

Miriam Vázquez Acevedo. Universidad Nacional Autónoma de México

O80

Determination of the molecular mechanism of interaction between hGOS2 protein and the antiapoptotic protein Bcl2

Ligia Vega Becerra. Instituto Potosino de Investigación Científica y Tecnológica A.C.

O81

Involvement of Human Papillomavirus 16 E5 Protein in the Activation of the Wnt/ β -catenin Pathway

Jimena Yadira Velásquez Hernández. Instituto Nacional de Cancerología/UNAM

O82

Poly(vinyl formal) membranes in catheter manufacturing: a study of crosslinking effects and ai-enhanced drug release predictions

Igor Garcia Atutxa. Instituto Politécnico Nacional

O83

In silico studies of the substrate scope of Glycerol Dehydrogenase from E. coli

Julian L. Wissner. Universidad Nacional Autónoma de México



PLENARY LECTURES

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

BAJA CALIFORNIA ROCK ART

Enah Montserrat Fonseca Ibarra¹

¹Instituto Nacional de Antropología e Historia. Calle Ryerson Núm., 99,
Col. Centro, C.P. 22800, Ensenada, Baja California
E-mail: montserrat_fonseca@inah.gob.mx

Rock art is one of the most important material expressions of the hunter-gatherer cultures who inhabited the Baja California Peninsula for at least the past 12,000 years and likely longer. This paper provides an overview of the rock art traditions of the peninsula through an analysis of the primary techniques (painting and engraving), pigments and color palette, and associations with landscape elements. Our focus is on observed patterns that allow the interpretation of related stylistic, biotic, and ethnoterritorial boundaries.

THE GENETIC HISTORY OF MEXICO SEEN THROUGH A PALEOGENOMICS LENS

María C. Ávila Arcos

Universidad Nacional Autónoma de México, Laboratorio Internacional de Investigación sobre el Genoma Humano, 3001
Boulevard Juriquilla, 76230 Querétaro, Qro. Mexico.
E-mail: mavila@liigh.unam.mx

The peoples that have inhabited the territory occupied by Mexico since the first humans arrived ca. 14k years ago, have experienced several demographic changes. Some of the most emblematic fluctuations involve the rise and fall of large pre-Columbian civilizations, followed by a population collapse due to European colonization and infectious diseases in the 16th century. These processes have been vastly studied through archaeological, and historical evidence. In recent years, the use of genomics and paleogenomics has allowed to broaden the perspective of how these changes occurred and helped paint a more detailed picture of our genetic history. In this talk, I will discuss how some paleogenomic studies have revealed past population processes in Mexico. Also, I will emphasize the contributions from my lab and how we use paleogenomic approaches to characterize the diversity of pathogens circulating during the colonial period of Mexico City. Our results illustrate the rich information paleogenomics can offer for the study of past humans and their pathogens, prompting a necessary discussion on the multifaceted consequences of European Colonization.

THE COMMON BEAN (*PHASEOLUS VULGARIS*)—*RHIZOBIUM ETLI* N-FIXING SYMBIOSIS: UNRAVELING NOVEL PLANT REGULATORS THROUGH GENETIC/GENOMIC APPROACH

Georgina Hernández Delgado*, Rocío Rejero-Saavedra, Gladys E. Jiménez-Nopala,
Litzy Ayra Pardo, Sara I. Fuentes Membreño, Alfonso Leija Salas, Mario Ramírez Yáñez, Mishael Sánchez-Pérez,
Pablo Peláez Hernández, Lourdes Girard Cuesy, Timothy G. Porch

Centro de Ciencias Genómicas, UNAM, Campus Morelos Av. Universidad S/N, Cuernavaca, Mor. 62210.

* 7773290864 gina@ccg.unam.mx

The legume family accounts for one-third of the crop production in the world. A key to success of this family is its evolution of symbiosis with microorganisms that facilitate plant nutrient acquisition. The nitrogen fixation symbiotic association (SNF) between legume plants and the soil bacteria commonly known as rhizobia, that occurs in N-limited soils, is responsible for most of the atmospheric N₂ incorporation into biological systems and has the potential to reduce the chemical fertilization used in agricultural management.

An efficient SNF initiates with the communication between the two symbionts through molecular signals and specific receptors, followed by rhizobial infection through root hairs and the formation of root nodules where rhizobia are allocated and fix N₂. This complex process is tightly regulated in both symbionts. Advances in legume genomics and genetics, especially during the last 20 years, have contributed to our understanding of legume genes required for the effective symbiosis with rhizobia. Such knowledge has derived mainly from the study of the model legumes.

Our research system is the SNF of common bean (*Phaseolus vulgaris*) that is the most important legume for human consumption, serving as the main non-animal protein source for humans. Despite the agronomical importance of the common bean, the genomics/genetic research and knowledge of common bean SNF remains scant. Our group contributed to the development of common bean functional genomics: transcriptomics and metabolomics. Based in global gene expression information, we have analyzed through reverse genomic approach, global regulators genes such as transcription factors and microRNAs demonstrating their role and relevance in the SNF.

The characterization of symbiotic mutants, mainly from the model legumes, has been instrumental for the understanding of legume symbiotic genes. However, to date, only one common bean symbiotic mutant has been genetic and molecularly characterized. It is evident that isolation, characterization and mapping of symbiotic common bean mutants is lacking for a deeper understanding of the SNF process. We have screened an EMS-generated common bean mutant population to search for mutants altered in nodulation. We then proceeded to characterize three non-nodulating mutants appearing to be monogenic and recessive. Our analysis has revealed that each mutant is altered in a different early step of the SNF. The whole genome sequence analysis approach led to predict candidate mutated genes for each mutant. Our current research aims to demonstrate the responsible mutated gene in each line and to analyze its role and relevance in the SNF process.

CHANNELS OF COMMUNICATION IN SPERM

Alberto Darszon¹, Claudia Sánchez-Cárdenas¹, Mariano Buffone², Pablo E. Visconti³, Gerardo Orta¹,
José Luis de la Vega-Beltrán¹, Ana Romarowski², Enrique I. Oliver^{1,4}

¹Departamento de Genética del Desarrollo y Fisiología Molecular, Instituto de Biotecnología, Universidad Autónoma de México, Cuernavaca, Morelos, México. ²Instituto de Biología y Medicina Experimental, Consejo Nacional de Investigaciones Científicas y Técnicas (IBYME-CONICET), Buenos Aires, Argentina. ³Department of Veterinary and Animal Science, University of Massachusetts, Amherst, MA, USA. ⁴Department of Physiology and Biophysics, Miller School of Medicine, University of Miami, Miami, USA.

Dr. Alberto Darszon

Depto. Genética del Desarrollo y Fisiología Molecular, Instituto de Biotecnología
Universidad Nacional Autónoma de México

E-mail: darszon@ibt.unam.mx

Apdo. Postal 510-3, Cuernavaca, Morelos 62250

tel (52-555)6227650, FAX (52-777)3172388

ABSTRACT. Ion channels regulate sperm swimming, capacitation, and the acrosome reaction (AR), key functions for fertilization. Capacitation is a maturational process mammalian sperm must undergo in the female genital track that prepares them for the AR and fusion with the egg. This process requires coordinated metabolic and signaling pathways. We have found that external substrate removal diminishes sperm intracellular ATP, elevates their intracellular concentrations of Ca^{2+} ($[\text{Ca}^{2+}]_i$) and H^+ (pHi), cells hyperactivate and hyperpolarize. Energetic substrate replenishment (SER) causes $[\text{Ca}^{2+}]_i$ to decrease and enhances sperm fertilizing capacity, corroborating the close link between metabolism and $[\text{Ca}^{2+}]_i$. We have been studying which sperm ion channels participate in these events and our recent advances will be presented. Interestingly, we find capacitation induces $[\text{Ca}^{2+}]_i$ oscillations that had not been described. We are exploring how cytoplasmic and organellar Ca^{2+} changes contribute to these oscillations and to progesterone responses.

The acrosome is a lysosome-related vesicular organelle located in the posterior region of the sperm head, that fuses with plasma membrane during the AR. Recent findings indicate acrosomal alkalinization is important for the AR. We have documented that amphipathic weak bases such as Mibefradil (Mib) and NNC 55-0396 (NNC) cause an acrosomal pH (pHa) increase by accumulating inside this organelle in mouse and human spermatozoa. This happens although these two compounds are blockers of CatSper, a sperm specific Ca^{2+} channel responsible for the main Ca^{2+} currents in sperm. The pHa elevation induces Ca^{2+} efflux from the acrosome and external Ca^{2+} influx, resulting in $[\text{Ca}^{2+}]_i$ increases and AR. To unravel how pHa triggers these changes we have used mouse sperm as a model, pharmacological tools, and single-cell Ca^{2+} imaging. Mouse sperm were exposing to Mib, NNC and the lysosomotropic agent Gly-Phe- β -naphthylamide (GPN). The three compounds increase pHa and release acrosomal Ca^{2+} without compromising acrosomal membrane integrity. Our GPN observations indicate that the osmotic component does not significantly contribute to acrosomal Ca^{2+} release caused by the pHa rise. Inhibition of two-pore channel 1 (TPC1) reduced the $[\text{Ca}^{2+}]_i$ increase stimulated by acrosomal alkalinization. On the other hand, Ca^{2+} uptake triggered by pHa alkalinization was diminished by blocking Ca^{2+} release activated Ca^{2+} (CRAC) channels.

Our findings contribute to understand the fundamental participation of sperm ion channels in sperm physiology and reproduction.

MITOCHONDRIA IN GLOBAL AND LOCAL INTRACELLULAR CALCIUM SIGNALING

György Hajnóczky, Benjamin Cartes Saavedra, Arijita Ghosh, Raghavendra Singh, Victor Hugo Sánchez-Vázquez, Maria Teresa Castromonte, Elena Berezhnaya, Mate Katona, György Csordas, Erin L. Seifert, Suresh K. Joseph and David Weaver

MitoCare Center, Department of Pathology, Anatomy and Cell Biology, Thomas Jefferson University, 1020 Locust str Ste 527 Philadelphia, PA 19107, USA

The importance of mitochondria in health and disease has been highlighted in a plethora of studies spanning decades. A major regulator of mitochondrial activities and contributions to cell functions is Ca^{2+} . With the molecular identification of the mitochondrial Ca^{2+} transport mechanisms, genetic strategies have become available to study the mechanisms and physiological relevance of mitochondrial Ca^{2+} signaling, and patients have been diagnosed with mitochondrial Ca^{2+} -linked genetic diseases in the last decade. Initially, mitochondria were linked to the global cytoplasmic Ca^{2+} . However, with the advance of technology to measure discrete organelles within the cell, mitochondria were shown to be localized at contacts with the endoplasmic reticulum, plasma membrane, and other organelles. At these close contacts, the generation of spatiotemporally confined $[\text{Ca}^{2+}]$ spikes that can be many times larger than the global cytoplasmic $[\text{Ca}^{2+}]$, drive mitochondrial Ca^{2+} signaling. Because mitochondria and other organelles undergo permanent redistribution, the source of mitochondrial Ca^{2+} can dynamically change. Importantly, recent studies have revealed the role of local Ca^{2+} transfer in a range of debilitating diseases ranging from neurodegeneration to metabolic disorders. This talk will summarize recent advances on both the global and local aspects of mitochondrial Ca^{2+} control and highlight relevant studies of our group.

HISTAMINE: THE (RELATIVELY) NEW KID IN TOWN

José Antonio Arias-Montaño

Department of Physiology, Biophysics and Neurosciences
Center for Research and Advanced Studies (Cinvestav), National Polytechnic Institute
Av. IPN, 2508, 07360 Mexico City, Mexico
antonio.arias@cinvestav.mx

Histamine was detected in the brain in 1919 by John J. Abel, but its role as a neuromodulator became evident only several decades later when immunohistochemical methods proved the existence of a histaminergic system in the mammalian central nervous system (CNS). Via pre- and post-synaptic mechanisms, histamine regulates a variety of CNS functions, such as wakefulness, feeding, drinking, hormone release, body temperature, analgesia, and motor activity.

All histaminergic neurons are located in the hypothalamus (~64,000 neurons in the human) and send diffuse projections to the entire CNS. Most histaminergic axons do not make typical synaptic contacts, allowing for the release of the neurotransmitter along the fibers and thus its action on a large number of cells. Histamine effects are mediated by four G protein-coupled receptors (GPCRs) that belong to the class A, rhodopsin-like family, and three of these receptors (H_1 , H_2 and H_3) are widely expressed in the CNS and couple to different G proteins ($G\alpha_{q/11}$, $G\alpha_s$ and $G\alpha_{i/o}$, respectively).

In this talk, I will present data on the biochemical pharmacology and function/dysfunction of CNS histamine receptors at the cellular, molecular, and systemic levels. Aspects to be addressed include interaction with receptors to other neurotransmitters, the effect of H_3 receptor activation in the prefrontal cortex in a rat model of schizophrenia, mutation-related dysfunction of H_2 and H_3 receptors, histamine receptors in astrocyte function, and the effect of H_1 receptor blockade during the fetal stage on brain development.

INNOVATION BY EVOLUTION: BRINGING NEW CHEMISTRY TO LIFE

Frances H. Arnold

California Institute of Technology, USA

The most powerful design process ever invented is evolution: it generates incomparable functionality and works at all scales, from molecules to entire ecosystems. There is nothing like it in the world of human engineering. Humans have used evolution for biological design for thousands of years, choosing who mates with whom and who goes on to parent the next generation. We can now use evolution to explore the future of chemistry by engineering the catalysts of life, enzymes. I will describe how we can direct enzyme evolution to solve challenging chemical problems once thought to be out of reach of biology, and even of chemistry.

ROTARY ENGINES OF LIFE: EXPLORING ATP SYNTHASE AND CRISTAE FORMATION IN *POLYTOMELLA PARVA*

Diego González-Halphen¹

Miriam Vázquez-Acevedo¹, Marcos Ostolga-Chavarría¹ and Héctor Miranda-Astudillo²

¹Departamento de Genética Molecular, Instituto de Fisiología Celular, UNAM. Apartado Postal 70-242 CP 04510. Tel. (5255)-5622-5620. E-mail: dhalphen@ifc.unam.mx; ²Instituto de Investigaciones Biomédicas, UNAM, Mexico City, Mexico.

Mitochondrial F₁F_o-ATP synthase is pivotal in cellular bioenergetics, utilizing the electrochemical proton gradient generated by the respiratory chain to produce ATP. This rotary motor-like enzyme is ubiquitous across eukaryotic lineages, except for those lacking mitochondria. Despite its ancient origin, ATP synthases display significant variations in polypeptide composition and structural diversity, reflecting the evolutionary divergence among eukaryotes. Notably, mitochondrial ATP synthases in various organisms, such as the colorless chlorophycean alga *Polytomella parva*, include lineage-specific subunits.

The *Polytomella* F₁F_o-ATP synthase, a dimer with an estimated molecular mass of 1600 kDa, comprises 18 polypeptides per monomer. Among these, eight subunits—alpha, beta, gamma, delta, epsilon, a (Atp6), c (Atp9), and OSCP—are conserved across most eukaryotes and form the enzyme's primary functional core. Additionally, the algal enzyme features ten unique polypeptides, designated Asa1 to Asa10 (ATP Synthase Associated proteins).

We investigated the topological arrangement of *Polytomella* mitochondrial ATP synthase components using diverse experimental methods: cross-linking to detect subunit interactions, the yeast two-hybrid system, reconstitution with recombinant subunits, and partial dissociation to form sub-complexes. Subunit stoichiometry was inferred through cysteine residue labeling, and the overall structural characteristics were modeled using small-angle X-ray scattering data and electron microscopy image reconstruction. Several high-resolution structures of the enzyme are now available.

We discuss the influence of these atypical subunits on the enzyme's oligomerization and their role in defining the architecture of mitochondrial cristae, providing new insights into the functional complexity and evolutionary adaptation of mitochondrial ATP synthases.

THE JOURNEY OF GLUCOSE IN THE BODY: FROM ABSORPTION TO GLUCOSE UPTAKE BY THE MUSCLE

Amira Klip

The Hospital for Sick Children, Toronto, ON, Canada
E-mail: amira@sickkids.ca

Glucose is the universal fuel of most mammalian cells, and it is largely replenished through dietary intake. Glucose availability to tissues is paramount for the maintenance of homeostatic energetics, and hence, supply should match demand by the consuming organs. In the first part of this presentation, we will discuss the cellular barriers encountered by glucose in its transit at the levels of the absorbing intestinal epithelial wall, the renal epithelium mediating glucose reabsorption, and the tight capillary endothelia (especially in the brain)¹. Glucose transiting through these cellular barriers must escape degradation to ensure optimal glucose delivery to the bloodstream or tissues. The liver, which stores glycogen and generates glucose *de novo*, must similarly be able release it intact to the circulation. The idea will be presented that the universal problem of sparing glucose from catabolism in favor of translocation across the barriers posed by epithelia and endothelia is resolved through common mechanisms involving glucose transfer to the endoplasmic reticulum, from where glucose exits the cells via unconventional cellular mechanisms.

In the second part, I will outline the most up-to-date models for the mechanism for glucose entry into the muscle fibers in response to insulin². Finally, how this process may fail when muscle is exposed to saturated fats will be briefly discussed.

References

1. Klip A, De Bock K, Bilan PJ, Richter EA (2024) *Annu. Rev. Physiol.* **86**, 149-173. <https://doi.org/10.1146/annurev-physiol-042022-031657>
2. Sylow L., Tokarz VL, Richter EA, Klip A (2021) *Cell Metab.* **33**, 758-780. <https://doi.org/10.1016/j.cmet.2021.03.020>

AI APPROACHES IN PROTEIN BIOINFORMATICS

Daisuke Kihara PhD

Department of Biological Sciences, Department of Computer Science.
Purdue University

Protein Bioinformatics studies the relationships between the 3D structures, sequences, and functions of proteins using computational approaches. It is a very exciting era for protein bioinformatics, as many AI techniques are now used in various topics, including protein 3D structure prediction, structure modeling with cryo-electron microscopy images, protein design, and drug design. We will overview the current capabilities and the forefront of research development in these fields. Additionally, we will discuss the areas that still require further development in the future.

MEMBRANE REMODELLING IN ENDOCYTOSIS AND PHAGOCYTOSIS

Sergio Grinstein^{1,2}, Trieu Le¹, Michelle Maxson^{1,2}, Spencer Freeman^{1,2}

¹The Hospital for Sick Children and ²University of Toronto
Toronto, Canada

E-mail: sergio.grinstein@sickkids.ca

Endocytosis and phagocytosis are related processes involving in both cases the internalization of sections of the plasma membrane. The resulting endomembrane vesicles and vacuoles, known as endosomes and phagosomes respectively, undergo a progressive maturation process culminating in fusion with degradative lysosomes. The presentation will deal with two distinct but related topics: 1) the obstacles encountered by targets of phagocytosis and how these are cleared by dynamic processes involving cytoskeletal remodelling and redistribution of transmembrane proteins and 2) the role of the luminal pH, Rab-family proteins and LRRK2 –a kinase implicated in the pathogenesis of Parkinson's disease– in the endosomal maturation process. The application of advanced imaging techniques to these biological problems will be discussed in detail.

PLENARY SYMPOSIA

XXXIV NATIONAL MEETING OF BIOCHEMISTRY



IMPACT OF NANOTECHNOLOGIES APPLIED TO HUMAN HEALTH

**BACTERIAL GROWTH INHIBITED
BY HYDROPHOBIC NANO EMULSIONS**

Jesús Carlos Ruiz Suárez, Alejandra Gabriela Valdez Lara, Angela María Jaramillo-Granada,
Daniel Ortega Zambrano, Hilda Mercado Uribe

Cinvestav-Monterrey, Parque PIIT, Apodaca, NL
E-mail: cruz@cinvestav.mx

Concerned about the increasing resistance of bacteria to antibiotics normally used to treat infections around the world, efforts are needed to discover new strategies against microbial diseases. Among the newest therapeutic targets, the bacterial cell membrane is an important one. The reason is because in the event of a drastic alteration, it can cause bacterial death. We propose the utilization of hydrophobic molecules, namely propofol (PFL) and cannabidiol (CBD), dissolved in nanodroplets of oil, to effectively strike the membrane of two well known pathogens: *Escherichia coli* and *Staphylococcus aureus*. First, we carried out calorimetric measurements to evaluate the effects of these drugs on model membranes formed by bacteria-like lipids. We found that the drugs modify their transition temperature, enthalpy of cohesion and cooperativity, which indicates a strong disruption of the membranes. Subsequently, we demonstrate, using atomic force and fluorescence microscopy, that the drugs produce a visible alteration in real bacterial membranes. Finally, we performed experiments to evaluate the inhibition of colony-forming units, finding strong inhibition. Our results may have useful implications in the global effort to discover new ways to effectively combat the growing threat of drug-resistant pathogens. Especially in skin infections.

IMPACT OF NANOTECHNOLOGIES APPLIED TO HUMAN HEALTH

CHALLENGES AND OPPORTUNITIES OF NANOTECHNOLOGY IN THE DEVELOPMENT OF NEXT-GENERATION VACCINES

Francisca Villanueva-Flores^{1*}

Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada (CICATA)
Unidad Morelos del Instituto Politécnico Nacional (IPN). Boulevard de la Tecnología No.1036, C.P. 62790.
Xochitepec, Morelos, México.
*fvillanuevaf@ipn.mx

Nanotechnology has transformed vaccine development through the innovative use of nanoparticles, enhancing antigen stability and immune response. Notably, lipid nanoparticle-based vaccines, such as those from Moderna and Pfizer/BioNTech for SARS-CoV-2, encapsulate mRNA and demonstrate efficacy rates exceeding 90%. These particles protect mRNA against degradation and facilitate its delivery to target cells, essential for producing viral proteins and robust immune responses. Despite the challenge of requiring low-temperature storage, these vaccines have been crucial in managing the pandemic.

Beyond lipid nanoparticles, various other nanotechnologies have made significant contributions to vaccine development. Virus-like Particles (VLPs), which simulate viral structures without containing genetic material, induce strong immune reactions without infection risk. VLPs have been effectively utilized in hepatitis B and human papillomavirus (HPV) vaccines, offering a highly immunogenic presentation of antigens. Saponin-based adjuvants are also integral, enhancing antigen presentation and immune activation. The Matrix-M adjuvant in the Novavax COVID-19 vaccine exemplifies how these adjuvants can augment immune responses and vaccine efficacy. Polymeric nanoparticles are designed to deliver antigens and adjuvants methodically, releasing their payloads over time to ensure prolonged immune responses. Similarly, inorganic nanoparticles, like gold and silica, are adapted for antigen and adjuvant functionalization, improving immune responses and delivery targeting. Their stability and modifiability make them suitable for diverse vaccine applications.

However, scalability, reproducibility, and safety challenges remain, necessitating tailored regulatory frameworks to ensure product safety and efficacy. Integrating nanotechnology into vaccine development demands interdisciplinary collaboration among scientists, engineers, clinicians, and regulators to optimize benefits and navigate challenges. Future innovations could lead to more targeted and personalized vaccines, reducing side effects and enhancing efficacy. Advances in scalable manufacturing processes are essential to address global needs, particularly in under-resourced regions. Ongoing research is critical, potentially yielding new vaccines against various pathogens and enhancing global health security and pandemic preparedness.

IMPACT OF NANOTECHNOLOGIES APPLIED TO HUMAN HEALTH

VIRUS-BASED ENZYMATIC NANOREACTORS FOR ENZYME REPLACEMENT THERAPY

Rafael Vázquez-Duhalt

Center for Nanosciences and Nanotechnology, Universidad Nacional Autónoma de México.
rvd@ens.cnyn.uanm.mx

Enzyme replacement therapy (ERT) has been used to treat a few of the many existing diseases which are originated from the lack of, or low enzymatic activity. Exogenous enzymes are administered to contend with the enzymatic activity deficiency. Enzymatic nanoreactors based on the enzyme encapsulation inside of virus-like particles (VLPs) appear as an interesting alternative for ERT. VLPs are excellent delivery vehicles for therapeutic enzymes as they are biodegradable, uniformly organized, and porous nanostructures that transport and could protect the biocatalyst from the external environment without much affecting the bioactivity. Consequently, significant efforts have been made in the production processes of virus-based enzymatic nanoreactors and their functionalization, which are here presented. The use of virus-based enzymatic nanoreactors for the treatment of lysosomal storage diseases such as Gaucher, Fabry, and Pompe diseases, as well as potential therapies for galactosemia, and Hurler and Hunter syndromes will be discussed.

References

1. González-Davis O., Villagrana-Escareño M.V., Trujillo M.A., Gama P., Chauhan K. and Vazquez-Duhalt R. (2023) Virus-like nanoparticles as enzyme carriers for enzyme replacement therapy (ERT). *Virology* 580: 73-87.
2. Gama P., Juarez P., Rodríguez-Hernández A.G. and Vazquez-Duhalt R. (2023) Glucoseoxidase virus-based nanoreactors for smart breast cancer therapy. *Biotechnol. J.* 18: 2300199.
3. Chauhan K., Olivares-Medina C.N., Villagrana-Escareño M.V., Juárez-Moreno K.O., Cadena-Nava R.D., Rodríguez-Hernández A.G. and Vazquez-Duhalt R. (2022) Targeted enzymatic VLP-nanoreactors with β -glucocerebrosidase activity as potential enzyme replacement therapy for Gaucher's disease. *ChemMedChem* 17: e202200384
4. Mejía-Méndez J.L., Vazquez-Duhalt R., Hernández L.R., Sánchez-Arreola E., Bach H. (2022) Virus-like particles: fundamentals and biomedical applications. *Int. J. Mol. Sci.* 23: 8579.
5. Gama P., Cadena-Nava R.D., Juarez-Moreno K., Pérez-Robles J. and Vazquez-Duhalt R. (2021) Virus-based nanoreactors containing GALT activity for classic galactosemia therapy. *ChemMedChem* 16: 1438–1445.

NEW ASPECTS ON CELLULAR REGULATION OF SENESCENCE AND DIFFERENTIATION

NEONATAL T CELLS

Santana Calderón, M.A., Galindo-Albarrán, A., Rodríguez Jorge, O., Labastida-Conde, R.G.,
Hernández-Acevedo, G.N., Sánchez-Villanueva, J.A., Gutiérrez-Reyna, D.Y., Kempis-Calanis, L.A.

Centro de Investigación en Dinámica Celular, UAEM.
santana@uaem.mx

Cell function is controlled through regulatory networks that depend on the interaction of the cell with its particular environment and developmental status. The perinatal period is characterized by changes in the immune system from a rather innate response to a stimulus-specific response, that continues its development in the neonate, both in composition and function. After birth, the sudden encounter with an antigen-full world causes a big immune challenge. Neonatal immune cells are prepared for this transition with a unique profile, that establishes a tolerant window and innate inflammatory rapid response to pathogen encounter. Our research focuses on the identification of the transcriptional and epigenetic signatures of neonatal CD4⁺ and CD8⁺ T lymphocytes, which are cells of the adaptive immune system. The major function of CD4⁺ T cells is to coordinate the adaptive immune response and of CD8⁺ T cells, the elimination of damaged or infected cells. The response to activation signals through the T cell antigen receptor and other receptors opened new horizons on the regulatory networks of the cells. We have also characterized the difference between T lymphocytes from neonates born through caesarian section or physiological delivery and from premature babies (27 to 32 weeks of gestation). Our results outline important differences with the adult lymphocytes that should be taken into consideration for medical management during this very vulnerable period of our lives.

NEW ASPECTS ON CELLULAR REGULATION OF SENESCENCE AND DIFFERENTIATION

FROM SINGLE-EXON TO MULTIOXONS: MAPPING THE IMPACT OF INTRONLESS GENES ON CANCER BIOLOGY

Katia Aviña-Padilla¹, José Antonio Ramírez-Rafael¹, Octavio Zambada-Moreno¹,
Gabriel Emilio Herrera-Oropeza², Javier De Las Rivas³ and Maribel Hernández-Rosales¹

¹CINVESTAV-Irapuato, Lib. Norte Carretera Irapuato León Km 9.6, 36821 Irapuato, Guanajuato, Mexico,

²King's College London, Strand, London WC2R 2LS, UK

³Centro de Investigación del Cáncer-IBMCC, CSIC/USAL Salamanca, España

E-mail: katia.avinap@cinvestav.mx

Intronless genes (IGs) are a ubiquitous feature across eukaryotes, distinguished by their lack of introns. Unlike their multi-exon counterparts, IGs are not regulated by the splicing machinery, potentially resulting in lower post-transcriptional expression variability. This characteristic makes IGs promising candidates for clinical biomarkers with higher predictability and more straightforward regulation, presenting potential targets for therapeutic intervention¹. In the same context, microproteins (miPs), small proteins encoded by small open reading frames have emerged as critical regulators in various biological processes, including cancer. Given the complex nature of cancer, characterized by uncontrolled cell division and multiple dysfunctional biological processes, the identification of stable biomarkers is crucial. Our study focuses on the functional characterization of IGs and miPs in the human genome and on identifying unique expression profiles and interactomes across eight different cancer types. We conducted a comprehensive analysis of the human genome, identifying 940 protein-coding IGs functionally enriched in histone, transmembrane, and transcription factor roles; as well as 535 miPs belonging to 29 gene families. Differential expression analysis was performed on cancer datasets to determine the prevalence and expression patterns in various tumors. Approximately 35% of the identified IGs were differentially expressed across the analyzed cancer datasets. Notably, 78% of these differentially expressed IGs exhibited elevated expression levels in tumor cells. A highly conserved induction of a group of deacetylase-histones located in a region of chromosome 6, enriched in nucleosome and chromatin condensation processes, was observed across all studied tumors. The study also identified microproteins encoded by IGs that were deregulated in cancerous tissues, suggesting their involvement in tumorigenesis. The findings suggest that IGs and human-encoded miPs play crucial roles in the tumor phenotype. This research emphasizes the potential of IGs and miPs as biomarkers and therapeutic targets in cancer. The creation of a descriptive data repository and the identification of unique expression profiles and interactomes those biomolecules could provide valuable insights into their functional roles in cancer.

References

1. Katia Avina-Padilla, Jose Antonio Ramirez-Rafael, Octavio Zambada-Moreno, Gabriel Emilio Herrera-Oropeza, Guillermo Romero, Ishaan Gupta, and Maribel Hernandez-Rosales (2022). Lecture Notes in Computer Science, *Comparative Genomics*, 311-337.

NEW ASPECTS ON CELLULAR REGULATION OF SENESCENCE AND DIFFERENTIATION

**REACHING NEW HORIZONS IN CELLULAR SENESCENCE
RESEARCH BY LEVERAGING A BRAND
NEW MULTI-SENESCENT CELL TYPE CATALOG**

Carlos Anerillas^{1,3}, Dimitrios Tsitsipatis¹, Krystyna Mazan-Mamczarz¹, Reema Banarjee², Gisela Altés¹, Martin Salamini-Montemurri¹, Ana Cunningham², Jen-Hao Yang¹, Rachel Munk¹, Martina Rossi¹, Yulan Piao¹, Jennifer L. Martindale¹, Jinshui Fan¹, Chang-Yi Cui¹, Supriyo De¹, Kotb Abdelmohsen¹, Nathan Basisty², Allison B. Herman¹ and Myriam Gorospe¹

¹Laboratory of Genetics and Genomics and ²Translational Gerontology Branch, National Institute on Aging Intramural Research Program, National Institutes of Health, Baltimore, Maryland, USA; ³Centro de Biología Molecular Severo Ochoa, Consejo Superior de Investigaciones Científicas (CSIC), Madrid, Spain.
canerillas@cbm.csic.es

More than 60 years after Hayflick and Moorhead's definition of cellular senescence, the search for reliable markers to identify this state, especially outside of cell culture, continues. There is an urgent need to develop a comprehensive catalog of cell senescence markers covering the variety of cell types that exist within an organism beyond the classic models of senescence, which have primarily focused on human fibroblasts. In this project, named the SenCat, we profiled over 30 models of senescence triggered by different means in 14 different primary cell types. Leveraging our expertise in culturing multiple cell types, we induced senescence in primary human epithelial, endothelial, immune, stem-like, muscle, and adipose cells, as well as astrocytes and fibroblasts. Through extensive transcriptomic and proteomic profiling, we identified both shared and unique markers for each senescence model. These markers can be used to reliably identify senescent cells in vivo through techniques such as immunostaining or single-cell RNA-seq. Our observations revealed that while senescent cells do not seem to fully conserve specific markers across different cell types, they do maintain the activation and repression of broader pathways. These include the activation of p53, Epithelial-Mesenchymal Transition, NF- κ B, and lysosomal pathways, as well as the repression of E2F and Myc pathways. Moreover, our pathway and shared-marker analysis suggests that cell senescence can be confidently defined as a damage response mechanism aimed at influencing tissue repair while exhibiting a unique metabolic profile. In conclusion, we believe this ambitious approach to identifying senescent cells within tissues will usher in a new era for senescence research, which is crucial for advancing our understanding of all the scenarios where cell senescence is relevant.

ANTIMICROBIAL DRUG RESISTANCE AND GENOMIC EPIDEMIOLOGY OF SUPERBUGS

EVOLUTIONARY DYNAMICS OF CARBAPENEM-RESISTANT *PSEUDOMONAS AERUGINOSA* IN CHILE: THE IMPACT OF THE ST654 'HIGH RISK' CLONE

Andrés Opazo-Capurro^{1,2}, Omar Alejandro Aguilar-Vera³, Paulina González-Muñoz^{1,2,4}, Luis Amsteins-Romero^{1,2}, Monserrat Quiroga¹, Antonia Encina¹, Néstor Herrera-Chávez⁵, Mario Quezada-Aguiluz^{1,2,6}, Alejandro Aguayo-Reyes^{2,6}, Felipe Morales-León^{1,2,7}, Vijna Illesca⁸, Rodrigo Vera⁹, Fabiola Salgado¹⁰, Patricio Suazo¹¹, Luz María Fuenzalida¹², Helia Bello-Toledo^{1,2}, Santiago Castillo-Ramírez³, Gerardo Gonzalez-Rocha^{1,2}

¹Laboratorio de Investigación en Agentes Antibacterianos (LIAA), Facultad de Ciencias Biológicas, Universidad de Concepción, Concepción, Chile. ²Grupo de Estudio en Resistencia Antimicrobiana (GRAM), Universidad de Concepción, Concepción, Chile.

³Programa de Genómica Evolutiva, Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México, Cuernavaca, México. ⁴Departamento de Ciencias Biológicas y Químicas, Facultad de Medicina y Ciencia, Universidad San Sebastián, Concepción, Chile. ⁵Laboratorio de Microbiología, Hospital Regional Guillermo Grant Benavente, Concepción, Chile.

⁶Grupo de Estudio de Enfermedades Infecciosas de la Universidad de Concepción (GrEEEn-UdeC), Departamento de Medicina Interna, Facultad de Medicina, Universidad de Concepción, Concepción, Chile. ⁷Departamento de Farmacia, Facultad de Farmacia, Universidad de Concepción, Concepción, Chile. ⁸Hospital Hernán Henríquez Aravena, Laboratorio Clínico, Temuco, Chile. ⁹Hospital de Urgencia Asistencia Pública, Santiago de Chile, Chile. ¹⁰Hospital Las Higueras, Talcahuano, Chile.

¹¹Hospital Regional de Talca, Talca, Chile. ¹²Hospital del Salvador, Santiago de Chile, Chile.

Email: andopazo@udec.cl

Antibiotic resistance represents a major threat to public health, with its clinical significance largely driven by its remarkable ability to acquire or upregulate various resistance determinants. Infections caused by antibiotic-resistant bacteria are normally associated with high mortality and morbidity rates, as well as elevated economic impact. The main highly resistant species involved in hospital-acquired infections are *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and Enterobacterales. Among them, carbapenem-resistant isolates, particularly carbapenemase producers, represent a clinical challenge since few therapeutic options are available, such as polymyxins and β -lactam/ β -lactamase inhibitors.

Due to the significance of carbapenem resistance, it is crucial to investigate the dissemination of 'high-risk' clones and the role of mobile genetic elements involved in the spread of carbapenem resistance genes. Accordingly, we have been investigating the molecular epidemiology and the mobile genetic elements involved in the evolution of carbapenem-resistant *P. aeruginosa* (CRPa) in Chile, where it is the main etiological agent of ventilator-associated pneumonia (VAP). In this sense, we have characterized the predominance of the ST654 'high-risk' clone in Chile, which is commonly associated with KPC carbapenemase. Specifically, most of the isolates belonged to a sub-clade including KPC producers that also clustered with strains from Argentina and the USA. Moreover, we determined that the bla_{KPC} gene was embedded in a Tn4401 transposon, suggesting that this genetic element plays an important role in the acquisition of this gene by this lineage in Chile. All these findings indicate a divergent evolutionary pathway of ST654 in the country.

Our results underscore the significance of 'high-risk' clones that are resistant to last-resort antibiotics, emphasizing the importance of monitoring highly relevant pathogens by whole-genome sequencing techniques. Such techniques could provide evidence that will allow the adoption of control measures to prevent the spread of highly resistant clinical pathogens.

ANTIMICROBIAL DRUG RESISTANCE AND GENOMIC EPIDEMIOLOGY OF SUPERBUGS

GENOMIC DIVERSITY AND ANTIMICROBIAL RESISTANCE GENES OF *SALMONELLA ENTERICA* IN MEXICO

Adrián Gómez-Baltazar¹, Angélica Godínez-Oviedo¹ and Montserrat Hernández-Iturriaga¹

¹Departamento de Investigación y Posgrado en Alimentos, Facultad de Química, Universidad Autónoma de Querétaro, C.P. 76010, Santiago de Querétaro, Querétaro, México.
E-mail: adrian.gomez@uaq.mx

Salmonella enterica is one of the most important foodborne pathogens worldwide. In Mexico, from 2017 to 2021, the National Epidemiological Surveillance System reported an annual average of 67,492 cases of NTS. However, it is estimated that 4.5 million cases of foodborne NTS occur each year in this country, with one out of every 27 individuals suffering from salmonellosis. In this study, we investigated the genomic diversity, antimicrobial resistance profiles, and population structure of *Salmonella enterica* strains isolated from raw chicken in three states of central Mexico. One hundred ninety-two strains were analyzed through whole-genome sequencing, revealing a diverse serovar distribution, with Infantis, Schwarzengrund, and Enteritidis predominating. The prevalence of antimicrobial resistance genes varied significantly among states, years, seasons, and retail establishments.

All strains were found to harbor genes encoding resistance to metals, while virulence gene analysis revealed diverse virulotypes among different serovars. SNP analysis indicated clonal relatedness among strains, with significant clusters associated with global isolates from various sources. Furthermore, a broader perspective was gained by evaluating the MLST population structure and frequency of antimicrobial resistance genes in 2561 *S. enterica* strains from Mexico, predominantly isolated from food sources. The study identified the most prevalent serovars and sequence types and detected 78 antimicrobial resistance genes belonging to 13 antimicrobial classes. These findings underscore the importance of genomic surveillance in guiding public health efforts and resources to combat foodborne diseases and antimicrobial resistance in Mexico, providing valuable insights for targeted control measures and global public health strategies.

ANTIMICROBIAL DRUG RESISTANCE AND GENOMIC EPIDEMIOLOGY OF SUPERBUGS

**ONE HEALTH AND PAN GENOMIC EPIDEMIOLOGY
OF A SUPERBUG**

Santiago Castillo-Ramírez

Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México.

E-mail address: iago@ccg.unam.mx

Antibiotic resistance is a major threat to human and animal health. In this regard, antibiotic-resistant *Acinetobacter baumannii* is one of the most important nosocomial pathogens nowadays. However, we know very little about this bacterial species outside the clinic and a clear global view of its resistome is also lacking. In the first part of the talk, I will show how we have provided one of the first global views of the resistome of this species. In the second part of the talk, I will show some data demonstrating that animal and grass clones are distantly related to the major human clones and appear to have limited antibiotic resistance potential. However, I will also show cases of non-human lineages of this species closely related to the major human clones and with clinically relevant antibiotic resistance genes. Key to our approach is an integrated framework in which we analyse at the same time the phylogeography/genomic epidemiology and molecular evolution of the lineages under study. Given our recent results, we suggest that surveillance of this species must go beyond the clinical settings and consider the environment in a clear One Health framework

WANDERINGS IN BIOCHEMISTRY

TALES OF A PATHOGENIC YEAST

Irene Castaño-Navarro¹¹IPICYT. Instituto Potosino de Investigación Científica y Tecnológica, A.C., División de Biología Molecular.
E-mail: icastano@ipicyt.edu.mx

The yeast *Candida glabrata* is part of the microbiota in many healthy people but can cause serious infections in severely immunocompromised patients. In the last decades *C. glabrata* has become the second most frequent species responsible for *Candida* infections in the bloodstream (candidemia) or other sterile organs (invasive candidiasis). We are interested in understanding some of *C. glabrata*'s virulence attributes that allow it to adapt to the harsh conditions within its host and successfully persist in it. One of the most striking features of *C. glabrata* is its large repertoire of genes encoding for proteins called adhesins, anchored to the fungal cell wall, many of which confer a strong ability to adhere to mammalian epithelial and endothelial cells. Our studies revealed that the majority of these genes encoding *bona fide* adhesins and adhesin-like proteins (ALPs), are localized close to all the telomeres in *C. glabrata*'s genome. This led to the discovery that adhesins and other ALP-encoding genes are subject to negative regulation of transcription called subtelomeric silencing in many *C. glabrata* strains. We found that subtelomeric silencing depends on *cis*-acting DNA elements and *trans*-acting factors including the Sir complex, γ Ku, Abf1, Rif1 and Rap1 proteins. All of these elements induce the formation of chromatin loop structures that profoundly impact expression of these genes, suggesting that non-linear interactions between distant *cis*-acting regions that result in the formation of chromatin loops, are required for the appropriate expression of virulence-related genes in *C. glabrata*. We have also studied the important ability of *C. glabrata* to acquire antifungal resistance, which make these infections very difficult to treat. At the heart of the antifungal resistance is a relatively high phenotypic and genotypic variability among *C. glabrata* clinical isolates obtained from single patients. This led to the finding that *C. glabrata* infections are composed of clonal but phenotypically and genotypically diverse individuals resulting in subpopulations of resistant strains among a majority of susceptible individuals within a clonal population. However, the standard practice in clinical microbiological laboratories is to analyse one individual from each clinical sample, leading to underestimation of possible resistant isolates.

We will discuss this work and some possible repercussions for appropriate treatment of patients and prompt diagnosis of *C. glabrata*. References

Reference

1. Irene Castaño, Shih-Jung Pan, Margaret Zupancic, Christophe Hennequin, Bernard Dujon and Brendan P. Cormack. (2005). *Molecular Microbiology*, 55 (4): 1246-1258.
2. Alejandro De Las Peñas, Shih-Jung Pan, Irene Castaño, John Adler, Robert Cregg and Brendan Cormack (2003). *Genes and Development*, 17(18): 2245-2258.
3. Lluvia L. Rosas-Hernández, Alejandro Juárez-Reyes, Omar E. Arroyo-Helguera, Alejandro De Las Peñas, Shih-Jung Pan, Brendan P. Cormack and Irene Castaño*. (2008). *Eukaryotic Cell*, 7(12): 2168-2178.
4. Eunice López-Fuentes, Grecia Hernández-Hernández, Leonardo Castanedo, Guadalupe Gutiérrez-Escobedo, Katarzyna Oktaba, Alejandro De Las Peñas and Irene Castaño* (2018). *Genetics*, 210: 113-128
5. López-Marmolejo AL, Hernández-Chávez MJ, Gutiérrez-Escobedo G, Selene Herrera-Basurto M, Mora-Montes HM, De Las Peñas A, Castaño I. (2024). *Fungal Genet. & Biol.* 172: 103891

WANDERINGS IN BIOCHEMISTRY

WANDERLUST: FROM GENOME REARRANGEMENTS TO CELL DIVISION CYCLE IN RHIZOBIUMDavid Romero Camarena¹¹Programa de Ingeniería Genómica, Centro de Ciencias Genómicas-UNAM. Cuernavaca, Mor., México.

E-mail: dromero@ccg.unam.mx

Over the years, work in my laboratory has been concentrated on genome structure and genome rearrangements in the nitrogen-fixing bacterium *Rhizobium etli* CFN42. Its genome is composed by a chromosome and six large plasmids (p42a-p42f), ranging in size from 180 to 610 kb. It is common in science that unexpected results pick our curiosity, leading to shifts in research emphasis. In my case, the shift was motivated by the unexpected stability of one of the “plasmids” (p42e). Deeper analyses of this compartment showed all the hallmarks of a secondary chromosome, being conserved in related species, containing over 15% of their genes participating in central metabolic processes and harbouring two essential genes of unknown function at the time (*rdsA* and *rdsC*)¹. Both genes encode possible transcriptional regulators, *rdsA* being a sensor histidine/kinase hybrid protein, participating in a two-component signal pathway of unknown structure and *rdsC* possessing a helix-turn-helix motif. To assess their roles, we generated conditional knockdown (cKD) mutants in both genes, characterizing its effect by growth tests, microscopy and RNAseq analyses. Both cKD mutants revealed striking variations in cell division and shape. Instead of the normal bacillary form, a large fraction of the cells (cKD::*rdsA*, 63%; cKD::*rdsC*, 23%) of the cells displayed a coccoid morphology. Both cell types can divide at a somewhat slower rate, but bacillary cells loss cell polarity for division, characteristic of *Rhizobium*. RNAseq analysis of cKD::*rdsA* revealed global changes, with downregulated genes in at least five biological processes: cell division, peptidoglycan biogenesis, respiration, translation, and motility². A similar analysis of cKD::*rdsC* revealed down regulated genes mainly involved in central metabolism, with a few downregulated genes shared with cKD::*rdsA*, mainly for peptidoglycan biosynthesis³. These analyses show that *rdsA* and *rdsC* control separate pathways that both impinge on the determination of cell division and shape.

Recent analyses succeeded in identifying the response element that interacts with the *rdsA* sensor kinase. This is gene RHE_CH03575, also an essential gene, which encodes the CpdR response regulator. A conditional knockdown mutant in gene RHE_CH03575 display a high number of spherical cells (ca. 45%) and problems in cell division. Both phenotypes are reversed upon restoring normal expression of this gene. In vitro phosphotransfer experiments with purified RdsA and CpdR reveal that RdsA can be phosphorylated, and is able to transfer the phosphate to CpdR, as expected in a two-component system. Experiments are underway to evaluate, by RNAseq, genes that are directly controlled by CpdR.

References

1. Landeta, C., et al.. (2011). Plasmids with a Chromosome-Like Role in Rhizobia. *J. Bacteriol.* 193, 1317–1326.
2. Martínez-Absalón, S., et al.. (2022) RdsA Is a Global Regulator That Controls Cell Shape and Division in *Rhizobium etli*. *Front Microbiol.* 13, 858440.
3. Martínez-Absalón S., et al.. RdsC is a gene on the secondary chromosome in *Rhizobium etli* determining cell division and shape. In preparation

WANDERINGS IN BIOCHEMISTRY

THE ROAD OF CELLULAR COMMUNICATION IN DIFFERENT MODELS

Teresa Hernández Sotomayor

Sociedad Mexicana de Bioquímica

E-mail: ths.hern@gmail.com

Aluminium (Al) is the most abundant metal on Earth. It represents 7% of the all elements. The toxicity produced by this metal is widely documented in tropical acid mineral soils. It is the major factor limiting the productivity of crop species. Coffee is one of the most important crops economically worldwide mainly due to the production of the secondary metabolite caffeine. In the other hand, the fruits of the habanero chili pepper belong to the species *Capsicum chinense* Jacq. Habanero chilies are relevant for the economy due to their high demand and agricultural production. However, this crop as well as coffee are affected by different types of biotic and abiotic stress. We have developed a biological model in which suspension cells of *Coffea arabica* and *Capsicum chinense* have been used.

Particularly, we paid attention to Phospholipase type C (PLC), and we evaluated the PLCs transcription profile in coffee suspension cells from 14 days of culture that were treated or not with 100 μM to AlCl_3 for 30s or 3h. *CaPLC1* and *CaPLC2* did not showed change after the treatment. In contrast, *CaPLC3* and *CaPLC4* showed specific profile, down- and up-regulated, respectively. We obtained coffee PLC´s by heterologous expression using pColdII vector in *E. coli* BL21star strain, they were purified by affinity using Ni-NTA column and we found that Al-treatment in vitro, also affects PLC2 activity but not PLC4. Also, we obtained PLCs constructions (promTUB::PLC::mGFP) to visualize PLCs subcellular localization during AlCl_3 treatments. When YFP-PH_{PLCII} (PIP₂ biosensor) was transfected into coffee protoplast, a fluorescence signal changes near to polar growing point was showed, but when they were treatment with Al^{3+} this signal was not detected. With these data, the PLC role into signal-transduction process in response to aluminum stress should be established. In the other hand, morphological changes were evident 24 hours after inoculation (hai) of *C. chinense* cells with the microbial consortium, which consisted primarily of *C. ignotum*. High levels of diacylglycerol pyrophosphate (DGPP) and phosphatidic acid (PA) were found around 6 hai. These metabolic changes could be correlated with high transcription levels of diacylglycerol-kinase (*CchDGK1* and *CchDG31*) at 3, 6 and 12 hai and also to pathogen gene markers, such as *CchPR1* and *CchPR5*.

SIMULTANEOUS SYMPOSIA

XXXIV NATIONAL MEETING OF BIOCHEMISTRY



SYMPOSIUM ON NEUROBIOLOGY

**ROLE OF AUTOPHAY IN NEURODEVELOPMENT
AND BRAIN AGING**

Susana Castro-Obregon

Departamento de Neurodesarrollo y Fisiología, Instituto de Fisiología Celular, UNAM.
Circuito Escolar SN, Ciudad Universitaria, Coyoacán 04510. CDMX. México
E-mail: scastro@ifc.unam.mx

Morphogenesis during embryonic development is the result of a fine balance among cell growth, proliferation, differentiation and death. Lately, it has been described that cellular senescence, a stage in which the cells no longer divide and acquire a secretory phenotype that influences the surrounding tissue, can also be programmed and contributes to morphogenesis. During tissue homeostasis and in aging, the same cellular processes occur, although specific molecular regulators participate at different life stages. Autophagy is involved in each of these cellular processes. As the efficiency of autophagy decreases with age, and interventions that delay aging stimulate autophagy, understanding how it contributes to each cell fate is fundamental not only to understand embryonic development, but also the biology of aging.

Autophagy is a process that, among other things, removes damaged cytoplasmic material such as organelles and protein complexes, maintains genome stability, and sometimes contributes to unconventional secretion. We study in mice the role of autophagy and programmed senescence during neural tube development, from which both the brain and the spinal cord will form. In contrast to the transient physiological role of cellular senescence during development and wound healing, during aging senescent cells persist and accumulate. Due to their secretory phenotype, they alter the surrounding tissue promoting carcinogenesis, chronic inflammation, and paracrine senescence. Neurons are particularly dependent on autophagy because it contributes to processes such as axon growth, synaptic formation and plasticity. We discovered that age-associated autophagy dysfunction contributes to neuronal senescence. Currently, we study why autophagy fails with aging and whether restoring its function in middle-aged mice prevents neuronal senescence and delays aging features.

This work is funded by UNAM-PAPIIT IN205324, CONAHCyT CBF2023-2024-1669 and Pew Innovation Fund 2022.

SYMPOSIUM ON NEUROBIOLOGY

CIRCADIAN RHYTHMS AS REGULATORS OF ENERGY HOMEOSTASIS

Lucía Mendoza Viveros

Instituto Potosino de Investigación Científica y Tecnológica. Camino a la Presa de San José 2055, Col. Lomas 4ta Sección, San Luis Potosí, S.L.P., México, 78216. Tel. 444-831-2000 ext. 2057
lucia.mendoza@ipicyt.edu.mx

Daily rhythms in behavior and physiology are regulated by the circadian system, an endogenous, self-sustaining mechanism that allows organisms to adapt to 24-hour cycles in environmental conditions (i.e. light/dark) due to the Earth's rotation. In mammals, the circadian system has a hierarchical organization, with a central pacemaker in the hypothalamic Suprachiasmatic nucleus (SCN). The SCN coordinates its activity to generate 24 hr oscillations encoding the time of day, and propagates this information *via* neuronal signals to other brain regions, which act as relays to synchronize rhythms in peripheral organs. The molecular clock, expressed virtually in every cell, is comprised by negative feedback loops in transcription and translation of "clock genes". The core loop is formed by the heterodimer BMAL1/CLOCK, which are transcription factors that bind to E-boxes in the promoters of *Period/Cryptochrome*. After being translated and a delay of several hours, PER/CRY translocate to the nucleus and prevent the activity of BMAL1/CLOCK, hence inhibiting their own expression and generating ~24-hour transcriptional and translational oscillations. Among the physiological processes regulated by the circadian system is energy homeostasis. Astrocytes have emerged as important players in organismal energy metabolism, sensing nutritional signals and participating in producing the appropriate response (i.e. in feeding behavior, thermogenesis). We used a mouse model with a conditional deletion of the clock gene *Bmal1* in astrocytes to examine the role of the astrocytic circadian clock in metabolism and the response to an obesogenic diet. We found that mice lacking *Bmal1* in GFAP⁺ cells (*Bmal1* cKO) challenged with a high fat diet (HFD) accumulated less fat than their wild-type (WT) counterparts, through an increase in energy expenditure and thermogenesis. We also found evidence that the brown adipose tissue (BAT) of *Bmal1* cKO animals was more active, therefore we examined the transcriptome of the Ventromedial Hypothalamus, a key hub controlling BAT activity. We found that the lack of astrocytic *Bmal1* was associated with increased cellular stress in the VMH, possibly underlying the over-activation of the highly thermogenic BAT.

SYMPOSIUM ON NEUROBIOLOGY

SOCIAL NEUROSCIENCE AND CULTURE OF PEACE

Lorena Paredes¹, Simona Mosco Aquino² and Roberto E. Mercadillo^{3,4}

¹National School of Anthropology and History, Periférico Sur y C. Zapote S/N, Tlalpan, 14030, Mexico City,

^{2,3}Metropolitan Autonomous University, San Rafael Atlixco 186, Iztapalapa, 09310, Mexico City, ⁴National Council for Humanities, Sciences and Technology, Insurgentes Sur 1582, Benito Juárez, 03940 Mexico City.

E-mail: emmanuele.mercadillo@gmail.com

This work presents the Psicocalle Colectivo initiative, a transdisciplinary university effort to understand the phenomena of street life and the use of psychoactive substances through neurosciences, anthropology, and psychology. The proposal is framed by the Declaration for the Transition towards a Culture of Peace in the 21st Century.

An action-research model is proposed based on a notion of a culture of peace that recognizes marginalization and exclusion as forms of subtle violence. It promotes education for peace and compassion as ways to connect with others and apply scientific knowledge in everyday life. This approach aims to develop empathetic skills, a sense of agency, and introspection, fostering social commitment, reflection on Human Rights, and politically responsible scientific practice, where university students are defined as peacebuilders.

The phenomenon of street life affects thousands of people in major cities, exposing them to diseases and neuropsychiatric disorders. The research of Psicocalle Colectivo originated at the Universidad Autónoma Metropolitana-Iztapalapa, focusing on the effects of psychoactive substance abuse on the nervous system and behavior.

The model is based on environmental enrichment, a strategy tested in animal models that improves cognitive and motor capacities affected by psychoactive substances. This methodology translates into psychosocial interventions that seek to improve public spaces and foster social interaction and community sense among homeless people.

Psicocalle Colectivo advocates for education for peace that integrates dissent, indignation, and responsible disobedience. It proposes community actions that promote reflection and dialogue, raising awareness about street life and the use of psychoactive substances.

The Psicocalle Colectivo initiative seeks not only to make structural violence visible and denounce it but also to accompany and train those affected, fostering an active and ethical citizenship. The proposal emphasizes the importance of collective action and social commitment for the construction of sustainable and equitable peace.

This proposal is presented at the Congress of the Mexican Society of Biochemistry, highlighting the relevance of a transdisciplinary approach and the integration of education for peace in scientific research and community actions.

References

1. Paredes L, Mosco-Aquino J, Mercadillo, RE. (2023). Revista CoPaLa. Construyendo Paz Latinoamericana. 8(18), 316-345.
2. Paredes L, Enciso F, Mercadillo RE. (2022). International Journal of Drug Policy 99, 103440.

GROWTH CONTROL AND METABOLISM IN FUNGI

**BRANCHED-CHAIN AMINO ACID PATHWAY REGULATION
AND ITS CONNECTION TO CARBON METABOLISM
DURING YEAST DIAUXIC SHIFT**

Ximena Escalera-Fanjul, Olufemi Bolaji, Bernhard Drotleff, Alexander De Luna,
Theodor Alexandrov and Edda Klipp

Humboldt-Universität zu Berlin, Alemania, Berlín

In *Saccharomyces cerevisiae* TORC1 controls the transition between exponential growth and stationary phase during the diauxic shift. This transition requires a balanced remodeling of metabolic fluxes through central carbon and amino acid metabolism. The appropriate distribution of the metabolites shared by these pathways ensures that energy and biomass production match cell growth during this transition. In yeast, the TORC1 pathway is regulated by leucine, but how this branched chain amino acid (BCAA) is regulated over the diauxic shift and to which extent the BCAA pathway plays a role in the crosstalk between central carbon and amino acid metabolism is only partially characterized. To systematically address this, we generated a strain library of the pathway GFP-fusions, in wild type and all the deletion strains that did not disrupt the biosynthesis of any BCAAs (isoleucine, valine and leucine). We assessed the protein and metabolite profiles of the pathway, as well as the metabolite cell context throughout the diauxic shift, by high-throughput flow cytometry and untargeted LC-MS, respectively. We reveal that BCAA response to the diauxic shift matches reported TORC1 activity, the fraction of the pathway committed to leucine biosynthesis displays a fermentative profile, opposed to the fermento-respiratory signature of the portion of the pathway shared by the three BCAAs. We identify the key elements regulating the pathway, and how the misregulation of the BCAA pathway affects distant metabolic circuits including central carbon metabolism.

GROWTH CONTROL AND METABOLISM IN FUNGI

THE CONVERGENCE OF MOLECULAR AND METABOLIC RESPONSES TO THERMO-ACIDIC STRESS IN YEAST

Alfredo Martínez Jiménez, Dana Montserrat Galarza Ávila,
Prisciluis Caheri Salas Navarrete, Luis Caspeta Guadarrama

Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, Universidad Nacional Autónoma de México.
Av. Universidad 2001, Col. Chamilpa, Cuernavaca, Morelos, 62210.
Tel: 777 329 1601; email: alfredo.martinez@ibt.unam.mx

The production of ethanol with the yeast *Saccharomyces cerevisiae* faces adverse conditions, such as acidic pH, presence of acetic acid, and supraoptimal temperatures. These stressful conditions can halt fermentation prematurely. Hence, knowledge on yeast responses to these conditions is essential to provide a tolerant phenotype to another strain by targeted genetic manipulation or to systematically modifying operational parameters in the benefit of yeast fermentations. The physiological and whole-genome analyses conducted on the thermotolerant TTY23, acid tolerant AT22, and thermo-acid tolerant TAT12 yeast strains, generated by adaptive laboratory evolution experiments in our laboratory, will be presented. Dominated biological processes to tolerate acidic conditions include proton export, response to high-osmolarity and acetate transport. Mayor responses for thermo-tolerance involve transcriptional regulation of genes associated with stress responses to reactive oxygen species and heat-shock. The results also suggest that converge of both responses is regulated by adjustments of fermentative growth and stress responses by glucose signaling pathways. Overall, these results suggest that evolved strains adjust their metabolic responses by controlling the pH by H⁺ and acetic acid transport, adapting their metabolism and stress responses via glucose signaling pathways, controlling cellular ATP pools by regulating the translation and the *de novo* synthesis of nucleotides, and direct the synthesis, folding and rescue of proteins throughout the heat-shock stress response. Moreover, the motifs analysis in mutated transcription factors suggested a significant association with differentially expressed genes found in thermoacidic tolerant yeast strains, which have several advantages to be used in industrial ethanologenic fermentations.

ADVANCES IN PROTEIN STRUCTURE, FUNCTION AND EVOLUTION

**REDOX BIOCATALYSIS AT THE IBT-UNAM:
MOLECULAR BASIS AND APPLICATIONS
OF OXIDOREDUCTASES**

Marcela Ayala Aceves

¹Institute of Biotechnology UNAM, Av Universidad 2001 Chamilpa, Cuernavaca Morelos.
E-mail: marcela.ayala@ibt.unam.mx

Redox enzymes with multiple potential applications, ranging from material synthesis, to medical and bioremediation alternatives, are studied in the redox biocatalysis laboratory at IBt UNAM. Several examples will be discussed, such as the enzymatic synthesis of polycyclic disulfides from aromatic dithiols, which may be further converted into lineal polymers through ring opening polymerization (ROP)¹; the enzymatic oxidation of pollutants such as ethers, hormones and volatile sulfides^{2,3,4}; and the enzymatic generation of antibactericidal compounds. These reactions are catalyzed by highly oxidizing metalloenzymes, such as fungal laccases, haloperoxidases and peroxygenases. The particular drawbacks of these enzymes will be presented, for example their autoinactivation due the formation of reactive enzyme-based intermediates but also substrate-derived oxidizing species. Knowledge of the molecular basis of these enzymes' catalysis allows finding alternatives to tackle these drawbacks. The strategies to address the inactivating phenomena are varied, from optimizing reaction conditions by balancing substrate concentration to reduce competing reactions², protecting the enzyme through site-directed immobilization in order to reduce mass transfer limitations⁵, and also modifying the primary structure of the enzyme through protein engineering in order to eliminate potentially oxidizing sites^{6,7}. Finally, a perspective of future works will be outlined.

References

1. Sierra E, Miranda A, Castillo E, Hu H, Ayala M. *J Chem Technol Biotechnol* 92 (2017) 2691-2698
2. Mireles R, Ramirez-Ramirez J, Alcalde M, Ayala M. *J Fungi* 7 (2021) 608
3. Undiano E, Miranda-Molina A, Roman R, Ayala M. *Nat Prod Res* 36 (2021) 5353-5357
4. Cruz JC, García J, Ayala M. *TIP* 18 (2015) 5-12
5. Salcedo K, Torres-Ramírez E, Haces I, Ayala M. *Biocatalysis* 1 (2015) 33-43
6. Avelar M, Pastor N, Ramírez J, Ayala M. *J Inorg Biochem* 178 (2018) 125-122
7. Ramirez-Ramirez J, Martin-Diaz J, Pastor N, Alcalde M, Ayala M. *Int J Mol Sci* 21 (2020) 5734

ADVANCES IN PROTEIN STRUCTURE, FUNCTION AND EVOLUTION

METAL-CRYSTALLIN INTERACTIONS: THE BIOINORGANIC FACET OF CATARACT DISEASE

Liliana Quintanar¹

¹Department of Chemistry and Center for Research in Aging, Center for Research and Advanced Studies (Cinvestav),
Mexico City, Mexico.

E-mail: lilianaq@cinvestav.mx

Cataract disease is the leading cause of blindness worldwide and it is caused by crystallin damage and aggregation. Essential metal ions, such as copper and zinc, can induce the non-amyloid aggregation of human lens β/γ -crystallin proteins.¹ In this presentation, the mechanisms of metal-induced aggregation of human lens β/γ -crystallin, associated to cataract disease, will be discussed. While zinc-induced aggregation of γ -crystallin involves mostly metal-bridging mechanisms,² copper-induced aggregation displays a more complex scenario. Specifically, copper ions activate different site-specific mechanisms, including: protein unfolding, metal bridging, disulfide bridging, and copper reductase activity that leads to formation of free radical species.³ The role of Cys residues in this interesting redox chemistry will be discussed.⁴ The case of metal-induced aggregation of γ -crystallins will be contrasted to that of β B2-crystallin, which displays an ATCUN-like high affinity copper binding site that is typical of copper transport proteins.⁵ Our spectroscopic studies provide deeper understanding of the nature of the metal binding sites found in human lens β/γ -crystallins and their impact in protein stability and aggregation, providing further insights into the bioinorganic chemistry of cataract disease. This research has been funded by the National Council of Science and Technology (CONACYT) through grant no. PN2076; and the Ministry of Education (SEP) through the PRODEP-CA program.

References

1. Quintanar, L. et. al. (2016). *ACS Chem. Biol.* **11**, 263-272.
2. Domínguez-Calva, J.A. et. al., (2018). *Biochemistry*, **57**, 4959-4962.
3. Palomino-Vizcaino, G. et. al., (2023). *J. Amer. Chem. Soc.*, **145**, 12, 6781-679.
4. Serebryany, E.; Thorn, D. C.; Quintanar, L. (2021). *Exp. Eye Res.* **211**, 108707.
5. Tovar, M. et. al., (2023). *Inorg. Chem.*, **62**, 27, 10592-10604.

ADVANCES IN PROTEIN STRUCTURE, FUNCTION AND EVOLUTION

RECENT PROGRESS IN PREDICTING THE STRUCTURE AND REACTIVITY OF PEPTIDES AND PROTEINS

Joel Ireta

¹Departamento de Química, Universidad Autónoma Metropolitana-Iztapalapa, México City, México
E-mail: iret@xanum.uam.mx

In recent years theoretical methods used to investigate the structure and reactivity of proteins and peptides have advanced significantly. Primarily due to the improvement of artificial intelligence and electronic structure methods together with more powerful computers. Nowadays it is possible to predict the structure of proteins with artificial intelligence, or to follow the atomic movement of small peptides and proteins at an electronic structure level of precision, using forcefields trained with electronic structure calculations and artificial intelligence. In this talk i will review some of these advances as well as the shortcomings of these methods. Moreover, I will present new ways to describe the protein structure and the application of electronic structure to investigate the properties of small peptides.

ADVANCES IN PROTEIN STRUCTURE, FUNCTION AND EVOLUTION

SEARCH FOR NEW BACTERIAL ENZYMES WITH POTENTIAL CLINICAL OR BIOTECHNOLOGICAL APPLICATIONS

Rosario A. Muñoz-Clares

Departamento de Bioquímica, Facultad de Química, UNAM
Ciudad Universitaria 04510, Ciudad de México, México.

E-mail: clares@unam.mx

Bacteria colonize every habitat on Earth, and many are important pathogens, due to their enormous metabolic versatility, which remains largely unexplored. They are therefore a significant source of novel proteins involved in undescribed metabolic pathways with potential clinical or biotechnological applications. To explore this potential, we chose to study the aldehyde dehydrogenases (ALDHs) from *Pseudomonas aeruginosa*, a soil and water bacterium and an important animal and plant pathogen. ALDHs catalyze the NAD(P)⁺-dependent oxidation of a wide variety of endogenous and exogenous aldehydes and play important roles in metabolism and detoxification. The *P. aeruginosa* strain PAO1 chromosome contains 23 genes coding for ALDH proteins, of which we have studied five: PA5372 (also known as *PaBADH*), PA5312 (also known as *PaPauC*), PA4189, PA0219, and PA2125. To determine their physiological functions and their structure-function relationships, both crucial for their potential use, we employed several experimental approaches: analysis of the genomic context and induction of their genes; growth analysis of PAO1 mutant strains defective in these genes; kinetic characterization and inhibition of the recombinant enzymes; determination of their tridimensional structure by crystallography or homology modeling; docking of putative aldehyde substrates into their active sites; obtention of these enzyme variants by site-directed mutagenesis; and phylogenetic relationships studies. PA5372, PA5312, PA4189, and PA0219 oxidize aldehydes bearing an amino group and can use most of them as “in vitro” substrates. However, despite their apparent promiscuity, their selectivity for specific aminoaldehydes exquisitely suits their “in vivo” roles. On the other hand, PA2125 preferentially oxidizes hydroxyl derivatives of benzaldehyde. In this talk, I will briefly present and discuss the results of my research group, which lead us to propose PA5372 and PA5312 as potential drug targets to combat *P. aeruginosa* infections, and PA2125 for possible use in bioremediation of contaminated soils and waters.

Acknowledgements. I acknowledge the financial support provided by PAPIIT-DGAPA, UNAM, grants IN204708, IN220317, IN222720, and IN203423, and CONAHCYT grants 16712 and 283524.

EDGES IN MITOCHONDRIAL KNOWLEDGE

MITOCHONDRIAL COMPLEMENT 1Q BINDING PROTEIN (C1QBP) INTERACTS WITH CYCLOPHILIN D AND REGULATES MOUSE HEART OXIDATIVE PHOSPHORYLATION AND PERMEABILITY TRANSITION

Manuel Gutiérrez Aguilar^{1,4*}, Paula J. Klutho¹, Lihui Song¹, Rodrigo Aguayo-Ortiz⁵, Christopher P. Baines^{1,2,3}

¹Dalton Cardiovascular Research Center, ²Department of Biomedical Sciences, ³Department of Medical Pharmacology and Physiology, University of Missouri, Columbia, MO 65211, U.S.A. ⁴Department of Biochemistry, ⁵Department of Pharmacy, Facultad de Química, Universidad Nacional Autónoma de México, México City 04510, México
bainesc@missouri.edu

Opening of the mitochondrial permeability transition (MPT) pore often underlies heart dysfunction. This pore is activated in response to calcium ions in a process dependent of Cyclophilin D (CypD) isomerase activity. CypD can bind to several proteins and enzymes inside the mitochondrial matrix including subunits of the ATP synthase and mitochondrial Slc25a carriers [1]. We have previously found Complement 1q-binding protein (C1qbp) as an in vitro CypD-binding target [2]. Here we further tested the interactions between C1qbp, CypD, and the MPT pore using molecular dynamics simulations and by generating mice with either C1qbp overexpression or gene silencing. Our results show that C1qbp binds to CypD and alters MPT. Moreover, mice with C1qbp downregulation show abnormal oxidative phosphorylation, cardiac dysfunction and cell death in a model of myocardial infarction. Overall, the results highlight the relevance of C1qbp as a regulator of the calcium-induced MPT pore, mitochondrial homeostasis and sensitivity to injury in the heart.

References

1. Zoratti M, Biasutto L, Parrasia S, Szabo I. Mitochondrial permeability transition pore: a snapshot of a therapeutic target. *Expert Opin Ther Targets*. 2024 Jan-Feb;28(1-2):1-3.
2. McGee AM, Baines CP. Complement 1q-binding protein inhibits the mitochondrial permeability transition pore and protects against oxidative stress-induced death. *Biochem J*. 2011 Jan 1;433(1):119-25.

EDGES IN MITOCHONDRIAL KNOWLEDGE

LRPPRC AND SLIRP SYNERGISE TO MAINTAIN SUFFICIENT AND ORDERLY MAMMALIAN MITOCHONDRIAL TRANSLATION

Diana Rubalcava-Gracia, Kristina Bubb, Fredrik Levander, Amelie V. August, Stephen Burr, Patrick Chinnery, Camilla Koolmeister, Nils-Göran Larsson.

Karolinska Institutet, Department of Medical Biochemistry and Biophysics, Suecia, Estocolmo

In mammals, the leucine-rich pentatricopeptide repeat protein (LRPPRC) and the stem-loop interacting RNA-binding protein (SLIRP) form a complex in the mitochondrial matrix that is required throughout the life cycle of most mitochondrial mRNAs. Pathogenic mutations in the *LRPPRC* and *SLIRP* genes cause mitochondrial diseases in humans, but the functions of the corresponding proteins are incompletely understood. We show that loss of SLIRP causes a decrease of complex I levels and increased interactions between mitoribosomes and translation factors in liver. We genetically disrupted complex formation between LRPPRC and SLIRP *in vivo* by generating mouse knock-in lines and found that LRPPRC is partially degraded and SLIRP disappears when both proteins cannot interact. Interestingly, livers from *Lrpprc* knock-in mice had reduced mitochondrial translation except for a marked increase in the synthesis of ATP8. We also report that introduction of a heteroplasmic mtDNA mutation (m.C5024T of the tRNA^{Ala} gene) into *Slirp* knockout mice has an additive effect leading to embryonic lethality and reduced growth of mouse embryonic fibroblasts. To summarize, we report that LRPPRC and SLIRP synergize to maintain complex I levels and coordinate the preferential translation of mitochondrial transcripts in a tissue specific manner to sustain mtDNA gene expression.

EDGES IN MITOCHONDRIAL KNOWLEDGE

THE DIVERGENT BIOGENESIS OF THE RESPIRATORY COMPLEX III IN MALARIA PARASITES

Aldo E. García Guerrero, Jessica Pita Aquino, Rebecca Marvin, Amanda Mixon-Blackwell and Paul Sigala

Department of Biochemistry School of Medicine, University of Utah
E-mail: aldo.garcia@biochem.utah.edu

Malaria is a heavy burden on public health, affecting millions of people and causing thousands of deaths every year. *Plasmodium falciparum* remains as the main causative agent of severe malaria. The lack of an effective vaccine and the surge of resistant parasites against frontline therapies highlight the urgency of understanding the unique parasite biology to identify novel vulnerabilities. Plasmodium parasites have a complex life cycle that involves two hosts: mosquitoes and humans. In humans, parasites initially invade hepatocytes and subsequently red blood cells. Our lab has focused its studies on the blood stage, as it is the symptomatic phase of malaria. During red blood cell invasion, parasites develop in a heme-rich environment. Therefore, heme homeostasis is critical for parasite survival, as it needs to properly contain the toxic labile heme and use it as a cofactor for the essential activity of the mitochondrial respiratory complexes. The respiratory Complex III is currently targeted by the antimalarial atovaquone, which specifically binds to the cytochrome *b* subunit. However, parasites resistant to atovaquone have emerged. In our lab, we aim to explore new vulnerabilities beyond cytochrome *b*. During my talk, I will discuss the divergency of Complex III and its biogenesis, focusing on the hemylation of cytochrome *c*₁ and the peculiarities in the biogenesis of the Rieske subunit. We propose that these distinct differences between malaria parasites and mammals in Complex III biogenesis could serve as potential targets to combat parasite infection.

References

1. Sigala PA, Goldberg DE. The peculiarities and paradoxes of Plasmodium heme metabolism. Annu Rev Microbiol. 2014;68:259-78. Epub 20140616. doi: 10.1146/annurev-micro-091313-103537.
2. Evers F, Cabrera-Orefice A, Elurbe DM, Kea-Te Lindert M, Boltryk SD, Voss TS, Huynen MA, Brandt U, Kooij TWA. Composition and stage dynamics of mitochondrial complexes in Plasmodium falciparum. Nat Commun. 2021;12(1):3820. Epub 20210621. doi: 10.1038/s41467-021-23919-x.
3. García-Guerrero AE, Marvin RG, Mixon-Blackwell A, Sigala PA. Biogenesis of cytochromes *c* and *c*₁ in the electron transport chain of malaria parasites. bioRxiv. 2024. doi: 10.1101/2024.02.01.575742.
4. Espino-Sanchez TJ, Wienkers H, Marvin RG, Nalder SA, Garcia-Guerrero AE, VanNatta PE, Jami-Alahmadi Y, Mixon Blackwell A, Whitby FG, Wohlschlegel JA, Kieber-Emmons MT, Hill CP, Sigala PA. Direct tests of cytochrome *c* and *c*₁ functions in the electron transport chain of malaria parasites. Proc Natl Acad Sci U S A. 2023;120(19):e2301047120. Epub 20230501. doi: 10.1073/pnas.2301047120.

EDGES IN MITOCHONDRIAL KNOWLEDGE

MITOCHONDRIAL RESPIRASOME: DIVERSITY AMONG CONSERVED FUNCTION

Héctor V. Miranda-Astudillo

Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas,
Universidad Nacional Autónoma de México, México.
hmiranda@iibiomedicas.unam.mx

Historically, eukaryotic organisms have been categorized into four still influential ‘kingdoms’: plants, animals, fungi and protists. These ‘kingdoms’ remain textbook representatives of the eukaryotic domain of life, but it has long been clear that they do not properly represent the high diversity among these organisms. Nowadays, it is clear that all extant organisms have been evolving for the same period of time and none are lower or higher. Currently, many well-established super-groups now encompass much more diversity than the traditional kingdoms¹. The current consensus on eukaryotic taxonomy englobes up to 13 different supergroups reaching from the eukaryotic origin., where the original ‘kingdoms’ are now distributed among all the supergroups, the former plant kingdom is now included in Chloroplastida supergroup, while animal and fungi kingdoms are encompassed in Opisthokonta, finally, the former protist kingdom is distributed among the rest of the actual supergroups². Nevertheless, the anthropogenic point of view in Science and the remnant idea of an “eukaryotic complexity scale” have influenced the way to study the bioenergetics through decades, focusing often only on a few representative species of plants, animals and fungi.

In recent years, several groups around the world have started to expand the horizon of bioenergetics, focusing in the study of new species belonging to different eukaryotic lineages. As consequence, clearly divergent structures has been described among all the studied supergroups, these structural features includes additional associated peptides, extra structural domains and even fully new architecture of several respiratory complexes. Here, we present the state of the art of the mitochondrial complexes structural biology, with special interest on the consequences over the ultrastructure of the mitochondrial cristae.

Acknowledgments. This research was supported by grant IA204524 (PAPIIT, DGAPA, UNAM) and by the Institutional Program (IIBO-UNAM) “Production of biomolecules of biomedical interest in microorganisms”.

References

1. Keeling, P. J. & Burki, F. Progress towards the Tree of Eukaryotes. *Curr. Biol.* 29, R808–R817 (2019).
2. Burki, F., Roger, A. J., Brown, M. W. & Simpson, A. G. B. The New Tree of Eukaryotes. *Trends Ecol. Evol.* 35, 43–55 (2020).

INSIGHTS INTO PLANT ADAPTATION: FROM GROWTH TO RESILIENCE

**DECIPHERING THE EPIGENETIC NETWORK
OF PLANT STEM CELL NICHE
MAINTENANCE AND PLASTICITY**

María de la Paz Sánchez, Diego Ornelas, Carlos Cortés, Edoardo Yañez,
Adriana Garay-Arroyo and Elena Álvarez-Buylla

Instituto de Ecología, Universidad Nacional Autónoma de México. 3er Circuito Ext.
Ciudad Universitaria. UNAM. México D.F. 04510.
Email address: mpsanchez@ieecologia.unam.mx

Plant Stem Cell Niches (SCN) are undifferentiated cells with the potential to self-renew and differentiate into progenitors and then into specific cell types. Two main SCNs, situated in the Shoot Apical Meristem (SAM) and Root Apical Meristem (RAM) are responsible for generating the aerial and root tissues of the plants. The Arabidopsis Root Stem Cell Niche (RSCN) is established and maintained by a complex gene regulatory network, where epigenetic processes play essential roles. The Trithorax Group (TrxG) is an epigenetic complex that regulates transcriptional activation through the deposition of H3K4me3 and H3K36me3 marks.

To gain insight into the role of TrxG proteins in the RSCN, this work focuses on the study of the role of TrxG proteins in the maintenance of RSCN and their implications in the RSCN plasticity during regeneration processes. Our results display different roles of some TrxG proteins in the maintenance of columella stem cells, regulation of quiescent center cell division and regulation of the auxin response in the RSCN. These functions also impact root regeneration processes, supporting the participation of TrxG in the RSCN plasticity.

Fundings. UNAM-DGAPA-PAPIIT: IN206223, IN213524 and CONAHCyT: CBF2023- 2024-433 and CBF2023-2024-1002.

INSIGHTS INTO PLANT ADAPTATION: FROM GROWTH TO RESILIENCE

VISUALIZING REACTIVE OXYGEN SPECIES IN LIVING PLANT CELLS UNDER BIOTIC AND ABIOTIC RESPONSES

Luis Cárdenas, Samantha E. Ryken, Janet Palacios-Martínez, Ana I. Chávez-Martínez, Diego Flores-Bautista¹, Juan E. Olivares, Gabriel Corkidi, Adán Guerrero and Magdalena Bezanilla

Instituto de Biotecnología, UNAM. Cuernavaca, Morelos, México. CP 62210.
luis.cardenas@ibt.unam.mx

Calcium and reactive oxygen species (ROS) play important roles in plant physiological processes, including development, hormonal signaling, biotic and abiotic stress responses. ROS activation of calcium channels influences functions such as stomatal opening and the maintenance of calcium gradients in *Arabidopsis* root hair cells, and pollen tubes. We have previously used “Hyper”, a hydrogen peroxide-specific probe in *Arabidopsis* plants to depict the ROS distribution during plant root and root hair growth. However, more recently, by utilizing a calcium-specific probe (GCamp) and Hyper, a hydrogen peroxide-specific probe, we simultaneously examined these dynamics in *Physcomitrium patens*. A multicolor spinning disk confocal imaging strategy enabled simultaneous visualization of calcium and ROS responses to chitin oligomers, a fungal-derived elicitors that activate plant defense pathways. Chitin oligomers induce similar signatures in calcium and ROS responses, characterized by a complex oscillatory landscape with up to two distinct yet intricate patterns. These patterns exhibit a principal oscillatory behavior where calcium and ROS responses are anti-phase locked. Treatment with an inhibitor of NADPH oxidase inhibitors resulted in a diminished response in ROS production. Furthermore, NADPH oxidase mutants in *P. patens*, display a ROS oscillatory landscape similar to that of the wild type, but with visually distinct residence times. This implies that factors beyond NADPH oxidase and calcium channels contribute to ROS and calcium generation in response to chitin, suggesting the involvement of alternative pathways for ROS production. This study depicts the intricate and adaptable signaling systems in plants.

Funded by DGAPA IN-210321, CV200519, and a DGAPA-UNAM sabbatical scholarship.

INSIGHTS INTO PLANT ADAPTATION: FROM GROWTH TO RESILIENCE

ROLE OF BENEFICIAL RHIZOBACTERIA IN PLANT ADAPTATION TO ENVIRONMENTAL STRESS

José López-Bucio, Kirán Rubí Jiménez-Vázquez, José López Hernández,
Jesús Salvador López Bucio, Joseline Suhail Alejo Guerra

Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo,
Edificio B3, Ciudad Universitaria, C. P. 58030, Morelia, Michoacán, México.
Tel. 4432452898; E-mail: jlbucio@umich.mx

Plants interact with a myriad of bacterial species that extend physiological functions, promote plant growth and their adaptation to challenging environments. Release of root exudates represents an effective strategy to recruit beneficial microbes that assist plants to take up nutrients, and resist stress and pathogen challenges. The plant-microbe relationships rely upon the emission of complex mixtures of volatiles, secondary metabolites, and plant and microbial hormones.

The rhizosphere represents an ecosystem where different groups of microbes interact via interspecific and trans-kingdom communication that involves mutual recognition. Recent advances indicate that roots are able to detect bacterial quorum-sensing molecules, mainly *N*-acyl-*L*-homoserine lactones (AHLs) and cyclodipeptides (CDPs), along with the more common root growth regulating phytohormones auxins and cytokinins that configure root morphogenesis and depend on the microbial species and culture conditions.

Research using *Arabidopsis thaliana* as target organism helped to clarify the specific the contribution of *Pseudomonas*, *Bacillus*, *Micrococcus*, and *Achromobacter* species isolated from different rhizospheres in plant growth promotion. These species may contribute to root branching, phosphate acquisition, and root waving and skewing. This root behavior is thought to contribute for a better soil exploration and to the search of water and nutrient patches for plant survival under extreme environments such as salty soils. Through modulating root branching and directional growth, bacterial metabolites account for a better adaptation of plants to the ever-changing environment.

INSIGHTS INTO PLANT ADAPTATION: FROM GROWTH TO RESILIENCE

PLANT SIGNALING MECHANISMS UNDER ENERGY STRESS

Iván Rodríguez-Cruz, Eleazar Martínez-Barajas, Patricia Coello*

Departamento de Bioquímica, Facultad de Química, UNAM.

Plants, as sessile organisms, must adapt to varying growing conditions, including nutrient deficiencies, water limitation, and fluctuations in temperature and day length.

They rely on sunlight to synthesize sucrose, the primary carbohydrate for energy distribution and growth. At night, energy is derived from the degradation of starch, a reserve material synthesized during the day. The transition between day and night creates an energy imbalance detected by Sucrose Non-fermenting Related Kinases (SnRK1), which act as energy sensor. By phosphorylating metabolic enzymes and transcription factors, SnRK1 can produce significant changes that allow plants to adapt to these fluctuating conditions. SnRK1 is a complex comprising a catalytic α subunit, and a β and γ regulatory subunits. The regulatory β subunits (SnRK1 β 1 and SnRK1 β 2) and a plant-exclusive γ subunit (SnRK1 $\beta\gamma$) contain a carbohydrate-binding module (CBM) at their N-terminus. We observed that these subunits bind maltose, a key product of starch degradation, and this binding positively regulate SnRK1 activity. Association of SnRK1 with starch metabolism is suggested in *snrk1* mutants, that accumulate starch at the end of night. DPE2 an enzyme involved in maltose metabolism during starch degradation, is phosphorylated by SnRK1 at three different sites, enhancing its activity. In this presentation, I will discuss how SnRK1 contributes to plant responses to energy stress induced by nutrient limitation during the day-night transition.

PAPIIT IN201922, PAIP 5000-9126

SYMPOSIUM ON BIOCHEMISTRY AND MOLECULAR BIOLOGY OF BACTERIA

PHENAZINES, BIOTECHNOLOGICAL COMPOUNDS PRODUCED BY *PSEUDOMONAS AERUGINOSA*: GENE REGULATION AND OVERPRODUCTION

Miguel Cocotl Yañez¹, Fernando Montelongo-Martínez¹, Misael Josafat Fabian del Olmo¹, Abigail González Valdez², Gloria Soberón-Chávez²

¹Departamento de Microbiología y Parasitología, Facultad de Medicina, ²Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas. Universidad Nacional Autónoma de México, Ciudad de México, México.
E:mail: mcocotl@comunidad.unam.mx

Pseudomonas aeruginosa produces five phenazines with redox activity: pyocyanin (PYO), phenazine-1-carboxamide (PCN), 1-hydroxyphenazine (1-OHPHZ), 5-methylphenazine-1-carboxylate (5-MPCA) and phenazine-1-carboxylic acid (PCA) [1] designated *phzA1B1C1D1E1F1G1* and *phzA2B2C2D2E2F2G2*, are homologous to previously studied phenazine biosynthetic operons from *Pseudomonas fluorescens* and *Pseudomonas aureofaciens*. Functional studies of phenazine-nonproducing strains of fluorescent pseudomonads indicated that each of the biosynthetic operons from *P. aeruginosa* is sufficient for production of a single compound, phenazine-1-carboxylic acid (PCA). Except for 5-MPCA, all other phenazines have shown antifungal and antibacterial activity. Moreover, PCA is used as a biopesticide that can prevent and control crop diseases. PYO is the most studied phenazine that serves as a virulence factor but also it displays antitumoral activity and it has been used as a bio-dye and biosensor. PYO synthesis and its gene regulation have been mainly studied in the type strains PAO1 and PA14. Also, due to the potential uses of phenazines in different biotechnology fields, there is a search to obtain overproducing strains [2]. With regards to this, *P. aeruginosa* ID4365 is an environmental strain that overproduces PYO compared with the type strains [3].

Initially, in this project, we determine how the posttranscriptional Rsm system controls phenazines production. We found that *rsmA* inactivation, which codes for the effector of the posttranscriptional Rsm system, reduces rhamnolipids and elastase synthesis, and virulence is abolished. However, PYO increases 5-fold compared with the wild-type strain in the deficient phosphate medium PPGAS. Since PYO is deleterious for other microorganisms, we studied the protective response against PYO overproduction. By a proteomic analysis, we found that levels of 194 proteins increase when *rsmA* is inactivated and some of these proteins are related to redox homeostasis including alkyl-hydroperoxides, catalases, and also heat shock proteins. Using transcriptional and translational fusions we determine how RsmA controls this protective response. Since *rsmA* inactivation increases PYO production we explored whether this strain can be suitable for phenazines production. HPLC analysis showed that *rsmA* mutation also increases OH-PHZ and PCA levels but preferentially PYO. Then, we generated additional mutations on this strain to obtain a PCA overproducing strain. Finally, PCA and PYO production was improved using different culture conditions.

References

1. Mavrodi DV, Bonsall RF, Delaney SM, Soule MJ, Phillips G, Thomashow LS. Functional Analysis of Genes for Biosynthesis of Pyocyanin and Phenazine-1-Carboxamide from *Pseudomonas aeruginosa* PAO1. *J Bacteriol.* 2001;183: 6454–6465. doi:10.1128/JB.183.21.6454-6465.2001
2. Yan J, Liu W, Cai J, Wang Y, Li D, Hua H, et. al. Advances in Phenazines over the Past Decade: Review of Their Pharmacological Activities, Mechanisms of Action, Biosynthetic Pathways and Synthetic Strategies. *Marine Drugs.* 2021;19: 610. doi:10.3390/md19110610
3. Grosso-Becerra M-V, Santos-Medellín C, González-Valdez A, Méndez J-L, Delgado G, Morales-Espinosa R, et. al. *Pseudomonas aeruginosa* clinical and environmental isolates constitute a single population with high phenotypic diversity. *BMC Genomics.* 2014;15: 318. doi:10.1186/1471-2164-15-318

SYMPOSIUM ON BIOCHEMISTRY AND MOLECULAR BIOLOGY OF BACTERIA

GENES REQUIRED FOR THE FORMATION OF VIRULENCE-PROVOKING BACTERIAL SPHINGOLIPIDS

Otto Geiger¹, Roberto Jhonatan Olea-Ozuna¹, Sebastian Poggio², Aurora Osorio², Jonathan Padilla-Gómez¹, Lourdes Martínez-Aguilar¹, Isabel M. López-Lara¹

¹Centro de Ciencias Genómicas, UNAM, Cuernavaca, Morelos. ²Instituto de Investigaciones Biomédicas, UNAM, Mexico City. Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México, Avenida Universidad s/n, Cuernavaca, Morelos, CP 62210, Mexico; otto@ccg.unam.mx; Tel. (+52) 777 3290815

In eukaryotes, sphingolipids are ubiquitous, participate in the formation of biological membranes, and are associated with important cellular functions. Although sphingolipids occur scarcely in bacteria, for some of them they are essential and, in other bacteria, they contribute to fitness and stability of the outer membrane, such as in the well-studied α -proteobacterium *Caulobacter crescentus*. This bacterium is widely distributed in freshwater lakes and streams and was considered to be non-virulent. We previously defined five structural genes for ceramide synthesis in *C. crescentus*. However, other mutants affected in genes of this same genomic region show cofitness with a mutant deficient in serine palmitoyltransferase, the enzyme catalyzing the committed step in sphingolipid biosynthesis. Here we show that at least two phospho-sphingolipids are produced in *C. crescentus* and that at least another six gene products are needed for the decoration of ceramide upon phospho-sphingolipid formation. All eleven genes participating in phospho-sphingolipid formation are also required in *C. crescentus* for membrane stability and for displaying sensitivity towards the antibiotic polymyxin B. The genes for the formation of complex phospho-sphingolipids are also required for *C. crescentus* virulence on insect larvae of the greater wax moth *Galleria mellonella*.

SYMPOSIUM ON BIOCHEMISTRY AND MOLECULAR BIOLOGY OF BACTERIA

TOWARDS ENHANCED PRODUCTION OF BIODEGRADABLE PLASTICS: STRATEGIES TO IMPROVE YIELD AND QUALITY IN *AZOTOBACTER VINELANDII*

Daniel Segura González

Departamento de Microbiología Molecular, Instituto de Biotecnología, Universidad Nacional Autónoma de México.
Av. Universidad 2001, Col. Chamilpa, CP 62210, Cuernavaca, Morelos.
Tel. (52)7773291629, daniel.segura@ibt.unam.mx

Polyhydroxyalkanoates (PHAs) are polyesters produced by various bacteria as reserve of carbon and reducing power. They are accumulated under conditions of excess carbon source and limitation of other nutrients. PHAs are well known as bacterial bioplastics, because they are thermoplastic materials that can be used industrially as biodegradable plastics, alternative to petroleum-based materials. This talk will present a review of the advances made at the Institute of Biotechnology for the production of PHAs through research in the following areas:

1. Genetics of PHAs synthesis. We have identified genetic regulatory systems that control PHAs synthesis, including alternative sigma factor RpoS, the global regulatory systems PTS-Ntr, and the two-component system GacS/GacA, which is linked to the post-transcriptional regulatory system RsmA/RsmZ. This has allowed for the construction of mutants in negative regulators such as RsmA and the IIA-Ntr protein, that show improved production.
2. Metabolic modifications that favor increased PHAs production or that alter the composition of the polyesters, improving their thermomechanical properties. In this regard, the introduction of metabolic blockades in pathways competing for metabolic precursors, has resulted in PHA overproduction. Two examples are the inactivation of the anaplerotic enzyme pyruvate carboxylase and enzymes of the biosynthesis of the polysaccharide alginate. Additionally, the introduction of the heterologous enzymes 3-hydroxyacyl-ACP thioesterase and 3-hydroxyacyl-CoA has allowed us to produce PHAs with different compositions by incorporating intermediates from metabolic pathways such as fatty acid synthesis into the bioplastic synthesis. This, in combination with the replacement of the PHA polymerase gene (*phbC*) of *A. vinelandii* with a gene (*phaC_{AC}*) from *Aeromonas caviae*, that codes for an enzyme with broader monomeric substrate specificity, allowed the production of materials with diverse and controllable thermomechanical properties.
3. Identification of new proteins involved in PHAs metabolism. By identifying proteins associated with the polymer granules we have found new proteins named phasins, and modifying their expression the accumulation of PHAs was improved because some of them control processes like the degradation of the polymer. Also, new enzymes responsible for the internal degradation of the PHAs (PHA depolymerases) have been identified, and their inactivation allows for the production of PHAs of a very high molecular weight, which improved the properties of the polymers produced.

ROS IN FUNGI, TURTLES, AND RATS: JANUS FAVORITE MOLECULES

EVIDENCE OF OXIDATIVE STRESS RESPONSES OF GREEN TURTLES (*CHELONIA MYDAS*) TO DIFFERENTIAL HABITAT CONDITIONS IN THE MEXICAN CARIBBEAN

Vanessa Labrada-Martagón^{1*}, Nadia Luisa Islas Madrid², Bárbara Cúmez Caté²,
Montserrat Solé⁴, Tania Zenteno-Savín²

¹Laboratorio Ecología de la Salud, *Facultad de Ciencias, UASLP*, Av. Chapultepec #1570, Col. Privadas del Pedregal, C.P. 78295, San Luis Potosí, San Luis Potosí, México. [vanessa.labraa@uaslp.mx]*; ²Laboratorio de Estrés Oxidativo, Programa de Planeación Ambiental y Conservación, Centro de Investigaciones Biológicas del Noroeste, Av. Instituto Politécnico Nacional #195, Playa Palo de Santa Rita Sur, C.P. 23096, La Paz, Baja California Sur, México. ³Institut de Ciències del Mar, CSIC, Psg. Marítim de la Barceloneta 37-49, 08003, Barcelona, Spain.
E-mail: vanessa.labrada@uaslp.mx

The Mexican Caribbean region located at the Mesoamerican Reef System is home to some of the most important foraging and nesting habitats for green sea turtles (*Chelonia mydas*) and is characterized by vast expanses of seagrass beds. During the last 25 years, urban development and tourism have increased in the Quintana Roo state. Additionally, this region has been ecologically affected in the last decade by pelagic *Sargassum* blooms. Information about the biochemical responses of green turtles from the Caribbean is limited, impeding the use of oxidative stress biomarkers for the evaluation of the health status of this population. This study aimed to assess if the oxidative stress indicators in the red blood cells of green turtles ($n=229$) are valuable biomarkers of the extent of the anthropic impact in this region during 2015-2019. Persistent organic pollutants were also measured in the plasma of free-living green turtles to characterize these habitats further. Oxidative stress indicators did not correlate with the handling time needed to withdraw the blood sample nor with the persecution time employed to capture the animals. The activities of GPx ($\rho=-0.26$, $p=0.006$) and SOD ($\rho=0.27$, $p=0.005$) were correlated with the size of the sea turtles (CCL); thus, the activity of SOD was higher in adults than in immature green turtles captured in the northern site (Punta Arenas, $p=0.02$). Green turtles inhabiting the southernmost foraging habitats (Punta Herrero and Xcalak) have higher plasmatic concentrations and more polychlorinated biphenyl congeners (PCBs) detected per individual and the higher activity of GPx than the other sites ($p<0.001$). Annual variability in oxidative stress responses of green turtles from Akumal and Punta Herrero was found (Wilks' Lambda=0.06, $p<0.001$), with modulation of oxidative stress responses over the years (2015-2019); individuals showed a shift from activation of the phase II detoxification (GST) and oxidative stress (TBARS) in 2015, followed by oxidative distress (SOD, CAT) in 2016, to a suggested oxidative eustress in the following years (2016-2017) which continued until 2019. Biochemical responses of green turtles captured during 2015 in Akumal and Punta Herrero Bays coincided with one of the first peaks reported of the atypical massive influx of pelagic *Sargassum*, which decreased in 2016-2019. The results of this study corroborate the utility of the oxidative stress indicators as biomarkers of environmental conditions in this sentinel species.

ROS IN FUNGI, TURTLES AND RATS: JANUS FAVORITE MOLECULES

CANDIDA GLABRATA (NAKASEOMYCES GLABRATUS) REQUIRES THE GSH/GLUTATHIONE REDUCTASE AND THIOREDOXIN/THIOREDOXIN REDUCTASE SYSTEMS FOR MITOCHONDRIAL FUNCTION AND OXIDATIVE STRESS RESPONSE

Ma. Guadalupe Gutiérrez Escobedo, Carlos Ricardo González Ruiz, Grecia Hernández-Hernández, Ana Belén Vargas Antillón, Irene Castaño-Navarro and Alejandro De Las Peñas

Redox homeostasis systems are primordial in all cellular biological processes. These systems are divided into two pathways: the glutathione pathway (GSH/Glr), which includes glutathione (GSH) and the glutathione reductase (Glr); and the thioredoxin pathway (Trx/Trr), which includes thioredoxin (Trx) and thioredoxin reductase (Trr) enzymes. Both pathways proceed through sequential oxidation-reduction reactions once they react with their target. As a result of the importance of maintaining redox balance within the cell, the glutathione and thioredoxin pathways function as background systems for each other. In the pathogen yeast *Candida glabrata*, the Trx/Trr system has two cytosolic thioredoxin reductases (Trr1 and Trr2) and a cytosolic (Trx2) and mitochondrial (Trx3) thioredoxins; while the GSH is present in all cellular compartments. Since the mitochondria is the organelle where the highest levels of reactive oxygen species are produced, we focus on the components that regulate the redox balance within the mitochondria. There are studies that points to an essential function of glutathione in mitochondria. We decided to generate a set of mutants where both redox systems were compromised. Interestingly, the simultaneous mutation of *TRR1*, *TRR2* and *GSH2* and *TRX2* and *GSH2* results in synthetic lethality, probably due to the lack of reduction of the RNR enzyme. However, the viable mutants (*trx3Δ gsh2Δ*), (*trr1Δ gsh2Δ*) and (*trr2Δ gsh2Δ*) have compromised growth on non-fermentable carbon sources such as glycerol or ethanol, with *trx3Δ gsh2Δ* being the most affected and *trx3Δ gsh2Δ* is more sensitive to H₂O₂ than single mutants or *trr1Δ gsh2Δ* and *trr2Δ gsh2Δ*. This result suggests that Trx3 and GSH participate cooperatively in mitochondria to maintain the redox balance. In addition to Trx3, the *C. glabrata* genome encodes for the mitochondrial peroxiredoxin Prx1 and other redoxins, such as glutathione reductase Glr1 and glutaredoxin Grx2. Both enzymes have cytosolic and mitochondrial localization due to the presence of an alternate transcription site. We confirmed the mitochondrial localization of Prx1 and the dual localization, cytosolic and mitochondrial, of Glr1 and Grx2 using translational fusions with GFP. Like *trx3Δ* mutant, the *prx1Δ*, *glr1Δ* and *grx2Δ* single mutants are not sensitive to exogenous H₂O₂. Surprisingly, *trx3Δ prx1Δ*, *glr1Δ prx1Δ* and *grx2Δ prx1Δ* double mutants are resistant to oxidative stress compared to the parental strain and single mutant. We used a bimolecular fluorescence complementation (BIFC) assay and observed that Prx1 physically interacted with Trx3, Glr1 and Grx2, whereas Trx3 interacted with Glr1 and Grx2. This result indicates that Prx1 may be the substrate for Trx3 or Grx2, whereas both Trx3 and Grx2 require GSH to maintain their reduced state. The absence of *PRX1* in *cta1Δ*, *trx3Δ*, *glr1Δ* and *grx2Δ* backgrounds decreases the viability against human polymorphonuclear cells, which suggests that Prx1 has an important role for the survival of *Candida glabrata* within the host.

ROS IN FUNGI, TURTLES AND RATS: JANUS FAVORITE MOLECULES

TARGETING SENESCENT CELLS TO DECREASE OXIDATIVE STRESS AND NEUROINFLAMMATION, AND PREVENT COGNITIVE IMPAIRMENT DUE TO CHRONIC OBESITY IN MIDDLE-AGED FEMALE RATS

Mina Königsberg^{1*}, Rosa Pamela Flores-Torres^{1,2,3}, Michel López-Teros^{1,3}, Verónica Salas-Venegas^{1,3}, Adriana Alarcón Aguilar¹, Norma E. López-Díazguerrero¹, Beatriz Gómez-González², Anahí Chavarría-Krauser⁴, Armando Luna-López⁵

¹Depto. Ciencias de la Salud. Universidad Autónoma Metropolitana, Iztapalapa (UAMI). ²Depto. Biología de la Reproducción. UAMI. ³Posgrado en Biología Experimental, UAMI. ⁴Facultad de Medicina. UNAM, ⁵Instituto Nacional de Geriatria.

*Av. Ferrocarril San Rafael Atlixco 186, CP 09310, CDMX.

mkf@xanum.uam.mx

Obesity has been declared a health emergency in Mexico and is a risk factor for developing cognitive decline that might lead to dementias. These conditions have a higher prevalence in women and constitute one of the most difficult challenges facing the Mexican health system due the growing elderly population. The cellular mechanism behind this phenomena has been related to increased inflammation that promotes the blood brain barrier (BBB) disruption generating neuroinflammation and oxidative stress. These factors induce the appearance of senescent cells (SC), which secrete a series of pro-inflammatory molecules called Senescent Associated Secretory Phenotype (SASP) that deteriorate the tissue. Currently, there are mainly two senotherapeutic approaches to contend with the accumulation of SC: senolytics, which promote their death, and senomorphics, which modify the secretory profile of these cells to reduce their negative impact. Molecules such as Quercetin and Dasatinib (D+Q) have been repositioned as senolytics, while sulforaphane (SFN) stands out as a senomorphic.

Our group has developed an in vitro model of primary astrocytes induced to SC with palmitic acid to study the effects of senolytics and senomorphics on cellular senescence and gliosis. We have also assayed the increased neuroinflammation and oxidative stress in key brain areas, leading to cognitive impairment due to a hypercaloric diet (HD) in middle-aged female Wistar rats. The effect of senotherapeutics (D+Q or SFN) on HD-rats was evaluated and our results showed that SFN did prevent cognitive decline, but D+Q did not. Currently, we continue to evaluate the effects of both treatments on BBB permeability and SC clearance in the brain.

We thank to Dr. Guerrero-Aguilera for the animals. The Project is financed by FORDECYT-PRONACES/263957/2020. RPFT, MLT, VSV are CONAHCyT scholarship holders.

RECENT PROGRESS IN SIGNAL TRANSDUCTION RESEARCH

REGULATION OF INSULIN SIGNALING: MULTIPLE FACTORS AND MOLECULAR MECHANISMS INVOLVED

Jesús Alberto Olivares-Reyes

Department of Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute
(CINVESTAV-IPN) Mexico City, Mexico City, ZIP 07360, Mexico
E-mail: jolivare@cinvestav.mx

Insulin is a hormone secreted by β -cells in the pancreas that plays a crucial role in regulating blood glucose levels by promoting the uptake and storage of glucose in muscle and adipose tissue, while simultaneously inhibiting glucose production in the liver. Furthermore, insulin regulates the metabolism of carbohydrates, lipids, and proteins, and promotes cell division and growth through mitogenic effects¹. The biological actions of insulin are initiated by the activation of insulin transmembrane receptors, which belong to the receptor tyrosine kinase (RTK) superfamily¹. Because insulin actions are highly regulated to promote proper metabolic function and energy balance, extensive research has revealed the involvement of multiple factors and molecular mechanisms in the regulation of insulin signaling². When these mechanisms are disrupted, a condition known as insulin resistance may occur, which is a consequence of deficient insulin signaling caused by mutations or post-translational modifications of its receptor or effector signaling proteins located downstream of it. Insulin resistance is one of the main characteristics of the pathological conditions associated with metabolic syndrome and obesity, both of which are critical promoters of type 2 diabetes mellitus (DM2), one of the leading causes of death in Mexico and worldwide². In recent years, various conditions, including inflammation, endoplasmic reticulum (ER) stress, and mitochondrial dysfunction, caused by diverse hormones, factors, and conditions, have been identified to promote insulin resistance^{3,4}. Our research focused on elucidating the molecular mechanisms associated with insulin resistance caused by hormones, lipids, and growth factors. This investigation has revealed a diversity of mechanisms that contribute to the regulation of insulin resistance, including the involvement of serine/threonine kinases, such as protein kinase C (PKC), mTOR/S6K, and JNK, as well as tyrosine phosphatases, which act particularly at the level of the insulin receptor and its substrate, IRS. Likewise, our research also suggests regulatory mechanisms at the level of critical kinases in insulin signaling, such as Akt. Finally, we will share new research that suggests an active role for insulin signaling in the regulation of GPCRs and RTK function.

This research is supported by a CONAHCYT research grant CF-2023-G-446.

References

1. Saltiel, A. R. (2021), *The Journal of Clinical Investigation*, 131, 1-12.
2. Gutierrez-Rodelo, C., Roura-Guiberna, A., Olivares-Reyes, J. A. (2017). *Gaceta Médica de México*, 153, 197-209.
3. Olivares-Reyes, J. A., Arellano-Plancarte, A., Castillo-Hernandez, J. R. (2009). *Molecular and Cellular Endocrinology*, 302, 128-139.
4. Vazquez-Jimenez, J. G., Roura-Guiberna, A., Jimenez-Mena, L. R., Olivares-Reyes, J. A. (2017). *Gaceta Médica de México*, 153, 773-783.

RECENT PROGRESS IN SIGNAL TRANSDUCTION RESEARCH

EPITHELIAL TO MESENCHYMAL TRANSITION IN BREAST CANCER CELLS: EGF SIGNALING

Eduardo Monjaraz Guzmán¹, Scarlett Gallardo-Arriaga¹, Laura González-González² and Ricardo Félix²

¹Physiology Institute-BUAP, Puebla, México; ²Department of Cell Biology, Cinvestav-IPN, Mexico City, Mexico.

E-mail: eduardo.monjaraz@correo.buap.mx

Breast cancer (BC) is the most frequently diagnosed with > 2.3 million new cases per year and the leading cause of death in women. BC can spread to other sites of the body and result in metastasis. Over time, 20–30% of patients with breast cancer develop metastases and die from causes related to the disease. Epithelial-mesenchymal transition (EMT) is one of the crucial steps that initiate cell progression, invasion, and metastasis. EMT is a complex and multi-functional transdifferentiation program by which epithelial cells acquire the structural and functional characteristics of mesenchymal cells, leading to reduced intercellular adhesion and increased motility. Under normal circumstances, EMT plays critical roles during embryogenesis, wound healing, and tissue remodelling. However, abnormal EMT contributes to cancer metastasis. The EMT involves an increased cell migration capacity, invasiveness, resistance to apoptosis, increased production of extracellular matrix components, activation of transcription factors, expression of specific cell surface proteins, reorganization and expression of cytoskeletal proteins, and changes in the expression of microRNAs. There are numerous EMT inducers including growth factors and cytokines, that activate signaling cascades that overlap with the tumor microenvironment and may lead to BC progression, such is the case of the Epidermal Growth Factor (EGF). EGF play an important role in mediating the interactions between the mesenchyme and the mammary bud epithelium promoting the formation of the mammary gland. However, the enhanced activity of the EGF receptor in cancer cells increases the activity of transcription factors that repress the expression of cell adhesion molecules. As a result, these cells change their number, morphology, express mesenchyme-associated molecules, and assume greater aggressiveness as they become more migratory, invasive and chemoresistance. EGF binds to a receptor tyrosine kinase (TRK) that is overexpressed in breast cancer. TRK act by affecting the activity of downstream signalling molecules including MAPK, FAK, Ras GTPases, and PI3K. We show that chronic treatment with EGF promotes the epithelial-mesenchymal transition of MDA-MB 231 cells.

RECENT PROGRESS IN SIGNAL TRANSDUCTION RESEARCH

SERCA PUMPS AS KEY REGULATORS OF CALCIUM SIGNALING IN BLOOD VESSELS

Angélica Rueda¹ and Cristian J. Arriero-Carrillo¹

¹Department of Biochemistry, Cinvestav-IPN, Av. IPN 2508. San Pedro Zacatenco, GAM. México City 07360, México.
E-mail: arueda@cinvestav.mx

Calcium (Ca^{2+}) is a fundamental intracellular messenger that participates in the transduction of a vast number of cellular signals. Transient increases in the cytoplasmic Ca^{2+} concentration ($[\text{Ca}^{2+}]_{\text{cyt}}$) play a key role in a wide variety of cellular functions, including the contraction of cardiac and vascular smooth muscle cells (VSMCs). The Sarco/Endoplasmic Reticulum Ca^{2+} ATPase (SERCA pump) participates in the dynamic regulation of the $[\text{Ca}^{2+}]_{\text{cyt}}$. This transmembrane protein belongs to the P-type ATPase family and uses the energy of ATP hydrolysis to transport Ca^{2+} from the cytoplasm to the intracellular Ca^{2+} stores, primarily located at the Sarcoplasmic Reticulum (SR) in VSMCs. In mammals, three different genes encode the SERCA pumps (ATP2A1-3) which by mRNA alternative splicing originate about twelve SERCA pump isoforms differentially expressed in a tissue-specific manner. In VSMCs of cerebral and mesenteric arteries (MA), SERCA2a and SERCA2b are the dominant isoforms. Previous data from our group showed that Aldosterone treatment of rat MA (Aldo 10 nM, 24 h) increased the expression of SERCA2, and the frequency of Ca^{2+} sparks¹, favouring the ignition of Ca^{2+} waves. By real-time qPCR and Western blot approaches, we corroborated that both SERCA2a and SERCA2b isoforms are expressed in rat MA, where SERCA2b was three times more abundant than SERCA2a. Aldo increased the expression of both SERCA isoforms at mRNA (2.4-fold for SERCA2a and 2.2-fold for SERCA2b) and protein levels (SERCA2a increased by 19% and SERCA2b by 20%) with respect to the control condition. SERCA2a was found mainly at the perinuclear region of VSMCs, and Aldo increased its expression at the superficial SR. Conversely, our findings showed that SERCA2b primarily resided at both superficial and perinuclear SR. Aldo treatment increased SERCA2b expression in superficial, cytoplasmic, and perinuclear SR regions, but the superficial SERCA2b showed the highest increment.

Our work provides novel evidence about Aldo-induced SERCA2a and SERCA2b upregulation and unveils their critical subcellular localization for buffering Ca^{2+} influx and the ignition of Ca^{2+} sparks and Ca^{2+} waves.

Work supported by Conahcyt project #A1-S-9082 (Fondo Sectorial de Investigación para la Educación, Convocatoria CB-2017-2018).

Reference

1. Salazar-Enciso R, et. al., *Frontiers in Physiology* (2022) 13:834220. doi:10.3389/fphys.2022.834220.

MITOCHONDRIA IN HEALTH AND DISEASE

ROLE OF MITOCHONDRIAL FUSION PROTEINS IN CELL HOMEOSTASIS

Antonio Zorzano

¹Institute for Research in Biomedicine (IRB Barcelona), The Barcelona Institute of Science and Technology, Barcelona Barcelona, Spain, ²Department of Biochemistry and Molecular Biomedicine, Faculty of Biology, University of Barcelona, Barcelona Barcelona, Spain, ³CIBER de Diabetes y Enfermedades Metabólicas Asociadas (CIBERDEM), Instituto de Salud Carlos III, Barcelona Barcelona, Spain.
E-mail: antonio.zorzano@irbbarcelona.org

Mitochondria are dynamic organelles that undergo fusion and division in a highly regulated manner. These adaptive morphological changes are known as mitochondrial dynamics and are crucial for mitochondrial function. The main players in mitochondrial dynamics belong to the dynamin superfamily and they can promote either organelle membrane fusion or membrane fission. Mitochondrial fusion is performed by the outer mitochondrial membrane proteins Mitofusin-1 (MFN1) and Mitofusin-2 (MFN2) and the inner mitochondrial membrane protein Optic atrophy-1 (OPA1). Mitochondria are able to establish physical interactions with other membranous components of the cell and some of these contacts are mediated by MFN1 or MFN2. MFN1 mediates mitochondrial contacts with the endoplasmic reticulum (ER) and the plasma membrane. MFN2 has been shown to participate in the apposition of mitochondria to ER, lysosomes, lipid droplets and melanosomes. Although classically known for their role in mitochondrial fusion, MFN1 and MFN2 are also involved in many other cellular processes, including mitochondrial bioenergetics, immune response, autophagy, cell cycle progression, cellular senescence and apoptosis. In the talk, we will analyze the functional implications of novel interactors of MFN proteins, and of MFN2 splicing variants and their link with cell homeostasis. This study will shed light into the function of MFN proteins in mitochondria-endoplasmic reticulum contact sites (MERCs) that could participate in metabolic disorders such as those found in insulin resistance, type 2 diabetes or in liver diseases.

Brief summary of CV. Antonio Zorzano is Full Professor of Biochemistry and Molecular Biology at the University of Barcelona, Director of Complex metabolic diseases and mitochondria laboratory at the IRB Barcelona, and Group Leader at CIBERDEM. Professor Zorzano received his PhD in Biology at the University of Barcelona, and did postdoctoral studies with Emilio Herrera (Hospital Ramon y Cajal, Madrid), Neil Ruderman (Boston University Medical Center), and Paul Pilch (Boston University Medical School). He was Visiting Professor at Boston University Medical School. He has supervised 47 Ph.D. theses, and has coordinated international consortia funded by different European agencies. He is co-inventor of 23 patents, and has published over 360 scientific articles (more than 49,000 citations), with key discoveries published in leading journals, and an h-index of 100 (Google Scholar). He has been founder of biotechnological companies in Spain and in UK.

Professor Zorzano's research focuses on the regulation of metabolism and its interplay with insulin resistance, obesity, type 2 diabetes, and liver diseases. His current interest links metabolism with mitochondrial dynamics, autophagy, and mitochondrial stress. A global goal of his group is to identify and validate molecular targets that permit the prevention or treatment of metabolic diseases by using cell-based systems, genetically modified mice, and translational approaches.

MITOCHONDRIA IN HEALTH AND DISEASE

**EXERCISE REGULATION OF MITOCHONDRIAL FUNCTION
IN METABOLIC DYSFUNCTION-ASSOCIATED
STEATOTIC LIVER DISEASE**

Rodrigo Troncoso

Laboratorio de Investigación en Nutrición y Actividad Física (LABINAF), Instituto de Nutrición y Tecnología de los Alimentos (INTA), Universidad de Chile.

Metabolic dysfunction-associated liver disease (MASLD), the new term that replaced nonalcoholic fatty liver disease (NAFLD), is a significant health concern. MASLD requires the presence of steatosis and at least one cardiometabolic risk factor. Mechanistically, disruption in mitochondrial function tends to provoke and aggravate metabolic dysregulation and likely contributes to the advancement of MASLD. On the other hand, lipid droplets (LD) are highly dynamic storage organelles; their accumulation causes MASLD. LD interacts with mitochondria, which impacts mitochondrial function. Understanding the role of exercise in modulating this interaction could have profound implications for the prevention and management of MASLD.

To examine the effects of aerobic exercise on the liver, two different MASLD models were used: a high-fat diet (HFD) to evaluate simple steatosis and an HFD-methionine choline-deficient diet (MCD) to evaluate steatohepatitis. Our results showed that exercise decreased disease severity and improved physical capacity compared to sedentary mice. Although exercise increased the number of LD in hepatocytes, LD were smaller than in the sedentary HFD mice. Notably, while sedentary HFD mice had increased hepatic lipid droplet (LD)-mitochondria interaction, in exercised HFD mice, there was a decreased interaction. The findings of the steatosis model are consistent with those of the steatohepatitis model. Aerobic exercise increased fatty acid oxidation and Mitofusin-2 abundance in peridroplet mitochondria. Taken together, our findings show that aerobic exercise reduced the progression of MASLD by promoting reduced lipid droplet-mitochondria interaction and increasing fatty acid β -oxidation.

MITOCHONDRIA IN HEALTH AND DISEASE

DEPRESSION-LIKE BEHAVIOUR IS COMORBIDITY IN A MODEL OF HEART FAILURE WITH PRESERVED EJECTION FRACTION INDUCED BY HIGH-FAT DIET AND L-NAME

Roger A. Maldonado-Ruiz¹, Victoria Palafox Sánchez¹, Judith Bernal-Ramírez¹, Omar Lozano García¹, Gerardo García-Rivas¹

1Av. Eugenio Garza Sada 2501 Sur, Tecnológico, 64849 Monterrey, N.L., C.P. 64700, tel. 8888216
E-mail: gdejesus@tec.mx.

Heart failure with preserved ejection fraction (HFpEF) is a syndrome with a 35% 2-year rate of hospitalization and 14% 2-year mortality. HFpEF patients have a prevalence of depression 20%¹. Several studies report that HFpEF and depression share proinflammatory cytokines, including IL-33, which plasma concentration has a positive correlation with left ventricle diastolic dysfunction² and depressive symptoms³. In addition, downregulation of the astrocyte-derived IL-33 in the amygdala prevents the development of depression in mice⁴. Even though, there is an under-recognition of depression in 50% of the HFpEF cases, that may be due to the absence of benefit from standard pharmacotherapy⁵. Thus, we propose that a mice model of HFpEF develops depression-like behavior and has an increased expression of IL-33 in the amygdala. To achieve this, we employ eight-week-old male C57BL/6 mice that were randomly assigned to the following groups. HFpEF group: mice fed with ad libitum HFD (60% Fat) and drinking water with L-NAME (0.5g/L) for 12 weeks, control: mice fed with ad libitum chow diet and tap water for 12 weeks. To determine cardiac hypertrophy, we measure the left ventricle cardiomyocyte cross-sectional area and, to identify the presence of depression we employ the open field test (OF), sucrose preference (SP) and novelty suppressed feeding (NSF). To determine inflammation at the amygdala, we measure IL-33, IL-10, IL-6, and IL-1 β expression by qPCR. Also, we measure GFAP gene expression as an indirect marker of gliosis. Finally, knowing that cholesterol homeostasis is critical for normal brain function, we quantify the ABCA1 expression. We found that the HFpEF group presents a 26% increase in cardiomyocyte area, a reduction of 11% (SP), a latency to eat of 53s (NSF) and spend 30% less time at the center and 20% more time at the edges of the arena in the OF test. Finally, the HFpEF group present an increased expression of ABCA1 (1.6-fold), GFAP (1.7-fold), IL-1 β (1.9-fold), and IL-33 (1.8-fold) as well as a 50% downregulation of IL-10 in the amygdala. With this evidence we can conclude that, an exposure to HFD+ L-NAME for 12 weeks promotes cardiac hypertrophy and depression-like behavior accompanied by an increased expression of IL-33 in the amygdala.

References

1. Dunlay, S. M., Roger, V. L. & Redfield, M. M. (2017). *Nat Rev Cardiol* 14, 591–602.
2. Yu Song, Fuhai Li, Yamei Xu, Yuan Liu, Yanyan Wang, Xueting Han, Yuyuan Fan, Juan Cao, Jie Luo, Aijun Sun, Kai Hu, Jingmin Zhou, Junbo Ge (2020). *Int J Cardiol* 304, 95–100.
3. Ewa A Oglodek, Marek J Just (2018). *Pharmacol Rep.* 70(6):1065-1072.
4. Xiao Zhuang, Bing Zhan, Yufeng Jia, Chaoze Li, Nan Wu, Ming Zhao, Nuo Chen, Yaxin Guo, Yingxin Du, Yi Zhang, Baihui Cao, Yan Li, Faliang Zhu, Chun Guo, Qun Wang, Yuan Li, Lining Zhang (2022). Volume 102, Pages 98-109.
5. Haider J Warraich, Dalane W Kitzman, David J Whellan, Pamela W Duncan, Robert J Mentz, Amy M Pastva, M Benjamin Nelson, Bharathi Upadhyya, Gordon R Reeves (2018). *Circ Heart Fail.* Nov;11(11):e005254.

MITOCHONDRIA IN HEALTH AND DISEASE

AVOCADO OIL: A PROMISING ALTERNATIVE AGAINST CHRONIC DEGENERATIVE DISEASES THROUGH ITS EFFECTS ON MITOCHONDRIA

Christian Cortés-Rojo¹, Marcela González-Montoya¹, Manuel Vargas Vargas¹, Olin Torres-Isidro¹

¹Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Edificio B-3, Ciudad Universitaria, Av. Fco. J Mujica S/N, Morelia, 58030, Michoacán, México.

E-mail: christian.cortes@umich.mx

The electron transport chain (ETC) is a hub in which several mitochondrial functions intersect including substrate oxidation, ATP synthesis, maintenance of the redox state of pyridine nucleotides, modulation of reactive oxygen species (ROS) levels, regulation of mitochondrial quality control processes and cell death through the maintenance membrane potential, among others. ETC dysfunction disrupts these processes, leading to exacerbated ROS production. High ROS levels activates inflammatory and fibrosis processes that together with uncontrolled cell death lead to tissue damage in chronic degenerative diseases such as diabetes or hypertension. Furthermore, ETC dysfunction plays a role in the development of insulin resistance in type 2 diabetes. Therefore, ETC dysfunction is considered an essential part of the development of diabetes and hypertension and several of their complications via excessive ROS production. Thus, mitochondria have been proposed as a therapeutic target for these diseases through the decrease of ROS production, which may block the induction of tissue damage and disease development. In this regard, we have investigated the beneficial effects of avocado oil intake on mitochondrial function and oxidative stress, and its impact on the development of diabetes and hypertension in animal models, as avocado oil is a source of bioactive compounds with antioxidant activity. We have found that avocado oil prevents ETC dysfunction of brain, liver, and kidney mitochondria from diabetic rats, as well as in kidney mitochondria from hypertensive rats. This decreases ROS production, cardiolipin loss, and lipid peroxidation and has resulted in decreased hypertensive and diabetic kidney damage, as well as decreased systemic blood pressure and blood glucose levels in hypertension and diabetes, respectively. Moreover, we have detected alleviation of non-alcoholic fatty liver disease in rats fed a high-fat, high-fructose diet. Regarding the latter, we believe that one mechanism by which avocado oil decreases insulin resistance is by increasing hepatic mitochondrial fatty acid oxidation, which decreases diacylglycerol levels and increases insulin sensitivity. This may be linked to increased NADH oxidation in complex I and decreased mitochondrial protein acetylation via induction of sirtuin-3 activity, which is currently under investigation in our group.

Acknowledgements. To Instituto de Ciencia, Tecnología e Innovación – Gobierno de Michoacán, (grants ICTI-PICIR23-063 and ICTI-PICIR23-028) and the funding by Coordinación de Investigación Científica – UMSNH.

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex of atoms, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

SIMULTANEOUS ORAL

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

EXPLORING THE QUATERNARY ASSEMBLY OF A HOMOMULTIMERIC CATECHOL 1,2-DIOXYGENASE: AN INTEGRATIVE STUDY

Arisbeth G. Almeida-Juárez¹; Liliana Pardo-López¹, Shirish Chodankar², Enrique Rudiño Piñera¹

¹Instituto de Biotecnología, UNAM. Cuernavaca, México. C.P.62210.

²Brookhaven National Laboratory NSLS-II. Upton, New York. 11973-5000

ariisalmeida@gmail.com

In 2015, the strain *S. stutzeri* GOM2 was isolated from marine substrate in the southwestern Gulf of Mexico¹. This strain exhibits the ability to grow in the presence of crude oil and harbors enzymes within its genome that facilitate the aerobic degradation of aromatic compounds, specifically benABC operon enzymes. One of the enzymes involved in the degradation of aromatic compounds is catechol 1,2-dioxygenase. This intradiol-type dioxygenase is Fe (III) dependent and participates in the oxidation of catechol or other catechol substituents^{2,3}. In contrast to the reported dimeric structures of catechol 1,2 dioxygenases in gram negative bacteria, in a previous study we revealed that under low ionic conditions, C12DO functions as a trimer¹. In this work, we endeavor to elucidate the quaternary structure of C12DO using various structural biology techniques. Our findings demonstrate the formation of larger oligomers than previously reported in low ionic conditions, after C12DO lyophilization and reconstitution. The size and quaternary structure of these oligomers were determined using Small-Angle-X-ray Scattering (SAXS), size exclusion chromatography (SEC), dynamic light scattering (DLS) and electron microscopy (MET). Notably, all these new oligomers forms of C12DO exhibit enzymatic activity, suggesting potential applications of C12DO in biorremediation.

References

1. Rodriguez-Salazar, J. Almeida-Juarez, A.G. Ornelas-Ocampo, K. Millan Lopez, S. Raga-Carbajal, E. Rodriguez-Mejia, J.L. Muriel-Millan, L.F. Godoy Lozano, E.E. Rivera-Gomez, N. Rudino-Pinera, E. Pardo-Lopez, L. 2020. Characterization of a Novel Functional Trimeric Catechol 1,2-Dioxygenase From a *Pseudomonas stutzeri* Isolated From the Gulf of Mexico *Frontiers in Microbiology*, 11, 1100
2. Dijk, J. A.; Stams A. J. M.; Schraa G.; Ballerstedt H.; De Bont J. A. M.; Gerritse J. 2003. Anaerobic oxidation of 2-chloroethanol under denitrifying conditions by *Pseudomonas stutzeri* strain *Applied Microbiology and Biotechnology*. 63:68-74
3. Micalella C., Martignon S., Bruno S., Pioselli B., Caglio R., Valetti F., et al. (2011). X-ray crystallography, mass spectrometry and single crystal microspectrophotometry: a multidisciplinary characterization of catechol 1,2 dioxygenase. *Biochim. Biophys. Acta* 1814 817–823

LINC01087/MIR-130A-3P/FOXA1 AXIS REGULATES THE CELL PROLIFERATION, MIGRATION AND VASCULOGENIC MIMICRY IN BREAST CANCER CELLS IN A THREE-DIMENSIONAL (3D) MICROENVIRONMENT

Sandra Lizeth Alvarado Ortigoza¹, Stephanie Iraiz Núñez Olvera², Mireya Cisneros Villanueva³, Alejandra Paola García Hernández¹, César López-Camarillo^{1*}

¹Program in Genomic Sciences, Autonomous University of Mexico City, Mexico City 03100, Mexico, ²Department of Cellular Biology and Physiology, Biomedical Research Institute, National Autonomous University of Mexico, Mexico City 04510, Mexico,

³Cancer Genomics Laboratory, National Institute of Genomic Medicine, Mexico City 14610, Mexico.

*Corresponding author. San Lorenzo 290 Col del Valle. 03100, Tel:5511070280,

E-mail: cesar.lopez@uacm.edu.mx

Long non-coding RNAs (lncRNAs) are regulatory molecules of >200 nt length that do not have an open reading frame, so they do not have the ability to encode proteins. lncRNAs function as master regulators of gene expression at the transcriptional and post-transcriptional levels and their deregulation has been linked to cancer. In recent years, the implementation of three-dimensional (3D) cell cultures has revolutionized the study of cancer. 3D cultures are characterized by using scaffolds of matrigel enriched in extracellular matrix proteins which promotes the formation of 3D structures of cancer cells. These 3D structures can form gradients of both nutrients and oxygen simulating a cellular environment like tumors *in vivo*. We recently reported a genome-wide expression profile of lncRNAs in BT-474 luminal B subtype breast cancer cells grown in a 3D microenvironment. Particularly, the lncRNA LINC01087 was overexpressed in 3D cultures in comparison to monolayer 2D cultures. In the present investigation, we studied the functional role of LINC01087/miR 130a-3p/FOXA1 co-regulation axis in breast cancer cells grown in 2D and 3D. The silencing of LINC01087 in 2D and 3D cultures using siRNAs resulted in decreased cellular viability, proliferation, and migration, as well as in the impairment of vasculogenic mimicry abilities. In 3D cultures, the LINC01087-deficient cells showed an abnormal phenotype; the cells were observed with a rough and tortuous contour. Moreover, LINC01087-deficient cells showed a significant change in the size and number of 3D structures in comparison to untreated cells. In addition, both LINC01087-deficient cells and cells that overexpress miR130a-3p showed a lower abundance of the FOXA1 protein, the predicted target of miR130a-3p. These data suggest that the affected cellular processes may be indirectly regulated by the LINC01087/miR-130a-3p/FOXA1 axis. In conclusion, LINC01087 is a key molecule important in the modulation of some hallmarks of breast cancer in 2D monolayer cultures as well as in 3D suggesting that it could be a potential therapeutic target in breast cancer therapies.

Acknowledgments: This work was funded by Consejo Nacional de Humanidades Ciencia y Tecnología CONAHCYT. Grant A3-S-33674.

PARTICIPATION OF EXON 18 VARIANTS OF THE SCN8A GENE IN THE PROLIFERATION AND METASTATIC BEHAVIOR OF CERVICAL CANCER CELL LINES

Tonantzin Guadalupe Anguheven Ledezma, Margarita Jacaranda Rosendo Pineda, Osbaldo López Charcas, Miguel Ramírez Aragón and Juan Carlos Gómora Martínez

Departamento de Neuropatología Molecular, Instituto de Fisiología Celular, UNAM.
E-mail: tonantzin@ifc.unam.mx

Cervical cancer (CeCa) is the fourth most common cancer and the fourth cause of cancer death in women over 15 years old worldwide¹. In the last 25 years, various types of ion channels have been associated with the hallmarks of cancer. Notably, voltage-gated sodium channels (Na_v channels), which are responsible for the generation and propagation of action potentials in excitable cells, have been related to cell migration and invasion processes in breast, ovarian, prostate, and cervical cancer, among others². Our research group reported a 40-fold higher expression level of Na_v1.6 channels in primary CeCa cultures compared to non-cancerous cervical tissue. Furthermore, through functional assays, a direct association was found between the activity of Na_v1.6 channels and invasiveness of this carcinoma³. The *SCN8A* gene that encodes the α -subunit of the Na_v1.6 channel generates three alternative splicing variants in exon 18: 18A, 18N and Δ 18⁴. The Δ 18 variant was localized in intracellular vesicles in macrophage cells (derived from leukemia) and melanoma cells mediating the formation of invadopodia⁵. Therefore, the goal of this project was to investigate the role of each Na_v1.6 variant in proliferation, migration, and invasiveness of CeCa cell lines which are positive to different types of human papillomavirus (HPV): SiHa (HVP-16), HeLa (HPV-18) and C33A (HPV negative). To evaluate the role of each variant, specific siRNAs that reduce its expression level were used. Our results showed a significant reduction in cell proliferation and invasiveness when the expression of 18A and Δ 18 variants was selectively decreased in the three cell lines. Cell proliferation is reduced between 70 and 80% by silencing 18A and Δ 18 in the three cell lines; cell migration was only affected in SiHa cells; and invasiveness was reduced by 50% in SiHa cells when the expression of 18A variant was reduced. The same trend was observed in C33A; however, in HeLa cells, which are the most invasive, it is the Δ 18 variant that influences invasiveness. Data suggest a differential effect of 18A and Δ 18 variants of the Na_v1.6 channel on CeCa cell proliferation, migration, and invasiveness. We propose the Δ 18 variant as a potential therapeutic target in CeCa treatment.

This work is supported by grants from CONAHCYT A1-S-19171 and DGAPA-PAPIIT-IN211123.

References

1. Globocan. (2024).
2. Lopez-Charcas, et al. (2021). *IScience*, 24(4).
3. Hernández-Plata, et al., (2012). *Int Journal Cancer*. 130, 2013.
4. Plummer, et al. (1997). *J Biol Chem*. 272, 24008. 5. Carrithers, et al. (2009). *J Biol Chem*. 284, 8114.

TARGETING ORNITHINE DECARBOXYLASE FOR CANCER THERAPY: ASSESSING INHIBITOR EFFICACY ON *IN VITRO* ASSAYS

Arroyo-Sánchez Beatriz Irene¹, Medina-Mendoza Gustavo Gerardo¹, Morales-Ríos Edgar², Olin-Sandoval Viridiana¹

¹Departamento de Biotecnología y Bioingeniería, CINVESTAV Zacatenco CDMX CP 07360,

²Departamento de Bioquímica, CINVESTAV Zacatenco CDMX CP 07360

bety.arroyos@cinvestav.mx; edgar.morales@cinvestav.mx y viridiana.olin@cinvestav.mx

Cancer is considered one of the leading causes of death worldwide¹. Therefore, the search for specific tumor growth inhibitors is a matter of great importance. One of the novel therapeutic targets is the enzyme ornithine decarboxylase (ODC), as it is overexpressed in several tumor cell types ². Currently, the inhibitors targeting the catalytic site of ODC, either have not been efficient, or they have toxic effects, thus, the search for molecules able to inhibit this enzyme is still an opportunity area. Previously, it has been reported that *S. cerevisiae*, rat and human ODC can use lysine as substrate (3). Considering this, in our workgroup, we have identified lysine analogues capable of inhibiting the recombinant ODC. Therefore, the aim of this work is to determine whether these compounds specifically inhibit the growth of tumor cell lines such as HepG2 (human hepatocellular carcinoma) by targeting ODC.

For this, seven ODC inhibitors were evaluated for their ability to inhibit cell proliferation of HepG2 cell line over a 72-hour period, using the crystal violet method. Out of the seven molecules tested, three exhibited inhibitory effects on cell proliferation: compound T (IC₅₀=633 μM), compound A (IC₅₀=300 μM), compound I (IC₅₀=513 μM). DFMO (IC₅₀= 90 μM), a well-established ODC inhibitor, was used as a positive control. In order to elucidate if the effects of these molecules on the proliferation of HepG2 are due to ODC inhibition, we are determining the intracellular polyamine concentration by HPLC coupled to a diode array detector.

References

1. WHO. (2020). Estimated age-standardized mortality rates (Word) in 2020, all cancers, both sexes, all ages. Globocan.
2. Dong, Y., Tu, R., Liu, H., & Qing, G. (2020). Regulation of cancer cell metabolism: oncogenic MYC in the driver's seat. *Signal Transduction and Targeted Therapy*, 5(1).
3. Casero, R. A., & Marton, L. J. (2007). Targeting polyamine metabolism and function in cancer and other hyperproliferative diseases. *Nature Reviews Drug Discovery*, 6(5), 373–390

CELL DIVISION AND MORPHOLOGY CHANGES CAUSED BY *CENR* OVEREXPRESSION AFFECT THE SYMBIOTIC NITROGEN FIXATION IN *R. ETLI* CFN42

María M. Banda, Ma. de la Paz Salas, Marisa Rodríguez, Sofía Martínez-Absalón, María del Rocío Reyero-Saavedra, Mishael Sánchez-Pérez, Ayari Fuentes, Georgina Hernández and Lourdes Girard Cuesy

Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México. Av. Universidad s/n. Chamilpa, 62210 Cuernavaca, Mor., México.
E-mail: bandahmm@ccg.unam.mx

Rhizobia are rod-shaped Gram-negative α -proteobacteria that can establish a nitrogen-fixing symbiosis with compatible legume hosts. The infection of a plant host by rhizobia consists of multiple developmental stages in which the bacteria modulate their cell proliferation in concert with the development of host cells. The bacteria penetrate the plant tissue in the root hairs through infection threads. During this interaction, the nodule, a newly induced plant organ, is colonized by the bacteria, from which bacteria are released and become enclosed in a plant-derived membrane. In the nodule, the bacteria differentiate into so-called bacteroids that provide combined nitrogen to the plant in exchange for nutrients.

Coordination between cell division and cell envelope synthesis is crucial for bacteria's growth, survival, and propagation. The transcription control of the cell cycle is highly conserved among α -proteobacteria. However, many rod-shaped α -proteobacteria species lack the proteins associated with cell elongation: MreB, MreC, MreD, RodZ, RodA, and PBP2. This situation is presented in bacteria belonging to the *Brucellaceae* and *Rhizobiaceae* families, where cell elongation occurs only from the new cell poles.

The gene transcription that determines cell envelope structure and function is commonly regulated by two-component signaling (TCS) systems, comprising a sensor histidine kinase (HK) and a cognate response regulator (RR). To identify TCS genes that contribute to cell division and cell envelope in the soil-dwelling bacteria *Rhizobium etli* CFN42, we focused on the 18 OmpR/PhoB family RRs encoded in its genome. Using a two-step recombination process to eliminate an *ompR* gene, we obtained a set of individual mutants. Using this methodology, we described that the *R. etli* OmpR regulator RetPC57 is critical in developing the *R. etli*-common bean symbiosis¹. However, we could not delete gene *RHE_CH03968*, which codified for an orthologous *cenR* gene, recently described as an essential regulator of cell envelope-related functions in α -proteobacteria. Based on its homology with these proteins, we named the *RHE_CH03968* gene *cenR*. In this work, we will discuss advances in the effect caused by the conditional overexpression of *cenR* in *R. etli* free-living growth and during symbiosis. Our results show that CenR regulates the expression level of essential genes from pathways controlling cell growth and cellular morphology in *R. etli*. Furthermore, during symbiosis, the overexpression of *cenR* resulted in nodules that showed lower nitrogen fixation activity and decreased expression of key genes directly involved in symbiotic nitrogen fixation.

Acknowledgments. This work was partially supported by PAPIIT-DGAPA (grant IN208823).

References

1. Rodríguez, S. et al (2020). *Frontiers in Microbiology*, 11, art. no. 615775.

THE ROLE OF THE REGULATORY PROTEIN PHbF IN THE CONTROL OF ACCUMULATION OF THE BIODEGRADABLE PLASTIC POLYHYDROXYBUTYRATE IN THE BACTERIUM *AZOTOBACTER VINELANDII*

Thalía Barrientos Millán, Libertad Adaya García, Josefina Guzmán Aparicio, Soledad Moreno, Guadalupe Espín, Daniel Segura González

Instituto de Biotecnología. UNAM. Av. Universidad 2001, Chamilpa, CP 62210, Cuernavaca, Morelos.
Tel. 7773291629 Ext. 27629, thalia.barrientos@ibt.unam.mx, daniel@ibt.unam.mx

Polyhydroxybutyrate (PHB) is a polyester produced by various archaea and bacteria as reserve of carbon, energy and reducing power. The importance of this compound in the industry lies in its ability to be used to manufacture biodegradable plastics to replace petroleum-based plastics. This polymer is accumulated intracellularly under conditions of excess carbon source and other nutrient limitations, and is mobilized (utilized) when the carbon source is scarce. *Azotobacter vinelandii* synthesizes PHB. Its synthesis starts from two molecules of acetyl-CoA, through three enzymatic steps catalyzed by β -ketothiolase, acetoacetyl-CoA reductase and PHB synthase, encoded by the *phbA*, *phbB* and *phbC* genes, respectively; while PHB degradation (mobilization) is carried out by the enzymes PHB depolymerases (*phbZ* genes), hydroxybutyrate dehydrogenase, succinyl Co-A transferase and, again, β -ketothiolase. Although both of these processes occur simultaneously in a constant synthesis/mobilization cycle, some mechanism is needed to control the balance of this cycle, favoring synthesis or degradation, depending on the metabolic conditions. While some regulators of the PHB biosynthetic genes are known in *A. vinelandii*, the genetic control of PHB degradation remains unknown. In this work, we showed that *A. vinelandii* PhbF is a regulator involved in both the biosynthesis and PHB mobilization processes, acting by repressing the expression of the *phbP1* gene, which encodes a phasin protein which covers the granule and is needed for full PHB production, and *phbZ1*, encoding a PHB depolymerase. PhbP1 phasin plays a role in PHB granule structure and PHB accumulation and has a PhbF-binding site on its promoter. PHB depolymerase PhbZ1 participates in the first step of the mobilization process, and the *phbZ1* gene shares a regulatory region with the *phbP2* gene (a second PHB granule-associated phasin); a PhbF binding site is found in the regulatory region shared by these two genes, which is located in the -35 region of the *phbZ1* promoter. In this study, the participation of the PhbF regulator in PHB granule formation and the mobilization process was demonstrated by analyzing the expression of the *phbP1*, *phbZ1*, and *phbP2* genes in both UW136 and UW136*phbF* strains through RT-qPCR and *gusA* transcriptional fusions. Additionally, analysis of PHB accumulation phenotypes was performed, and EMSAs were conducted to determine if PhbF can bind to the sites found in the aforementioned promoters. In the UW136*phbF* mutant, the decrease in PHB accumulation, the increase in *phbZ1*, *phbP1*, and *phbP2* gene expression, together with the binding of the PhbF protein to the regulatory regions of *phbP1* and *phbZ1-phbP2*, indicate that PhbF is a negative regulator of these genes, acting directly. This is something new in *A. vinelandii*, especially the regulation exerted over *phbZ1*, which has not been reported before.

References

1. Pötter, M. & Streinbüchel, A. (2005) Poly(3-hydroxybutyrate) granule-associated proteins: Impacts on poly(3-hydroxybutyrate) synthesis and degradation. *Biomacromolecules* 6, 552-560.
2. Adaya, L., Millán, M., Peña, C., Jendrossek, D., Espín, G., Tinoco-Valencia, R., Guzmán, J., Pfeiffer, D. & Segura, D. (2018) Inactivation of an intracellular poly-3-hydroxybutyrate depolymerase of *Azotobacter vinelandii* allows to obtain a polymer of uniform high molecular mass. *Applied Microbiology and Biotechnology* 102, 2693-2707.

EFFECT OF DENGUE VIRUS SEROTYPE 2 (DENV2) ON THE PREGNANE X RECEPTOR (PXR) SIGNALING PATHWAY IN MOUSE PERITONEAL MACROPHAGES

Carlos Daniel Bautista Olivier¹, Jorge Humberto Limón Pacheco¹ and Guillermo Elizondo^{1*}

¹Centro de Investigación y de Estudios Avanzados of I.P.N. (CINVESTAV), Av. Instituto Politécnico Nacional 2508, Col. San Pedro Zacatenco, Del. Gustavo A. Madero, Mexico City, P.C. 07360. Tel: +52 (55) 5747 3800 Ext. 5555

*Email: gazuela@cinvestav.mx

Dengue virus is the main mosquito-borne virus in the world with approximately 5.2 million cases reported in 2023. The infection can develop severe forms of the disease, such as dengue hemorrhagic fever (DHF) or dengue shock syndrome (DSS), which in the last year led to the death of 7,300 people. Most severe forms of the disease are due to reinfection with a different serotype, making vaccine preparation difficult. Among the strategies to treat this disease are those that interfere with the signaling pathways that the virus uses to infect its target cells at the beginning of the infection, such as macrophages and dendritic cells. Recently, we identified that after infecting mouse peritoneal macrophages (MΦm) with dengue virus serotype 2 (DENV2), the Pregnane X Receptor (PXR) is activated. PXR is an important xenosensor in the induction of xenobiotic-metabolizing enzymes and enzymes that promote lipid metabolism, in addition to being a negative regulator of the inflammatory response. Due to the above, in the present work, we investigate whether DENV2 uses PXR to facilitate its replication. Using a PXR antagonist, ketoconazole (KTZ), we identified that, in MΦm, inhibition of PXR results in a decrease in the number and size of lipid droplets and of lipid metabolism genes, essential for the assembly of the virus. On the other hand, PXR inhibition caused the restoration of the expression and secretion of inflammatory cytokines and interferons type I and II. The above resulted in a decrease in virus replication and viral load. Therefore, PXR could be a therapeutic target for the treatment of Dengue.

PHYSIOLOGICAL EFFECT OF THE INTERACTION BETWEEN THE MITOCHONDRIAL MAIZE HEXOKINASE 4 AND THE BETA-GLUCOSIDASE AGGREGATING FACTOR 1

Burgos-Palacios Andrés¹, Sánchez-Nieto Sobeida²

^{1,2}Laboratorio de Transporte y Percepción de Azúcares en Plantas, Departamento de Bioquímica Facultad de Química, Universidad Nacional Autónoma de México.

¹aburgosbp@gmail.com, ²sobeida@unam.mx.

Hexokinases (HXKs) are moonlighting proteins that have hexose phosphorylating activity and glucose sensor or protein kinase activities. The moonlighting protein activity depends on different factors such as their location in the cell or the interaction with proteins, nucleic acids, metabolites or ions¹. For example, AtHXK1 a mitochondrial protein that is an enzyme, is also able under high glucose concentration to be a glucose sensor and form a complex with two proteins at the nucleus; the trimeric complex represses the expression of photosynthetic genes.

Several studies have been made to identify novel interactors², in maize we identified putative interactors for ZmHXK4 through pull-down assays. Beta Glucosidase Aggregating Factor 1 (ZmBGAF1) a biotic stress related protein interacts *in vitro* and *in vivo* with the mitochondrial glucose sensor ZmHXK4. In mammals, the hormone Metil Jasmonate (MeJA) can break the HXK-mitochondria interaction, leading to cell death³. In maize, MeJA induced the transcription of ZmHXK4 and is involved in the defence responses of ZmBGAF1 to pathogen attack. The main goal of this work is to determine if MeJA promotes the interaction between ZmHXK4 and ZmBGAF1.

Transient expression of ZmHXK4 and ZmBGAF1 in *Nicotiana* leaves produced an increase in the permeability of the Evans Blue dye and affects the texture but not the color of the leaves. Both proteins are expressed mainly in the stomata, affecting its morphology. The addition of MeJA also affects the subcellular localization of ZmBGAF1. The interaction of ZmHXK4 and ZmBGAF1 occurs inside the cell. The results suggest that the interaction of both proteins is needed to affect the stomata morphology, and MeJA is part of the pathway that promotes the interaction between both proteins.

References

1. Rodríguez-Saavedra C, Morgado-Martínez LE, Burgos-Palacios A, King-Díaz B, López-Coria M, & Sánchez-Nieto S (2021) *Frontiers In Molecular Biosciences*, 8 (701975), 1-23
2. Hu DG, Sun CH, Zhang QY, An JP, You CX, & Hao YJ (2016) *PLoS genetics*, 8 (480), 1-15.
3. Godbole A, Dubey AK, Reddy PS, Udayakumar M, & Mathew MK (2012) *Protoplasma*, 250(4), 875-884.

TRANSCRIPTOMIC PROFILE OF SEQ-1 PEPTIDE IN STEATOTIC HEPATOCYTES: IMPLICATIONS IN LIVER FIBROSIS

Sandra Calixto-Tlacomulco¹, Jaime Mas-Oliva² and Juan Pablo Reyes Grajeda¹

¹Instituto Nacional de Medicina Genómica, Periférico Sur No. 4809, 14610, Tlalpan, CDMX, ²Instituto de Fisiología Celular, UNAM, Cto. Exterior s/n, C.U., Coyoacán, 04510, CDMX
E-mail: jreyes@inmegen.gob.mx

The Seq-1 peptide, key component of the HB-ATV-8 immunotherapy (US Patent No. US9539312, MX347400B, EP2868327), is derived from the Cholesterol Ester Transfer Protein. Administered intranasally, HB-ATV-8 has been shown to prevent liver fibrosis and atherosclerotic lesions in hypercholesterolemic rabbits and pigs^{1,2}.

This study investigates the molecular mechanisms of Seq-1 in extracellular matrix remodeling, utilizing an *in vitro* steatosis model with human HepG2 cells. We examined the transcriptomic profile of Seq-1-treated steatotic HepG2 cells, using functional enrichment analyses to reveal gene regulatory mechanisms influenced by Seq-1.

HepG2 cells were treated with a mixture of oleic and palmitic acid in a 2:1 ratio at a concentration of 0.6 mM. The experimental groups were as follows: Ct (untreated cells), FFA-Tx (cells stimulated with fatty acids), and FFA-Tx+Seq-1 (cells stimulated with FFA plus 100 µg Seq-1)³.

Using Clariom-D Affymetrix arrays we identified 9,191 differentially expressed genes across the groups. PCA and hierarchical clustering revealed distinct gene expression patterns. Functional enrichment and protein-protein interaction analyses identified molecular processes linked to fatty acid stimulation and Seq-1 peptide treatment. In the FFA-Tx group, 121 genes showed differential expression related to membrane depolarization, matrix adhesion regulation, and ribonuclease activity. In contrast, Seq-1 treatment influenced 105 genes associated with adiponectin signaling, vascular wound healing, and lipid oxidation processes. Seq-1 treatment impacted several molecular functions, including N-methyltransferase activity and histone binding. Protein-protein interaction networks indicated Seq-1's potential role in fatty acid oxidation and hepatic fibrosis pathways, suggesting promising anti-fibrotic mechanisms.

In summary, Seq-1 peptide treatment induces significant gene expression changes that modulate pathways involved in lipid metabolism, inflammation, and fibrosis in steatotic HepG2 cells. These findings highlight the potential of the Seq-1 peptide as a key component of the HB-ATV-8 vaccine in regulating molecular pathways implicated in hepatic fibrosis.

References

1. García-González, V., Delgado-Coello, B., Pérez-Torres, A., & Mas-Oliva, J. (2015). Reality of a vaccine in the prevention and treatment of atherosclerosis. *Archives of medical research*, 46(5), 427-437.
2. Gutiérrez-Vidal, R., Delgado-Coello, B., Méndez-Acevedo, K. M., Calixto-Tlacomulco, S., Damián-Zamacona, S., & Mas-Oliva, J. (2018). Therapeutic intranasal vaccine HB-ATV-8 prevents atherogenesis and non-alcoholic fatty liver disease in a pig model of atherosclerosis. *Archives of Medical Research*, 49(7), 456-470.
3. Calixto-Tlacomulco, S., Luna-Reyes, I., Delgado-Coello, B., Gutiérrez-Vidal, R., Reyes-Grajeda, J. P., & Mas-Oliva, J. (2024). CETP-derived Peptide Seq-1, the Key Component of HB-ATV-8 Vaccine Prevents Stress Responses, and Promotes Downregulation of Pro-Fibrotic Genes in Hepatocytes and Stellate Cells. *Archives of Medical Research*, 55(2), 102937-102937.

EFFECTS OF POLLUTANT PHENANTHRENE ON GLUTATHIONE-DEPENDENT ENZYMES AND GLUTATHIONE CONTENT IN THE SHRIMP *PENAEUS VANNAMEI*

Laura Camacho-Jiménez¹, Lilia Leyva-Carrillo¹, Alma Beatriz Peregrino Uriarte¹,
Silvia Gómez Jiménez¹ and Gloria Yepiz-Plascencia^{1*}

¹Group of Comparative Biochemistry and Physiology. Centro de Investigación en Alimentación y Desarrollo (CIAD), A.C., Carretera Gustavo Enrique Astiazarán Rosas, No. 46, Col. La Victoria, Hermosillo, Sonora C.P. 83304, México
E-mail: laura.camacho@ciad.mx, gyepiz@ciad.mx

Phenanthrene (PHE) is a polycyclic aromatic hydrocarbon (PAH) ubiquitous in coastal ecosystems. The shrimp *Penaeus vannamei* is native of the Northwestern coast of Mexico, where is also extensively farmed. PAHs can affect shrimp health and survival and endanger natural populations and aquaculture production. Crustaceans defend themselves from xenobiotics such as PAHs through enzymatic systems that metabolize the compounds and neutralize the reactive oxygen species (ROS) produced during their biotransformation. Glutathione (GSH)-dependent enzymes as glutathione S-transferases (GSTs), glutathione peroxidases (GPx) and some peroxiredoxins (Prx) are key enzymes to neutralize organic electrophiles and ROS that can cause oxidative damage to cells. Even though these enzymes have been studied in shrimp exposed to diverse stressors, information about their responses to PAHs and other organic pollutants is scarce. Herein we analyzed the effects of an acute (96 h) exposure of juvenile *P. vannamei* to PHE on GST delta class (GSTD), GST theta class (GSTT), GPx4 and Prx6 expression in hepatopancreas, as well as total GST and GSH-dependent activity. PHE significantly increased GSTD (24, 48 and 96 h) and Prx6 (48 h) expression, but no induction was found for the GPx4 and GSTT genes. GST activity increased after 48 and 96 h exposure, in contrast to GSH-dependent peroxidase activity, that decreased after 72 and 96 h of challenge. Moreover, total, reduced, and oxidized glutathione (GT, GSH and GSSG) were analyzed in response to the pollutant. GSH concentrations were reduced during all the exposure times, indicating its utilization for PHE metabolism or ROS neutralization. However, the GSH/GSSG ratio did not change, suggesting that hepatopancreas can maintain redox balance during short-term sub-lethal exposure to PHE. These results also indicated that GSH-dependent enzymes have different sensitiveness to PAHs, and probably different roles in their metabolism and neutralization of ROS in *P. vannamei*.

MULTIVALENT DYNAMIC COLOCALIZATION OF AVIAN INFLUENZA POLYMERASE AND NUCLEOPROTEIN BY INTRINSICALLY DISORDERED ANP32A REVEALS THE MOLECULAR BASIS OF HUMAN ADAPTATION

Aldo R. Camacho-Zarco¹, Lefan Yu¹, Tim Krischuns², Selin Dedeoglu³, Damien Maurin¹, Guillaume Bouvignies⁴, Thibaut Crépin¹, Rob W. H. Ruigrok¹, Stephen Cusack⁵, Nadia Naffakh², and Martin Blackledge^{1*}

¹Institut de Biologie Structurale, Université Grenoble Alpes, Grenoble, France; ²Institut Pasteur, Université Paris Cité, Paris, France; ³Centré de RMN à Très Hauts Champs, FRE 2034, University of Lyon, France; ⁴Laboratoire des Biomolécules, Département de Chimie, École Normale Supérieure, Université Paris 06; ⁵European Molecular Biology Laboratory, Grenoble, France; *71 Avenue des Martyrs, IBS, 38000 Grenoble, France; E-mail: martin.blackledge@ibs.fr

Adaptation of avian influenza RNA polymerase (FluPol) to human cells requires mutations on the 627-NLS domains of the PB2 subunit^(A). The E627K adaptive mutation compensates a 33-amino-acid deletion in the acidic intrinsically disordered domain of the host transcription regulator ANP32A, a deletion that restricts FluPol activity in mammalian cells^(B, C, D). The function of ANP32A in the replication transcription complex and in particular its role in host restriction remains poorly understood.

Here we characterize ternary complexes formed between ANP32A, FluPol, and the viral nucleoprotein, NP, supporting the putative role of ANP32A in shuttling NP to the replicase complex^(E). We demonstrate that while FluPol and NP can simultaneously bind distinct linear motifs on avian ANP32A, the deletion in the shorter human ANP32A blocks this mode of colocalization. NMR reveals that NP and human-adapted FluPol, containing the E627 K mutation, simultaneously bind the identical extended linear motif on human ANP32A in an electrostatically driven, highly dynamic and multivalent ternary complex.

This study reveals a probable molecular mechanism underlying host adaptation, whereby E627K, which enhances the basic surface of the 627 domain, is selected to confer the necessary multivalent properties to allow ANP32A to colocalize NP and FluPol in human cells.

Bibliography

1. Wandzik J.M. *et al.*, Structure and function of influenza polymerase. *Cold Spring Harb Perspect Med.* 2021, 11(9).
2. B. Long, J. S. *et al.* Species Difference in ANP32A Underlies Influenza A Virus Polymerase Host Restriction. *Nature* 2016, 529 (7584), 101–104.
3. C. Fan, H. *et al.* Structures of Influenza A Virus RNA Polymerase Offer Insight into Viral Genome Replication. *Nature* 2019, 573 (7773), 287–290.
4. D. Camacho-Zarco, A. R., *et al.* Molecular Basis of Host-Adaptation Interactions between Influenza Virus Polymerase PB2 Subunit and ANP32A. *Nat. Commun.* 2020, 11 (1), 3656.
5. E. Camacho-Zarco A.R., *et al.* Multivalent dynamic colocalization of avian influenza polymerase and nucleoprotein by intrinsically disordered ANP32A reveals the molecular basis of human adaptation. *J. Am. Chem. Soc.* 2023, 145, 20985–21001.

TRICHODERMA ATROVIRIDE SMALL RNA1 TARGETS THE ARABIDOPSIS PRIM2 GENE TO ESTABLISH A MUTUALISTIC RELATIONSHIP

Sergio Casas-Flores¹, Eyra Judith Hernández-Hernández¹, Mitzuko-Dautt Castro¹,
Fernanda Padrón-Rodríguez¹, Luis Fernando García Ortega¹, María del Carmen González-López¹,
Magnolia Estrada-Rivera¹ and Saúl Jijón Moreno¹

¹IPICYT, División de Biología Molecular. Camino a la presa San José No. 2055, Colonia Lomas 4a sección.
C.P. 78216. San Luis Potosí, Mexico
E-mail: scasas@ipicyt.edu.mx

The establishment of symbiotic associations between *Trichoderma* fungi and plants positively influence plant growth and resistance to pathogens. During this interaction, *Trichoderma* spp. release small RNAs (sRNAs) that potentially target host genes, aiding in the development of a beneficial relationship. However, the involvement of sRNA-mediated gene silencing in plant-*Trichoderma* interaction remains unclear. In this study, using sRNA deep-sequencing, we identified 31 potential sRNAs produced by *Trichoderma atroviride* that exhibit significant changes in accumulation levels when grown in the presence of the plant *Arabidopsis thaliana*. Among these, we focused on *Ta_sRNA1*, which shows high accumulation during *Trichoderma*-*Arabidopsis* co-culture. *Ta_sRNA1* was found to be transferred to plant roots, where it associates with *Arabidopsis* ARGONAUTE-1 (AGO1) and AGO2 proteins, targeting the *Arabidopsis* gene *PRIM2*, encoding the large subunit of DNA primase. Treatment of *Arabidopsis* roots with a *Ta_sRNA1*-overexpressing *Trichoderma* strain resulted in enhanced downregulation of *PRIM2* compared to wild-type *Trichoderma*-treated plants. Consistently, overexpression of *Ta_sRNA1* in *Arabidopsis* led to downregulation of *PRIM2*, indicating that *Ta_sRNA1* negatively regulates *PRIM2* transcripts. Furthermore, *Ta_sRNA1*-overexpressing *Arabidopsis* lines exhibited increased resistance to the necrotrophic pathogen *Botrytis cinerea*, along with enhanced expression of defense-related genes and accumulation of hydrogen peroxide in leaves, compared to wild-type *Arabidopsis* plants. Overexpression of *Ta_sRNA1* in *Arabidopsis* compromised *Trichoderma*-induced systemic resistance, suggesting a complex interplay between *Ta_sRNA1*, *PRIM2*, and plant defense responses. Additionally, we found that *Ta_sRNA1* plays a role in plant growth regulation and affects plant root colonization by the fungus, although it is not required for *Trichoderma*-induced plant growth promotion. A T-DNA insertional mutant of *PRIM2* and *PRIM2*-overexpressing *Arabidopsis* lines exhibited enhanced resistance and increased susceptibility to *Botrytis*, respectively, indicating that the *Ta_sRNA1* target *PRIM2* functions as a susceptibility gene in *Arabidopsis*. Overall, our findings shed light on the involvement of *Ta_sRNA1* in establishing a mutualistic relationship between *T. atroviride* and *Arabidopsis* by targeting *PRIM2*, influencing root colonization and priming response, ultimately impacting *Arabidopsis* growth and susceptibility to *B. cinerea*.

DESIGN AND PRODUCTION OF A H7N3 HIGHLY PATHOGENIC AVIAN INFLUENZA RECOMBINANT VACCINE

Miguel Ángel Castillo Martínez*, Leandro Alberto Núñez Muñoz*, Berenice Calderón Pérez*, América Padilla Viveros*, Roberto Ruiz Medrano*, Beatriz Xoconostle Cázares*

*Depto. de Biotecnología y Bioingeniería, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional. Av. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, 07360 Ciudad de México, CDMX. E-mail: bxoconos@cinvestav.mx

The avian industry is one of the largest in Mexico, contributing 0.75% of the gross domestic product in 2021 and it is estimated to grow by 2% compared to 2023¹. Fresh eggs and poultry meat are an important protein source for the population. Among the problems associated to this industry, pest and diseases threat its stability and production.

In 2012, an outbreak of highly pathogenic avian influenza was reported in Jalisco, affecting the production of eggs and poultry meat to the extent of increasing inflation². While there have been many strategies to mitigate the effects of influenza, such as improved biosafety protocols, sanitary actions for cleaner facilities, and financial subsidies to producers, the best alternative is the production and application of a poultry vaccine as it can be used as a protective and preventive measure¹.

Nowadays, highly pathogenic influenza H7N3 has evolved to five clades, restricted to the most economically important productive regions³.

In this work, by employing bioinformatic tools, three open reading frames encoding hemagglutinin, neuraminidase, and M2 were selected for a rational design of a recombinant vaccine candidate. Conserved, immunogenic domains were identified on the influenza virus and by reverse genetics, a synthetic gene with codon preference of *Escherichia coli* was design. The heterologous expression in the bacterial systems allow the production of a non-glycosylated antigen. The use of this recombinant protein as an elicitor of the immune system to produce potential neutralizing antibodies will be presented.

References

1. www.siap.gob.mx SIAP (Consulted, May 21, 2024).
2. www.banxico.org.mx Banxico (Consulted, May 21, 2024).
3. Navarro-Lopez, R., Xu, W., Gomez-Romero, N., Velazquez-Salinas, L., Berhane, Y. (2022). *Pathogens*, 11: 1284

SEARCH FOR LEADING MOLECULES TARGETING THE REPLICATIVE CORE OF THE SARS-COV-2 POLYMERASE FOR THE DEVELOPMENT OF SPECIFIC ANTIVIRAL DRUGS

Armando Cruz-Rangel^{1,2}; Victor Manuel Luna Pineda;
Juan Pablo Reyes Grajeda¹ and Enrique García Hernández^{*2}

¹Instituto Nacional de Medicina Genómica. Periferico Sur 4809, Arenal Tepepan, Tlalpan, 14610 Ciudad de México, CDMX;

²Instituto de Química, circuito exterior s/n Circuito de la, Investigación Científica, C.U., 04510 Ciudad de México, CDMX.

Tel. 5556224424, Email: acruz@inmegen.gob.mx, jreyes@inmegen.gob.mx, egarciah@unam.mx.

The SARS-CoV-2 genome encodes 16 non-structural proteins (nsps 1-16) that form the virus's replicative complex. Nsp12 possesses RNA-dependent RNA polymerase (RdRp) activity and, along with the nsp7 and nsp8 subunits, constitutes the minimal unit for genome replication and RNA transcription. According to Cryo-EM structures, nsp12 associates with two copies of nsp8 and one of nsp7, facilitating contact between nsp12, nsp7, and the viral RNA. These proteins form complexes among themselves and with viral RNA, suggesting the presence of transient complexes during the assembly of the replicative nucleus. Notably, nsp7 and nsp8 increase nsp12's affinity for viral RNA. Since nsp7 and nsp8 have no homology to human proteins, their contact interfaces with nsp12 are promising targets for designing molecules aimed to inhibit viral replication. This study aims to identify molecules capable of disrupting the assembly of the nsp7 and nsp8 subunits and inhibiting the activity of the SARS-CoV-2 RNA polymerase. Nsp7 and nsp8 subunits were overexpressed and purified using a bacterial heterologous system. Size-exclusion chromatography (SEC) and cross-linking experiments showed that nsp7 exhibits several oligomerization states (tetramer, trimer, dimer, and monomer), while nsp8 remains dimeric. Circular dichroism spectroscopy revealed a high content of repetitive secondary structure for nsp7, whereas nsp8 showed little secondary structure. At physiological pH, nsp7 formed high molecular weight oligomers, but when co-incubated with diferuloylmethane, dimer formation occurred. Ligand absorption coincided with protein peaks in SEC, suggesting co-elution. Fluorometric titration of the ligand with nsp7 showed fluorescence quenching, revealing at least two binding events with apparent K_d values of 0.36 μM ± 0.09 and around 5 μM ± 5, affirming the interaction. Overall, these findings propose that diferuloylmethane could be a lead molecule for the development of new antiviral drugs.

LINKING ELEVATED PROTEOLYSIS WITH CPXR REGULATORY MECHANISMS IN *SERRATIA MARCESCENS* HU1848

Lizeth De Anda¹, Carlos Alvarez¹, Samantha Barahona¹, Faviola Tavares-Carreón², Miguel A. Becerril¹, Rogelio de J. Treviño¹, Angel Andrade¹

¹Departamento de Microbiología, Facultad de Medicina, UANL, ²Facultad de Ciencias Biológicas, UANL, E-mail: angel.andradet@uanl.edu.mx

Serratia marcescens, a Gammaproteobacteria commonly found in soil and water, is globally recognized as an opportunistic multi-resistant pathogen causing nosocomial outbreaks. In *S. marcescens*, the production of secondary metabolites along with degradative enzymes, including the main extracellular protease, PrtS, is positively regulated by the two-component system, EepRS. Accordingly, regulation of the response regulator EepR is tightly controlled. Here we describe the *S. marcescens* clinical isolate HU1848, displaying higher proteolytic activity and *prtS* expression, as evidenced through azocasein degradation and qRT-PCR, respectively. Notably, we found that *eepR* expression is significantly higher in strain HU1848 compared to other *S. marcescens* isolates. An *in vivo* transcriptional assay further confirmed the increased expression of *eepR* in HU1848. Analysis of the *eepR* regulatory region revealed two potential CpxR binding motifs. The interaction of CpxR with both motifs was validated through EMSA. A *S. marcescens* HU1848 *cpxR* deletion strain was created, and the evaluation of *eepR* transcription (qRT-PCR) in this background indicated that CpxR negatively regulates *eepR*. Sequence conservation suggests that regulation of *eepR* by CpxR is common among *S. marcescens* species. Furthermore, we determined that deletion of *cpxR* in strain HU1848 causes one to two log reduction in MIC for ampicillin, nalidixic acid, and different aminoglycosides. Direct binding of CpxR to efflux pumps belonging to SMR, RND and MFS families was also confirmed by EMSA. In addition, a spontaneous CpxR point mutant (L208P), unable to interact with *prtS* promoter, was isolated. Structural mapping of this mutation places it on α -helix 8, near the CpxR DNA-binding domain. To better characterize the interaction of *S. marcescens* CpxR with its DNA target sequences, structural models of both CpxR and the CpxR DNA-binding motif were created. Through *in silico* evaluations of molecular docking key residues on α -helix 8 were revealed. Molecular dynamics, along with site-directed mutagenesis, are currently underway to validate the contribution of identified residues in CpxR interaction with its DNA target sequences. Overall, our work will provide a comprehensive understanding of the CpxR regulatory mechanisms in *S. marcescens*.

THE ACETOGENIN LAHERRADURIN AND THE ALKALOID LIRIODENINE PROMOTE APOPTOSIS AND AUTOPHAGY INDUCTION THROUGH MITOCHONDRIAL DYNAMICS DYSREGULATION IN COLORECTAL CANCER *IN VITRO*

Izamarly Delgado-Waldo^{1,2,4}, Svetlana Dokudovskaya¹, Carlos Pérez-Plasencia³, Nadia Jacobo Herrera⁴

¹CNRS UMR9018, Université Paris-Saclay, Institut Gustave Roussy, Villejuif, France. ²Posgrado en Ciencias Biológicas, UNAM, CDMX, México. ³Laboratorio de Genómica, Instituto Nacional de Cancerología, CDMX, México. ⁴Unidad de Bioquímica, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán, CDMX, Mexico.
e-mail: izz.waldo1@gmail.com

Colorectal cancer (CRC) ranks as the third most common neoplasm worldwide, with Mexico reporting 16,802 cases and 8,603 deaths annually. Despite available treatment options such as surgery, chemotherapy, and radiation therapy, prolonged exposure to conventional chemotherapeutic agents often leads to drug toxicity or resistance. Plant-derived compounds, such as alkaloids and Annonaceous Acetogenins (ACGs), offer promising alternatives. ACGs like Laherradurin (LAH) inhibit complex I of the electron transport chain, inducing cell growth inhibition in CRC models. Alkaloids like Liriodenine (LIR), a topoisomerase II inhibitor, exhibits cytotoxic activity in various cancer cell lines, including CRC. Treatment with these plant-derived compounds individually induced programmed cell death and autophagy. Our investigation revealed LAH and LIR exposure induce alterations in mitochondrial function. Notably, LAH and LIR were found to increase glycolysis and fragment mitochondria, leading to impaired OXPHOS and contributing to the induction of p-H2AX. This finding suggests that LAH and LIR exert their cytotoxic effects on CRC cells, in part by inducing apoptosis that triggers DNA damage. Moreover, dysregulation of the mTOR signaling pathway was observed, affecting cell proliferation and increasing CRC cell sensitivity to autophagy. These findings underscore the potential of LAH and LIR as innovative therapeutic agents for CRC treatment. Further investigation into their mechanisms of action is warranted to fully harness their therapeutic potential and develop more effective, targeted treatments for CRC.

PLANT CELL WALL DEGRADING ENZYMES IN THE SECRETOME OF COLLETOTRICHUM LINDEMUTHIANUM PATHOTYPES

Karla Morelia Díaz-Tapia¹, María Guadalupe Zavala-Páramo¹, Ma. Irene Morelos-Martínez¹,
Everardo López-Romero², Horacio Cano-Camacho¹

¹Centro Multidisciplinario de Estudios en Biotecnología, FMVZ, Universidad Michoacana de San Nicolás de Hidalgo, Km 9.5, Carretera Morelia-Zinapécuaro, Posta Veterinaria, Morelia, Michoacán, México, ²Departamento de Biología, División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Noria Alta SN, Guanajuato, Gto. México.

E-mail: morelia.diaz@umich.mx

The ascomycete *Colletotrichum lindemuthianum* is a phytopathogenic fungus that causes the anthracnose disease in the common bean plant (*Phaseolus vulgaris*). Presents a great diversity of pathotypes with different levels of virulence against bean varieties worldwide. *C. lindemuthianum* has a hemibiotrophic lifestyle that includes the secretion of plant cell wall degrading enzymes (PCWDEs) that function in a coordinated and synergistic to degrade the cell wall (PCW) of its host¹. The PCW is mainly composed of cellulose, hemicellulose, pectin and lignin. Knowledge about fungal PCWDEs comes mainly from studies focused on their isolation, characterization, and application; furthermore, also due to its importance in multiple biotechnological applications in industrial processes². The objective of this study was evaluating the PCWDEs in the secretomes of *C. lindemuthianum* pathotypes with different virulence level. Four pathotypes of *C. lindemuthianum* were used under culture conditions supplemented with glucose or green bean tissue for 10 days. The proteins were identified and quantified by Label-free based Quantitative Proteomic Analysis on the nano LC-MS/MS platform through the Creative Proteomic Inc. service. The results showed 400 identified proteins, of which 59 are PCWDEs (15 pectinases, 19 hemicellulases, 10 debranching enzymes, 12 cellulases and 3 auxiliary enzymes), belonging to 30 CAZy families. PCWDE secretion profiles were different between the four pathotypes and between the two treatments.

References

1. Mendgen, K.; Hahn, M. (2002). Trends Plant Sci., 7, 352-356.
2. Conejo-Saucedo et al. (2011). Curr. Trends Microbiol., 7, 1-13.

MOLECULAR HANDCRAFT OF CHIMERIC PROTEINS

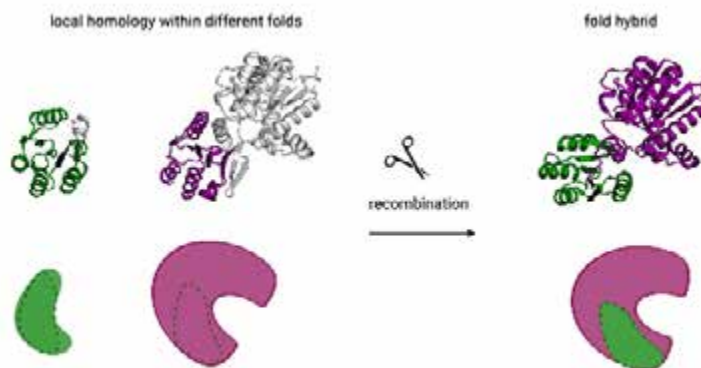
Saacnicteh Toledo-Patiño, Sara Kathrin Goetz, Sooruban Shanmugaratnam, Birte Höcker*,
José Arcadio Farías-Rico*

Corresponding Authors*

Center for Genome Sciences UNAM, Av. Universidad s/n, Universidad Autonoma del Estado de Morelos, 62209
Cuernavaca, Morelos, Mexico.

Tel: +52 7773291777; ext: 38479 E-mail: jafarias@ccg.unam.mx

Modular assembly is a compelling pathway to create new proteins, a concept supported by protein engineering and millennia of evolution. Natural evolution provided a repository of building blocks, known as domains, which trace back to even shorter segments that underwent numerous 'copy-paste' processes culminating in the scaffolds we see today. Utilizing the subdomain-database Fuzzle, we constructed a fold-chimera by integrating a flavodoxin-like fragment into a periplasmic binding protein. This chimera is well-folded and a crystal structure reveals stable interfaces between the fragments. These findings demonstrate the adaptability of α/β -proteins and offer a stepping stone for optimization. By emphasizing the practicality of fragment databases, our work pioneers new pathways in protein engineering. Ultimately, the results substantiate the conjecture that periplasmic binding proteins originated from a flavodoxin-like ancestor.



Reference

1. <https://doi.org/10.1002/1873-3468.14856>

OAEVOB: ONLINE-ADJUSTED EVOLUTIONARY BICLUSTERING ALGORITHM TO ANALYZE GENE EXPRESSION DATA

Edgardo Galán Vásquez¹, Raúl Galindo Hernández¹ and Katya Rodríguez-Vázquez¹

¹Instituto de Investigaciones en Matemáticas Aplicadas y en Sistemas, UNAM, Escolar 3000, C.U., Coyoacán, 04510 Ciudad de México, CDMX.
E-mail: edgardo.galan@iimas.unam.mx

Gene expression data analysis has been efficient using evolutionary and biclustering algorithms that help identify meaningful biological relationships in genes and detect condition-specific functional gene clusters.

In this work, we propose OAEVOB, a new evolutionary-based biclustering algorithm that efficiently handles large gene expression datasets. The algorithm performs preprocessing using quantification and normalization matrices to identify genes with null values. Subsequently, an initial scan is performed to identify highly correlated genes in each biclustering that serves as seed, these biclustering are subsequently entered into an evolutionary algorithm that performs mutations and crossovers to guarantee a set of biclustering with the highest fitness. Furthermore, OAEVOB incorporates an online adjustment function that efficiently identifies significant biclusters by updating mutation probability and crossover parameters, as well as the integration of different gene similarity metrics in biclustering such as Pearson correlation, distance correlation, biweight midcorrelation, and mutual information.

OAEVOB allows you to analyze different genetic expression data technologies that include DNA microarrays, RNA sequencing, and single-cell RNA sequencing. Therefore, we evaluated its behavior with expression data from these three technologies and OAEVOB demonstrates its versatility by successfully identifying highly correlated gene expression biclusters in all the similarity measures analyzed. Furthermore, when biclusters are evaluated by functional enrichment analysis, they exhibit biological functions, suggesting that OAEVOB effectively identifies biclusters with specific biological functions.

References

1. Huang Q., Huang X., Kong Z., et al. (2019). *IEEE Transactions on Evolutionary Computation*, 23, 803-814.
2. Liu X., Li D., Liu J., et al. (2020). *Bioinformatics*, 36, 5054-5060.

STRUCTURAL DESIGN OF A GENETIC CIRCUIT TO OBTAIN A BIOSENSOR

America Selene Gaona Mendoza¹, Julio Armando Massange Sánchez² and Luz Edith Casados Vázquez¹

¹División de Ciencias de la Vida, Universidad de Guanajuato, C.P. 36500, ²Unidad de Biotecnología Vegetal, Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A.C. C.P. 44270
E-mail: as.gaonamendoza@ugto.mx

Whole cell microbial biosensors (WCMB) are genetic tools used to detect target molecules, for example, biological or chemical contaminants¹. The functionality of the biosensor is based on the design of genetic circuits that allow the biosensor cell to transcribe and translate the elements correctly². Therefore, the objective of this work is to design a functional genetic circuit that contains the genetic elements necessary to be recognized by the cellular chassis. The biosensor model was based on the *Listeria monocytogenes* (Lm) Agr system, which consists of a histidine kinase receptor (*agrC*) that recognizes the communication molecule AIP, a regulatory element (*agrA*), and the inducible promoter to AIP, PII, which in the biosensor circuit regulates the *mCherry* reporter gene. Previously, parameters such as the codon adaptation index (CAI) and the efficient number of codons (ENc) of *agrC*, *agrA* and *mCherry* were evaluated to determine the probability of expression in *L. lactis*, which participates as a cellular chassis. Biosensor B1 was generated by Gibson assembly and was the template for obtaining biosensors B2, B3 and B4 with changes in regulatory elements such as RBS and length of spacer sequences. Subsequently, the functioning of the circuit was evaluated at the transcriptional and translational level by RT-PCR and SDS-PAGE or Western blot, respectively. As a result of the analysis of *agrC*, *agrA* and *mCherry*, genes with optimized sequences were generated that present CAI values of 1.0, 1.0 and 0.56, respectively, in addition to an ENc value of 21 for all three. Considering that an optimal CAI value is 1.0 and an ENc value of 21 represents a bias in codons towards those most used by *L. lactis*, a high translation of the mRNA into protein can be inferred³ and corroborated by the protein profile and the biosensor efficiency. Once the biosensors were generated, the changes with respect to biosensor B1 were confirmed based on a sequence alignment; for biosensors B2 and B4, when analyzing the sequence in the RBS-*agrA* region, an addition of 13 bp is presented between sites 1516 and 1517, which corresponds to the RBS "AAGGAG" that belongs to *L. lactis* and a spacing sequence of 7 bp before the ATG. While, for biosensors B3 and B4, the sequence in the RBS-*mCherry* region presents a 23 bp deletion between sites 2752 and 2776. These results will allow expanding the repertoire of possibilities to obtain a biosensor with greater detection sensitivity and speed in detecting the target molecule.

References

1. Hasan, A., Nurunnabi, M., Morshed, M., Paul, A., Polini, A., Kuila, T., Jaffa, A. A. (2014). *BioMed research international*, 2014.
2. Kosuri, S., Goodman, D. B., Cambray, C., Mutalik, V. K., Gao, Y., Arkin, A. P., Church, G. M. (2013). *Proceedings of the National Academy of Sciences*, 110, 14024-14029.
3. Puigbo, P., Guzmán, E., Romeu, A., Garcia-Vallvé, S. (2007). *Nucleic acids research*, 35, W126-W131.

DECODING THE MECHANISM GOVERNING THE STRUCTURAL STABILITY OF WHEAT GERM AGGLUTININ

Enrique García Hernández¹, Luis Fernando Cofas-Vargas², Jesús Antonio Rauda-Ceja¹
and Jorge Luis Medrano-Cerano¹

¹Universidad Nacional Autónoma de México, Instituto de Química, Ciudad Universitaria, Ciudad de México, 04510,

²Biosystems and Soft Matter Division, Institute of Fundamental Technological Research, Polish Academy of Sciences, Pawińskiego 5B, 02-106 Warsaw, Poland.

E-mail: egarciah@unam.mx

The exceptional ability of lectins to recognize glycoconjugates has been increasingly exploited for the advancement of research tools and clinical applications (1). As a potential drug carrier, wheat germ agglutinin (WGA) stands out for its significant structural stability, high water solubility, specific recognition of cell lineages, especially malignant ones, and ability to surmount barriers such as the blood-brain barrier and the enterocytes of the intestinal epithelium (2). Notwithstanding these advantages and the research endeavors conducted thus far, the utilization of WGA in human and animal therapeutics is yet in its nascent stages of development. This study delves into the basis of the structural stability of WGA (3). Utilizing differential scanning calorimetry and molecular dynamics simulations, we characterized the thermal unfolding of the full protein as well as each of its four isostructural domains independently. Additionally, we determined the 3D structures of the isolated domains using nuclear magnetic resonance. Comparative analysis of the domains, both as part of the complete protein and expressed independently, allowed us to identify key factors influencing the WGA's complex unfolding process. Crucially, we pinpointed interactions that could serve as a blueprint to enhance the protein's stability, aiming to improve its resilience against extreme acidity and proteolytic degradation during the transit through the digestive tract. These insights hold significant potential for fine-tuning the effectiveness and safety of WGA in drug transport and delivery applications.

Acknowledgements. This work was financed by DGAPA, UNAM [PAPIIT, IN206221, IN209624].

References

1. Solís D, Bovin N V, Davis AP, Jiménez-Barbero J, Romero A, Roy R, Smetana K, Gabius H-J (2015). A guide into glycosciences: How chemistry, biochemistry and biology cooperate to crack the sugar code. *Biochim. Biophys. Acta* 1850, 186–235.
2. Balčiūnaitė-Murzienė, G., Dzikaras, M. (2021). Wheat Germ Agglutinin—From Toxicity to Biomedical Applications. *Appl. Sci.* 11, 884.
3. Medrano-Cerano, J.L., Cofas-Vargas, L.F., Leyva, E., Rauda-Ceja, J.A., Calderón-Vargas, M. Cano-Sánchez, P., Titau-Delgado, G., Melchor-Meneses, C.M., Hernández-Arana, A., del Río-Portilla, F., García-Hernández, E. (2024) Decoding the mechanism governing the structural stability of wheat germ agglutinin and its isolated domains: A combined calorimetric, NMR, and MD simulation study. *Protein Sci.* 33(6), e5020.

JOINT MICROBIOME IN A SPONDYLOARTHRITIS MURINE MODEL: EXPLORATION OF GUT-JOINT AXIS

Susana Aideé González-Chávez¹, María Fernanda Alvarado-Jáquez¹,
Joan Sebastian Salas-Leiva², Jonathon E. Mohl³, César Pacheco-Tena¹

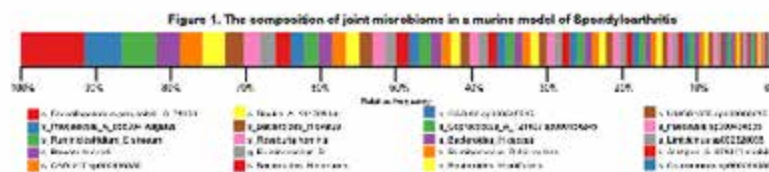
¹Universidad Autónoma de Chihuahua, Chihuahua 31125, México; ²CIMAV, Chihuahua 31136, México;
³UTEP, El Paso Tx 79968, USA.
E-mail: sagonzalez@uach.mx

Introduction. Spondyloarthritis (SpA) are characterized by inflammation and abnormal bone proliferation, leading to loss of joint function. The mechanism of bacterial dissemination in the gut-joint axis in SpA is not clearly defined¹. Joints have been considered sterile; however, a joint microbiome has recently been described, and its dysbiosis could contribute to SpA.

Objective. To confirm the existence of a joint microbiome in a murine model of SpA and compare it with the gut microbiome. Also, explore the link between joint microbiome and joint inflammation.

Materials and Methods. The SpA murine disease (SpAD) was induced in DBA/1 mice. Bacterial DNA and RNA were purified from the mice's knee joints and gut. The 16S V3-V4 region was sequenced and compared between the joint and gut. RT-qPCR evaluated bacterial RNA expression. The co-localization of bacterial components and inflammatory cytokines was assessed by immunofluorescence (IF) in mice's joints.

Results. The amount of bacterial DNA and the expression of its RNA was greater in SpAD mice than in healthy mice. Figure 1 shows the species of the joint microbiome of SpAD mice. The joint and gut microbiomes shared similarities, including their most abundant classes, *Clostridia* and *Bacteroidia*; however, the species *Victivallis plana*, *Victivallis vadensis*, and *Cloacibacillus porcorum* were only found in the joint. The IF demonstrated the colocalization of TNF- α , IL-17, IL-23, and IL-6 with Gram+ and -bacterial components in different joint structures.



Conclusion. We confirmed the joint microbiome in SpAD and healthy mice. The presence of bacteria's RNA advised their viability. Joint bacteria are shared with those of the gut, which could suggest their translocation in the gut-joint axis. The pro-inflammatory bacteria *Prevotella copri* and *Faecalibacterium prausnitzii* in joints of SpAD mice are consistent with findings in human arthritides. The co-localization of inflammatory cytokines with bacterial components in joint structures links the joint inflammation and the joint microbiome.

References

1. Qaiyum Z, Lim M, Inman RD (2021). The gut-joint axis in spondyloarthritis: immunological, microbial, and clinical insights. *Semin Immunopathol*, 43(2):173-192.

HIS-TAGGED PROAPOPTOTIC KLAKE PEPTIDE: A DUAL PURPOSE STRATEGY AND THEIR ANTICANCER ACTIVITY *IN VITRO*

Aldo O. González-Cruz¹, José Juan Pérez-Trujillo², Ulises Edgardo de León-Covarrubias²,
Elizabeth Ochoa-Muñoz¹, Mónica A. Ramírez-Cabrera¹, Isaías Balderas-Rentería¹, Eder Arredondo-Espinoza¹

¹Universidad Autónoma de Nuevo León, UANL, Facultad de Ciencias Químicas, Laboratorio de Farmacología Molecular y Modelos Biológicos. Av. Guerrero S/n, Treviño, 64570 Monterrey, N. L., México. ²Universidad Autónoma de Nuevo León, UANL, Facultad de Medicina, Departamento de Histología. Av. Dr. José Eleuterio González 235, Mitras Centro, 64460 Monterrey, N.L., México.

E-mail: eder.arredondosp@uanl.edu.mx

Approximately 20 million new cancer cases and 10 million deaths from this disease are recorded worldwide annually. Chemotherapy is generally a cost-effective regimen, however, it has associated drawbacks as drug resistance and side effects.¹ Hence, the improvement of anticancer molecules is needed. An attractive strategy is the proapoptotic peptides, such as the KLAKE peptide to induce apoptosis by disrupting the mitochondrial membrane in cancer cells. Sadly, these peptides have poor internalization activity on their own and require the coupling of a drug-delivery system or modification of their amino acid sequence.² Therefore, this study aimed to evaluate a new His-tagged KLAKE peptide by assessing, its cytotoxic and proapoptotic activities, as well as its internalization capacity, *in vitro*. The MTT method was used to evaluate the cytotoxicity of the proapoptotic peptide KLAKE in comparison with KLAKE fused to the EGFR-targeting CPP NRPDSAQFWLHH, both with and without a His-tag, using MCF-7, A-549, CT26, and Vero cell lines. Additionally, TUNEL and immunofluorescence assays were performed to confirm the proapoptotic effect and peptide cell uptake, respectively. The results show that the His-tagged KLAKE peptide can induce cell death preferentially in A-549 and CT26 cancer cells, with IC₅₀ values of 33.33±2.87 µM and 42.28±3.21 µM, respectively. In contrast, KLAKE without the His-tag had no significant effect on any cell line. This may be due to interactions between the histidine residues and the lipid bilayer under the acidic balance in the cancer cell microenvironment,³ which facilitate cell internalization, as observed in the immunofluorescence assay. In summary, a His-tag modification to the KLAKE peptide can be a useful tool for identifying its presence through immunological methods while also enhancing the cell penetration capability.

References

1. Chhikara BS, Parang K. Global Cancer Statistics 2022: the trends projection analysis. *Chemical Biology Letters*. 2023;10(1).
2. Wang D, Chen L, Wang M, et al. Delivering Proapoptotic Peptide by HSP Nanocage for Cancer Therapy. *Macromol Chem Phys*. 2020;221(12):2000003. doi:10.1002/macp.202000003
3. Xiang B, Jia XL, Qi JL, et al. Enhancing siRNA-based cancer therapy using a new pH-responsive activatable cell-penetrating peptide-modified liposomal system. *Int J Nanomedicine*. 2017;Volume 12:2385-2405. doi:10.2147/IJN.S129574

RESVERATROL INHIBITS THE PI3K/AKT INSULIN PATHWAY BY ACTIVATING CLASSICAL PKCS

Karla Daniela Hernández González¹ & Jesús Alberto Olivares-Reyes^{2*}

^{1,2}Laboratory of Signal Transduction, Department of Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute, Av. Instituto Politécnico Nacional No.2508, Colonia San Pedro Zacatenco, C.P 07360, CDMX, México.

*E-mail: jolivare@cinvestav.mx

Resveratrol (RSV) is a polyphenolic compound reported for its multiple benefits in treating cardiovascular diseases, obesity, diabetes mellitus, and cancer. The positive effects of RSV have been mainly associated with its antioxidant activity and with the increased expression of proteins that metabolize free radicals and reactive oxygen species (ROS)¹. However, there is controversy regarding the effect of RSV on the insulin pathway in the liver, where inhibition of PI3K and Akt has been reported^{2,3}. Inhibition of the PI3K/Akt metabolic pathway by RSV was investigated in liver C9 cells and Hepa1-6 cells, and it was shown that RSV affects Tyr phosphorylation of the insulin receptor (IR) and causes inhibition of Akt and glycogen synthase (GS). The effects of RSV on p-Akt and p-GSK3 α/β were prevented with the use of the PKC inhibitors, BIM-I (for classical and new PKCs) and Gö697 (for PKC α and β), with more significant recovery of p-Akt-Ser⁴⁷³ with Gö697, suggesting that classical PKCs predominantly mediate the effects of RSV. We subsequently analyzed the effect of RSV and insulin on the activation of classical PKC isoforms that are associated with the condition of insulin resistance in the liver and observed an increase in the phosphorylation of PKC α (Ser⁶⁵⁷), PKC β II(Thr⁶⁴¹) and total serines of PKC β I. Furthermore, we demonstrate that RSV can activate PKC α independently of insulin and promotes its interaction with IR. Although the negative regulation of the insulin pathway at the IR level appears to be regulated by classical PKC, RSV also promotes the phosphorylation of new PKC isoforms, such as PKC δ (Ser⁶⁴³) and PKC ϵ (Ser⁷²⁹), and the interaction of the latter with IR. The activation of new PKCs could be associated with regulatory mechanisms downstream of the IR, such as the activation of GSK3 α and MAPK proteins, which RSV positively regulated through a mechanism that also depends on PKCs. Furthermore, we also demonstrated that RSV could increase the phosphorylation in Tyr¹⁵² of phosphatase PTP1B, which could be participating in the RSV-induced IR dephosphorylation. These observations allow us to conclude that RSV inhibits the PI3K/Akt pathway at the level of the IR by activating classical isoforms of PKC and PTP1B. Furthermore, the effects on GSK3 α/β and ERK1/2 suggest that RSV regulates other mechanisms downstream of IR, in which new PKCs could be involved.

Funding. This project was supported by CINVESTAV-IPN and KDH-G hold a CONAHCYT graduated scholarship (925174).

References

1. Li, Z. *et al.* Antioxidant Activity and Mechanism of Resveratrol and Polydatin Isolated from Mulberry (*Morus alba* L.). *Molecules* **26**, (2021).
2. Fröjdö, S., Cozzone, D., Vidal, H. & Pirola, L. Resveratrol is a class IA phosphoinositide 3-kinase inhibitor. *Biochem. J.* **406**, 511-518 (2007).
3. Zhang, J. Resveratrol inhibits insulin responses in a SirT1-independent pathway. *Biochem. J.* **397**, 519-527 (2006).

INDIVIDUAL miRNAS REGULATE SETS OF TRANSCRIPTS THAT ENCODE FOR PHYSICALLY INTERACTING SPECIFIC PROTEINS WITH EACH OTHER

Alma Lilia Hernández-Olvera¹, Mariana Flores-Torres¹, Felipe Vadillo-Ortega^{1,2},
Erika Chavira-Suárez^{1,2}, Humberto Gutiérrez³

¹Unidad de Vinculación Científica de la Facultad de Medicina, UNAM en el Instituto Nacional de Medicina Genómica. 4809 Periferico Sur, Arenal de Tepepan, 14610 Tlalpan, Mexico City, Mexico. ²Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México. 411^a. Circuito Escolar, Copilco Universidad, 04360 Coyoacán, Mexico City, Mexico.

³División de Genómica Computacional, Instituto Nacional de Medicina Genómica. 4809 Periferico Sur, Arenal de Tepepan, 14610 Tlalpan, Mexico City, Mexico. Tel: 5545446213

E-mail: lilia.olvera.99@gmail.com

microRNAs (miRNAs) are small non-coding transcripts that play a central role in the regulation of cognate target messenger RNAs at the post-transcriptional level. miRNAs have been implicated in the regulation of numerous biological processes through their ability to regulate a wide variety of gene targets. On the other hand, individual transcripts (messenger RNAs) can be regulated by multiple miRNAs. Despite the recent advancements in miRNA target prediction, it remains unclear what are the common properties associated with the targets of individual miRNAs. By examining sets of predicted targets for all currently annotated human miRNAs (using the TargetScan and miRTarBase repositories) and their protein-protein interactions, we found that protein targets of miRNAs sets associated with distinct functions display a statistical excess of protein-protein interactions when compared to equally sized background protein samples. This enrichment holds equally true for sets of functionally related miRNAs as well as for random sets and/or individual miRNAs. Overall, our results demonstrate that miRNAs regulate coding transcripts whose protein products engage in direct protein-protein interactions and suggest that miRNAs in general regulate cellular pathways involving clusters of physically interacting proteins.

β -HAIRPIN OF *THERMUS THERMOPHILUS* LACCASE: A REGULATOR OF ENZYMATIC ACTIVITY?

Leticia León-Luna¹, Enrique Rudiño Piñera¹, Paloma C. Gil-Rodríguez¹

¹Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología, Universidad Nacional Autónoma de México, 2001 Universidad Av., Cuernavaca, Morelos 62210, México.
E-mail: leticia.leon@ibt.unam.mx

Laccases (EC 1.10.3.2) are members of a family of enzymes known as multicopper oxidases (MCOs), representing the largest subgroup of blue multicopper oxidases. These enzymes exploit the redox properties of copper ions to catalyze the oxidation of a wide variety of substrates, while simultaneously reducing molecular oxygen to water¹. MCOs can be classified into two broad categories: those with high substrate specificity and those with low substrate specificity². Laccases, which fall into the latter category, oxidize a diverse array of organic molecules to facilitate various processes in plants, fungi, and bacteria. Additionally, plant and bacterial laccases typically exhibit low redox potential, whereas high redox potential laccases are more commonly found in fungi. Consequently, the laccases currently used in industry are primarily derived from fungi³. However, these fungal laccases are sensitive to environmental changes, leading to significant interest in discovering robust proteins capable of functioning under extreme conditions. This has directed attention towards extremophilic proteins for potential industrial applications. Thus, we chose to study the laccase from *Thermus thermophilus*, an extremophilic organism isolated from hydrothermal vents in Japan, where temperatures can exceed 100 °C⁴. This enzyme has a distinctive structural feature rich in methionines at the substrate oxidation site entry, termed the β -hairpin⁵. The β -hairpin exhibits a dynamic, pH-dependent behavior that alters the exposed surface of the entry cavity to the T1Cu site, impeding substrate proximity to the active site. This region is the most flexible and mobile in the enzyme's structure⁶. Two enzyme mutants were created by removing the β -hairpin at different positions ($\Delta 1$ and $\Delta 2$) to assess the impact of these deletions on laccase activity. Both mutants proved to be functional despite the removal of 16 and 25 residues in the $\Delta 1$ and $\Delta 2$ variants, respectively. Additionally, the enzymes retained their activity even after exposure to temperatures above 60 °C, indicating that the deletions did not affect thermostability. Although the $\Delta 1$ mutant did not show apparent improvements in catalytic constants, the $\Delta 2$ mutant exhibited significant changes in kinetic parameters, demonstrating higher substrate affinity and improved specificity constants in most cases. Regarding pH, the activity range of the enzyme was broadened in both $\Delta 1$ and $\Delta 2$, despite the pH dependency on the substrate used.

Reference

1. doi: 10.1007/s00018-009-0169-1 (2010).
2. doi: 10.1007/s00018-014-1826-6 (2015).
3. doi: 10.1016/j.biotechadv.2014.12.007 (2015).
4. doi: 10.1007/s00792-005-0458-z (2005).
5. doi: 10.1107/S174430911103805X (2011).
6. doi: 10.1016/j.jsb.2021.107740 (2021).

AT PHYSIOLOGICAL PH AND TEMPERATURE, L-TYROSINE CAN INHIBIT THE FORMATION OF AMYLOID FIBERS OF HUMAN LYSOZYME

Santos López^{1,2}, Arturo Rojo-Domínguez², Hugo Nájera^{2*}

¹Posgrado en Ciencias Naturales e ingeniería, Universidad Autónoma Metropolitana–Cuajimalpa, Mexico City, México;

²Departamento de Ciencias Naturales, Universidad Autónoma Metropolitana–Cuajimalpa, Mexico City,

Av.Vasco de Quiroga 4871, Colonia Santa Fe Cuajimalpa, Alcaldía Cuajimalpa de Morelos, México, Ciudad de México, C.P. 05300.

Tels. (52 55) 2636 3800 Ext. 3853 e mail: hnajera@cua.unam.mx

The amyloid fibers are related to many diseases. Investigating this area is essential for discovering compounds that help to treat these affections. The present research demonstrates three critical aims: 1. The formation of amyloid fibers at physiological pH and temperature. 2. The capacity of L-tyrosine to inhibit the formation of amyloid fibers of human lysozyme. 3. A proposal of the action mechanism of this compound.

The effect of L-tyr is observable with a molar ratio (1:1) on physiological conditions pH 7.4 at 37 °C. Reduced to half the fluorescence signal of thioflavin T, *i.e.*, decreased amyloid formation. On the other hand, in chemical and temperature denaturation, a change in the spectrum of the intrinsic fluorescence of the lysozyme is visible when the L-tyrosine is present; besides, the differential scanning calorimetry experiment shows a significant difference in C_p values when the compound is present, these results suggest a real interaction between them. The docking MOE evidence shows that L-tyr is in the same place as the natural substrate, but the interactions are with different amino acids, which could be the key to the inhibition effect. This work demonstrates that an essential amino acid like L-tyrosine has a good inhibition power in forming amyloid fibers through its stabilizer effect on the protein.

IDENTIFICATION OF DIFFERENTIAL PROTEINS PRODUCED IN HYPO- AND HYPER-VIRULENT MUTANTS GROWN IN WHEAT COLEOPTILES COMPARED TO THE WILD TYPE STRAIN OF *FUSARIUM GRAMINEARUM*

Ana Lilia Martínez-Rocha¹*, Bruno Guillermo Hidalgo-Hernández¹, Jorge Eduardo Hernández-Espinosa¹, J. Félix Gutiérrez-Corona¹, Estela Ruiz-Baca²

¹University of Guanajuato, Division of Natural and Exact Sciences, Department of Biology. *Noria Alta s/n, Col. Noria Alta, Guanajuato, Gto. 36050, México. Tel: +52 473 7320006 ext.8296.

²University Juárez of Durango State, Faculty of Chemistry Sciences.

*E-mail: anamartinez@ugto.mx

The ascomycete fungus *Fusarium graminearum*, is one of the principal and most devastating agronomically important plant pathogens, since it is the causal agent of Fusarium head blight in wheat (FHB) and corn ear rot¹. In *F. graminearum*, overexpression of the gene coding for the enzyme deoxyhypusine synthase (DHS_{oex}) produces a hypervirulent phenotype. In contrast, overexpression of the gene coding for deoxyhypusine hydroxylase (DOHH_{oex}) produces a hypovirulent phenotype compared to the wild type strain (WT)². The enzymes DHS and DOHH are involved in the activation of the eucaryotic translation initiation factor 5A (eIF5A) by synthesizing the unique amino acid hypusine³. In this work, we evaluated the infection and proliferation of the mutants DHS_{oex}, DOHH_{oex}, and the WT strain of *F. graminearum* expressing GFP constitutively, inside wheat coleoptiles by fluorescence microscopy. Results revealed that DHS_{oex} mutant proliferated faster than the WT strain inside the coleoptiles, while DOHH_{oex} mutant only grew outside the coleoptiles without penetration. Furthermore, we determine the amount of genomic DNA of the mutants and WT strain inside the coleoptiles using qPCR and the gene TRI5 coding for a trichodiene synthase, a one-copy gene exclusive of *F. graminearum*. Finally, we identified proteins with differential expression produced in DHS_{oex} and DOHH_{oex} mutants compared to WT, involved in the pathogenesis and proliferation of the fungus inside wheat coleoptiles, through the absolute quantification of proteins using mass spectrometry LC-MS. Identification of such proteins could be the key to determining specific targets necessary to control the growth of this pathogen.

References

1. Mentges, M., Glasenapp, A., Boenisch, M., Malz, S., Henrissat, B., Frandsen, R. J., ... & Martínez-Rocha, A. L. (2020). Molecular Plant Pathology, 21(8), 1070-1087.
2. Martínez-Rocha, A. L., Woriedh, M., Chemnitz, J., Willingmann, P., Kröger, C., Hadelers, B., ... & Schäfer, W. (2016). Scientific reports, 6(1), 24698.
3. Park, M. H. (2006). Journal of biochemistry, 139(2), 161-169.

EFFECT OF CO-CULTURE OF *A. FLAVUS* AND *P. OSTREATUS* ON THE EXPRESSION OF GENES ASSOCIATED WITH THE BIOSYNTHESIS OF AFLATOXINS AND DETOXIFYING ENZYMES

Luis Jesús Martínez Tozcano¹, Soley Berenice Nava Galicia¹, Martha Dolores Bibbins Martínez¹

¹Centro de Investigación en Biotecnología Aplicada, Instituto Politécnico Nacional. Ex-Hacienda San Juan Molino Carretera Estatal Tecuexcomac-Tepetitla Km 1.5, Tlaxcala C.P. 90700, Mexico.

Email: lmartinezt2102@alumno.ipn.mx, mbibbinsm@ipn.mx

Aflatoxins are secondary metabolites produced mainly by the saprophytic plant pathogen *Aspergillus flavus*, in particular, aflatoxin B₁ is classified as the mycotoxin with the highest toxicity to humans and animals. The infection and production of aflatoxins by *A. flavus* affects numerous crops of agroeconomic interest worldwide, so the quality and safety of grains are a priority for both the health and economic sectors. There are different methods for the control and/or regulation of the aflatoxin production, among them biological methods stand out because they represent safer, environmentally friendly and economically accessible methods compare with the chemical methods. White rot fungi, such as *Pleurotus ostreatus*, produce non-specific enzymes with the ability to degrade xenobiotics and recalcitrant agents such as aflatoxins. The aim of this research was to evaluate at the transcriptional level the effect of co-culture on the differential expression of oxidase genes and aflatoxin biosynthesis of *P. ostreatus* and *A. flavus*, respectively. Co-cultures were established in liquid medium and expression levels were monitored using RT-qPCR. RT-qPCR analysis showed a differential relative regulation (\log_2) dependent on co-culture time for genes related to aflatoxin biosynthesis and oxidase genes studied. After 78 h, negative regulation of the genes *aflR*, *aflD*, *aflM*, *BrlA*, *laeA* was observed, with expression levels of -3.85, -2.16, -0.61, -3.31, -1.87 respectively, on the other hand, *aflS* presented an induction level of 3.98. On the other hand, induction was observed in the expression levels of all oxidase genes evaluated, with the highest expression level detected for the gene coding for the dye peroxidase enzyme (*dyp4*) at 78 h with an induction level of 2.51. In co-culture, aflatoxin production was not detected, in contrast to *A. flavus* monoculture, where AFB₁ levels of 65 ppm were reached after 78 h of culture. According to the previous results, the biosynthesis/degradation of aflatoxins is affected in the presence of *P. ostreatus* with the possible participation of oxidases such as dye peroxidase and other metabolites with inhibitory activity produced by this organism.

ENERGETIC IMPAIRMENT IN CARDIOMETABOLIC HEART FAILURE: FROM TRANSCRIPTOMICS AND MITOCHONDRIAL COMPLEXOME PROFILING TO CARDIOMYOCYTE RESPIRATION

Abraham Méndez-Fernández¹, Luis A. Luévano-Martínez¹, Alfredo Cabrera-Orefice², Ángel Fernández-Mora¹, Felipe Salazar-Ramírez¹, Bianca Nieblas-León¹, Noemí García¹, Víctor Treviño¹, Guillermo Torre-Amione¹, Gerardo García-Rivas^{1*}

¹Tecnológico de Monterrey, Institute for Obesity Research, Experimental Medicine and Advanced Therapies Unit, Hospital Zambrano Hellion, TecSalud, Monterrey, Nuevo León, Av. Batallón de San Patricio 112, Real San Agustín, 66260 San Pedro Garza García, N.L., ²Functional Proteomics Center, Institute for Cardiovascular Physiology, Goethe University, Frankfurt am Main, Germany *8115163692; gdejesus@tec.mx

Background. Heart failure stands as a top cause of death worldwide. In particular, cardiometabolic heart failure with preserved ejection fraction (HFpEF) can be conceived as the cardiac manifestation of a systemic metabolic disturbance, typically driven by obesity. In the present study, transcriptomic data suggested mitochondrial impairment, confirmed by functional assays and mitochondrial complexome profiling. **Methods.** HFpEF mice were fed with a high-fat diet and exposed to L-NAME for eight weeks. Gene expression of the heart was assessed via next-generation-based RNA-seq. Activity of respiratory complexes was studied in BN-PAGE in-gel activity assays. High-resolution respirometry was used to measure the oxygen consumption of ventricular cardiomyocytes. The abundance of the subunits of respiratory complexes was explored via mass-spectrometry-based complexome profiling. **Results.** HFpEF animals were characterized by visceral fat accumulation, hypertension, and diastolic dysfunction. RNA-seq uncovered 1535 differentially-expressed genes (DEGs) for HFpEF vs. CTRL. 146 DEGs (142 down-regulated in HFpEF) were contained in the GO-CC “mitochondrion” term, mainly affecting respiratory complexes I, IV, and V. High-resolution respirometry revealed that the spare respiratory capacity was compromised in HFpEF myocytes (CTRL 2.3 ± 0.23 , HFpEF 1.6 ± 0.12) along with a 1.5-fold increase of leak respiration. In-gel activity of complex V was decreased to 88% in solubilized mitochondrial membranes of HFpEF, compared with CTRL. Mass spectrometry revealed hyperacetylation affecting several complex V subunits. **Conclusion.** Our findings suggest impaired mitochondrial bioenergetics in the heart of a mouse model of cardiometabolic HFpEF, probably due to ATP-synthase hyperacetylation and changes in complexome profile.

STRUCTURAL INSIGHTS INTO THE GLUTATHIONE TRANSFERASE SIGMA CLASS FROM TAENIA SOLIUM: CRYSTALLOGRAPHIC ANALYSIS AND FUNCTIONAL IMPLICATIONS

Ricardo Miranda Blancas¹, Cesar Sanchez², María Cristina Cardona Echavarría¹, Roberto Flores López³, Ponciano García Gutiérrez², Rafael Arturo Zubillaga², Enrique Rudiño Piñera¹, Abraham Landa³

¹ Instituto de Biotecnología, UNAM, Cuernavaca, Morelos, C. P. 62215, México. ² Departamento de Química, UAM-I, Ciudad de México, C. P. 09340, México. ³ Facultad de Medicina, UNAM, Ciudad Universitaria, Ciudad de México, C. P. 04510, México.
E-mail: ricardo.miranda@ibt.unam.mx

Taeniasis and cysticercosis are health problems listed among the 20 neglected diseases prevalent in tropical areas. Additionally, *T. solium* has developed strategies involved in the modulation of the host immune response. In this context, glutathione transferase enzymes play an important role in the detoxification process to maintain the homeostasis of the parasite. The glutathione transferase sigma class of *T. solium* (Ts24GST) was identified in the genome project, and the present work shows its expression in different life cycle stages. Furthermore, the gene was characterized and used to develop a recombinant expression system. The overexpressed protein was purified and identified by Western Blot (WB). Kinetic parameters were determined, and the pH and temperature stability were described. Finally, the crystallographic structure was determined at a resolution of 1.3 Å. This structure was used for molecular dynamics focused on the conformational stability of Ts24GST and structural analysis. The results obtained in this work reinforce the idea that sigma-type glutathione transferases could be involved in functions beyond detoxification systems.

References

1. Miranda-Blancas R, Rodríguez-Lima O, García-Gutiérrez P, Flores-López R, Jiménez L, Zubillaga RA, Rudiño-Piñera E, Landa A. (2024). FEBS Open Bio, 14(5), 726-739.
2. Nguyen HA, Bae YA, Lee EG, Kim SH, Díaz-Camacho SP, Nawa Y, Kang I, Kong Y. A novel sigma-like glutathione transferase of *Taenia solium* metacestode. (2010). Int J Parasitol. 40(9), 1097-1106.

BACTERIAL MICROBIOTA DURING EMBRYONIC DEVELOPMENT: A MATERNAL INHERITANCE

Yendi E. Navarro Noya¹, Nina Montoya Ciriaco¹, Elizabeth Selene Gómez Acata¹, Luc Dendooven¹,
Stephanie Hereira Pacheco¹, Anibal H. de la Vega Pérez²

¹Laboratorio de Interacciones Bióticas, Centro de Investigación en Ciencias Biológicas, Universidad Autónoma de Tlaxcala, Tlaxcala, Mexico, ²Estación Científica La Malinche, Centro Tlaxcala de Biología de la Conducta, Universidad Autónoma de Tlaxcala, Tlaxcala, Mexico.
E-mail: yendiebenezer.navarro.n@uatx.mx

The bacterial microbiota plays a crucial role in different aspects of animal hosts, including nutrient acquisition, energy balance, physiology, reproduction, immunity, behavior, and organ development. However, our understanding of the microbiota's role during embryonic development in non-mammalian viviparous vertebrates remains limited.

In this study, samples from the intestine, mouth, cloaca, and the aseptic ventral skin from maternal individuals of the viviparous lizard *Sceloporus grammicus* Wiegmann, 1828 were taken to compare their microbiota with that found in the embryo's intestinal tract and amniotic environment (amniotic fluid, membrane and extraembryonic yolk) at the last stage of development. We used metabarcoding of the 16S rRNA to explore the bacterial microbiota in the gastrointestinal tract and amniotic environment of lizard embryos and mothers.

We found that bacterial 16S rRNA genes were present in the embryos of *S. grammicus*, albeit with low diversity and similar community structures, suggesting strong controls on maternal transmission of microorganisms to the embryo. Furthermore, 78% of the embryonic amplicon sequence variants (ASVs) were found in the maternal bacterial microbiota, indicating that the remaining 28% may have been transmitted during earlier stages of pregnancy. The embryonic bacteria showed greater similarity to those found in the mother's mouth and aseptic ventral skin than those in the cloaca and intestine, suggesting a possible maternal origin of the transmitted bacteria. Additionally, the bacterial composition was more alike among embryos, even across different mothers.

These findings provide evidence of vertical microbiota transfer during embryonic development in non-mammalian viviparous vertebrates, suggesting that the transmission of bacteria from the mother to the embryos is a controlled process that begins during embryonic stages.

OLIGOMERIC REGULATION OF CYSTATHIONINE BETA-SYNTASE MEDIATED HYDROGEN SULFIDE PRODUCTION MODULATES UPR INDUCTION

Elías Nieto Zaragoza^{1*}, Emilio Espinoza-Simon¹, Damián Ramirez-Robles¹, Nayeli Torres-Ramírez², Albert Guskov³, Nuria Sanchez-Puig⁴, Francisco Torres-Quiroz^{1*}

¹Instituto de Fisiología Celular, Universidad Nacional Autónoma de México (UNAM), ²Facultad de Ciencias, Universidad Nacional Autónoma de México (UNAM), ³Groningen Institute for Biomolecular Sciences and Biotechnology, University of Groningen, Instituto de Química, ⁴Universidad Nacional Autónoma de México (UNAM)
E-mail: +enieto@ifc.unam.mx *ftq@ifc.unam.mx

Proteins can form heteromeric complexes with other interacting partners or homomeric complexes by interacting with identical proteins. The latter interaction can lead to novel properties, such as the exposure of allosteric sites, stabilization in high-order forms, and alterations in the catalytic site. Recent advances in protein structure prediction suggest that approximately 20% of eukaryotic proteomes form homomers, ranging from dimers to high-order filaments.

The promiscuous enzyme cystathionine beta-synthase (CBS) catalyzes the condensation of serine and homocysteine to cystathionine, but also the production of hydrogen sulfide (H₂S) from cysteine. This conserved enzyme has been characterized as a dimer and a tetramer. However, there is little information about the dynamics behind these two conformations. Recently, a filament model for human CBS was reported, shedding light on the diverse oligomeric regulation of this enzyme.

H₂S is a gasotransmitter capable of generating post-translational modifications on cysteine residues. An imbalance of hydrogen sulfide metabolism has been linked to chronic degenerative diseases such as diabetes, Alzheimer's disease, and cancer. In such pathologies, cells exhibit the development of ER stress.

We found that hydrogen sulfide metabolism is crucial for the unfolded protein response (UPR) during ER stress. We also described a novel response to ER stress involving global post-translational modification of protein thiols. CBS, encoded by CYS4, is a central node in this regulation. Kar2 and Ire1 expressions are downregulated, and UPRE transcription is upregulated in a CYS4 mutant. Using point mutations, we discovered that H₂S production, but not the canonical activity, is responsible for this regulation.

Purifying the wild-type γ CBS and the C301S mutant, we found that a large supramolecular structure forms in the wild-type protein in a concentration-dependent manner, increasing protein stability. Atomic force microscopy revealed that a filament-like structure forms when treated with L-cysteine at high protein concentrations. In contrast, using the single-particle assay Mass Photometry, L-cysteine destabilized the protein into dimers at nanomolar concentrations.

Finally, we found that this mechanism is conserved, as CBS from rat hepatocytes formed foci and filament-like structures in vivo when treated with L-cysteine.

This project was financed by UNAM-DGAPA-PAPIIT through the research program IN208922 and by the Consejo Nacional de Humanidades, Ciencias y Tecnologías through the Ciencia de Fronteras 2023 program CF-2023-I884.

UNVEILING THE UPTAKE AND THE EARLY GENETIC RESPONSES OF *ARABIDOPSIS* TO GROWTH PROMOTING MICROBIAL VOLATILES

Z. Fernandina Nieves-López¹, Robert Winkler¹, Héctor Guillén-Alonso^{1,2}, Raúl Alcalde-Vázquez¹, Camilo Pérez-Salazar¹, Laila Pamela Partida-Martínez^{1*}

¹Centro de Investigación y de Estudios Avanzados (Cinvestav), Irapuato, 36824, México,

²Instituto Tecnológico Nacional, 38010 Celaya, Guanajuato, México.

E-mail: laila.partida@cinvestav.mx

Chemical communication between plants and their microbiome via volatile organic compounds (VOCs) influences plant development, physiology, and stress responses. While some VOCs can enter plant tissues and induce genetic changes in plants, the specific genetic responses remain unclear. The microbial VOCs ethyl isovalerate (EV) and camphene (CAMP), emitted by *Bacillus* and *Beauveria pseudobassiana* strains, respectively, had beneficial effects in *Arabidopsis thaliana* when exposed to bioactive doses in a closed system for 14 days. In this work, *Arabidopsis* seedlings were exposed to bioactive doses of EV and CAMP for two hours. Using a modular biological mass spectrometer (MoBiMS) and low-temperature plasma coupled to mass spectrometry, we demonstrated that *Arabidopsis* absorbs these volatiles, taking up approximately 0.703 and 3.19 ppm per 100 mg biomass of EV and CAMP, respectively. This exposure increased the number of siliques and seeds without affecting seed germination. mRNA-seq analysis revealed early genetic responses in roots in exposed plant to EV and CAMP, including signalling pathways of auxin, gibberellin, abscisic acid, WRKY transcription factors, lncRNAs, and miRNAs. In shoots, genes related to pathogen attack, stress responses, flowering, and growth were induced. CAMP-induced miRNAs in roots repressed fungal defense responses, while EV-induced miRNAs modulated transcription factors affecting flowering time and cell wall assembly. These findings will help identify, through the use of *Arabidopsis* mutants, which genes are regulated by EV and CAMP, enhancing our understanding of chemical signal perception between plants and microbial VOCs.

EFFECT OF PROTEIN OVEREXPRESSION AND SILENCING EIMYB23 ON THE ENCYSTMENT OF ENTAMOEBIA INVADENS

Jatziry Daniela Ocampo Ulloa^a, Alan Jesé Rodríguez Almaraz^b, Patricia Areli Cuellar Silva^b, Elizabeth J. Castañeda-Ortiz^b, Abigail Betanzos Fernández^c, Jesús Valdés Flores^a, Elisa Irene Azuara Liceaga^{b*}

^aDepartamento de Bioquímica, CINVESTAV-IPN, Ciudad de México C.P. 07360. ^bLicenciatura y Posgrado en Ciencias Genómicas, Universidad Autónoma de la Ciudad de México, Ciudad de México C.P. 03100. ^cDepartamento de Infectómica y Patogénesis Molecular, Cinvestav-IPN, Ciudad de México C.P. 07360. *S. Lorenzo 290, Col del Valle Sur, Benito Juárez, Ciudad de México, C.P. 03100. Tel: 54886661 Ext.15302. Elisa.azuara@uacm.edu.mx

Entamoeba histolytica is a protozoan that causes human amoebiasis, the third cause of death due to parasitic infections which increases to one hundred thousand deaths annually.¹ The infection begins with ingesting cysts, which is the transmission stage of the parasite. Trophozoites can invade and damage the intestinal mucosa through various virulence factors whose transcriptional expression is regulated by several transcription factors (TFs).²⁻⁴ The most abundant TFs in this parasite are of the Myb family, which has a highly conserved MYB DNA binding domain (DBD-Myb). Three families comprise the EhMyb proteins. Family I includes EhMyb10, which has two repeats (R1 and R2) in the DBD Myb. Interestingly, an increase in the EhMyb10 expression was reported in trophozoites interacting with mouse intestinal cells or when they are incubated with human colon explants.⁵ On the other hand, because *E. histolytica* cannot be encyst *in vitro*, it has been challenging to analyze this process; yet, *E. invadens* has been employed as a study model to clarify the cellular mechanisms behind encystment.⁶ that may be indirectly related to the virulence of the parasite. An RNA-seq analysis of 11,549 genes in *E. invadens* showed global transcriptional changes occurring upon encystment and excystment. In another transcriptomic study of *E. invadens* encystment using DNA microarrays, some upregulated genes encoded Myb transcription factors.^{7,8} However, 48 genes that encode proteins of *E. invadens* that contain the Myb domain were found and categorized by bioinformatic analysis. These genes had distinct patterns of expression under basal, encystment, and excystment conditions. Interestingly, the *eimyb23* gene, an ortholog of EhMYB10, was observed to have an increase in expression during trophozoite to cyst conversion.⁹ This study aims to evaluate the effect of overexpression and silencing of EiMyb23 on the encystment of *E. invadens*. We obtain stable transfectants of *E. invadens* trophozoites that overexpress or inhibit the expression of the *eimyb23* gene. Modification of gene expression in the *E. invadens* transfectants was monitored by semi-quantitative RT-PCR and immunofluorescence. Our analysis showed an increase or decrease in the expression of the mRNA of the *eimyb23* gene in the overexpression or silencing conditions. In the immunofluorescence assays, we observed similar results, where the perinuclear localization of the overexpressed EiMyb protein can be seen, compared to the empty control. Interestingly, during encystment, the *eimyb23* gene was expressed more frequently in *E. invadens* (8, 24, 48, and 72 hours). The development of trophozoites into cysts was confirmed using Calcofluor White staining.

Referencias

1. Ralston, K.S *et al.*, 2011. Trends Parasitol.27, 254
2. Azuara-Liceaga, E. *et al.*, 2005. Exp. Parasitol. 110, 280–285
3. Rawat, A. *et al.*, 2020. Biol. Drug Des. 96, 731–744.
4. Romero-Díaz *et al.*, 2007. BMC Mol. Biol.
5. Meneses, E. *et al.*, 2010. Gene 455, 32–42.
6. Ehrenkaufer, G.M., Singh, U., 2012. Mol. Biochem. Parasitol. 184, 59–62.
7. De Cádiz, A.E. *et al.*, 2013. PloS One 8, e74840.
8. Ehrenkaufer, G.M. *et al.*, 2013. Biol. 14, R7.
9. Cuellar, P. *et al.*, 2024. Genes 15, 201.

HPGROEL PROTEIN IS SECRETED BY *HELICOBACTER PYLORI* AND BINDS OTHER SECRETED PROTEINS, ITS CHAPERONIN ACTIVITY IS MAINTAINED AT EXTREME PH

*José de Jesús Olivares-Trejo¹, Máximo B. Martínez Benítez¹, Yaneli Martínez Sotelo¹, Héctor Quezada Pablo², Ana Laura Guzmán Ortiz², Laura Isabel Vázquez-Carrillo¹, Erica Jasmín Sánchez Hernández¹

¹Posgrado en Ciencias Genómicas, Campus del Valle, Colegio de Ciencia y Tecnología, Universidad Autónoma de la Ciudad de México. ²Laboratorio de Investigación en Inmunología y Proteómica, Hospital Infantil de México Federico Gómez. Universidad Autónoma de la Ciudad de México, San Lorenzo 290, Col del Valle Delegación Benito Juárez CP 03100.

Proyecto: UACM CCYT-CON-08

*Corresponding author: jose.olivares@uacm.edu.mx, olivarestrejo@yahoo.com

Helicobacter pylori is a Gram-negative bacterium that is considered a causal agent of peptic ulcer and gastritis and its chronic infections increase the risk of stomach cancer. This bacterium has the capacity of invading the gastric epithelium because it can resist the acidic environment of the stomach, in part because it secretes the enzyme urease. This enzyme produces ammonium, which creates an alkaline micro-environment to neutralize the acidic pH. Other proteins such as VacA, CagA and adhesins are also secreted and their functions in the extracellular environment are known. However, the role of the secreted protein HpGroEL (*H. pylori* Chaperonin) is not yet clear. To investigate the function of the HpGroEL protein, it was purified by affinity chromatography on a hemin-agarose resin and its activity was tested at different pH values, using the same chaperonin of *E. coli* (EcGroEL) as a control. It was found that HpGroEL was active at pH values between 4 and 6. In another experiment, the proteins secreted by *H. pylori* were incubated with recombinant HpGroEL (rHpGroEL) and the interacting proteins were identified by gel electrophoresis and mass spectrometry. Based on the functions of these proteins, we propose that HpGroEL is secreted by *H. pylori* to maintain the proper folding of its extracellular interactors, and in this way, this chaperonin contributes to conquer the hostile environment of the stomach.

Keywords. *Helicobacter pylori*, GroEL protein, Chaperonin activity, Secreted proteins, pH

EXPOSURE TO SUPERPARAMAGNETIC IRON OXIDE NANOPARTICLES INDUCED HEPATORENAL TOXICITY IN BALB/C MICE

Babatunde O. Adetuyi¹, Elizabet Estrada-Muñiz¹, Daniela Pérez-López¹,
Margarita L. Alvarado-Noguez², Jaime Santoyo-Salazar², Libia Vega^{1*}

¹Department of Toxicology and ²Department of Physics, Centre for Research and Advanced Studies of the National Polytechnic Institute, Ave. IPN 2508, Zacatenco, 07360, Mexico City, Mexico.

Email: lvega@cinvestav.mx

Cancer is a major public health challenge, with chemotherapy as its primary treatment. The widespread delivery of these drugs can harm healthy tissues, and thus, exploring alternative treatments that minimize adverse effects is crucial. Many chemotherapeutic agents are difficult to administer because of their low solubility in water. Hence, the search of drug carriers of these compounds is necessary to minimize damage to healthy tissues. Superparamagnetic iron oxide nanoparticles (NPs) are being investigated as potential drug carriers, and their toxicity assessment, are essential to ensure safety administration of NPs and further development as clinically used treatment/delivery system.

NPs were synthesized via the coprecipitation method and characterized for their physiochemical properties. Toxicity assessments of iv administration of NPs were performed at doses of 250, 500, and 700 mg/kg body weight (bw) following a modified version of the OECD 407 guideline, in BALB/c female mice aged 6-8 weeks. Mortality rate, food and water consumption, bw change and systemic toxicity signs were recorded for all the evaluation period. Before sacrificing the animals, an overnight urine sample and blood samples were obtained. Gross necropsy was performed. Biochemical analysis was assessed in urine and blood samples, the white blood cell counts and histological evaluations of liver and kidney were conducted. NPs administration did not result in mortality but caused systemic toxicity signs over the 28-day period and reduced bw gain at 700 mg/kg. NPs at 250 mg/kg decreased total protein (TP) and glucose levels and lymphocyte proportion, and increased bilirubin, alanine aminotransferase (ALT) and lactate dehydrogenase (LDH) levels, and increased eosinophils. At 500 mg/kg, NPs decreased blood and urinary glucose, TP levels and lymphocyte count, and elevated ALT, LDH levels and neutrophil in the blood. At 700 mg/kg, NPs elevated blood TP, creatinine, ALT, LDH levels, with decreased blood albumin, creatinine, monocytes, and urine TP levels. Histological examination revealed degeneration of liver hepatocytes and central veins and kidney glomeruli with NPs accumulation in the organs.

The administration of NPs, at the doses evaluated, produce systemic toxicity and blood cells, liver and kidney affections.

Funding by CONAHCyT, grant 21067.

FUNCTIONAL CHARACTERIZATION OF A NON-SPECIFIC PHOSPHOLIPASE C (PVNPC4) IN BEAN-RHIZOBIA SYMBIOSIS AND ROOT DEVELOPMENT

Ronal Pacheco¹, M.A. Juárez-Verdayes², Marlen Delgado¹, Alfonso Leija³, Javier Montalvo-Arredondo², Noreide Nava¹ and Carmen Quinto^{1*}

¹Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Universidad 2001, Colonia Chamilpa, Cuernavaca 62210, Morelos, México. ²Departamento de Ciencias Básicas, Universidad Autónoma Agraria Antonio Narro, Calzada Antonio Narro 1923, Buenavista, 25315, Saltillo, Coahuila, México.

³Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México, Av. Universidad 2001,

*carmen.quinto@ibt.unam.mx Tel: 7773291666

Plant phospholipases C (PLC) are phospholipids-degrading enzymes that are classified into two subfamilies: phosphoinositide-specific PLC (PI-PLC) and 2) non-specific PLC (NPC). PI-PLCs have been widely studied in various biological processes, such as responses to stress and pathogen attack as well as in plant development; however, NPCs have received less attention. Neither PI-PLCs nor NPCs have been analyzed in the symbiotic interaction between Fabaceae species (commonly legumes) and soil nitrogen-fixing bacteria called rhizobia. The establishment of the legume-rhizobia symbiosis leads to the development of a new root organ called nodule, where nitrogen fixation takes place, which is, along with photosynthesis one of the most important biological processes on earth. In addition to PLC degrading phospholipids, they play a key role in lipid-mediated signaling and metabolism. Recently, the participation of lipid metabolism in the development of nodules in soybean has been reported, this being a legume model phylogenetically close to the common bean (*Phaseolus vulgaris* L.). In this work, we functionally characterized bean *PvNPC4* during legume-rhizobia symbiosis. Surprisingly, our findings provide evidence that *PvNPC4* plays an important role in root development. The number of primary and lateral roots and the length of primary roots were reduced due to RNAi silencing of *PvNPC4*. In the legume-rhizobia symbiosis, *PvNPC4* transcript levels increased three days post-inoculation (dpi) with *Rhizobium tropici* and decreased in inoculated roots and nodules at 14 dpi. Furthermore, rhizobial infection and nodule number decreased in *PvNPC4*:RNAi transgenic roots. On the other hand, the expression of *PvEnod40*, a regulatory gene of the early stages of symbiosis, was drastically decreased due to the silencing of *PvNPC4*. These results indicate that *PvNPC4* is a key regulator of root and nodule development and-constitute the first evidence for the involvement of PLC in the symbiosis between legume and rhizobia.

Acknowledgements. This work was supported by grant IN204024 from PAPIIT to CQ and CF-I-297 from CONAHCyT. RP received the grant 749422 from CONAHCyT

BENZOPYRENE DEGRADATION INDUCES CHANGES IN ANTIOXIDANT AND DETOXIFYING METABOLISM IN *DEBARYOMYCES HANSENI*

Francisco Padilla-Garfias, Minerva Araiza-Villanueva, Norma Silvia Sánchez, Martha Calahorra and Antonio Peña

Departamento de Genética Molecular, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Circuito Exterior s/n, Ciudad Universitaria, México City 04510, México.
E-mail: fpadilla@ifc.unam.mx, apd@ifc.unam.mx

Polycyclic aromatic hydrocarbons (PAHs), such as benzopyrene (BaP), are regarded as contaminants of soils and aquatic niches. These contaminants are ubiquitous and are considered extremely toxic. The necessity for novel remediation strategies employing microorganisms has prompted our search for a yeast species capable of growth in the presence of BaP. The marine and extremophilic yeast *Debaryomyces hansenii* has been demonstrated to be capable of grow in the presence of BaP. Initial findings indicate that the degradation of BaP is contingent upon the concentration and quantity of yeast, which employs a cytochrome P450 (CYP)-mediated oxidation process to metabolise BaP. Consequently, the understanding of the *D. hansenii* xenome (the biosystem responsible for the detection, transport, and metabolism of xenobiotics) or the genes and pathways involved in BaP oxidation could facilitate the utilisation of this yeast in mycoremediation processes. An RNA-Seq analysis revealed that 1179 genes were overexpressed in *D. hansenii* when grown in the presence of BaP. Among the genes, some were found to be related to xenobiotic degradation, glutathione homeostasis, and oxidative stress. These results were corroborated by qPCR and by monitoring the activities of the principal detoxifying enzymes, showing a correlation with the expression levels of their respective genes.

References

1. Padilla-Garfias, F., Sánchez, N. S., Calahorra, M., & Peña, A. (2022). DhDIT2 encodes a *Debaryomyces hansenii* cytochrome P450 involved in benzo(a)pyrene degradation-A proposal for mycoremediation. *Journal of fungi*, 8(11), 1150. <https://doi.org/10.3390/jof8111150>
2. Aranda E. (2016). Promising approaches towards biotransformation of polycyclic aromatic hydrocarbons with Ascomycota fungi. *Current opinion in biotechnology*, 38, 1-8. <https://doi.org/10.1016/j.copbio.2015.12.002>

RESTORATIVE EFFECTS OF (+)-EPICATECHIN IN A RODENT MODEL OF AGING INDUCED MUSCLE ATROPHY: UNDERLYING MECHANISMS

Carlos Palma Flores^{1,2}, Israel Ramirez-Sanchez^{1,2}, Viridiana Navarrete-Yañez², Lucia Ramirez¹, Leonor Galera², Enrique Mendez-Bolaina³, Veronica Najera¹, Guillermo Ceballos², Francisco Villarreal^{1,4}

¹School of Medicine, UCSD, La Jolla, California, USA. ²Seccion de Estudios de Posgrado e Investigacion, Escuela Superior de Medicina, IPN, Plan de San Luis y Diaz Miron s/n, Col. Casco de Santo Tomas, Del. Miguel Hidalgo C.P. 11340, Mexico City, Mexico.

³Facultad de Ciencias Quimicas, Universidad Veracruzana, Orizaba, Mexico. ⁴VA San Diego Health Care, San Diego, CA, USA. israel.ramirez14@hotmail.com.

Sarcopenia is a progressive skeletal muscle (SkM) disorder characterized by the accelerated loss of muscle mass (atrophy) and function. SkM atrophy is associated with increased incidence of falls, functional decline, frailty and mortality. In its early stage, SkM atrophy is associated with increased pro-inflammatory cytokine levels and proteasome-mediated protein degradation. These processes also link to the activation of atrophy associated factors and signaling pathways for which, there is a lack of approved pharmacotherapies. The objective of this study, was to characterize the capacity of the flavanol (+)-epicatechin (+Epi) to favorably modulate SkM mass and function in a rat model of aging induced sarcopenia and profile candidate mechanisms. Using old male Sprague-Dawley rats, an 8 weeks oral administration of the +Epi was implemented while control rats only received water. SkM strength, treadmill endurance, muscle mass, myofiber area, creatine kinase, lactate dehydrogenase, troponin, α -actin, tumor necrosis factor (TNF)- α and atrophy related endpoints were quantified in plasma and/or gastrocnemius. We also evaluated effects on insulin growth factor (IGF)-1 levels and downstream signaling (AKT/mTORC1). Treatment of aged rats with +Epi, led to significant increases in front paw grip strength, treadmill time and SkM mass vs. controls as well as beneficial changes in makers of myofiber integrity. Treatment significantly reversed adverse changes in plasma and/or SkM TNF- α , IGF-1, atrophy and protein synthesis related endpoints vs. controls. In conclusion, +Epi has the capacity to reverse sarcopenia associated detrimental changes in regulatory pathways leading to improved SkM mass and function. Given these results and its recognized safety and tolerance profile, +Epi warrants consideration for clinical trials.

AIM11: A NOVEL YEAST ASSEMBLY FACTOR FOR COMPLEX IV AND SUPERCOMPLEXES

Ulrik Pedroza-Dávila¹, Yolanda Camacho-Villasana¹, Alfredo Cabrera-Orefice^{2,3}, Madhurya Lutikurti³, Ulrich Brandt³ and Xochitl Pérez-Martínez^{1*}

¹Departamento de Genética Molecular, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, México City, México. ²Functional Proteomics, Institute of Cardiovascular Physiology, Goethe University, Frankfurt am Main, Germany.

³Medical BioSciences Department, Radboud University Medical Center, Nijmegen, the Netherlands

*E-mail: xperez@ifc.unam.mx

In *Saccharomyces cerevisiae*, the formation of the electrochemical gradient used for ATP synthesis is due to the proton-pumping activity of the respiratory complexes III₂ and IV. In addition, these complexes associate forming supercomplexes (SC). This last conformation is made of the complex III dimer flanked by one or two complex IV monomers¹. Some of the proposed functions of supercomplexes are substrate channeling, respiratory complex stability, respiratory activity enhancing and adaptation to the energetic requirements of the cell². However, the mechanism of SC assembly and respiratory enzymes is still undercharacterized. We recently identified Aim11 as a novel supercomplexes and complex IV assembly factor³. The $\Delta aim11$ mutant showed a deficient growth in respiratory media. Complex III subunits' levels were not affected but complex IV subunits' levels were diminished. Moreover, supercomplexes amount were decreased. Further characterization showed that Aim11 not only affected SC assembly, it has a role in complex IV assembly, specifically affecting Cox3 and proteins related to its biogenesis.

References

- Berndtsson, J., Kohler, A., Rathore, S., Marin-Buera, L., Dawitz, H., Diessl, J., ... & Ott, M. (2020). Respiratory supercomplexes enhance electron transport by decreasing cytochrome c diffusion distance. *EMBO reports*, 21(12), e51015.
- Eldeeb, M. H., Camacho Lopez, L. J., & Fontanesi, F. (2024). Mitochondrial respiratory supercomplexes of the yeast *Saccharomyces cerevisiae*. *IUBMB life*.
- Flores-Mireles, D., Camacho-Villasana, Y., Lutikurti, ... & Pérez-Martínez, X. (2023). *Life Science Alliance*, 6(7).

CYTOTOXIC PROPERTIES AND GENE EXPRESSION MODULATING EFFECTS OF CINNAMON ESSENTIAL OIL (*CINNAMOMUM ZEYLANICUM*) IN CERVICAL CANCER CELLS

Ruben Piña-Cruz^{1,2}, Andrea Molina-Pineda³, Aleyda G. Zamudio-Martínez², Andrea Navarro-Zazueta²,
Luis Felipe Jave-Suárez² and Adriana del Carmen Aguilar Lemarroy^{2*}

¹Programa de Doctorado en Ciencias en Biología Molecular en Medicina, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara, Guadalajara, Jalisco. ²División de Inmunología, Centro de Investigación Biomédica de Occidente-IMSS, Guadalajara, Jalisco. ³Consejo Nacional de Humanidades, Ciencias y Tecnologías, CONAHACYT, CDMX.

*E-mail: ruben.pinacruz@gmail.com

Cervical cancer is a significant global health problem affecting millions of women worldwide including Mexico. Current treatments such as surgery, radiation, and chemotherapy have notable limitations and adverse effects, requiring the search for new therapeutic compounds. Cinnamon essential oil (CEO), derived from *Cinnamomum zeylanicum* bark, is recognized for its beneficial properties, including anticancer effects. However, its potential against cervical cancer remains unexplored. Thus, this study aimed to evaluate the effects of CEO on cervical cancer cell lines HeLa and SiHa, and the non-tumorigenic HaCaT cell line as a non-tumorigenic control. The assessment involved screening for cytotoxic properties using WST-1 and Annexin-V/propidium iodide assays, as well as examining gene expression modulation through RNA sequencing and bioinformatic analysis. The results showed that CEO treatment decreased the viability of HeLa and SiHa cells in a dose-dependent manner, while exerting a minor effect on HaCaT cells, suggesting a selective targeting of cancer cells. CEO also induced cell death in the cancer cell lines. RNA sequencing and bioinformatic analysis revealed that CEO modulates the expression of more than 1500 genes. Through enrichment analysis, more than 50 pathways modulated by CEO were identified, including the Spliceosome, Ribosome, Cell Cycle, Proteosome, and Apoptosis pathways. In conclusion, CEO could have anti-cancer properties against human cervical cancer cells by decreasing cell viability and inducing cell death, potentially through the modulation of multiple genes and pathways. These findings suggest that CEO could be a promising therapeutic agent for cervical cancer, although further studies are necessary to confirm these results and establish the safety and efficacy of CEO in humans.

TESTOSTERONE AND ESTRADIOL INDUCE THE EXPRESSION OF *ACTINOBACILLUS SEMINIS* SECRETED PROTEASES

Gerardo Antonio Ramírez Paz y Puente¹, Candelario Vázquez Cruz²,
Tomas Villamar Duque¹ y Erasmo Negrete Abascal¹

¹Facultad de Estudios Superiores Iztacala, UNAM, Av. De Los Barrios 1, Los Reyes Iztacala, 54090 Tlalnepantla, Edo de México;

²Centro de Investigaciones en Ciencias Microbiológicas, ICUAP, BUAP, Puebla, México.

E-mail: tonypazypuente@gmail.com

Actinobacillus seminis is a Gram-negative bacterium member of the Pasteurellaceae family, part of the ruminant's prepuce microbiota. Still, also the causal agent of epididymitis, infertility, and sterility, pathologies present mainly when ruminants get sexual maturity and there are increase in hormones¹. Sexual hormones can participate in host-microorganisms communication and regulate the virulence factor expression². Besides improving its growth, testosterone and estradiol favor the *A. seminis* adhesins expression and biofilm formation. This work aimed to determine the effect of testosterone (1–5 ng/ml) and estradiol (5–25 pg/ml) in *A. seminis* proteases expression. As was observed using zymograms, the presence of steroid hormones increases the expression of 50 kDa metalloprotease and 65 kDa serine proteolytic activities. The metalloprotease activity is optimal at a pH of 6-7 and stable until 80°C. The serin protease activity is optimal at a pH of 6-8 and stable until 70°C. This last protease can also degrade bovine and horse hemoglobin and casein. Both proteolytic activity bands presented immune cross-reactivity with sera against metalloproteases from *A. seminis*⁴ and *A. pleuropneumoniae*⁵ and with a serum against a *Mannhemia hemolytica* serine protease⁶. Changes in host hormone concentrations control the expression of different *A. seminis* virulence factors, including proteases.

Project supported by DGAPA-UNAM PAPIIT IN204122. GARPP is a CONAHCYT (1321742) scholarship and a Programa de Maestría y Doctorado en Ciencias de la Producción y Salud Animal, UNAM student.

References

1. Al-Katib WA y Dennis SM (2009). New Zealand Veterinary Journal, 57:6, 352-358.
2. Gümüş D, Kalaycı Yüksek F, Sefer Ö, Yörük E, Uz G, Anđ Küçük M. (2019). Microbial Pathogenesis, 132:319-324.
3. Ramírez-Paz y Puente GA, Chávez-Florez CI, Montes-García JF, Sánchez-Alonso P, Cobos-Justo ME, Vázquez-Cruz C, Zenteno, E y Negrete-Abascal E. (2023). FEMS Microb. Letters, 370:1-8.
4. De la Cruz Montoya AH, Ramírez-Paz y Puente GA, Vazquez-Cruz C, Montes-García JF, Negrete-Abascal E. (2023). Pak Vet J, 43(2): 276-282.
5. Negrete-Abascal E, Tenorio RV, Guerrero AL, et al. (1998). Ca J Vet Res 62:183-90.
6. Rosales-Islas V, Ramírez-Paz-y-Puente GA, Montes-García F, Vázquez-Cruz C, Sánchez-Alonso P, Zenteno E y Negrete-Abascal E.(2024). Microbial Pathogenesis, 106706.

DESIGN AND IMPLEMENTATION OF A CELL SURFACE DISPLAY SYSTEM ON *NEUROSPORA CRASSA* USING NATIVE CELL WALL PROTEIN, ACW-1, AS ANCHOR

Ana Sofía Ramírez-Pelayo, Claudia Cecilia Vega-García, Lorena Amaya-Delgado,
Jorge Rodríguez, Jorge Verdín*

Unidad de Biotecnología Industrial, Centro de Investigación y Asistencia en Tecnología
y Diseño del Estado de Jalisco, A. C. (CIATEJ). Camino Arenero 1227, El Bajío, 45019, Zapopan, Jalisco.

*jverdin@ciatej.mx

Cell surface display allows the expression of proteins on the cell surface of a host by molecularly fusing a gene that encodes for a protein of interest (passenger) to the gene of a protein that naturally resides on its plasma membrane or the cell wall (anchor, CWP). When the CWP is expressed, it anchors the passenger protein to the surface, where both proteins may perform their functions. Therefore, the selection of an adequate anchor is instrumental to the display's success. Surface display systems have been implemented in bacteria and yeast for library screening, biosensor development, and consolidated bioprocessing, among others; however, it has been poorly exploited in biotechnological workhorses such as filamentous fungi. This work aimed to design and implement a cell surface display system on the filamentous fungus *Neurospora crassa*. ACW-1 (NCU08936), one of its cell wall proteins, was functionally characterized to assess its suitability as the anchor. Its structure was predicted by AlphaFold as a highly ordered protein composed of β -sheets. More characterized homologous proteins in other fungi perform functions related to the maintenance of the cell wall, so ACW-1 might have a similar role, considering its highly structured configuration. Growth experiments in normal and cell wall inhibitory conditions (Congo Red, Calcofluor White, osmotic stress, temperature stress) using an *acw-1* knock-out (KO) mutant indicated this putative function was not vital for the survival of the fungus, since the KO strain behaved mostly like the wild-type (WT) strain. The KO strain was more sensitive to glucan-disrupting Congo Red, so this suggested that ACW-1 might interact with β -1,3 glucans. Since the absence of ACW-1 does not compromise cell viability, it was further evaluated as an anchor to display GFP. Fluorescent signals were observed by laser-scanning confocal microscopy far from the hyphal tip, localized on the outermost region and septae of hyphae. The expression of this fusion was confirmed by Western Blot, mostly on the plasma membrane. Together these results suggested that ACW-1 is a suitable anchor for a display system. A whole-cell biocatalyst displaying a heterologous peroxidase was further developed on the surface of *N. crassa* using ACW-1 as an anchor. This work was supported by SENER-CONACYT Sustentabilidad Energética, grant 245750; and CONAHCYT-Ciencia de Frontera 2019, grant 552259.

EFFECT OF PH ON THE GSSG-DEPENDENT SUBSTRATE INHIBITION OF THIOREDOXIN GLUTATHIONE REDUCTASE. AN INITIAL VELOCITY AND PROGRESS CURVE-BASED STUDY

Juan Luis Rendón*, José de Jesús Martínez González, Alberto Guevara Flores and Patricia del Arenal Mena

Departamento de Bioquímica, Facultad de Medicina, UNAM. Ciudad Universitaria; Ciudad de México, CP 04510

Tel. +52(55) 5623-2169

*Correo electrónico: jrendon@bq.unam.mx

Thioredoxin-glutathione reductase (TGR) represents an interesting isoform of thioredoxin glutathione reductase. The enzyme is an atypical NADPH-dependent disulfide reductase with the ability to reduce either thioredoxin or oxidized glutathione (GSSG). This last activity is due to the presence of a glutaredoxin-like domain appended to the N-terminal end of the thioredoxin reductase module. A kinetic study of TGR based in initial velocity data, as well as product inhibition studies, revealed the enzyme follows a two-sites ping pong bi bi kinetic mechanism, in which substrates are bound under rapid equilibrium conditions. Interestingly, TGR shows substrate inhibition as well as complex profiles of the full progress curves with GSSG as substrate, which are dependent on the concentration of NADPH, enzyme and the product GSH. A mechanism-based model which predicts all the kinetic features of the GSSG-dependent substrate inhibition was previously developed.

In the present work a detailed kinetic study based on both initial velocity and full progress curves on the effect of pH on the substrate inhibition at 25 °C is presented. Results revealed the GSSG-dependent substrate inhibition is present in the pH range 5.8 to 9.5. The value of the apparent inhibition constant (K_i) was dependent on pH. Interestingly, the profile of the full progress curves was strongly dependent on pH. In the pH range 7 to 10.2 the expected complex profile described at pH 7.8 was present, albeit the magnitude of the apparent lag segment was variable. By contrast, experiments carried at acid pH values (5.8-6.5) in the presence of high GSSG concentrations revealed an initial burst stage followed by an apparent steady-state segment, without trace of the lag segment of the progress curves observed at higher pH values. These data suggest under these conditions GSSG behaves as a slow-binding inhibitor. The results described in the present work are discussed under the general model describing the kinetic behavior of the enzyme.

EXPLORING CHROMATIN DYNAMICS DURING IN VITRO DECIDUALIZATION OF HUMAN ENDOMETRIAL STROMAL CELLS

Alejandra Monserrat Retis-Resendiz¹, Martha Paloma Domínguez Mora¹, Yesenia Cid Cruz¹,
Jessica Romero Reyes¹, Dora María Velázquez Hernández, Elizabeth García Gómez¹, Moisés Leon-Juárez²,
Ignacio Camacho-Arroyo¹, Edgar Ricardo Vázquez-Martínez^{1*}

¹Unidad de Investigación en Reproducción Humana, Instituto Nacional de Perinatología, Facultad de Química, UNAM, Mexico City. ²Departamento de Inmunobioquímica, Instituto Nacional de Perinatología, Mexico City. *Corresponding author. Unidad de Investigación en Reproducción Humana, Instituto Nacional de Perinatología, Facultad de Química, UNAM. Montes Urales 800, Lomas de Virreyes 11000, Mexico City, Mexico. Tel. 55-55209900 ext. 346, email: vamer@comunidad.unam.mx

Introduction. Decidualization, crucial for establishing and supporting pregnancy, involves the differentiation of endometrial stromal cells (ESCs) orchestrated by estradiol (E2), progesterone, and the intracellular increase of cAMP. The progesterone receptor (PGR) plays a pivotal role in decidualization. Previous studies have linked decidualization to changes in gene expression and chromatin state. However, the first steps in chromatin regulation and 3D genome structure during decidualization have not been explored. This project aimed to investigate the effects of early decidualization on chromatin state and the regulation of PGR expression in immortalized human ESCs.

Methods. Immortalized human ESCs (T-hESC, ATCC CRL-4003) were treated with E2, medroxyprogesterone (MPA), and cAMP (EMC) or vehicle at different time points. Gene expression of decidualization biomarkers PRL and IGFBP1 was monitored over time by RT-qPCR. RNA-seq and ChIP-seq experiments were performed at 24 h post-treatment to analyze transcriptome changes and the genomic enrichment of histone modifications (H3K4me3, H3K27ac, and H3K4me1), respectively. Then, we chose the PGR gene to evaluate the local chromatin dynamics and to detect potential cis-regulatory elements by 4C-seq. Finally, T-hESC underwent different treatments with individual/combined EMC components, hormone receptor antagonists, and inhibitors of cAMP downstream effectors to dissect mechanisms governing PGR expression.

Results. Decidualization biomarkers, IGFBP1 and PRL, were significantly induced at 24 h of EMC treatment compared to the vehicle. Transcriptome analysis identified 7,588 differentially expressed transcripts. Gene ontology analyses identified the up-regulation of processes related to angiogenesis, insulin response, and extracellular matrix (ECM) up-regulated, while DNA metabolism and cell cycle was down-regulated. Chromatin changes involve gaining or losing histone modification enrichment. Many genes retained this enrichment: 24,260 with H3K4me3, 3,322 with H3K4me1, and 42,793 with H3K27ac. Some lost (6,559 H3K4me3, 14,769 H3K4me1, 35,763 H3K27ac), while others gained (2,445 H3K4me3, 6,371 H3K4me1, 7,506 H3K27ac) enrichment post-EMC treatment. H3K4me3 regions were primarily enriched in promoters and introns; H3K27ac and H3K4me1 were mainly found in intronic and distal intergenic regions. Genes up-regulated in association with H3K4me3/H3K27ac were related to hormone responses, while down-regulated genes with retained enrichment were

associated with DNA metabolism. Lost enrichment was associated with genes related to cell migration, while genes associated with regions that gained H3K4me3/H3K27ac were related to cell adhesion and ECM. H3K4me1-conserved regions in up-regulated genes during EMC-decidualization were related to glucose transport, while those down-regulated were associated with DNA metabolism. Regions that lost H3K4me1 were associated with DNA metabolism, while those that gained it were associated with angiogenesis. Interestingly, four distal regions enriched with H3K4me1/H3K27ac interacted with the PGR promoter, exhibiting increased H3K4me3/H3K27ac enrichment upon EMC treatment. Moreover, cAMP induced PGR expression via PKA pathway, while MPA down-regulated it through the PGR.

Conclusion. At 24 h of EMC treatment, T-hESC underwent extensive shifts in transcriptome and histone modifications enrichment, affecting both promoter and distal regulatory regions. EMC exposure modulated histone modifications enrichment associating with gene expression changes, particularly impacting angiogenesis, insulin regulation, and DNA metabolism. PGR gene expression regulation is associated with local chromatin dynamics and PKA signaling.

Acknowledgments. We acknowledge CONACYT (Grant Number A1-S-26749), INPer (Grant Number 571,3000-20109-01-571-17), and UNAM-PAPIIT (Grant Number IA209520)

SPONTANEOUS CALCIUM TRANSIENTS IN STRIATAL ASTROCYTES: EVIDENCE FROM A PRECLINICAL MODEL OF AUTISM

Daniel Reyes-Haro, Hugo Saavedra-Bonilla and Durairaj Ragu Varman

Departamento de Neurobiología Celular y Molecular, Instituto de Neurobiología, Universidad Nacional Autónoma de México, Campus Juriquilla. Blvd. Juriquilla #3001, Juriquilla, Querétaro, Qro. CP 76230, México.
Tel. 55 5623 4062; email: dharo@unam.mx

Autism spectrum disorder (ASD) is known as a group of neurodevelopmental conditions including stereotyped and repetitive behaviors, besides social and sensorimotor deficits. Anatomical and functional evidence indicates atypical maturation of the striatum. Astrocytes regulate the maturation and plasticity of synaptic circuits, and impaired calcium signaling is associated with repetitive behaviors and atypical social interaction. Spontaneous calcium transients (SCT) recorded in the striatal astrocytes of the rat were investigated in the preclinical model of ASD by prenatal exposure to valproic acid (VPA). Our results showed sensorimotor delay, augmented glial fibrillary acidic protein -a typical intermediate filament protein expressed by astrocytes- through development, and increased frequency of SCT, with a reduced latency that resulted in a diminished amplitude in the VPA model. The convulsant picrotoxin, a GABA_A (γ-aminobutyric acid type A) receptor antagonist, reduced the frequency of SCT in both experimental groups but rescued this parameter to control levels in the preclinical model of ASD. The amplitude and latency of SCT were decreased by picrotoxin in both experimental groups. Nipecotic acid, a GABA uptake inhibitor, reduced the mean amplitude only for the control group. Nevertheless, nipecotic acid increased the frequency but diminished the latency in both experimental groups. Thus, we conclude that striatal astrocytes exhibit SCT modulated by GABA_A-mediated signaling, and prenatal exposure to VPA disturbs this tuning.

Acknowledgements. This work was supported by Consejo Nacional de Humanidades, Ciencias y Tecnologías (CONAHCYT-319209) and Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica -Dirección General de Asuntos de Personal Académico -Universidad Nacional Autónoma de México (PAPIIT-DGAPA-UNAM IN209121 and IN214324) to DR-H. The authors are grateful to R. Arellano, A. Martínez-Torres and E. Garay for academic support. Special thanks to E. N. Hernández-Ríos, M. Mendoza-Baltazar, A. Castilla, M.C. Mendoza-López, N. Aranda and M. García-Servín for technical support.

CONSORTIUM OF MICROORGANISMS FOR POLYSTYRENE BIODEGRADATION

Marcela Robles-Machuca^{1*}, Iris-Rocío Tirado-Lucas¹, Kassandra-Selene Reyna-Gaeta¹,
Yoselin-Karely Rodríguez-Reyes¹

¹Universidad Autónoma de Nayarit, Secretaría de Investigación y Posgrado. Ciudad de la cultura s/n centro, 63000,
Tepic, Nayarit, Mexico.

*marcela.robles@uan.edu.mx

Introduction. The biodegradation of plastics by microorganisms has emerged as an alternative ecological solution to their accumulation problem. Some microorganisms have developed the capacity to express and secrete enzymes that depolymerize different types of plastics to be used as carbons and energy sources. In nature, bacteria, fungi, and yeast interact within the same niche, where their association can form a consortium that synergistically enhances plastic biodegradation. Polystyrene (PS) has a significant environmental impact due to its high resistance to natural degradation.

Aims. To analyze the biodegradation of polystyrene by a consortium of microorganisms.

Methodology. 2x2cm of PS was exposed to environmental conditions for 21 days. *Bacillus cereus*, *Acidovorax delafieldii*, *Saccharomyces cerevisiae*, and no-identified (NI) fungus were used to form consortia of bacteria-bacteria (BB), bacteria-yeast (BY), and yeast-fungus (YF). Consortia and individual microorganisms were grown in a liquid minimal media (MM) with PS for five weeks at room temperature. Every week, the plastic weight loss was analyzed by gravimetry; the growth was monitored at 600nm; and, the esterase/lipase (E/L) activity was quantified using p-NPB 40mM at 405nm for 20 minutes.

Results. The best PS weight loss was found with *A. delafieldii* alone and NI-fungus consortium with 0.6mg. However, the PS weight loss was not shown with the other consortia of *A. delafieldii*. The growth within the consortia was enhanced by combining *A. delafieldii* with NI-fungus achieving 0.9 OD₆₀₀ in the first two weeks. The consortium of *A. delafieldii* with NI-fungus demonstrated the best E/L activity with 22 U/L constantly by four weeks. The consortium of *B.cereus* with NI-fungus showed the highest E/L activity with 31 U/L at two weeks but decreased significantly in the next weeks.

Conclusion. The consortia bacteria-fungus enhanced the growth and the expressed extracellularly E/L activity, showing a capacity for PS biodegradation. *A. delafieldii* and NI-fungus were the best consortium by the growth in an MM with PS, E/L activity, and PS weight loss. Further analysis of biofilm formation, hydrophobicity, and microplastic release will be carried out.

BIOCHEMICAL STUDY OF MDM2 AND RB MRNA INTERACTION

Andrés Usiel Rodríguez Rodríguez and Vanesa Olivares Illana

Laboratorio de Interacciones Biomoleculares y Cancer. Instituto de Física, Universidad Autónoma de San Luis Potosí, Avenida Parque chapultepec1570, Privadas del Pedregal, 78295, San Luis Potosí, SLP. México.
Tel. 4448262300. Email: vanesa@ifisica.uaslp.mx.

The process by which cancer develops continues to be a topic of great interest. It is well known that there are different signaling pathways that allow deregulated proliferation by silencing or amplifying genes, which generate a change in the expression of their proteins (1, 2). The most studied are the pathways involving p53, Rb, MDMX, and MDM2. Recently, our working group reported that under conditions of DNA damage, MDM2 increases the expression levels of the tumor suppressor Rb. This effect is due to an interaction between the MDM2 protein and the *Rb* mRNA. Rb has been seen to increase its expression under the condition of DNA damage and the presence of MDM2 (3). It is of our interest to find the domain or region of MDM2 responsible for the interaction with *Rb* mRNA. Using different techniques such as RNA ELISAs, mRNA co-immunoprecipitation, among others, we are analyzing the different domains of MDM2 in its interaction with *Rb* mRNA. Currently, we have observed that under conditions of DNA damage, the N-terminal domain of MDM2 significantly increased the expression of the Rb protein in H1299 MDM2^{-/-} cells.

Reference

1. Liu, X., Zhang, Y., Wang, Y., Yang, M., Hong, F., & Yang, S. (2021). Protein Phosphorylation in Cancer: Role of Nitric Oxide Signaling Pathway. *Biomolecules*, 11(7), 1009. <https://doi.org/10.3390/biom11071009>
2. Dhillon, A. S., Hagan, S., Rath, O., & Kolch, W. (2007). MAP kinase signalling pathways in cancer. *Oncogene*, 26(22), 3279–3290. <https://doi.org/10.1038/sj.onc.1210421>
3. Hernández-Monge, J., Rousset-Roman, A. B., Medina-Medina, I., & Olivares-Illana, V. (2016). Dual function of MDM2 and MDMX toward the tumor suppressors p53 and RB. *Genes & cancer*, 7(9-10), 278–287. <https://doi.org/10.18632/genesandcancer.120>

OVERVIEW OF CIRCULATING MICRORNAS AND GENOMICS VARIANTS IN PREDIABETES PATIENTS PRESENTING DIFFERENT CLINICAL COURSES: SEARCH FOR NOVEL INFORMATIVE MOLECULES

Sandra Romero-Córdoba^{1,2}, Diego Ruiz Ruiz¹, Karina Torres-Ortiz¹, Fabiola del Razo Olvera², Cesar Lam Chung², Carlos Aguilar Salinas², Gabriela Galan-Ramirez², Imperio Sánchez-Pérez², Daniel Cuevas Ramos²

¹Instituto de Investigaciones Biomédicas, UNAM, Circuito, Mario de La Cueva s/n, C.U., Coyoacán, 04510 Mexico City, Mexico,

²Instituto Nacional de Ciencias Médicas y Nutrición "Salvador Zubirán", Vasco de Quiroga 15, Belisario Domínguez Secc 16, Tlalpan, 14080, Mexico City.

sromero@iibiomedicas.unam.mx

Prediabetes (preDM), occurs when blood glucose levels are higher than normal but not high enough to be considered type 2 diabetes (DM), is a risk factor for DM. The prevalence of preDM is high, highlighting the need for early action to prevent disease progression. However, it must be recognized that there is great heterogeneity in individual risk among people with prediabetes for developing DM. Therefore, we must identify new informative molecules that will allow to stratify patients with preDM who require early intervention for a progressive disease state.

Therefore, the aim of this study is to characterize genomic variations, particularly the frequency of the risk SNP rs13342692, as a proxy for DM risk haplotype in the SLC16A11 gene and mitochondrial DNA copy number, both measured by real-time PCR. Together with biochemical parameters (glucose metabolism, lipid profile, renal and hepatic function) and anthropometric characteristics in a cohort of 261 volunteers with different glucose metabolic states. The frequency of the rs13342692 polymorphism was 44.4% (10.7 in homozygous and 33.7% in heterozygosity) in the study cohort. Particularly in individuals with preDM the frequency was 45.7%, similar to our validation cohort (N=144) of 55.23%. Likewise, we observed a higher copy number in the mitochondrial genome in volunteers with preDM and DM levels.

The second objective was to investigate the longitudinal differential expression of circulating microRNAs in patients with preDM who did or did not develop DM during a follow-up of at least one year, allowing statistical identification of the clinical course. To this end, we recruited 66 patients from the initial cohort, diagnosed with preDM, who were followed biochemically and molecularly for at least one year with monthly visits and nutritional intervention. In this work we employed a time series strategy to define the most informative miRNAs that describe early changes that will lead to diabetes progression. To study the circulating miRNAome, we performed massive sequencing of small RNAs with a read depth of 20 million from 158 samples, corresponding to 5 times in 42 patients. We identified a set of 12 miRNAs whose temporal expression over 1 year is associated with DM progression. These results will provide new informative molecules, which could allow a more precise and early identification of those prediabetic patients at high risk of developing diabetes.

DESIGN OF *DE NOVO* PROTEINS AND ENZYMES USING GENERATIVE LANGUAGE MODELS

Sergio Romero-Romero^{1,2}, Alexander E. Braun², Timo Kossendey², Max Paulfranz², Paul Jacobs², Noelia Ferruz³, Sabrina Wischt², Sooruban Shanmugaratnam² and Birte Höcker²

¹Department of Biochemistry and Structural Biology, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, 04510 Mexico City, Mexico, ²Department of Biochemistry, University of Bayreuth, 95447 Bayreuth, Germany, ³Department of Structural and Molecular Biology, Barcelona Institute of Molecular Biology, 08028 Barcelona, Spain.
E-mail: sromero@ifc.unam.mx

Protein design aims to build novel proteins tailored for specific purposes, holding the potential to tackle many environmental and biomedical problems. Recent progress in Transformer-based architectures has enabled the implementation of large language models (LLMs) capable of generating text with human-like capabilities. These LLMs models have shown a great relevance for protein science due to the analogies between human languages and protein sequences, whereby protein sequences have the similar hierarchical order of building blocks as found in human languages. Motivated by this success and as a shift of paradigm in protein design, we assessed the ability of unconditional and conditional language models to navigate new protein sequence space outside the boundaries of natural-occurring ones. Using two generative language models, ProtGPT2 and ZymCTRL, which generate new protein sequences following the principles of natural ones, we experimentally characterized an extensive number of *de novo* proteins and enzymes to investigate their biochemical, biophysical, and functional properties. Experimental validation of a set of novel proteins generated by ProtGPT2 showed that many of these are well expressed in bacterial host cells and contain the ability to fold and function as their natural counterparts. Additionally, some of the enzymes generated by ZymCTRL and fine-tuned ProtGPT2 performed the intended triosephosphate isomerase activity, with catalytic efficiency orders of magnitude lower in comparison to natural enzymes. Finally, proteins generated by ProtGPT2 and ZymCTRL were subjected to a directed evolution approach with different screening and selection methods to evaluate the possibility of improving the solubility and foldability of some of these language-model-generated proteins. Overall, the results support the idea that protein language models are a promising alternative for creating tailor-made proteins and expanding the boundaries of our known protein sequence space, resulting in robust and well-behaved macromolecules, and enhancing the applications of protein design.

ADAPTIVE STRESS RESPONSE TO NITROGEN LIMITATION IN THE ANTARCTIC YEAST *RHODOTORULA MUCILAGINOSA* M94C9

Miguel Rosas-Paz¹, Alberto Zamora-Bello¹, Nayeli Torres-Ramírez¹, Diana Villarreal-Huerta¹,
Ofelia Méndez-Romero², Lucero Romero-Aguilar³, Juan Pablo Pardo-Vázquez³, Viviana Escobar-Sánchez¹,
Claudia Segal-Kischinevsky¹ and James González^{1*}

¹Departamento de Biología Celular, Facultad de Ciencias, ²Departamento de Genética y Biología Molecular, Instituto de Fisiología Celular, ³Departamento de Bioquímica, Facultad de Medicina, UNAM, Ciudad Universitaria, CDMX, México.
E-mail: miguel1295rp@ciencias.unam.mx

The polyextremophile red yeast *Rhodotorula mucilaginosa* displays tolerance to diverse environmental stressors, including cold, osmolarity, salinity, and oligotrophic conditions. Particularly, this yeast exhibits a remarkable ability to accumulate lipids and carotenoids in response to nitrogen starvation stress conditions. However, research into lipid and carotenoid biosynthesis has been hampered by limited genetic tools and a scarcity of studies on adaptive responses to nutrient stressors stimulating lipogenesis and carotenogenesis. This study explores the impact of nitrogen stress on the adaptive response in the Antarctic yeast *R. mucilaginosa* M94C9. Variation of nitrogen availability reveals a nitrogen-dependent modulation of biomass and lipid droplets production, accompanied by significant ultrastructural changes to withstand nitrogen starvation stress. Further investigation into the expression profiles of *RmACC1*, *RmFAS1*, *RmFAS2*, *RmDGA1*, *CAR0*, and *CAR1* genes under nitrogen stress revealed that the prolonged up-regulation of the *RmDGA1*, *RmCAR0*, and *CAR2* genes serves a molecular indicator of lipogenesis and carotenogenesis. Subsequent fatty acid profiling unveiled an accumulation of oleic and palmitic acids under nitrogen limitation during the stationary phase. Finally, the production of carotenoids, such as β -carotene, torulene, and torularhodin increased as nitrogen availability decreased. This investigation enhances our understanding of nitrogen stress adaptation and lipid-carotenoid biosynthesis in *R. mucilaginosa*. The insights gained from this study can contribute to the development of more efficient biotechnological processes for the production of valuable compounds.

WNT SIGNALING IN CANCER STEM CELL: TOWARDS THE NON-CANONICAL PATHWAY

Miguel Angel Sarabia Sánchez¹, Angela Patricia Moreno Londoño¹, María Cristina Castañeda Patlán¹,
Eduardo Alvarado Ortiz¹, Juan Carlos Martínez Morales¹ and Martha Robles Flores¹

¹Department of Biochemistry, Faculty of Medicine, National Autonomous University of Mexico (UNAM), Mexico City 04510, Mexico
E-mail: rmartha@unam.mx; Tel: +5255 5623 2258

Cancer is a multifactorial disease featured by intra- and inter-tumoral cell heterogeneity. The research on the intra-tumoral heterogeneity has revealed the existence of Cancer Stem Cells (CSC), which are mainly responsible for maintaining tumor growth, chemoresistance, and metastasis. Therefore, understanding CSC characteristics is critical to progress in cancer therapy¹. Wnt signaling has been well established for the functionality of CSC, however the majority of studies have been focused on β -catenin-dependent mechanisms, referred as canonical Wnt pathway.

Recent previous findings have demonstrated that a single Wnt ligand trigger signaling beyond β -catenin, wherein canonical and non-canonical Wnt pathways are simultaneously activated in colon cancer cells. In addition, the stimulation with Wnt3a or Wnt5a (prototypic ligands engage in eliciting canonical and non-canonical Wnt pathway, respectively) triggers PLC-dependent calcium mobilization². These data were obtained from cultures with cellular heterogeneity, where CSC are underrepresented, so it remained to know whether the Wnt/calcium pathway is crucial for CSC.

The aim of the present research was to investigate the role of the Wnt/calcium pathway in CSC of colorectal cancer. We employed sphere-forming cultures as a tool to augment the proportion of CSC upon *in vitro* conditions. Moreover, the use of colorectal cell lines with responsive or constitutive activity of the Wnt/ β -catenin pathway allowed to discern the contribution of Wnt/calcium signaling in the functional effects of Wnt ligands on CSC.

The results have shown that the sphere-formation capacity, a typical CSC-related characteristic, was promoted by Wnt3a or Wnt5a, without necessarily stimulate β -catenin-dependent transcription.

Upregulation of sphere formation by Wnt3a or Wnt5a required the downstream activation of PLC, the intracellular calcium mobilization, and the transcription factor NFAT³.

Therefore, our results indicate that both types of Wnt ligands activate Wnt/calcium signaling axis to induce and maintain CSC of colorectal cancer. These findings open the outlook towards the possible involvement of Wnt/calcium in other features of CSC related to the persisting challenge in disease eradication.

References

1. Qiuping L, et al. (2023). *Cancer cell International*, 23(1):305
2. Flores-Hernández E, et al. (2020). *Cell Signaling*, 72, 1-14.
3. Sarabia-Sánchez MA, et al. (2023). *Frontiers in Oncology*, 13, 1-14

BORIS: A NOVEL ONCOGENE AND MOLECULAR PLAYER IN GLIOBLASTOMA PATHOGENESIS

Ernesto Soto-Reyes¹, Sofía Plata-Burgos¹, Gerardo Ramirez-Mejia¹,
Aylin del Moral Morales, Thalia E. Sánchez-Correa²

¹Departamento de Ciencias Naturales, Universidad Autónoma Metropolitana-Cuajimalpa (UAM-C), Mexico City, Mexico. cp05348, phone 55 5558146500 ext. 3879. ² Servicio de Neurocirugía, Hospital General de Xoco, Mexico City, Mexico. Corresponding author: esotoreyes@cua.uam.mx

Glioblastoma Multiforme (GBM) stands as one of the most aggressive and lethal forms of primary brain tumors, characterized by its relentless infiltration, rapid proliferation, and resistance to conventional therapies, which often result in poor patient outcomes. Despite extensive research efforts, the molecular intricacies underlying GBM pathogenesis remain elusive. Unlike other malignancies, where significant strides have been made in deciphering molecular drivers, GBM presents a challenge due to its inherent heterogeneity and complex interplay of genetic and epigenetic alterations. Interestingly, recent studies have shed light on potential molecular players in GBM pathogenesis. It has been reported that Brother of the Regulator of Imprinted Sites (BORIS) may compete with its paralog, CTCF, leading to impaired gene expression. BORIS has emerged as a significant factor in various tumors, with studies suggesting its potential role in conferring stemness characteristics to cancer cells, thus contributing to cancer formation and progression. Consequently, BORIS has been proposed as a promising cancer marker, offering insights into the molecular landscape of GBM and paving the way for novel diagnostic and therapeutic strategies. To address this, we investigated the expression levels of BORIS and its paralog, CTCF, in T98G and U87MG cell lines. Additionally, we employed chromatin immunoprecipitation assay followed by Next Generation Sequencing (ChIP-seq) to identify BORIS binding sites on the genome. Subsequently, we conducted gene ontology analysis to elucidate the functions of the genes potentially regulated by BORIS. Our findings unveil an inverse expression pattern between BORIS and CTCF, suggesting potential competition for gene binding sites. ChIP-seq analysis revealed that BORIS predominantly binds to promoter regions. Furthermore, gene ontology analysis suggests that BORIS may regulate genes involved in developmental processes, anatomical morphogenesis, various cellular functions, and cell communication, which could elucidate how glioblastoma cells acquire stemness characteristics. Also, structural biology analyses were conducted to demonstrate the capacity to compete for consensus sequences between BORIS and CTCF. These findings shed light on the intricate molecular mechanisms driving GBM, offering promising avenues for identifying prognostically significant biomarkers.

CROSSTALK BETWEEN WNT/ β -CATENIN AND PI3K/AKT PATHWAYS MEDIATED BY HOTAIR/HIF1 α AXIS IN CERVICAL CANCER

Samuel Trujano Camacho^{1,2}, Nadia Jacobo Herrera³, Concepción Gutiérrez Ruiz^{5,6}, Jossimar Coronel Hernández², Oliver Millan Catalan² and Carlos Pérez-Plasencia^{2,4}

¹Experimental Biology Ph Program, DCBS, Universidad Autónoma Metropolitana- Iztapalapa, Iztapalapa, Mexico. ²Genomics Laboratory, National Cancer Institute (INCan), Mexico City, Mexico. ³Biochemistry Unit, National Institute of Medical Sciences and Nutrition, Salvador Zubirán (INCMNSZ), Mexico City, Mexico. ⁴Biomedicine Unit, Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México (UNAM), Tlalnepantla, Edo de México. ⁵Laboratory of Experimental Medicine, Translational Medicine Unit, Instituto de Investigaciones Biomédicas, UNAM / Instituto Nacional de Cardiología Ignacio Chávez, Tlalpan, Mexico City C.P. 14080, Mexico. ⁶Department of Health Sciences, Universidad Autónoma Metropolitana-Iztapalapa, Iztapalapa, Mexico City C.P. 09340, Mexico.

Corresponding author: Carlos Perez-Plasencia Ph. D.
Av. San Fernando 22, 5533572955, carlos.pplas@unam.mx

One of the main characteristics of cervical cancer (CC) is the deregulation of cellular responses through interaction networks known as signaling pathways. The recent research has pointed to long non-coding LncRNAs, as complex molecules involved in sustained activation of cancer-related signaling pathways. Research suggests that the long non-coding RNA HOTAIR acts as a positive regulator of the PI3K/AKT and Wnt/ β -catenin signaling pathways in several types of cancer. Despite this, little has been described about the mechanisms that allow sustained activation of the PI3K/AKT and Wnt/ β -catenin pathways mediated by HOTAIR in CC. In this way, we evaluated the crosstalk between PI3K/AKT and Wnt/ β -catenin pathways when HOTAIR was knocked and overexpressed evaluated its effect over the transcriptional activity of both signaling pathways in HeLa cell line derived from cervical cancer, demonstrating that HOTAIR is an essential molecule in the sustain activation of both signaling pathways. Finally, we demonstrated that HIF1 α transcribes HOTAIR which entails to methylation of the PTEN promoter by means of DNMT1, thus maintaining the activation of the PI3K/AKT and Wnt/ β -catenin pathways. These results suggest a new mechanism where HOTAIR maintains positive crosstalk between PI3K/AKT and Wnt/ β -catenin activation as well as HOTAIR/HIF1 α axis in cervical cancer.

NEW SMALL-MOLECULE CD36 ANTAGONISTS

Marco Velasco-Velázquez¹, Sandra L. Guerrero-Rodríguez¹, Fabiola Cortés-Mendoza¹, Karla Pineda-Medina¹, Nohemí Salinas-Jazmín¹, Karen Bahena-De Dios¹, Mayra Pérez-Tapia²

¹Department of Pharmacology, School of Medicine, UNAM. ²UDIBI, ENCB, IPN
Ciudad Universitaria, Coyoacán, CDMX CP 04510. Tel. 55 56232282 marcovelasco@unam.mx

Lipid metabolic reprogramming in cancer cells is key for tumor progression. CD36 is overexpressed in several types of cancer where it increases the fatty acids (FA) uptake. CD36⁺ cancer cells display increased stemness and functional properties associated to malignancy, including clonogenicity, chemo- and radioresistance, and metastasis-initiating capability. Thus, CD36 has been pointed as a potential therapeutic target in cancer.

In this work we aimed to identify small-molecule CD36 antagonists by combining virtual screening and *in vitro* experimental evaluation. In our computational assays, we identified a pocket within the fatty acid channel of CD36 that: i) is druggable; ii) includes key functional residues; iii) contains “hot spots” for the binding of small molecules; and iv) is likely the binding site for reported CD36 inhibitors. Molecular docking of +25,000 compounds from a chemical library of drug-like compounds found fifteen potential antagonists that bind CD36 with high ligand efficiency. Based on molecular dynamics simulations and commercial availability, six compounds were purchased for experimental evaluation in cultures of HepG2 cells. Four compounds reduced the uptake of fluorescent palmitic acid (PA) in by flow cytometry assays. The effect of the compounds on clonogenicity was evaluated in tumorsphere assays. Five of the compounds reversed the increase in tumorsphere-formation efficiency caused by PA. However, two of those compounds were highly cytotoxic; suggesting that their effect on clonogenicity is not mediated by CD36 blockage. Interestingly, one compound stimulated both PA uptake and tumorsphere formation, indicating that it activates CD36. In conclusion, our discovery platform allowed the identification of CD36 antagonist that can become lead compounds for the development of new therapies.

Supported by CONAHCYT A1-S-18285 and PAPIIT UNAM IN206622.

REGULATORY RELATIONSHIPS BETWEEN GENES ARE NOT LINKED TO DIRECT TRANSCRIPTION FACTOR-TARGET INTERACTIONS IN GENE REGULATORY NETWORKS

Diego Velázquez Trejo^{1,2}, Adrián Islas Ortíz^{1,2}, Daniela Del Toro¹, Humberto Gutiérrez¹

¹Instituto Nacional de Medicina Genómica, Periférico Sur 4809, 14610 CDMX, ²Posgrado en Ciencias Biológicas, UNAM.
+525630001076, d.velazquez.mc@gmail.com

Gene expression patterns are governed by interactions between numerous transcriptional regulators and their target genes. Most gene regulatory interactions reported in the literature have been inferred from gene knock down/out experiments, based on the general assumption that the expression of transcriptional regulators is directly or indirectly associated to the expression of the corresponding target genes. This assumption, however, has never been formally tested. Using a computational gene regulatory network model we investigate the statistical association between the expression levels of regulatory genes and their direct targets. We find that regulators tend to display little to no correlation with their direct targets and this remains independent on whether the regulatory influence is positive or negative. Using available human expression data for a set of 260 known transcription factors and their experimentally confirmed targets, in combination with expression perturbation data derived from transcription factor knock-down measurements compiled in the C Map project database, we confirmed the lack of association between gene regulators and direct targets observed in synthetic regulatory networks. We define “effective” regulators as genes with the highest influence on a target gene, after the regulator’s knock down, in opposition to “direct” regulators (that is, direct transcription factor-target pairs). Along these lines, we find that when an “effective” regulator is knocked out, the association between the target gene and its highest correlate is significantly disrupted, suggesting that correlated expression in transcriptional networks is the result of coregulation, rather than direct interaction between coexpressed genes. In contrast, and in line with the above findings, knocking out the “direct” regulator does not affect the association between the target gene and its highest correlate beyond what would be expected by chance. Our results suggest that, at the transcriptome level, regulatory relationships between genes cannot be attributed to direct transcription factor-target interactions but are instead diffusely distributed throughout the transcriptional network.

A MODULAR TOOLBOX TO STUDY BIOLOGICAL SYSTEMS: REAL-TIME VOLATILOMICS AND MASS SPECTROMETRY IMAGING UNDER AMBIENT CONDITIONS

Robert Winkler^{1,2}, Raúl Alcalde-Vázquez¹, Rosina Torres-Ortega¹, Héctor Guillén-Alonso^{1,2}, Abigail Moreno-Pedraza³, Ignacio Rosas-Román⁴, Jens Riedel⁵, Enrique Ramírez-Chávez¹, Jorge Molina-Torres¹, Cesaré Ovando-Vázquez⁶, Laila Pamela Partida-Martínez¹

¹CINVESTAV Irapuato, 36824 Irapuato, México, ²Department of Biochemical Engineering, Nacional Technological Institute, 38010 Celaya, Mexico, ³Leibniz-Institut für Gemüse- und Zierpflanzenbau (IGZ), 14979 Großbeeren, Germany, ⁴Universidad de Guanajuato, División de Ciencias e Ingenierías, 37150 León, Mexico, ⁵Bundesanstalt für Materialforschung und -prüfung (BAM), 12205 Berlin, Germany, ⁶CONACYT IPICYT, 78216 San Luis Potosí, Mexico.
E-mail: *robert.winkler@cinvestav.mx

Biological systems exhibit highly complex and dynamic networks of metabolites. However, conventional analytical technology usually requires extensive sample manipulation, which results in the loss of relevant temporal and spatial information about living organisms.

The profile of volatile organic compounds (VOCs) of plants can change in seconds, for example, as a response to plant wounding, and act as mediators and for chemical communication. To overcome limitations of existing devices, i.e., slow response (GC-MS: minutes to hours/sample), high cost (PTR-MS: ~1 million USD), and bias towards detectable molecule classes (PTR-MS, SPME), we build the 'Modular Biological Mass Spectrometer' (MoBiMS) which can measure a broad range of VOCs and provides spectra compatible with the NIST database. The low-cost mass analyzer (~60,000 USD) permits real-time VOC analyses of biological samples with a time resolution of less than 1 second.

Further, we developed a low-temperature plasma probe to probe biological surfaces directly. The 6-pentyl- α -pyrone (6-PP) signal during the 10-day interaction between *Trichoderma atroviride* and *Arabidopsis thaliana* demonstrated a regular pattern correlating with the day-night cycle. Time series analyses confirmed 6-PP as a physiological variable promoting the homeostasis of the plant-fungal interaction [2].

In addition, the localization of compounds indicates their biological function and biosynthesis. Thus, we built the 'Open LabBot/ RmsiGUI' hardware/software platform for the chemical imaging of fresh plant tissue under ambient conditions [3]. This system allowed us to visualize the distribution of bioactive alkaloids in native tissues [4] and to investigate the time course of metabolic profiles during the mesquite-mistletoe parasitism.

Altogether, we built a modular toolbox for studying molecules in their biological context under realistic conditions and provided the research community with open hardware and software solutions.

Funding: Conacyt-DFG 2016/277854

References

1. Alcalde-Vázquez, R. et al. (2022), *Microchemical Journal* 175, 107090.
2. Torres-Ortega, R. et al. (2022), *Metabolites* 12, 1231.
3. Rosas-Román, I. et al. (2020), *Microchemical Journal* 152, 104343.
4. Moreno-Pedraza, A. et al. (2019), *Analytical Chemistry* 91, 2734.

CHARACTERIZATION OF THE GLYOX FROM *AZOTOBACTER VINELANDII* AND ITS APPLICATION FOR GLYCINE BIOSENSING

Andrés Zárate-Romero^{1,2}, Aarón Mena-Rodríguez¹, Raúl García-Morales³, Rafael Vázquez-Duhalt¹

¹Centro de Nanociencias y Nanotecnología UNAM, Ensenada, B. C., ²Consejo Nacional de Humanidades, Ciencias y Tecnologías, CDMX ³División Académica de Ciencias Básicas, Universidad Juárez Autónoma de Tabasco, Cárdenas, Tabasco.
E-mail: azarate@ens.cnyn.unam.mx

Glycine level is an indicator of diseases related to the central nervous system and metabolic disorders¹. It is therefore relevant to have tools that can detect glycine at physiological concentrations. Current methods to measure glycine are slow, invasive, expensive and hard to access. The use of biosensors promises to be a reliable alternative with low cost and high response speed². Carbon paper biosensors in conjunction with Prussian blue as a mediator have shown favourable results in the production of amperometric biosensors. This type of biosensor usually relies on enzymes to have specificity in the detection of analytes. In the case of glycine detection, a glycine oxidase can be used. The glycine oxidase (GlyOx) from *Azotobacter vinelandii* is an uncharacterized enzyme, the sequence analysis suggests that is a monomeric protein, in difference to most of the characterized GlyOx which are tetramers. The monomeric structure is favorable for oriented immobilization of active site in a biosensor for further applications. In this work, the GlyOx from *A. vinelandii* was characterized and a glycine biosensor was assembled. The enzyme was expressed in *E. coli* in and purified by chromatographic methods. The saturation kinetics characterization of the enzyme with glycine as substrate was carried out, and the optimum pH value, thermal stability, and oligomerization state were obtained. Maximum activity was observed at pH 9, K_m , k_{cat} , and K_{si} values of 4.65 mM, 0.57 s⁻¹ and 32.13 mM were obtained, respectively. The enzyme was confirmed as a monomer, according to DLS and gel filtration chromatography. For the fabrication of the biosensor, a composite of chitosan GlyOx was deposited on carbon paper added with Prussian blue. The electrochemical characterization for the biosensor for detection of glycine was carried out by cyclic voltammetry and chronoamperometry. The LOD and LOQ of the biosensor were 2.66 mM and 8.05 mM. The fabricated glycine biosensor is sensitive to slightly higher concentrations than plasma/blood normal levels. This prompts further investigation of fabrication conditions that improve the efficiency of GliOx-based biosensors for use in glycine detection.

References

1. Wang Q, Liu Y, Campillo-Brocal JC, Jiménez-Quero A, Crespo GA, Cuartero M. (2021) Biosens Bioelectron; 182:113154.
2. Pérez-Ràfols C, Liu Y, Wang Q, Cuartero M, Crespo GA. (2020) Sensors (Basel). 20(14):4049.



TECHNICAL TALKS

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

LET'S OPTIMIZE YOUR PURIFICATION PROCESS

Carlos Bravo

Bio-Rad

carlos_bravoalberto@bio-rad.com

Achieving a successful method for protein purification usually is a slow, complicated, and frustrating process that consumes plenty of time and resources. Developing a new protein purification process represents a complex challenge requiring the integration of multiple unit operations to ensure high purity, yield, and safety of the final product. Design of Experiments (DoE) offers a systematic approach to optimize these processes, enhancing both efficiency and robustness. In this talk, we will delve into the principles of DoE coupled to Stain-free electrophoresis and Image Lab analysis and its application in the downstream purification of viral vaccines. Through real-world viral vaccine purification example, we will demonstrate how Bio-Rad's DoE in just a few hours can uncover interactions between variables, enhance process robustness, and ensure scalable and cost-effective protein production according to your own goals.

ESTRATEGIAS PARA LA PURIFICACIÓN DE PROTEÍNAS

Carlos Chávez Castañeda

Especialista de aplicaciones para Cytiva en Latinoamérica.
(Científica Senna)

¿Cuáles son los factores más importantes que hay que tener en cuenta a la hora de desarrollar un protocolo de purificación? Sabemos que el desarrollo de un protocolo no es sencillo y debemos plantearnos si hacer marcación o no, las técnicas cromatográficas y cómo combinarlas, cuál es la mejor fase estacionaria para cada fase del proceso, y los métodos analíticos necesarios. En esta presentación se abordarán algunos aspectos fundamentales, como las resinas para la purificación de proteínas recombinantes y los flujos de trabajo de purificación.

What are the most important factors to consider when developing a purification protocol? We know that protocol development is not easy and we need to consider whether to label or not, chromatographic techniques and how to combine them, the best stationary phase for each stage of the process, and the analytical methods required. This talk will cover some key aspects, such as resins for recombinant protein purification and purification workflows.

ESPECTROMETRÍA DE MASAS CON TECNOLOGÍA ORBITRAP Y SUS PRINCIPALES APLICACIONES EN CIENCIAS DE LA VIDA

Filippo Bedani

Instrumentación y Servicios Analíticos, S.A.

www.isasa.com.mx

La espectrometría de masas es una técnica analítica poderosa que, en el lapso de unas décadas, de ser una herramienta para “las élites” académicas e industriales se ha vuelto en una herramienta accesible a un número creciente de laboratorios en todo el mundo. Este aumento en el número de espectrómetros de masas instalados se debe a aspectos como avances en tecnología, expansión en el número de aplicaciones, requerimientos regulatorios, miniaturización, reducción de costos, y la posibilidad de alto rendimiento, para mencionar algunos.

Cuando se habla de espectrometría de masas, una de las distinciones más importantes es entre los espectrómetros de masas de alta y de “baja” resolución. Mientras estos últimos son más adecuados para análisis “dirigidos”, los espectrómetros de masas de alta resolución permiten también análisis “no dirigidos”, y se han vuelto herramientas imprescindibles en muchas investigaciones, y en particular en las ciencias de la vida.

En esta presentación se hablará de la espectrometría de masas de alta resolución con analizadores de masas con tecnología Orbitrap, y de cómo espectrómetros de masas con esta tecnología acoplados a técnicas cromatográficas han revolucionado la investigación en disciplinas de grande actualidad como lo son la metabolómica y la proteómica.

QUALITY ANALYSIS USING THE AGILENT PROTEOANALYZER SYSTEM AND SDS-PAGE. A COMPARISON OF SIZING AND QUANTIFICATION PERFORMANCE

Daniel Favato

Genomics Field Application Scientist, Latin America, Agilent Technologies
(Patrocinado por Química Valaner)

Quality control (QC) provides valuable information about the integrity of samples such as proteins before use in assays, experiments, or product release. Ensuring that samples are of high quality enhances the reproducibility of workflows, reduces variability and minimizes the risk of data inconsistencies. Among the different attributes of proteins, the size and quantification of samples can be assessed with electrophoretic separations using sodium dodecyl sulfate (SDS). SDS denatures the sample and provides the proteins with a consistent mass-to-charge ratio, allowing for size-based separation. Traditionally, protein QC is performed with sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). Alternatively, capillary electrophoresis sodium dodecyl sulfate (CE-SDS) uses the same principles of SDS denaturing and size-based separation but uses a gel-filled capillary instead of a slab gel. In doing so, CE-SDS provides faster separation times, higher resolution, accurate sizing, and consistent quantification. The Agilent ProteoAnalyzer system is an automated CE-SDS instrument that uses a 12-channel capillary array to analyze multiple samples in parallel and allows for multiple runs to be programmed at once. The system is designed to facilitate precise and accurate measurements of proteins and allow for the detection of impurities while only requiring 1 μ L of sample.

In this seminar, quality assessment of different proteins will be compared between automated CE-SDS using the ProteoAnalyzer system and conventional SDS-PAGE.

TOOLS AND SOLUTIONS FOR 3D CELL CULTURE

David Sebastián Hernández Toledano

Uniparts

Cuitláhuac 46, Col. Anáhuac. Ciudad de México, C.P. 11320.

Tel. 5548221157; asesoriabiocel@uniparts.com.mx

The evolution of cell cultures toward tridimensional (3D) systems has marked a significant advance in biomedical research, allowing the creation of more realistic and functional models that better resemble the natural microenvironment of *in vivo* tissues.

These systems offer an effective alternative for the research of cell-cell and cell-extracellular matrix (ECM) interactions, which is essential for several applications, such as studies in disease progression, response to xenobiotics, and developmental biology.

The development of technologies and products, such as hydrogels, synthetic scaffolds, and scaffold-free systems, has allowed the overcoming of technical barriers in 3D cell cultures. These innovations help us achieve better replication of the tissue microenvironment, optimize cell growth and differentiation, and provide more precise control of cell culture conditions, thereby increasing the reproducibility and scalability of experiments.

Through the integration of diverse new technologies, we present workflows, solutions, and tools for the establishment, improvement, and evaluations of 3D cell cultures.

MACS TECHNOLOGY: CELL SEPARATION

UNIPARTS

Miltenyi Biotec's Magnetic Cell Separation (MACS) technology, allows magnetic separation of cell populations using specific antibodies conjugated to magnetic microbeads to label surface epitopes. This technology offers various cell separation options that can be performed in 3 simple steps: 1) magnetic labeling with MACS reagents, 2) magnetic separation of labeled and unlabeled cells with MACS Columns, and 3) elution of labeled cells. With Miltenyi, you can perform different types of cell separation, positive cell isolation, negative cell isolation or label-free selection. This method is fast and gentle, ideal for isolating viable and functional cells, which allows continuing with the workflow, you can make cell culture, cell analysis, molecular analysis or downstream sorting.

MACS technology, whether in manual, automated or semi-automated versions, is widely used for basic, translational and clinical research, for example in biomedical research, cell therapies and stem cell studies, providing effective tools for the manipulation and analysis of specific cell populations.

The background of the entire page is a composite image. On the left, there is a detailed cross-section of a leaf, showing the intricate network of veins and the cellular structure of the epidermis and mesophyll. On the right, there is a glowing, golden-yellow molecular model, possibly representing a protein or a complex carbohydrate, with various atoms and bonds visible. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

BASIC BIOCHEMISTRY

ARE ZEA MAYS HXK7-8 GLUCOSE SENSORS?

David Acosta Muro and Sobeida Sánchez Nieto

Dpto. de Bioquímica, Fac. De Química, UNAM. Ciudad Universitaria, Coyoacán, Cd.Mex. México, C.P. 04510

E-mail: 317153794@quimica.unam.mx and sobeida@unam.mx

Hexokinases (HXK) are key enzymes for glucose metabolism, they phosphorylate hexoses as the first step in the glycolysis, but besides their catalytic function several HXKs have glucose sensor activity. In *Arabidopsis thaliana* plants, *AtHXK1* is a catalytic enzyme and also a glucose sensor that downregulates the expression of the photosynthetic genes when the protein senses high glucose concentration (1, 2). Nine HXKs have been found in maize, three are HXK-like (*ZmHXK3a*, *ZmHXK3b* and *ZmHXK10*), four are mitochondrial with catalytic and glucose sensor function (*ZmHXK4*, 5, 6 and 9), and two cytosolic with catalytic activity (*ZmHXK7* and 8) (1, 3). However, until this moment it is not known whether these cytosolic isoforms are glucose sensors as the cytosolic hexokinase 7 (*OsHXK7*) found in rice (*Oryza sativa*) (4). *A. thaliana* null mutant *gin2-1*, insensitive to glucose, presents a nonsense mutation in the HXK1 gene (*Q432**), this mutant is often used to test the sensor activity of different HXKs. To investigate if *ZmHXK7* and 8 have sensor activity, in this work *gin2-1* plants were complemented with these genes. The genes were subcloned from *Escherichia coli* strains obtained previously (3). The presence of the genes in the clones were confirmed by PCR and sequence analysis before the *gin2-1* were subject to the floral dip transformation protocol. The T2 plants were able to grow in selection medium. The plants will be tested for the glucose sensor activity.

References:

1. Rodríguez-Saavedra, C., Morgado-Martínez, L. E., Burgos-Palacios, A., King-Díaz, B., López-Coria, M., & Sánchez-Nieto, S. (2021). *Frontiers in Molecular Biosciences*, 8, 1-23.
2. Moore, B., Zhou, L., Rolland, F., Hall, Q., Cheng, W. H., Liu, Y. X., Hwang, I., Jones, T., & Sheen, J. (2003). *Science* (New York, N.Y.), 300, 332–336.
3. Aguilera-Alvarado, G. P., & Sánchez-Nieto, S. (2017). *Plant & Cell Physiology*, 58, 1151–1160.
4. Kim, H. B., Cho, J. I., Ryoo, N., Shin, D. H., Park, Y. I., Hwang, Y. S., Lee, S. K., An, G., & Jeon, J. S. (2016). *Journal of Integrative Plant Biology*, 58, 127–135.

EVALUATION OF SPECIFIC L-GLUTAMINASE ACTIVITY IN THE UNCONVENTIONAL YEAST *RHODOTULA MUCILAGINOSA*

Paola Itzel Acosta-Valdelamar, Paulina Castañeda-Tamez, Ofelia Méndez-Romero, Carolina Ricardez-García and Salvador Uribe-Carvajal¹

¹Mexico. Cto. Exterior s/n, C.U., Coyoacán, 04510 Ciudad de México, CDMX.
Tel: 5556225632; suribe@ifc.unam.mx

Some organisms grow in environments that are inhospitable to others. Among other conditions, the lack of essential nutrients prevents growth. Glutamine (Gln) is essential in many organisms as it may be used both, as a nitrogen source for the biosynthesis of amino acids and nucleotides, and as a carbon source to build lipids, carbohydrates and tricarboxylic acid cycle (TCA) intermediates. In undernourished mammals and in tumour cells, the glutaminolysis mitochondrial anaplerotic pathway is essential for energy production. At the start of the Gln-dependent anaplerotic pathways, the amidohydrolase enzyme L-glutaminase (GLS) catalyzes the conversion of glutamine to glutamate, releasing ammonium ions. The extremophile yeast *R. mucilaginosa* was cultivated in a minimal medium containing Gln as the sole carbon and nitrogen source and efficient growth was observed. To further analyze its Gln-handling properties, the activity of L-glutaminase was measured in *R. mucilaginosa* grown in minimal medium plus 2% dextrose. Glnase activity was higher in the stationary phase than in the log phase.

OVEREXPRESSION OF TIMP3 DECREASES MIGRATION IN A549 CELL LINE

Arantxa Melissa Aguilar López^{1,2}, Marco Antonio Espina Ordóñez^{1,2}, Remedios Ramírez³, Annie Pardo³,
Moisés Selman¹, José Cisneros¹, Mariel Maldonado¹

¹Instituto Nacional de Enfermedades Respiratorias "Ismael Cosío Villegas", Calz. De Tlalpan 4502, sección XVI, Tlalpan, 14080, CDMX, México. ²Posgrado en Ciencias Biológicas, UNAM, ³Facultad de Ciencias, UNAM, Investigación Científica, C.U., Coyoacán, 04510, CDMX, México.
E-mail: maldonadomariel@outlook.com

Matrix metalloproteinases (MMPs) are zinc dependent proteins that degrade components of the extracellular matrix, and activate other MMPs or diverse mediators. One way these proteins are regulated is through the tissue inhibitors of metalloproteinases (TIMPs), although there are cases where TIMP2 can activate proMMP2¹. MMP28 induces an invasive and migratory phenotype in alveolar epithelial cells², in addition with an increase of TIMP3. Based on this background, the aim of this study is to analyze the role of TIMP3 in the migratory phenotype induced by MMP28 in A549 cells and its relationship with MMP2. Results: qPCR demonstrated that the increase of TIMP3 in MMP28 overexpressing cells is independent of TGFB. In addition, overexpression of TIMP3 in A549 cells, resulted in an increase of TIMP3 confirmed by qPCR and western blot. These cells exhibited reduced migration in wound healing assays, with a significant difference observed at 24 hours, indicating decreased migration in A549 cells overexpressing TIMP3. Besides, in MMP28 overexpressing cells and at the same time overexpressing TIMP3 via lentiviral particles, TIMP3 protein levels in total lysate were initially elevated but decreased with passaging, correlating with more migration. Furthermore, through an ELISA an increase in TIMP3 was confirmed in conditioned medium from cells that exhibited higher TIMP3 in the total lysate and slower migration phenotype. Based on these results, it was concluded that in these scenarios, TIMP3 is not associated with a faster migration in MMP28 overexpressing cells. These findings aligns with the literature, where the overexpression of TIMP3 is correlated with the decrease of migration.

References

1. Itoh, Y. (2015) *Matrix Biology*. **44-46**:207-23
2. Maldonado, M., Salgado-Aguayo, A., Herrera, I., Cabrera, S., Ortíz-Quintero, B., Staab-Wejnitz, C., Eickelberg, O., Ramírez, R., Manicone, A., Selman, M., Pardo, A. (2018) *American Journal of Respiratory Cell and Molecular Biology*. **Vol. 59**: 77-86.

INHIBITION OF P62 REDUCES MOBILITY AND INTRACELLULAR CALCIUM IN MOUSE SPERMATOZOA

Charmina Aguirre Alvarado^{1,2}, Vianey Flores González¹, Lorena Rodríguez Páez¹,
Guadalupe Elizabeth Jiménez-Gutiérrez³ y Joaquín Cordero Martínez¹

¹Laboratorio de Bioquímica Farmacológica. Escuela Nacional de Ciencias Biológicas. Instituto Politécnico Nacional, México City, México. ²Unidad de Investigación Biomédica en Inmunología e Infectología, Hospital de Infectología “Dr Daniel Méndez Hernández”, CMN “La Raza”, Instituto Mexicano del Seguro Social (IMSS), México City, México.

³Departamento de Genética, Laboratorio de Medicina Genómica, Instituto Nacional de Rehabilitación Luis Guillermo Ibarra Ibarra, Ciudad de México, Mexico
E-mail: charmina_burana@hotmail.com

Autophagy is a homeostatic and bioenergetic regulatory mechanism to ensure survival. This highly conserved eukaryotic cellular recycling process refers to cytoplasmic structures and even complete organelles, which are contained in a vesicle called autophagosome, which when fused with lysosomes will form the autolysosome. The p62 protein has been related to the degradation of protein complexes and in the formation of the autophagosome through its interaction with the LC3 protein. LC3 has been described as a biomarker of autophagy in mammals as it is part of the autophagosome membrane. In some studies it has been observed that p62 levels decrease when autophagy is induced and when autophagy is inhibited p62 levels increase¹. Autophagy plays a very important role in ensuring that spermatozoa fulfill their function of successfully fertilizing the egg². To date, the mechanism of p62-mediated autophagy in spermatozoa is not fully understood.

In this work, total and progressive sperm motility, sperm viability, as well as intracellular calcium $[Ca^{2+}]_i$ concentrations were evaluated. The results indicate that the kinetic parameters of motility, total and progressive motility decreased dramatically after 15 min of incubation in the presence of K67, suggesting that p62 may play an important role in mouse sperm motility. Low concentrations of the inhibitor K67 did not affect sperm viability (1.25 and 2.5 μ M), suggesting that the previously observed effect on sperm motility is due to p62 inhibition and not to any toxic effect of the inhibitor. The presence of the inhibitor K67 did not induce a statistically significant decrease in $[Ca^{2+}]_i$, suggesting that p62 inhibition is not related to modulation of Ca^{2+} channels, whose activity is similarly related to sperm capacitation. Taken together, the results suggest that p62 activity plays an important role in the maintenance of sperm motility and that its inhibition results in a drastic decrease in mouse sperm motility and most likely in the induction of autophagy.

References

1. Runwal, G, et al. (2019) *Sci Rep* **9**: 10147.
2. Zhu Y, et al (2019). *Syst Biol Reprod Med* **65**: 265–272.

EFFECT OF CHITOSAN ON THE PERMEABILITY OF DIFFERENT YEASTS STRAINS

Minerva Araiza-Villanueva, Francisco Padilla-Garfias, Mariana Maldonado, Martha Calahorra, Norma Silvia Sánchez and Antonio Peña

Departamento de Genética Molecular, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Circuito Exterior s/n, Ciudad Universitaria, México City, 04510, México
E-mail: maraiza@ifc.unam.mx, apd@ifc.unam.mx

Chitosan is an oligosaccharide derived from chitin that at acidic pH is protonated and forms a polycation which, upon binding to membrane phospholipids, by its negative charge destabilizes the membrane, causing cell death in microorganisms such as yeast and filamentous fungi. Although its efficacy is lower in comparison with antifungals, chitosan has been shown to inhibit the growth of various microorganisms, affecting the permeability of the membrane and the organization of its cell wall. In this work, we propose the use of chitosan to permeabilize different yeast strains to study metabolic functions *in situ*. This shows an advantage to methods where cell extracts are prepared, since cell functionality is preserved when cells are treated with low concentrations of chitosan. Therefore, we analyzed the effect of chitosan in *Saccharomyces cerevisiae*, *Candida albicans* and *Debaryomyces hansenii* yeast strains and its impact on cell wall and cell membrane integrity as well as determine the enzymatic activity of enzymes such as hexokinase, glucose-6-phosphate dehydrogenase, aldolase, enolase, pyruvate decarboxylase and alcohol dehydrogenase.

References

1. Aranaz, I., Alcántara, A. R., Civera, M. C., Arias, C., Elorza, B., Heras Caballero, A., & Acosta, N. (2021). Chitosan: An overview of its properties and applications. *Polymers*, 13(19), 3256. <https://doi.org/10.3390/polym13193256>
2. Peña, A., Sánchez, N. S., & Calahorra, M. (2013). Effects of chitosan on *Candida albicans*: conditions for its antifungal activity. *BioMed Research International*, 2013. <https://doi.org/10.1155/2013/527549>
3. Li, R., Zhu, L., Liu, D., Wang, W., Zhang, C., Jiao, S., Wei, J., Ren, L., Zhang, Y., Gou, X., Yuan, X., Du, Y., & Wang, Z. A. (2022). High molecular weight chitosan oligosaccharide exhibited antifungal activity by misleading cell wall organization via targeting PHR transglucosidases. *Carbohydrate Polymers*, 285. <https://doi.org/10.1016/j.carbpol.2022.119253>

THE C-TERMINAL FRAGMENT OF ONE OF THE ASPARAGINYL ENDOPEPTIDASES (AEPs) OF *TRICHOMONAS VAGINALIS* WORKS AS AN AEP PROTEOLYTIC ACTIVITY INHIBITOR

Rossana Arroyo^{1*}, Esly Alejandra Euceda-Padilla¹, Claudia Ivonne Flores Pucheta², Octavio Montes-Flores², Miriam Guadalupe Mateo-Cruz¹, Jaime Ortega López^{2*}

Centro de Investigación y de Estudios Avanzados del IPN (CINVESTAV). ¹Depto. de Infectómica y Patogénesis Molecular, ²Depto. de Biotecnología y Bioingeniería, Ciudad de México, CP 07360
rarojo@cinvestav.mx

The asparaginyl endopeptidases (AEPs) are cysteine peptidases that belong to family C13 of clan CD. AEPs play key roles in multiple biological processes and the pathogenicity of some microorganisms, such as cytoadherence in *Trichomonas vaginalis*. *T. vaginalis* is a sexually transmitted protozoan parasite that encodes multiple CPs, particularly 10 AEP genes, which are synthesized as inactive zymogens processed before activation. This type of zymogen is organized in different domains, such as a signal peptide (SP), peptidase domain (PD), activation peptide (AP), and a prodomain at the carboxy-terminal region (CD). The last two domains act as proteolytic activity inhibitors that block the access of the substrate to the catalytic domain. This work aimed to evaluate whether the C-terminal fragment of trichomonad AEP-2 acts as an inhibitor of AEP activity in *T. vaginalis*. Thus, the C-terminal domain of AEP-2 (C-AEP-2) of *T. vaginalis* was cloned and expressed in *E. coli* as a recombinant protein. The C-AEP-2 was purified, and its inhibitory capacity in the AEP proteolytic activity of trichomonad extracts was evaluated. Our data show that the C-AEP-2r partially inhibited the asparaginyl endopeptidase activity of clan CD *T. vaginalis* peptidases, suggesting that this fragment is only inhibiting the proteolytic activity of the cognate peptidase and no other AEPs.

Agradecimientos: Premio Para las Mujeres en la Ciencia L’Oreal-UNESCO-AMC-CONALMEX 2023.

PHENOTYPICAL RESPONSES OF SEEDLINGS FROM *ARABIDOPSIS THALIANA* MPK3 AND MPK6 MUTANTS TO HIGH TEMPERATURES

David F. Barrera-Gómez¹, A. Arturo Guevara-García², Berenice García Ponce de León³, Mariana Saucedo-García⁴, Laura Carmona-Salazar¹ and Marina Gavilanes-Ruiz¹

¹Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México, Mexico City 04510. 5556225376. gavilan@unam.mx; ²Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos 62210; ³Departamento de Ecología Funcional, Instituto de Ecología, Universidad Nacional Autónoma de México, Mexico City 04510; ⁴Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo, Hidalgo

As global temperatures rise, the growth, production, and survival of plants are increasingly affected. Plants have developed different strategies to cope with high temperatures. It has been reported that the activation of MPK3 and MPK6 contributes to signaling in the response to high temperatures. However, this contribution is not fully understood. Therefore, we studied the effects of high temperature exposure on *mpk3*, *mpk6* and *OVMPK6* mutant plants analyzing their phenotypic alterations. *Arabidopsis thaliana* wt and MPK3/MPK6 mutant seedlings and adult plants were treated at 22°C (control), or 37°C (moderate high temperature), or 45 (severe high temperature) or a combination of a pre-exposure at 37°C followed by an exposure at 45°C (considered as a combination of acclimation and challenge condition). Then, phenotypes were assessed by picture recording and imaging processing to quantify the number of emerging leaves, lateral roots, the root length and leaf chlorosis. Seedlings were unaffected by 37 °C exposure compared to the control (22°C). In contrast, 45°C exposure produced a dramatic inhibition of root elongation and the number of lateral roots in all genotypes. In addition, the number of emerging leaves decreased, and the seedlings developed a chlorotic phenotype. In the case of adult plants, no serious damage at 45°C was observed in any genotype, suggesting an effective basal tolerance. However, under exposure at 50°C, plants of all genotypes showed extensive damage that was not observed in the MPK6 adult plants. These results suggest that MPK6 is a positive contributor to heat tolerance, and that MPK3 may have a redundant effect.

This work has been financed by DGAPA, UNAM (PAPIIT IN213124) and Facultad de Química, UNAM (PAIP 5000 9115). DFBG is recipient of the CONAHCYT fellowship number 1317285.

CHEMICAL REMOTION OF GLYCAN MOIETIES TO ENHANCE PROTEIN IDENTIFICATION BY LC-MS/MS

Esaú Bojórquez-Velázquez*, José M. Elizalde-Contreras, Francisco A. Reyes-Soria, Eliel Ruiz-May*

Red de Estudios Moleculares Avanzados, Instituto de Ecología A.C. (INECOL), Xalapa, Veracruz, Mexico.
Carretera antigua a Coatepec No. 351, El Haya, 91073.
esau.bojorquez@inecol.mx, eliel.ruiz@inecol.mx

The proteomic analysis of samples such as saliva or nasopharyngeal exudates implies a series of technical problems that obstruct the proteomic workflow. The most relevant challenging fact for this type of biological samples is the presence of hyperglycosylated proteins, mainly mucins, a major component of mucus and mucous membranes. The abundant high molecular weight glycan moieties reduce the performance of processing steps such as tryptic hydrolysis and chromatographic separations; in the case of the latter, we noticed “poisoning” of the reverse phase nanoscale analytical columns by highly glycosylated peptides, due to their extremely polarized amphipathic nature that difficult being eluted, and causing permanent damage to the chromatographic system, then the disruption of the analysis processes and increase in operating costs. To solve these problems, we propose an alternative method for the chemical remotion of glycosylations through breaking ether glycosidic bonds by using Trifluoromethanesulfonic acid (Triflic acid, TFMS), allowing the integrity of the polypeptide chains and the conservation of the innermost N-linked GlcNAc bound by an amide bond to asparagine residues. Salivary proteins were treated with additives like acetone, saturated phenol, or sodium dodecyl sulphate before the Triflic acid reaction, and remotion of glycosylations was monitored by mass shift in SDS-PAGE and by Periodic acid–Schiff staining. Control and TFMS-treated samples were subjected to a standardized proteomic workflow that includes reduction, alkylation, tryptic hydrolysis, and fractionation by high pH reversed-phase chromatography before LC-MS/MS. The RAW MS data were analyzed by four search engines (MaxQuant, MS AMANDA, MSFragger, and SEQUEST-HT) allowing the identification of 978 unique proteins. The search of HexNAc as dynamic modification showed an increment in the identification of glycosylation sites in the TFMS-treated samples in the conserved domain Asn-X-S/T, MSFragger and SEQUEST-HT algorithms had the best performance for glycopeptides identification.

DEVELOPMENT OF A TOOL FOR THE ASSESSMENT OF CRITICAL THINKING IN MEDICAL STUDENTS

Oliva Briz Tena, Juan Pablo Pardo-Vázquez, Teresa Imelda Fortoul Van der Goes,
Nancy Fabiola Pérez Herrera, Deyamira Matuz Mares

Universidad Nacional Autónoma de México
Pallares y Portillo 199, Col. Barrio San Lucas, Coyoacán, CDMX, 04040
5544724715; olibt.unam@gmail.com

Introduction. Critical thinking promotes rational autonomy, intellectual freedom, and evidence-based investigation for problem-solving. The lack of critical thinking has been linked to medical errors, which in some countries represent the third leading cause of death. Therefore, it is essential to have evidence that students develop this competency from the early years of their undergraduate studies.

Objective. This study aimed to design a tool for the assessment of critical thinking in medical students that provides validity evidence.

Method. A developmental, quantitative, descriptive, cross-sectional study was conducted. An evaluation tool was designed using the method of case-based clinical reasoning tests (CBCRT).

Participants. included professors of Biochemistry and Molecular Biology, General Practitioners, Family Physicians, Emergency Physicians, Internists, Nephrologists, and first-year medical students.

Results. A tool with 24 items was created, and psychometric analysis was conducted based on Classical Test Theory. A Cronbach's alpha of 0.43 was obtained, attributed to a small sample size and consequently high variance since it was the first time students were responding to this type of tool. Seven items did not meet the established parameters and will be reviewed again by the corresponding committees.

Conclusion. The tool received good acceptance from the students, and the time required to complete it was less than that needed for traditional exams. Additionally, the tool can be used remotely through a simple platform like Google Forms, and it is possible to identify the basic knowledge that needs to be reinforced for solving clinical cases.

Keywords. Critical thinking, assessment tools, validity evidence.

RELATIONSHIP BETWEEN THE ENZYME GLUCOSE-6-PHOSPHATE DEHYDROGENASE (ZWF1) AND HYDROGEN SULFIDE PRODUCTION IN THE YEAST SACCHAROMYCES CEREVISIAE

María Jatziry Camacho Campos, Elías Nieto Zaragoza, Francisco Torres-Quiroz

Instituto de Fisiología Celular. Departamento de Bioquímica y biología estructural.

Universidad Nacional Autónoma de México, Ciudad de México.

Circuito. Exterior s/n, C.U., Coyoacán, 04510 Ciudad de México, CDMX

55 562573 ftq@ifc.unam.mx

Hydrogen sulfide (H_2S) is a colorless gas that becomes toxic at high concentrations. It is permeable to membranes, and there are multiple physiological processes in which it regulates the cell; two of them are oxidative stress and alcoholic fermentation. The cell produces H_2S through two distinct pathways: the sulfur fixation pathway and transsulfuration, where the CYS4-encoded cystathionine beta synthase synthesizes cystathionine.

The sulfur binding pathway requires four molecules of NADPH. This molecule functions as an electron carrier and can capture free radicals involved in oxidative stress. The main source of NADPH in the cell is the pentose phosphate pathway (PPP). PPP has multiple branches, but we will focus on NADPH production. The first enzyme of the PPP is glucose-6-phosphate dehydrogenase, which is encoded by *ZWF1*. The Zwf1 protein catalyzes the reaction between glucose-6-phosphate, which comes from glycolysis, and D-6-phosphogluconoleacetone, resulting in NADPH.

To study the relationship between *ZWF1* and H_2S , we first confirmed the strain lacking *ZWF1* ($\Delta zwf1$). Next, we generated the $\Delta cys4\Delta zwf1$ strain. Next, we induced oxidative stress with hydrogen peroxide and found that $\Delta zwf1$ is highly sensitive to 3 mM H_2O_2 . On the other hand, $\Delta zwf1\Delta cys4$ recovered the growth phenotype when the hydrogen peroxide concentration was increased with respect to $\Delta zwf1$. Then, we grew the *WT*, $\Delta zwf1$, $\Delta cys4$, and $\Delta cys4\Delta zwf1$ strains in YPD medium and observed that $\Delta zwf1$ grows faster than *WT*, but $\Delta cys4$ and $\Delta cys4\Delta zwf1$ grew slowly, confirming a genetic interaction between both genes. After that, we measured H_2S production with a methylene blue assay. We observed that $\Delta zwf1$ produces less H_2S compared to the wild type. We performed a methionine auxotrophy test and observed that *zwf1* is not methionine auxotrophic. Finally, the respiratory and fermentative metabolisms of the strains of interest were also evaluated.

We conclude that $\Delta zwf1$ deregulates hydrogen sulfide production. The production of NADPH and hydrogen sulfide appears to have dichotomous behavior. Based on the above information, *ZWF1* plays a crucial role in regulating physiological processes in cells where H_2S is involved.

This project was financed by UNAM-DGAPA-PAPIIT through the research program IN208922 and by the Consejo Nacional de Humanidades, Ciencias y Tecnologías through the Ciencia de Fronteras 2023 program CF-2023-I884.

ROLE OF THE *KLNrg1* AND *KLRtg3* PROTEINS FROM *K. LACTIS* IN GLUCOSE REPRESSION, RETROGRADE RESPONSE AND MITOCHONDRIAL DNA INTEGRITY

Carlos Campero-Basaldúa¹, Karina Robledo-Márquez², Lina R. Riego-Ruiz², Edgar Ramírez¹, Yael González Tinoco¹ and Alicia Gonzalez-Manjarrez¹

Instituto de Fisiología Celular, UNAM¹. Cto. Exterior s/n, C.U., Coyoacán, 04510 Ciudad de México, CDMX, División de Biología Molecular, Instituto Potosino de Investigación Científica y Tecnológica (IPICYT), San Luis Potosí, SLP, México².
E-mail: jbasaldua@ifc.unam.mx

In *Saccharomyces cerevisiae*, *ScNrg1* and *ScRtg3* respectively mediate glucose repression and signaling from the mitochondria to the nucleus. We have also shown that *ScNrg1* and *ScRtg3* constitute a novel hybrid transcriptional regulator, which determines mitochondrial DNA integrity. This work considers the construction of the appropriate tagged strains to carry out *K/Nrg1* and *K/Rtg3* co-immunoprecipitation analysis and determine formation of the *K/Nrg1-K/Rtg3* hybrid in *K. lactis*. We are also, addressing the individual roles of *K/Nrg1* and *K/Rtg3*, which have not been analyzed in *K. lactis*. We are working on qPCR analysis to determine whether as well as in *S. cerevisiae* the orthologous *K/Nrg1*, works as transcriptional repressor, preventing expression of genes involved in the utilization of alternative carbon sources in the presence of glucose. *ScNrg1* regulates a variety of stress responses, some of the *Nrg1* target genes are *SUC2* (extracellular invertase) *RSB1* (lipid translocase) *DIT1* (spore wall maturation), and *SGA1* (glucoamylase;). We have identified the corresponding *K. lactis* orthologues for these genes, and will determine the regulatory role of *K/Nrg1* on these four genes expression. To analyze the role of *K/Nrg1* on the retrograde response we have identified the *ScCIT2* (citrate syntgase) *K. lactis* orthologous gene, which is known to be positively regulated by *ScRtg3*. This will allow us to determine whether the *K. lactis* *K/Nrg1* and *K/Rtg3*, which are *S. cerevisiae* orthologous genes, play a similar physiological role to that found in *S. cerevisiae*.

ScNRG1 and *ScNRG2* are paralogous genes, regulating of various stress responses. *ScNrg1* neofunctionalization has allowed its interaction with *ScRtg3* eliciting formation of the *ScNrg1-ScRtg3* hybrid, which is not formed between *ScNrg2* and *ScRtg3*. One of the questions we are addressing is whether *K/Nrg1* has also subfunctionalized and can form heterodimers with *ScRtg3* and with *K/Rtg3*.

Thus, our study addresses the regulatory role of *K/Nrg1* and *K/Rtg3* as independent modulators, and formation of the *K/Nrg1-K/Rtg3* hybrid.

THE MOONLIGHT PROTEIN ALT1

Yamile Paredes-Chiquini¹, Honza Ameli Cazarin-Mendoza¹, Eduardo Cardona-Guerrero¹,
Jose Carlos Campero-Basaldua¹ and Alicia Gonzalez-Manjarrez¹

¹Instituto de Fisiología Celular, Universidad Nacional Autónoma de México.
Circuito Exterior s/n Ciudad Universitaria, Coyoacán, 04510 México D.F.
E-mail: amanjarr@ifc.unam.mx

Paralogous genes are duplicated genes in the same organism that evolved from either a partial or total duplication of the genome. The study of paralogous genes in the yeast *Saccharomyces cerevisiae* showed that the retention and functional diversification of duplicated genes played an important role in the acquisition of facultative metabolism.

ALT1 and *ALT2* are nuclear paralogous genes that codify for Alt1 and Alt2 proteins in *Saccharomyces cerevisiae*. Alt1 is a mitochondrial alanine aminotransferase which degrades alanine to pyruvate using pyridoxal phosphate as a cofactor. Alt1 and Alt2 have a 65% aminoacidic identity, despite this Alt2 is not an aminotransferase and its function still undetermined. Alt1 carries out transamination in a catabolic and biosynthetic way in *S. cerevisiae*. It has been reported that an *alt1Δ* mutant in glucose and ammonia (biosynthetic media) shows a reduced growth rate compared to a wild type strain, this phenotype confirmed the existence of alternative pathways to synthesize alanine. Whereas in glucose and alanine (catabolic media) the *alt1Δ* mutant is incapable of growing, indicating that utilization of alanine as nitrogen source is exclusively carried out by the Alt1 enzyme.

In previous studies we reported that an *alt1Δ* mutant shows a petit phenotype incapable of growing in ethanol as unique carbon source due to a decrease in the expression of mitochondrial genes involved in oxidative phosphorylation. We also found that the mtDNA vs nDNA ratio is diminished in the *alt1Δ* mutant compared to a wild type strain. Taken together, these results suggest that Alt1 is a moonlight protein with a structural function regulating the expression and integrity of mitochondrial genes.

We suggest that Alt1, plays a role in the mitochondria, forming part of the mitochondrial system that organizes the mitochondrial-DNA nucleoids. To test this hypothesis, we propose to relocate Alt1 from de mitochondria to the cytosol by removing the mitochondrial localization peptide and to transform an Alt1 catalytic site mutant allele in an *alt1Δ* mutant strain. The phenotypes of these modified strains, will be tested by the expression of mitochondrial genes, the mtDNA vs nDNA ratio and growth rate analysis.

HEPATOPROTECTIVE EFFECT OF BACTERIAL CYCLODIPEPTIDES IN OBESITY RATS MODEL

Angela Regina Castillo Jimenez¹, Citlali Figueroa Guzmán¹, Lorena Martínez Alcantar¹, Jesús Campos García¹

¹Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo.
F. J. Mújica s/n, Edificio U3, Ciudad Universitaria, Morelia, Michoacán, México, C.P. 58030
E-mail: lorena.martinez@umich.mx

The liver is an organ crucial in energy metabolism; it performs a key role in maintaining glucose and lipid homeostasis and energy balance (e.g., *de novo* lipogenesis, fatty acid uptake, fatty acid oxidation, and triacylglycerol uptake). An imbalance between these processes may result in abnormal hepatic lipid accumulation, commonly called non-alcoholic fatty liver disease (NAFLD) (1). NAFLD occurs once the adipose tissue and liver exceed its capacity to store lipids (2). Cyclodipeptides (CDPs) are organic compounds produced mainly through bacteria. They have attracted research attention for potential new drug discovery; they have been studied for their antidiabetic, antibacterial, and anticancer activities (3). In this work, we analyzed the activity of bacterial CDPs in a model of obesity induced by a high-fat diet designed for this experiment using rats of the Wistar strain. The experimental period lasted 37 weeks, and dietary obesity was diagnosed using anthropometric methods such as body mass, Lee's index, body mass index, thoracic and abdominal circumference, and weight gain percentage. From week 25, the rats were treated for 10 weeks with CDPs administered intraperitoneally every 3 or 6 days, depending on the group. The results indicated that the bacterial CDPs alleviate liver fat accumulation and liver damage and increase cellular function markers (AST and ALT). Hepatic aquaporin expression levels in liver tissue were determined by immunodetection. The findings revealed that protein expression levels of the AQP1, AQP5, AQP8, and AQP9 were significantly decreased in the liver of obese rats compared with the control rats fed with a regular diet. Notably, in the liver of obese CDPs-treated groups, the protein expression levels of AQP1, AQP5, AQP8, and AQP9 significantly reverted to similar levels of the regular diet group. The findings revealed that the expression levels of aquaporins as obesity markers significantly decreased their expression in the liver of the obese animals treated with the bacterial CDPs, showing their anti-obesogenic potential.

References

1. Nguyen, P. et al. (2008). Liver lipid metabolism. *Journal of Animal Physiology and Animal Nutrition*, 92(3), 272-283.
2. Seebacher, F., et al. (2020). Hepatic lipid droplet homeostasis and fatty liver disease. *In Seminars in Cell & Developmental Biology*. 108:72-81.
3. Prasad, C. (1995). Bioactive cyclic dipeptides. *Peptides*, 16(1), 151-164.

PHLOEM EXUDATE PROTEINS OF GRAFTED AND NON-GRAFTED BEANS (*PHASEOLUS SP.*) CULTIVATED UNDER IRRIGATION AND WATER RESTRICTION DURING GRAIN FILLING

Martha Itsel Cayetano-Marcial¹, Daniel Padilla-Chacón^{2*}, Amelia López-Herrera¹, Cecilia Beatriz Peña Valdivia¹, Ebandro Uscanga-Mortera¹, Antonio García-Esteva¹

¹Colegio de Postgraduados, Campus Montecillo, Km. 36.5 Carretera. México-Texcoco, Montecillo 56264, Texcoco, Estado de México. ²Consejo Nacional de Humanidades, Ciencias y Tecnologías (CONAHCYT). Programa IIXM del CONAHCYT. Campus Montecillo, Km. 36.5 Carretera. México-Texcoco, Montecillo 56264, Texcoco, Estado de México.

*Autor de correspondencia: danielpadilla30@yahoo.com.mx

Abstract. The common bean (*Phaseolus vulgaris* L.) constitutes a major staple crop in Latin American countries and North Africa; however, the major limitation is drought which causes more than 80% of the annual losses. To obtain cultivars with greater tolerance to drought, breeding programs attempt to generate intraspecific and interspecific crosses but the process is low and requires prolonged periods. Grafting is an alternative technique with results in the short term. Here, we investigated the protein content of phloem sap exudates by SDS-PAGE and Shotgun proteomics and explored how this reflects in physiological responses to drought in ungrafted and grafted bean plants among *P. vulgaris* cv. Pinto Saltillo (scion) and with *P. acutifolius* cv. Tepary café (rootstock) subjected in irrigation with 100% field capacity (FC) and moisture restriction at 50% of FC for 4 days. Initially, we found that water restriction has significant stimulation on stomatal conductance (*g_s*) in grafting plants. The exudate sap from grafting plants show a protein profile by SDS-PAGE two principal bands with molecular weight of 50 KDa and 25 KDa. The shotgun analysis searched against UniProt's legume database identified a GDSL esterase/lipases protein was identified in the exudate of grafted plants under water restriction. In addition, subcategories and pathways annotated with Gene Ontology (GO) classification and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analysis were analyzed and through protein-protein interaction (PPI) network analysis we found that other proteins participated in drought responses in the phloem. Over all, this study offers an imperative role in plant development and stress tolerance and could be considered as a potential marker of events mediating drought response.

A BIOCOMPUTATIONAL AND SITE-DIRECTED MUTAGENESIS APPROACH TO RECOVER THE ACID PHOSPHATASE/PHYTASE ACTIVITY OF EHHAPP49, AN ATYPICAL AMOEBIC ENZYME

Guadalupe A. Chávez-Melendrez¹, Patricia L.A. Muñoz-Muñoz¹, Samuel G. Meléndez-López¹, Pablo A. Madero-Ayala¹, Marco A. Ramos-Ibarra¹, Rosa E. Mares-Alejandre¹

¹Facultad de Ciencias Químicas e Ingeniería, Universidad Autónoma de Baja California, Tijuana, BC 22390, México.
E-mail: a1246332@uabc.edu.mx; rmares@uabc.edu.mx.

The histidine phosphatase (HP) superfamily is a diverse and large group of enzymes that share the conserved catalytic loop RHxxRxP, in which the His residue is phosphorylated during protein function.¹ This superfamily comprises two major classes: branch-1 collects a variety of enzymes, including fructose-2,6-bisphosphatases and phosphoglycerate mutases, and branch-2 comprises acid phosphatases and phytases.¹ Currently, several HP enzymes are biomolecules with medical and biotechnological applications. Most organisms encode a wide collection of phosphatases, including the protozoan parasite *Entamoeba histolytica* (the causative agent of amoebiasis in humans). A comprehensive in-silico analysis revealed 250 non-redundant phosphatases, including 19 belonging to the HP superfamily.² In particular, a protein encoded by the EHI_146950 gene, named EhHAPP49, caught our attention because of the lack of information about its precise function. Initial biochemical studies (using a recombinant protein) demonstrated that EhHAPP49 is an atypical branch-2 HP phosphatase that lacks acid phosphatase/phytase activity but exhibits alkaline pyrophosphatase activity.³ In this study, a computational-experimental approach was used to generate site-specific mutants of EhHAPP49, aimed at expanding the substrate-binding pocket conformation, thus recovering the HP enzyme function. Based on the current results, a single mutation appears to increase pyrophosphatase activity, suggesting that the pocket entry is broader and enhances substrate diffusion. Additional data indicated that certain mutations may restore phosphatase activity to some extent. Thus far, our findings support the hypothesis that active site entrance plays a decisive role in the substrate selectivity of EhHAPP49 by acting as a molecular sieve.

References

1. Rigden DJ (2008). *Biochemical Journal*, 409(2), 333–348.
2. Anwar T, Gourinath S (2013). *PLoS ONE*, 8(11), e78714.
3. Terán-Ramírez C, Mares-Alejandre RE, Estrada-González AL, Muñoz-Muñoz PLA, Ramos-Ibarra MA (2021). *International Journal of Molecular Sciences*, 22(4), 2164.

FUMARATE REDUCTASE IN RHODOTORULA MUCILAGINOSA AND ITS POSSIBLE PHYSIOLOGICAL ROLES

Natalia Chiquete-Félix¹, Paulina Castañeda-Tamez¹, Méndez-Romero Ofelia¹, Carolina Ricardez-García¹,
Natalia Copitin-Niconova¹, Paola Acosta-Valdelamar¹
and Salvador Uribe-Carvajal¹

¹Depto. De Genética Molecular, Instituto de Fisiología Celular, Universidad Nacional
Autónoma de México
E-mail: nchiquete@ifc.unam.mx

Under anaerobic conditions, different organisms express an electron transport chain that uses different molecules as final electron acceptors. Some of these are trimethylamine oxide, sulphur compounds, nitrate and fumarate. Fumarate reductase (FRed) is a key component of anaerobic respiration which catalyzes fumarate reduction. Thus, in absence of oxygen, fumarate can be used as a final acceptor in bacteria such as *Helicobacter pylori* and *Escherichia coli*, in protozoa such as *Trypanosoma*, *Plasmodium*, and *Leishmania*, and in helminths including *C. elegans*. In eukaryotes, cytosolic FRed maintains the redox balance in the cell when oxygen is deficient, as it oxidizes flavin adenine dinucleotide (FADH₂) and nicotinamide adenine dinucleotide (NADH) (Kim et al., 2018). The extremophile yeast *Rhodotorula mucilaginosa* may be found in environments such as ice glaciers, heavily contaminated waters, and soils, where oxygen concentrations vary widely. *Rhodotorula* has a branched respiratory chain (RC) expressing some alternative NADH dehydrogenases type II (NDH2) and an alternative oxidase (AOX) (Castañeda-Tamez P et al., 2024). In isolated and solubilized mitochondria from *R. mucilaginosa*, FRed activity was detected by clear-activity PAGE, and its presence was confirmed by mass spectroscopy. When *R. mucilaginosa* was subjected to hypoxia or anaerobiosis, a decrease in cell growth and oxygen consumption was observed. Here, FRed activity was likely needed to control the redox balance in the cell, helping survival in extreme environments.

References

1. Kim S, Kim CM, Son YJ, Choi JY, Siegenthaler RK, Lee Y, Jang TH, Song J, Kang H, Kaiser CA, Park HH (2018). *Nat Commun.* 19;9(1):4867.
2. Castañeda-Tamez P, Chiquete-Félix N, Uribe-Carvajal S, Cabrera-Orefice A (2024). *Biochim Biophys Acta Bioenerg.* 1;1865(2):149035.

INTROGRESSION OF DROUGHT TOLERANCE OF *PHASEOLUS ACUTIFOLIUS* A. GRAY TO *PHASEOLUS VULGARIS* L

Marcela Cilia García¹, Cecilia Beatriz Peña Valdivia¹, José Cruz Jiménez Galindo², Antonio García Esteva¹, Ebandro Uscanga-Mortera¹, Daniel Padilla-Chacón³

¹Posgrado de Botánica, Campus Montecillo, Colegio de Postgraduados, 56230, Montecillo, Texcoco, Estado de México.

²Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias, 31500, Cd. Cuauhtémoc, Chihuahua.

³Facultad de Ciencias Químicas, Universidad Veracruzana, 94340; Orizaba, Veracruz.

The common bean (*Phaseolus vulgaris* L.) is the most consumed legume in the world. In Mexico, its production is mainly produced in erratic rainfed environments where drought causes low grain yields. The tepary bean (*Phaseolus acutifolius* A. Gray) is a desirable genetic resource for incorporation of improved resistance into common bean (*P. vulgaris* L.). Among the strategies to obtain drought-resistant bean crops, the transfer of adaptive traits through interspecific crosses has increased in recent years. The objective of this work is to identify morphological, molecular and biochemical markers of drought resistance in a bean population obtained by interspecific crossing between *P. vulgaris* (susceptible) and *P. acutifolius* (tolerant). The crossing was carried out by emasculation, obtaining an F1 seed and segregating it to the F4 generation with 48 independent lines. The germination capacity was evaluated with low water potential simulated with polyethylene glycol (PEG-6000) at -0.6 MPa and contrasting lines were chosen in the development of the root and hypocotyl, with the purpose of correlating these data with allelic sequences that indicate tolerance to drought, genotyping analysis was carried out by diversity matrix technology (DARtseq) in eight lines and two parents. The results showed that 154 single nucleotide polymorphisms (SNP) were obtained with a polymorphism information content (PIC) greater than 0.468. In addition, the concentration of soluble sugars, starch, protein, amylose/amylopectin ratio and some minerals were quantified. The protein percentage was similar in the lines and parents ($\approx 25\%$), in the same way in the fiber content ($\sim 5\%$), ash ($\sim 4\%$), zinc (~ 25 mg/kg). Regarding magnesium concentration, line 44_1 showed higher levels compared to the other lines, but similar to the parents (150 mg/kg). Furthermore, the results revealed that line 44_1 showed higher concentrations of glucose and fructose content ($\sim 100\%$) compared to the parents and the other lines. Line 44_1 also presented higher levels ($\sim 80\%$) compared to the other lines and a 20% iron concentration compared to its parents ($p \leq 0.05$). These findings allow us to identify markers that may be key to developing varieties that have drought tolerance traits in common bean genetic improvement programs.

Keywords: bean, drought tolerance, climate change, DARtseq, SNPs, soluble sugars, iron.

ρ BR-2-APB IS AN INEFFICIENT ENDOPLASMIC RETICULUM CALCIUM RELEASING AGENT

M. Sc. Rodrigo Contreras-Gaytán¹ & PhD. Agustín Guerrero-Hernández¹

¹Department of Biochemistry, CINVESTAV-Zacatenco, Av. Instituto Politécnico Nacional 2508, Col. San Pedro Zacatenco, del. Gustavo A. Madero, CP: 07360
rodrigo.contreras@cinvestav.mx

A plethora of cellular processes are regulated by transient elevations of cytosolic calcium (Ca^{2+}) concentration ($[\text{Ca}^{2+}]_i$). Cell $[\text{Ca}^{2+}]_i$ responses involve an extensive signaling toolkit comprising ion channels, calcium-binding proteins, and pumps¹.

In non-excitabile cells, the inositol 1,4,5-trisphosphate receptor (IP_3R) is the principal ion channel, increasing the $[\text{Ca}^{2+}]_i$. Agonists of PIP_2 -specific PLC-coupled GPCRs transiently increase the $[\text{Ca}^{2+}]_i$ by generating IP_3 and allowing Ca^{2+} release from the endoplasmic reticulum (ER) through the IP_3Rs ².

The ER is the major intracellular Ca^{2+} store, and cellular events that induce ER store depletion, like activation of IP_3R , trigger a refilling process known as store-operated Ca^{2+} entry (SOCE), which involves activation of plasma membrane Orai1 channels by STIM1, an ER membrane Ca-binding protein^{1,2}. 2-Aminoethyl diphenylborinate (2-APB) is a SOCE modulator since, at low, it increases while at high concentrations, inhibits SOCE. In previous studies, León-Aparicio *et al.* proved that 2-APB reduces the luminal ER Ca^{2+} concentration ($[\text{Ca}^{2+}]_{\text{ER}}$) by activating Orai3 channels³. In collaboration with Dr. Mancilla from Cinvestav's Chemistry dept, we synthesized ρ Br-2-APB, a more potent ER Ca^{2+} releasing agent than 2-APB.

In the presence of 1.8 mM extracellular Ca^{2+} , histamine (an IP_3 -producing agonist) and ρ Br-2-APB showed similar reduction of the $[\text{Ca}^{2+}]_{\text{ER}}$ as determined with G-CEPIA1er indicator but a completely different $[\text{Ca}^{2+}]_i$ increase since the cytoplasmic response induced by ρ Br-2-APB was only 20% of the one produced by histamine. However, dominant negative mutant of Orai channels inhibit 2-APB Ca^{2+} release but not the one induced by ρ Br-2-APB indicating that they have different targets to induced ER Ca^{2+} release. Since the G-CEPIA1er indicator uses calmodulin to detect the $[\text{Ca}^{2+}]_{\text{ER}}$, we verified that saturating concentrations of ρ Br-2-APB do not reduce the G-CEPIA1er signal.

ρ Br-2-APB induces ER Ca^{2+} release without the participation of IP_3Rs . In conclusion, IP_3R is an efficient ER Ca^{2+} -releasing channel, while the one activated by ρ Br-2-APB is not.

References:

1. Luan, S., & Wang, C. (2021). *Annual Review of Cell and Developmental Biology*, 37, 311-340.
2. Berridge, M. J. (2016). *Physiological reviews*, 96(4), 1261-1296.
3. León-Aparicio, D., *et al.*, (2017). *Cell Calcium*, 65, 91-101.

DPE2 ACTIVITY IN BEAN FRUIT PERICARP IS POST-TRANSLATIONALLY REGULATED

Gerardo Contreras-Ruiz¹, Lilia Angélica Bernal-Gracida¹ and J. Eleazar Martínez-Barajas¹

¹Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México: Av. Universidad N° 3000, Col. UNAM, Ciudad Universitaria, Alcaldía Coyoacán, Ciudad de México, C.P. 04510
Telefono: 5556225276 E-mail: emtz@unam.mx

The disproportionating enzyme 2 (DPE2 EC 2.4.1.25) is a 4- α -glucanotransferase that catalyses the transference of a glucose unit from maltose to a soluble polysaccharide. This activity is crucial in plants for the conversion of starch degradation products into hexose phosphates, used in various metabolic processes or in sucrose synthesis, which is exported throughout the plant. Arabidopsis plants with silenced DPE2 genes accumulate starch and maltose². In leaves the activity of DPE2 is increased during the night¹ as a consequence (at least in part) to the phosphorylation at serine 786². Bean fruit pericarp accumulates a significant amount of starch that is degraded during the accelerated growth stage of the seeds⁵. Starch degradation in the bean fruit pericarp is characterized by increased phosphorylation, improving utilization by β -amylase. The activities of the DPE2 and PHO2 enzymes, which convert starch degradation products into sucrose are also increased. The independence between DPE2 activity and gene expression suggests that the activity is post-translational regulated. *In silico* analysis indicates several possible post-translational modifications in DPE2: acetylation at M1, R219, and K916; ubiquitination at R227 and K358; and phosphorylation at S449 and T555. DPE2 tertiary structure was modelled and these modifications were located. In addition, protein extracts of bean fruit pericarp were fractionated on anion exchange chromatography and two peaks were identified. We observed significant differences in their kinetic parameters, suggesting the existence of two distinct states of the protein. The identification of the molecular bases of these differences will provide basis to understand how DPE2 activity is regulated in the bean fruit pericarp.

Acknowledgements. This research was supported by DGAPA (IN205323) and FQ (PAIP 5000-9127). GCR is a recipient of CONAHCYT scholarship.

References

1. Chia, T., Thorneycroft, D., Chapple, A., Messerli, G., Chen, J., Zeeman, S. C., & Smith, A. M. (2004). *The Plant Journal*, 37(6), 853-863.
2. Malinova, I., Mahlow, S., Alseekh, S., Orawetz, T., Fernie, A. R., Baumann, O., Steup, M., & Fettke, J. (2014). *Plant physiology*, 164(2), 907-921.
3. Lu, Y., Steichen, J. M., Yao, J., & Sharkey, T. D. (2006). *Plant physiology*, 142(3), 878-889.
4. Ruiz-Gayosso, A., Rodríguez-Cruz, I., Martínez-Barajas, E., & Coello, P. (2022). *Plant Physiology and Biochemistry*, 193, 70-77.
5. Belmont, R., Bernal, L., Padilla-Chacón, D., Coello, P., & Martínez-Barajas, E. (2022). *Plant Science*, 316, 111163.
6. Bernal, L., Luján-Soto, E., Fajardo-Hernández, C. A., Coello, P., Figueroa, M., & Martínez-Barajas, E. (2022). *Physiologia Plantarum*, 174(6), e13836.

CHARACTERIZATION OF SPHENOTOXIN FROM OPHRYACUS SPHENOPHRYS AND ITS COMPARATION WITH CLASSIC CROTOXIN

Tania Corkidi¹, Edgar Neri² and Alejandro Alagón³

¹Institute of Biotechnology, Av. Universidad 2001, Chamilpa, 62210 Cuernavaca, Mor.
E-mail: tania.corkidi@ibt.unam.mx

Sphenotoxin is a β -neurotoxin with phospholipase A2 (PLA2) activity found in the venom of the snake *Ophryacus sphenophrys*¹. It has one acidic subunit (A) that doesn't have catalytic activity and a basic subunit which has catalytic activity, just like in the case of the classic crotoxin (CB) present in *Crotalus durissus terrificus*'s venom (rattlesnake)². Even though the basic subunit of sphenotoxin has a high percent of identity in its amino acid sequence in relation to the subunit B of crotoxin (86 %), there are some differences concerning the recognizing and neutralizing capacities of commercial antivenoms, PLA2 activity and lethal potency. In this project, we identified 2 different isoforms of the basic subunit of sphenotoxin (B1 and B2) being B1 the isoform that has less lethal potency, more recognition by the two antivenoms tested, and more PLA2 activity in contrast with CB and B2. CB is 1.5 times more lethal than B2, and 3.6 times more lethal than B1. Also, B2 has 2.5 times more PLA2 activity than CB and B1 has 6 times more PLA2 activity than CB. Using two different Mexican antivenoms (Antivipmyn and BIRMEX), we saw that the recognition capacity of Antivipmyn has slightly different titles when comparing CB, B1 and B2, being B1 4 times less recognized and 7 times less recognized by BIRMEX comparing to the recognition capacity of CB. On the other hand, B2 is 7 times less recognized by Antivipmyn and 9 times less recognized by BIRMEX in contrast to the recognition capacity for CB. Using computerized models, we observed that the principal chain of amino acids of CB and the isoforms B1 and B2 does not have an obvious change in their tridimensional structure. However, certain changes of amino acids within the N-terminal and within the interface between the two subunits (A and B) could explain the subtle changes in PLA2 activity, lethal potency, and recognition of antivenoms. Certain changes in the lateral amino acid chains as in B1 (H1-D1 y W30-G30) could modify the interaction with A subunit. Also, the mayor number of changes are found along the N-terminal and C-Terminal, and these two parts are exposed to the solvent. With this information, we believe that these may explain the differences in recognition and neutralization by the antivenoms used.

References

1. Neri-Castro, E., Lomonte, B., Valdés, M., Ponce-López, R., Bénard-Valle, M., Borja, M. (2019). *Proteomics*, **192**, 196-207.
2. Faure, G., Bon, C. (1987). *Toxicon*, **25**, 229-234.

LACTATE DEHYDROGENASE AND ITS RELEVANCE IN CANCER BIOLOGY: REGULATION, ISOENZYMES, AND INHIBITION MECHANISMS

Carla Isabela Cuevas Bañuelos, Andrés Reyes Chaparro, Dulce Estefanía Nicolás Álvarez

National Polytechnic Institute, National School of Biological Sciences, México City (11350), Extension Manuel Carpio and Plan de Ayala, Col. Santo Tomás, Miguel Hidalgo.

E-mail: dnicolas@ipn.mx*

Lactate dehydrogenase (LDH) is a cytoplasmic enzyme present in all cells of all tissues, composed of four subunits of two different types of polypeptide monomers, referred to as H and M. Research on LDH is significant as tumors have survival advantages due to lactate secretion. LDH consists of two main types of polypeptide subunits, LDHA and LDHB. The LDHA gene is located on chromosome 11, while LDHB is on chromosome 12. Additionally, chromosomes 1, 2, 4, 9, and 10 contain sequences related to LDHA, and chromosomes X and 13 contain sequences related to LDHB. LDHA shows high expression levels in all tumor cells, while LDHB levels vary between different tumor types. This expression pattern may contribute to the divergent dynamics of lactate and oxidative capacities in tumor cells, as seen in MDA-MB-231 adenocarcinoma cells for LDHA and MCF-7 adenocarcinoma cells for LDHB. Both isoforms, LDHA and LDHB, are regulated by various mechanisms at the transcriptional, post-transcriptional, and post-translational levels. LDHA is regulated by factors such as FOXM1, c-Myc, hypoxia, and estrogens, while LDHB is regulated by mTORC1, STAT3, HMGB2, and miRNAs such as miR-375. Lactic acid confers invasive properties to tumor cells, affecting normal tissue structure. LDH also plays a crucial role in altering cancer cell metabolism, known as the Warburg effect, where cancer cells favor glucose fermentation to lactate even in the presence of sufficient oxygen. This phenomenon allows cancer cells to rapidly produce energy, essential for their growth and survival. The drug under study, gossypol, a polyphenolic diterpene found in cottonseed (*Gossypium* sp.), is used to treat metastatic endometrial carcinoma of the ovary. Gossypol inhibits LDH-C4 due to the presence of 4-isopropyl salicylaldehyde in its molecular structure. Gossypol anchors to the active site, with one end containing 4-isopropyl salicylaldehyde, while the other end blocks the coenzyme entry, explaining why gossypol competes with the coenzyme (NADH) for the same binding site on the LDH-C4 active site. It is hypothesized that only the 4-isopropyl salicylaldehyde parts of the gossypol molecule are responsible for inhibiting this isoenzyme.

References

1. Marcial-Reyes, D. B. (2022). Exploración a la EC. 1.1. 1.27 Lactato Deshidrogenasa.
2. Herrera-González, Norma Estela; Martínez-García, Felipe; Mejía-Jiménez, Emanuel El efecto Warburg: la mano derecha en el desarrollo del cáncer. *Revista de Especialidades Médico-Quirúrgicas*, vol. 20, núm. 2, abril-junio, 2015, pp. 171-177 Instituto de Seguridad y Servicios Sociales de los Trabajadores del Estado Mexico, México.
3. Urbańska, K., & Orzechowski, A. (2019). Unappreciated Role of LDHA and LDHB to Control Apoptosis and Autophagy in Tumor Cells. *International Journal Of Molecular Sciences*, 20(9), 2085

CHLOROTRACKER: A PHOTOACTIVABLE 1,8-NAPHTHALIMIDE DERIVATIVE AS A PROBE FOR CHLOROPLAST IMAGING

B. Areli Carranza Correa¹*, Karen M. Dahlman Cabrera¹*, Ixsoyen F. Vázquez Sandoval², Arturo Jiménez Sánchez² and Manuel Gutiérrez Aguilar¹

¹Department of Biochemistry, Facultad de Química, Universidad Nacional Autónoma de México, México City, 04510, México; ²Department of Organic Chemistry, Instituto de Química, Universidad Nacional Autónoma de México, México City, 04510, México
E-mail: manu@unam.mx

The development of subcellular compartment-specific dyes has significantly enhanced cellular biology tools. For example, potentiometric dyes, such as rhodamine derivatives, efficiently bind to mitochondria¹, while BODIPY sensors can mark lipid droplets within cells, bind to protein receptors, and are also used for evaluating the viscosity inside cells². In this study, we tested the targeting properties of “chlorotracker”—a 1,8-naphthalimide derivative—on isolated chloroplasts and leaf mesophyll cells from *Arabidopsis thaliana*. The results show that chlorotracker can efficiently label isolated chloroplasts. Additionally, we found that chlorotracker fluorescence progressively increases upon fluorochrome excitation, indicating that it is a photoactivable probe. Therefore, the results presented herein highlight the utilization of chlorotracker as a novel probe for chloroplast tracking and morphology assessment.

References

1. Desai S, Grefte S, van de Westerlo E, Lauwen S, Paters A, Prehn JHM, Gan Z, Keijzer J, Adjobo-Hermans MJW, Koopman WJH. Performance of TMRM and Mitotracker in mitochondrial morphofunctional analysis of primary human skin fibroblasts. *Biochim Biophys Acta Bioenerg.* 2024 Apr 1;1865(2):149027.
2. Jurgutis D, Jarockyte G, Poderys V, Dodonova-Vaitkuniene J, Tumkevicius S, Vysniauskas A, Rotomskis R, Karabanovas V. Exploring BODIPY-Based Sensor for Imaging of Intracellular Microviscosity in Human Breast Cancer Cells. *Int J Mol Sci.* 2022 May 19;23(10):5687.

ENGINEERING OF NANOCOMPARTMENTS BASED ON ENCAPSULIN A FROM MYXOCOCCUS XANTHUS FOR USE AS A PROTEIN RELEASE SYSTEM

Willy Angel Delgado Tapia¹, Ismael Bustos Jaimes¹

¹National Autonomous University of Mexico (UNAM), Department of Biochemistry, Faculty of Medicine, Mexico City 04510, Mexico
E-mail: angel.93feu@hotmail.com

Encapsulins (ENCs) have recently been used in biotechnology, biomedicine, and synthetic biology. One of the most studied has been *M. xanthus* because of its chemical and physical features. Here, we work with chimeric ENCs from *M. xanthus* to realize a nanocarrier capable of delivering the system CRISPR-Cas in the form of RNPs to approach the advantage of these particles to tolerate elevated temperature and extreme pH with a mechanism of targeting delivery.

So, we use the Spy Tag/ Spy Catcher system to give ENCs the ability with a peptide to enter a cancer human cell line. Also, in this work we used, the intrinsic capacity of the ENC to encapsulate the RNP Cas9 through the cargo-loading protein (CLP) that gives the ability to any protein that gets this short peptide to go inside of it, of course depending on the steric hindrance. In consequence, we did the co-expression of the protein EncA, which is in charge of building an icosahedral nanoparticle throughout 180 protomers by the assembly of itself, and RNP Cas9 from *S. pyogenes* was carried out recombinant in *E. coli*. The RNP was synthesized in pET 28b plasmid, modifying the sequence of them adding an NLS to allow easy entry into the cell and the CLP to take the protein for interacting with the inner surface of the ENC. Also, together with the RNAsg targeting to NSD1 gene overexpressed in liver cells, NSD1 is a protein from histone lysine-methyltransferases that has been associated with invasion, migration, and proliferative capabilities, so we use this gene to prove if our system based on ENCs with the RNP Cas9/RNAsg NSD1 inside, decorated with a short peptide denominated , for targeting these nanoparticles to a HepG2 cellular line, it was efficient to deliver the charge inside of the desired line cell and provoke proliferation changes and INDELS in the NSD1 gene. However, one problem was the steric hindrance regarding the RNP Cas9 has 1397 amino acid residues being a fairly large protein to encapsulate more than three. It might bring problems for the correct folding of the particle. Then, we used a miniature RNP Cas to avoid the steric hindrance problem, promote the correct folding of the ENCs, and house a greater number of RNPs because this protein has 565 amino acid residues, which is less than half of Cas9.

According to this, we purify the ENCs with Cas9 or CasMINI RNAsg NSD1 produced in *E. coli* by IMAC, then we analyzed with different techniques such as Dynamic Light Scattering (DLS), Transmission Electron Microscopy (TEM), SDS-PAGE to verify the adequate folding and the charge inside of them as well as their activity of the RNPs alone and inside of ENCs.

References

1. Xu X, Chemparathy A, Zeng L, Kempton HR, Shang S, Nakamura M, Qi LS. (2021). Engineered miniature CRISPR-Cas system for mammalian genome regulation and editing. *Mol Cell*. 81(20):4333-4345.

EVALUATION OF THE ANTIFUNGAL EFFECT OF THE EXTRACT OF MORINGA OLEIFERA ON ASPERGILLUS PARASITICUS

Edmar de Jesus Díaz-García¹, María del Socorro Pina Canseco², Marco Antonio Sánchez Medina³, Alma Dolores Pérez Santiago³

¹CONAHCYT-Universidad Autónoma Benito Juárez de Oaxaca, Carretera Antigua a San Felipe del Agua S/N. Oaxaca. C.P. 68020. ²Universidad Autónoma Benito Juárez de Oaxaca, Carretera Antigua a San Felipe del Agua S/N. Oaxaca. C.P. 68020. ³Tecnológico Nacional de México/ Instituto Tecnológico de Oaxaca, Av. Víctor Bravo Ahuja No. 125 Esq. Calz. Tecnológico, Oaxaca, Oax. C.P. 68030. E-mail: edg.89@live.com.mx

Aspergillus flavus and *Aspergillus parasiticus* are fungi that produce secondary metabolites known as aflatoxins, highly carcinogenic, teratogenic and mutagenic compounds, causing great damage to the health of animals and humans, as well as great economic losses worldwide. Currently, inhibitors of *A. parasiticus* development and aflatoxin production are being investigated (Caceres et al, 2020; EL-Sayed et al, 2022; Martinez 2023). On the other hand, it has been reported that some plant extracts can modify the normal development of fungi of the *Aspergillus* genus (Oladeji et al, 2020). Therefore, the objective of this work is to evaluate the interaction of Moringa oleifera extract with *A. parasiticus*.

The crude extract prepared from *Moringa oleifera* (*M. oleifera*) and ethanol was characterized by Liquid Chromatography coupled to Mass Spectrometry, and total polyphenols were measured. The effect of the *M. oleifera* extract was a decrease in biomass. produced based on the control used, which was dimethyl sulfoxide (DMSO). The percentages of biomass decrease were not significant for the concentrations of 100, 90 and 80 mg of *M. oleifera* extract /mL of DMSO, with respect to the control without treatment. Microscopy of the extract interacting with the *A. parasiticus* strain was also performed, observing that the spores treated with the extract presented morphological modifications, which may involve damage to the cell wall of *A. parasiticus*. Likewise, antifungal activity tests were carried out with inhibition zone techniques and the viability of *A. parasiticus* spores using colony forming units (CFU), finding that there is no effect on the *Aspergillus parasiticus* strain.

References

1. Caceres, I., Khoury, A. A., Khoury, R. E., Lorber, S., Oswald, I. P., Khoury, A. E., & Bailly, J. D. (2020). Aflatoxin biosynthesis and genetic regulation: a review. *Toxins*
2. El-Sayed, R. A., Jebur, A. B., Kang, W., & El-Demerdash, F. M. (2022). An overview on the major mycotoxins in food products: Characteristics, toxicity, and analysis. *Journal of Future Foods*, 2(2), 91-102.
3. Martínez Miranda, M. M. (2023). *Evaluación de riesgo de aflatoxinas en arroz*. Editorial Universidad de Caldas.
4. Oladeji, O. S., Odelade, K. A., & Oloke, J. K. (2020). Phytochemical screening and antimicrobial investigation of Moringa oleifera leaf extracts. *African Journal of Science, Technology, Innovation and Development*

MCT11 TRANSPORTER OVEREXPRESSION IN VITRO LEADS TO LIPID ACCUMULATION

Natalí Domínguez-Velázquez¹, Julio García Cordero², Gustavo G. Medina-Mendoza¹, Jocelyn C. Pérez-Lara², Gabriela R. Luna-Palencia¹, Leticia Cedillo Barrón², Viridiana Olin Sandoval¹

¹Departamento de Biotecnología y Bioingeniería, Centro de Investigación y de Estudios Avanzados, del Instituto Politécnico Nacional, Ciudad de México, México. ²Departamento de Biomedicina Molecular, Centro de Investigación y de Estudios Avanzados, del Instituto Politécnico Nacional, Ciudad de México, México.
E-mail: viridiana.olin@cinvestav.mx

The MCT11 transporter is encoded by the *slc16a11* gene. MCT11 is a member of the monocarboxylate transporter family SLC16A. It has been reported that almost all the transporters of the SLC16A family are located in the plasma membrane, and that some of them require chaperones like basigin or embigin for their localization¹.

Recently, single nucleotide polymorphisms in the coding region of MCT11 have been associated with the development of type 2 diabetes in Mexican and Latin American people². MCT11 is mainly expressed in liver, and changes in its expression promotes alterations in lipid metabolism^{2,3}. However, the mechanisms involved in this process are unknown. Therefore, we designed a model of MCT11 overexpression in HEK293 cells, fused in the N-terminal to the green fluorescent protein (eGFP), allowing us to evaluate the subcellular localization and function of the transporter.

The expression of MCT11 was indirectly estimated by measuring the levels of green fluorescent protein (GFP) via flow cytometry, and the overexpression was confirmed by measuring the mRNA levels of the *slc16a11* gene.

In order to determine its cellular localization, we performed immunofluorescence assays where we determine that MCT11-eGFP is located in endoplasmic reticulum, mitochondria and plasma membrane. Moreover, we have determined by Oil Red O staining, that the MCT11 overexpression, promotes the accumulation of intracellular lipids. We are in the process of identifying them by gas chromatography.

Thus, our results suggest that the MCT11 transporter is localized in the endoplasmic reticulum, mitochondria, and in the plasma membrane, and that its associated to the accumulation of lipids.

References

1. Felmllee, M. A., Jones, R. S., Rodriguez-Cruz, V., Follman, K. E. & Morris, M. E. Monocarboxylate transporters (SLC16): Function, regulation, and role in health and disease. *Pharmacological Reviews* **72**, 466–485 (2020).
2. Williams Amy, A. L. *et al.* Sequence variants in SLC16A11 are a common risk factor for type 2 diabetes in Mexico. *Nature* **506**, 97–101 (2014).
3. Rusu, V. *et al.* Type 2 Diabetes Variants Disrupt Function of SLC16A11 through Two Distinct Mechanisms. *Cell* **170**, 199-212.e20 (2017).

EVALUATION OF THE CHANGES IN CARBON FIXATION AND CARBOHYDRATE CONTENT BETWEEN DIVERSE BREAD WHEAT GENOTYPES TOLERANT AND SENSITIVE TO HEAT STRESS

Ana L. Encinas-Montes¹, Andrea Romero-Reyes¹, Sergio G. Hernandez-Leon^{1*}

¹Centro de Investigación en Alimentación y Desarrollo, A.C. Carretera G.E. Astiazarán Rosas No. 46, Col. La Victoria, C.P. 83304, Hermosillo, Sonora, México.

Corresponding: sergio.hernandez@ciad.mx, (+52) 662 289-2400 ext 509 and 518

Wheat (*Triticum aestivum* L.) has become an important staple food in the world, currently ranking third in cereal production worldwide. Agriculture faces a major problem, including abiotic stress in plants, especially heat stress has become one of the main causes of crop losses worldwide, significantly affecting production and yield. Heat stress (HS) affects growth and development stages, leading to morphological, physiological, and biochemical alterations. Climate change makes this situation worse, as high temperatures have an adverse impact on wheat cultivation, which is sensitive to heat stress due to its nature as a cool-season crop. Therefore, it led us to search for strategies to find genotypes tolerant to climate change that may contribute to wheat production in such temperature increasing conditions. Herein, two experiments were performed: control and HS during the vegetative stage with four bread wheat genotypes (two sensitive and two tolerant to HS which were previously classified according to grain yield reduction) in a greenhouse at the Food and Development Research Center (CIAD) in Hermosillo, Sonora, Mexico. It was observed that genotypes 6 (tolerant), 18 (sensitive) and 26 (tolerant) increased the chlorophyll a, b, total and carotenoids content, while in genotype 24 (sensitive) was maintained. The activity of RuBisCO was maintained in all genotypes. The content of starch increased only in sensitive genotypes while decreases in the tolerant under heat stress. On the other hand, sucrose decreased in sensitive and increased in tolerant genotypes under heat stress. Furthermore, the glucose and fructose content were maintained in all genotypes under heat stress. In conclusion, these results suggest that sensitive and tolerant may possess different mechanisms to enhance chlorophyll content, maintain or increase RuBisCO activity and modulate carbohydrate metabolism in response to heat stress, sugars synthesis and translocation might be potentially contributing to a better performance of tolerant genotypes under high temperature conditions compared to the sensitive ones.

IDENTIFICATION AND EVALUATION OF TANNINS AND FLAVONOIDS OF *NEPHELIUM LAPPACEUM* OF INDUSTRIAL INTEREST

Adelma Escobar Ramírez^{1*}, José Guadalupe de la Cruz Hernández¹, Evelyn de Dios Vásquez, José A. Lorenzo Alcudia, Erick Sierra Campos⁵, Oswaldo I. Hernández Abreu¹, José A. González Garrido¹, Carlos J. López Victorio¹, Claudia I. Avitia Domínguez⁵, Alfredo Téllez Valencia⁵, Cuauhtémoc Alvarado Sánchez¹, Edgar Zenteno Galindo[‡], Vanessa Dehennaut[†]

^{*}Universidad Juárez Autónoma de Tabasco, División Académica de Ciencia Básicas, Grupo de investigación de bioquímica y biología molecular. Carretera Cunduacán-Jalpa Km1. Col. La Esmeralda, CP 86690, Cunduacán, Tabasco, México, adelma.escobar@ujat.mx, 9933892285, [†] Instituto del cáncer ONCOLille, Cancer Heterogeneity, Plasticity and Resistance to Therapies UMR 9020 CNRS, Bd du professeur Jules leclercq 59000 Lille, France. [‡]Universidad Nacional Autónoma de México, Facultad de Medicina, Dpto. Bioquímica, Av. Universidad 3000, C.U, 04510 Ciudad de México. ⁵Universidad Autónoma Benito Juárez de Oaxaca, Facultad de odontología, Oaxaca, Oaxaca, ⁵ Facultad de Ciencias Químicas GP, Facultad de Medicina y Nutrición, Universidad Juárez del Estado de Durango.

Nephelium lappaceum is a fruit rich in vitamins, minerals, proteins, and other diverse metabolites such as phenolic acids, phenols, including flavonoids and tannins, which offer multiple health benefits.

Some reports relate the content of flavonoids and tannins with antioxidant capacity and their effect in preventing the effects of cellular aging, cancer, diabetes, and cardiovascular diseases, as is the case of tannins present in grapes (*Vitis vinifera*), and tannins obtained from precious wood trees such as *Quercus robur* oak, which are also used in the wine industry.

Tannins are used to tanning the skins, useful in the manufacture of shoes, belts, bags and other products made from this material.

Nephelium lappaceum is a little studied fruit of Asian origin, that was recently introduced in Mexico in the region of the Soconusco in Chiapas, and currently its cultivation has spread to other states such as Tabasco, Michoacán, Nayarit, and Oaxaca. *Nephelium lappaceum* (rambután), has a uniform red or yellow pericarp, with long spines (similar to flexible thorns), and a seed covered by a white or translucent aril that is the edible part of the fruit. Currently this edible part is the most appreciated part of the fruit, not the peel or pericarp that in most cases is discarded, without considering that it is a usable resource because in the peel we can find a large amount of anthocyanins, N, P, K, Ca, Mg, tannins and flavonoids, among others.

Previous results obtained by our team have shown that *Nephelium lappaceum* possesses considerable total phenols, a potent antioxidant effect, and also antiproliferative effects in colon cancer cell lines HCT116, and prostate Du145, so its use as a phytomedicine, in the first place, is visualized. On the other hand, the present project also visualizes the use of flavonoids and tannins present in *Nephelium lappaceum* for its use in the wine industry and in leather tanning.

Currently, total extracts have been carried out with organic solvents for the extraction of phenols including Flavonoids and Tannins. The total extract has been purified by low pressure chromatography on a LH-20 column and has also been subjected to HPLC separation. The separation of components present in the extract has also been visualized by thin layer chromatography. Currently we have qualitative results of the presence of Flavonoids and Tannins present in the total extract of *Nephelium lappaceum* peel, by Shinoda technique and the gelatin method. Tests are planned to identification of tannins and flavonoids by mass HPLC, as well as to perform tanning tests on cattle skin for their potential use in the leather tanning industry.

IMPACT OF THE W165F MUTATION IN PKBADH ON THE FORMATION OF THE PKBADH-NAD⁺ COMPLEX

Anabel Félix Arredondo¹, José Guadalupe Soñanez Organis¹, César Muñoz Bacasehua¹, Norma Aurora Stephens Camacho², Elisa M. Valenzuela Soto¹, Jesús Alfredo Rosas Rodríguez¹

¹University of Sonora. Blvd. Lázaro Cárdenas No. 100, Col. Francisco Villa, Navojoa, Sonora. Postal Code 85880, ²State University of Sonora. Blvd. Manlio Fabio Beltrones 810, Col. Bugambilia, Postal Code 85875, Navojoa. a217204113@unison.mx

Betaine aldehyde dehydrogenase (BADH, betaine aldehyde: NAD⁺ oxidoreductase, EC 1.2.1.8) catalyzes the conversion of betaine aldehyde (BA) to glycine betaine (GB)(1). The synthesis and accumulation of GB have been associated with osmoregulation and osmoprotection functions, the reduction of an oxidative environment, and regulation in the expression of stress response genes (2). The formation of the pkBADH-NAD⁺ complex induces changes at the secondary structure level due to a rearrangement at the active site and is favored by the presence of potassium (3). It is known that pkBADH contains four tryptophan residues (W) per subunit, where the proximity of tryptophans 156 and 165 allows for the study of structural changes using intrinsic fluorescence emission techniques (2). In this work, it was demonstrated that the W165F mutation in pkBADH does not affect enzymatic activity. However, fluorescence emission studies revealed a greater exposure of tryptophan 156, with the accessible fraction of tryptophan (α) showing values 1.82 times higher than pkBADH. Fluorescence quenching highlights the involvement of tryptophan 156 in forming the pkBADH-NAD⁺ complex. Additionally, the W165F mutation caused structural alterations, increasing compactness in the tertiary structure and reducing the number of available active sites (n). The mutant presented one available active site, unlike the two present in pkBADH, demonstrating the crucial role of W156 in forming the complex.

References

1. Guzmán-Partida, A. M., & Valenzuela-Soto, E.M (1998). *Comp Biochem Physiol B Biochem Mol Biol.* 119: 485-491.
2. Rosas-Rodríguez, J. A., & Valenzuela-Soto, E. M (2011). *Chem Biol Interact.* 91(1): 159-164.
3. Muñoz-Bacasehua, C., Rosas-Rodríguez, J. A., López-Zavala, A. A., & Valenzuela-Soto, E. M. (2021). *Luminescence: the journal of biological and chemical luminescence.* 36(7): 1733-1742.

DEFINING ESSENTIAL AND NON-ESSENTIAL CHLOROPLAST RIBOSOMAL PROTEINS IN *NICOTIANA TABACUM* AND *ARABIDOPSIS THALIANA*

Emanuel Osmar Flores Camargo¹, Tzvetanka Dimitrova Dinkova², Noé Valentín Duran Figueroa¹, María del Carmen Oliver Salvador¹ and Jesús Agustín Badillo Corona¹

¹Unidad Profesional Interdisciplinaria de Biotecnología, Instituto Politécnico Nacional, Av. Acueducto SN Col. Barrio la Laguna Ticomán, 07340 México City, México, ²Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México, 04510 México City, México
E-mail: jbadillo@ipn.mx

Ribosomes are complexes of rRNA and proteins that carry out the translation process for protein synthesis. This process can occur in different cell compartments: the cytoplasm and organelles such as mitochondria and plastids (e.g. chloroplasts).

Unlike the knowledge generated about the essentiality and function of ribosomal proteins in bacteria (*Escherichia coli*), the essentiality and function of a large number of ribosomal proteins in plant chloroplasts is still unknown. Between 2001 and 2022, studies of the essentiality of chloroplast ribosomal proteins have been performed by *knocking-out* genes with selection markers in tobacco; and the use of T-DNA insertion lines, RNA interference and/or CRISPR-Cas9 in *A. thaliana*^{1,2,3}. This has paved the way for the characterization of 43 proteins out of a total of 58 proteins that make up the chloroplast ribosome.

In this work, we aim to elucidate the essentiality of the chloroplast ribosomal proteins that are still uncharacterized. In tobacco, for those genes encoded in the chloroplast genome, we are following the approach of deleting by substitution, the coding sequences of the genes of interest with the selection marker gene *aadA*, which confers resistance to spectinomycin. In *A. thaliana*, we are using the CRISPR/Cas9 system to target the chloroplast ribosomal protein genes encoded in the nucleus.

We currently have different transplastomic tobacco lines with putative deletions of the genes *rp14*, *rp16* and *rps8*. Preliminary results suggest that these genes are essential, because the state of homoplasmy, that is the state where all copies of the plastid genome have been eliminated, has not been achieved. Generation of additional transplastomic lines to delete *rps11*, *rps19* and the double copy of *rpl2* and *rps7* is still in progress.

Also, 10 vectors have been designed and constructed with the CRISPR/Cas9 system and used to transform *A. thaliana* to target the site direct *knock-out* of the genes that code for the proteins encoded in nucleus (*rps6*, *rps10*, *rpl5*, *rpl9*, *rpl12*, *rpl15*, *rpl17*, *rpl19*, *rpl29* and *rpl34*). Transformation and editing of these lines are soon to be confirmed.

The knowledge generated from the study of chloroplast ribosomal proteins could not only be useful to discover the function of these proteins beyond the clearly relevant function as structural part of the ribosome but also used for the design of ribosomes with a reduced number of proteins or to develop ribosomes with certain modifications that confer them with new functional properties.

Keywords: chloroplast, essentiality of proteins, CRISPR/Cas9, transplastomic plants, ribosomal proteins.

References

1. Tiller, N., and Bock, R. (2014), The translational apparatus of plastids and its role in plant development. *Mol. Plant*, 7: 1105–1120.
2. Bobik, K., Fernandez, J.C., Hardin, S.R., Ernest, B., Ganusova, E.E., Staton, M. and Burch-Smith, T.M. (2019), The essential chloroplast ribosomal protein uL15c interacts with the chloroplast RNA helicase ISE2 and affects intercellular trafficking through plasmodesmata. *New Phytol*, 221: 850-865.
3. Dupouy, G., McDermott, E., Cashell, R. *et al.* (2022), Plastid ribosome protein L5 is essential for post-globular embryo development in *Arabidopsis thaliana*. *Plant Reprod*, 35: 189–204.

THE COMBINATION OF THREE DRUGS MODIFIES THE METABOLISM IN COLORECTAL CANCER CELLS

Laura C. Flores García, Jossimar Coronel Hernández, Verónica García Castillo y Carlos G. Pérez Plascencia

Facultad de Estudios Superiores Iztacala (UNAM), Instituto Nacional de Cancerología

Av. De los Barrios 1, Hab. Los Reyes Iztacala Barrio de los Árboles/Barrio de los Héroes, 54090 Tlalnepantla, EdoMex.

Currently, colorectal cancer represents a public health problem worldwide, as it ranks third in both incidence and mortality (GLOBOCAN (2022), Liu *et al.* (2022)). Colorectal cancer is characterized by the excessive proliferation of epithelial cells of the colon or rectum, forming a benign adenoma until progression to a carcinoma that can lead to metastasis (Mármol *et al.* (2017), Hon *et al.* (2021)). On the other hand, among the characteristics of tumor cells is the reprogramming of cellular metabolism to promote cell growth and division (Hanahan & Weinberg, 2011). For example, reprogramming of glycolysis is associated with an increase in glucose uptake, a result of mutations that converge in PI3K/Akt signaling, which promotes energy generation through the formation of lactate and the production of intermediates metabolic processes of biosynthetic pathways (Pavlova *et al.*, 2022). Likewise, the increase in glutamine metabolism is another of the altered metabolic pathways in cancer cells that favors proliferation, by generating precursors of amino acids such as glutamate, aspartate, serine, alanine and proline, in addition to being a source in the production of α -ketoglutarate (Pavlova *et al.*, 2022). Both metabolic pathways converge in the tricarboxylic acid cycle (TCA), causing the surpluses produced by both metabolic pathways to generate a “vent” for the TCA carbon atoms, buffering the mitochondrial electron charge (Pavlova *et al.*, 2022). Therefore, these characteristics of tumor cells are promising therapeutic targets, so it is feasible to assume that the use of selective drugs to inhibit glycolysis and ATP generation, as well as increasing DNA damage and production of species reactive oxygen, can promote apoptosis. Previously, our work group established a new pharmacological therapy focused on attacking the metabolism of the tumor cell through the use of metformin (it inhibits complex I of the respiratory chain, which increases the concentration of AMPK and allows the inhibition of the Akt/mTOR pathway), sodium oxamate (inhibitor of lactate dehydrogenase A, decreasing ATP production) and doxorubicin (intercalant in DNA, inhibitor of topoisomerase II and inducer of reactive oxygen species) (Figueroa *et al.* (2016)). Previous results demonstrated the reduction in mTOR and HIF-1 α activation, which are master coordinators of metabolism by promoting the synthesis of glycolytic enzymes (Coronel *et al.* (2021)). Thus, in the present study the effect of triple therapy (3Tx) in HCT-116, RKO and SW-620 cells was evaluated, where we determined the IC50 to be used in the combinatorics and observed that each line showed a behavior different in response to 3Tx, that is, for the HCT-116 line the drugs had an additive effect, in the SW-620 line the effect was synergistic and in the RKO line the observed effect was competitive. On the other hand, when evaluating cellular metabolism, we observed a decrease in glucose uptake in both lines; however, the effect was greater in the RKO line compared to the HCT-116 cells. Whereas there was an increase in ATP production due to treatment with 3Tx, however, the effect was like that of doxorubicin (individual) in the HCT-116 line, contrary to the effect reported in RKO cells where we obtained a lower response of 3Tx compared to doxorubicin. For its part, lactate synthesis was not affected by treatment with the combination of the three drugs in any of the two lines evaluated. With these results, we demonstrate that the use of triple therapy induces changes in metabolism, where we observe mechanisms of action dependent on the cell line and how these drugs establish their interaction (additive or competitive), which ultimately promotes the death of tumor cells. However, it remains to be determined whether this metabolism modification is consistent with the modification of PI3K/Akt/mTOR pathway signaling.

THE TRANSLATION FACTOR EIF4E IS A KEY MEDIATOR OF DOXORUBICIN RESISTANCE: INSIGHTS FROM A TRIPLE-NEGATIVE BREAST CANCER MODEL

Héctor Frayde-Gómez^{1,2}, Brenda Chimal-Vega^{1,2}, Ángel Pulido Capiz^{1,2}, Octavio Galindo-Hernández^{1,2}, José Luis Vique-Sanchez^{3,4}, Ana Gabriela Leija Montoya⁴, Marco A. Ramos-Ibarra⁵, Víctor García González^{1,2*}

¹Departamento de Bioquímica, Facultad de Medicina Mexicali, Universidad Autónoma de Baja California, Mexicali 21000, México;

²Laboratorio Multidisciplinario de Estudios Metabólicos y Cáncer, Universidad Autónoma de Baja California, Mexicali 21000, México; ³Centro de Ciencias de la Salud Mexicali, Universidad Autónoma de Baja California, Mexicali, BC, México;

⁴Facultad de Medicina Mexicali, Universidad Autónoma de Baja California, Centro Cívico, Mexicali 21000, BC, México;

⁵Grupo de Investigación en Biotecnología y Biociencias, Facultad de Ciencias Químicas e Ingeniería, Universidad Autónoma de Baja California, Tijuana 22390, México

E-mail: vgarcia62@uabc.edu.mx

Breast cancer (BC) is the most common cancer type and is among the leading causes of cancer-related deaths in women worldwide. BC tumors with triple-negative (TNBC) are highly aggressive and associated with a relatively short life expectancy. Chemotherapy with anthracyclines is one of the standard treatments for TNBC¹. Chemoresistance development represents a major challenge. This condition is associated with alterations in mechanisms of translational capacity increase of cancer cells, such as overexpression of drug efflux pumps². A key component of the initial stage of protein translation is the eukaryotic initiation factor 4F (eIF4F) complex. A subunit of this complex, the eIF4E factor, has been reported to play a critical role in chemoresistance³. A TNBC model of chemoresistance to Doxorubicin (Dox), based on the MDA-MB-231 cell line, was generated using its IC₂₅ value as a selective dose. Moreover, effective drug processing linked to a high capability of migration and invasion was found in MDA_R cells. Overexpression and activation of the eIF4E factor. Primary results indicate that eIF4E-p^{Ser209} dysregulation was associated with increased expression levels of ABCB1. In addition, a potential dependence of nuclear factor erythroid 2-related factor 2 (Nrf2) was activated under high concentrations of Dox in MDA_R cells. Overall, our findings suggest that chemoresistance increased cellular invasiveness and revealed a possible mechanism of DOX detoxification, which involves the ABCB1 transporter and the transcription factor Nrf2 with possible eIF4E-dependent modulation.

References

1. Won, K. A., & Spruck, C. (2020). Triplenegative breast cancer therapy: Current and future perspectives (Review). *International journal of oncology*, 57(6), 1245–1261.
2. Piska, K., Koczurkiewicz-Adamczyk, P., Jamrozik, M., Bucki, A., Kołaczkowski, M., & Pękala, E. (2023). Comparative study on ABCB1-dependent efflux of anthracyclines and their metabolites: consequences for cancer resistance. *Xenobiotica; the fate of foreign compounds in biological systems*, 53(6-7), 507–514.
3. Yang, X., Zhong, W., & Cao, R. (2020). Phosphorylation of the mRNA cap-binding protein eIF4E and cancer. *Cellular signalling*, 73, 109689

ISOLATION OF INTACT *P. PARVA* AND *C. REINHARDTII* PLASTIDS FOR THE STANDARDIZATION OF AN *IN VITRO* PROTEIN IMPORT MODEL

Sergio Fuentes-Hernández¹, Miriam Vázquez-Acevedo¹ and Diego González-Halphen¹

Instituto de Fisiología Celular, UNAM. Circuito Exterior Universitario s/n, Ciudad Universitaria, 04510 CDMX
(5556225620-sfuentes@ifc.unam.mx)

Protein import is a phenomenon that occurs in eukaryotic cells. Thus, nuclear-encoded proteins that are synthesized in the cytosol are directed towards their target organelle (i.e., mitochondria and chloroplasts). In general, the chloroplast import protein machinery is known to be composed of membrane translocases (TOC/TIC, Sec, SRP & Tat), peptidases (SPP, PreP & TPP) and targeting signals (TP & SS)¹.

The chlorophycean alga *Polytomella parva* lost its photosynthetic machinery and adapted a heterotrophic lifestyle. Thus, *P. parva* now exhibits starch-containing, colorless plastids (also called amyloplasts) that have lost their photosynthetic capabilities but are still metabolically active. *P. parva* amyloplasts lack a plastid genome⁴, which is usually present in free-living organisms. The transcriptome of *P. parva* and its amyloplast proteome have revealed the presence of several protein import components (TOC/TIC, Sec, SRP, PreP, SPP & TPs)². Therefore, it has been suggested that all the proteins present in the algal colorless plastids must be synthesized in the cytosol and internalized into the plastid. *P. parva* and *Chlamydomonas reinhardtii* share a common ancestor, and the import machinery of both algae exhibit homologous components of the TOC/TIC translocators. Possibly, both chloroplast and amyloplasts could indistinctly recognize the targeting sequences of the protein precursors targeted to these plastids. Although the import of proteins into the chloroplast has been studied in *C. reinhardtii*³, protein import into the amyloplast has not been explored in *P. parva*. Here, we propose to standardize an *in vitro* model of protein import in isolated plastids of both *C. reinhardtii* and *P. parva* plastids and to test whether the *C. reinhardtii* import machinery is capable of internalizing *P. parva* proteins and vice versa. An important challenge for the development *in vitro* protein import models is obtaining an intact plastid preparation. Here, we explored different methods to obtain intact chloroplasts and amyloplasts from both chlorophycean algae. We obtained a sample enriched with intact amyloplasts that was characterized by transmission electron microscopy: starch granules surrounded by membranes were observed after breaking the *P. parva* cells and isolating plastids by differential centrifugation. These amyloplasts could be competent for protein import assays. On the other hand, we have tried to break *C. reinhardtii* cells to obtain intact chloroplasts using detergent, mechanical disruption and sonication. The integrity of the obtained chloroplasts still needs to be confirmed.

This work was supported by grants PAPIIT (IN207023) from DGAPA-UNAM and CF2019-21856 from CONACyT.

References

1. Ballabani, et al. (2023) Front. Physiol., 14: 1213866
2. Fuentes-Ramírez, et al. (2012) Microbiol. Res., 243: 126649
3. Rochaix, J.-D. (2022) FEBS J, 289: 6908-6918
4. Smith & Lee. (2014) Plant physiol. 164: 1812-1819

DETERMINATION OF THE ACTIVITY OF THE ENZYMES IN THE PRIMARY ASSIMILATION OF NITROGEN IN THE EMBRYOGENIC SYSTEM OF *COFFEA CANEPHORA*

Laura C. Fuentes-Vázquez¹, Rosa María Galaz-Avalos² and Víctor M. Loyola-Vargas^{3*}

¹⁻³Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, Calle 43, 130, Colonia Chuburná de Hidalgo, Mérida, Yucatán, C.P. 97200.

*E-mail:vmloyola@cicy.mx; Phone (52) 999 942 83 30, Ext. 243

Somatic embryogenesis (SE) is valuable for plant propagation and genetic improvement. It allows the study of morphological, biochemical, and molecular events in several species, such as *C. canephora*. During this *in vitro* biological process, bipolar structures regenerate from somatic cells, which can then produce complete plants. The cells respond to endogenous stimuli that induce different signals, thus modifying the cell's genetic program¹. Several factors are crucially involved in somatic to embryogenic cell differentiation and morphogenesis, including explant type, culture medium, growth regulators, and nitrogen source². Undoubtedly, nitrogen (N) plays an essential role in SE. Ammonium, nitrate, and amino acids are three forms of N used to add in vitro culture mediums. The balance between the nitrogen sources could be a determinant in the production of embryos during morphological differentiation. The knowledge about the biochemical and molecular mechanisms that trigger the signal to form the embryo by modifying the N source is scarce. Duarte-Aké et al. (2022) hypothesized that the balance between auxins (Aux) and cytokinins (CKs), which are related to N availability, could generate the signals that induce SE³. This study investigates how the nitrogen source influences the enzymatic activity of primary N assimilation and its relationship with Aux and CK biosynthesis during SE of *C. canephora*. We will present the data supporting this hypothesis.

References

1. Elhiti, M., & Stasolla, C. (2022). Transduction of signals during somatic embryogenesis. *Plants*, **11**(2), 178.
2. Nic-Can, G. I., Avilez-Montalvo, J. R., Aviles-Montalvo, R. N., Márquez-López, R. E., Mellado-Mojica, E., Galaz-Ávalos, R. M., & Loyola-Vargas, V. M. (2016). The relationship between stress and somatic embryogenesis. *Somatic Embryogenesis: Fundamental Aspects and Applications*, 151-170.
3. Duarte-Aké, F., Márquez-López, R. E., Monroy-González, Z., Borbolla-Pérez, V., & Loyola-Vargas, V. M. (2022). The source, level, and balance of nitrogen during the somatic embryogenesis process drive cellular differentiation. *Planta*, **256**(6), 113.

FLUORIDE CONCENTRATION IN THE BREAST MILK OF LACTATING WOMEN IN THE CITY OF DURANGO

Sergio Galindo Najera¹, Omar Tremillo Maldonado¹, Leslie Gamboa Guerrero¹, Oscar Almeda Ojeda¹, Gamaliel Ortiz Sarabia¹, Marcelo Gomez Palacio Gastélum¹, Sergio Manuel Salas Pacheco², José Manuel Salas Pacheco², Adda Agustina Sandoval Carrillo²

¹Facultad de Odontología, Universidad Juárez del Estado de Durango; ²Instituto de investigación Científica.
Correo electrónico: sergio-alberto2014@hotmail.com

Introduction. Fluoride plays an important role in biological processes such as osteogenesis and odontogenesis, maintaining the hardness of tissues and providing greater resistance against acids, but there are also several conditions caused by chronic fluoride consumption, among the best known are dental fluorosis. The World Health Organization recommends that fluoride intake in 1, 2 and 3 year olds should be limited to 0.5, 1.0 and 1.5 ppm/day. Human breast milk is universally known as the optimal source of postnatal nutrition for infants and it has been shown that the concentration of fluoride in water can have a direct impact on the concentration of fluoride in breast milk and these findings highlight the importance of monitoring and regulating fluoride levels in drinking water in order to ensure optimal and safe fluoride exposure to infants through breast milk.

Objective. To determine the concentrations of fluoride present in breast milk of lactating women in the city of Durango.

Materials and Methods. Samples of breast milk, urine, tap water and bottled water were collected from 10 lactating mothers. Quantification of the samples was performed using potentiometry with ion selective electrode.

Discussion and Conclusions. All samples had detectable levels of fluoride, however, 30% of the population showed levels higher than those established by the WHO, finding a correlation between drinking water consumption and its increase in breast milk levels.

Results. The levels in urine ranged from 1.85 to 9.64 ppm with a mean of 3.59 ppm of fluoride, tap water ranged from 0.77 to 4.76 ppm with a mean of 2.48, in tap water we had .00 and .96 respectively and .13 ppm mean. The concentration of fluoride in breast milk fluctuated between 0.00 and 0.17 with a mean of 0.02, yielding very varied data taking into account the concentration of fluoride in tap water, thus demonstrating a correlation between fluoride intake and its presence in breast milk.

CHARACTERIZATION OF MITOCHONDRIAL Ca^{2+} REGULATION IN HELA CELLS

Leonardo Gallegos¹, Itzel Lievano¹, Lizeth Sandoval¹, Manuel Arias² and Agustín Guerrero-Hernández¹

¹Cinvestav, IPN Avenue 2508 Mexico City and ²Facultad de Estudios Superiores Iztacala-UNAM, Tlalnepantla de Baz 54090, State of Mexico.

E-mail: mlgallegos@cinvestav.mx

Calcium ion (Ca^{2+}) is a second messenger involved in cellular processes such as proliferation, differentiation, senescence, autophagy, among others. The steady activity of pumps, ion channels, and Ca^{2+} binding proteins results in a resting intracellular Ca^{2+} concentration ($[\text{Ca}^{2+}]_i$) of 100 nM. Different physical and chemical stimuli increase $[\text{Ca}^{2+}]_i$ by inducing the entry of this ion either through the plasma membrane or from intracellular Ca^{2+} reservoirs. This ion alters the activity of different organelles, such as the mitochondria where Ca^{2+} drives ATP production, but also induces the opening of the mitochondrial permeability transition pore (mPTP), culminating in cell death. Due to these antagonistic roles of Ca^{2+} , studying its regulation in mitochondria is very important. The Ca^{2+} uptake into the mitochondrial matrix involves the activity of ion channels in two membranes: first, the Voltage-Dependent Anion Channel 1 (VDAC1) present in the outer mitochondrial membrane (OMM), and the Mitochondrial Calcium Uniporter (MCU) multiprotein complex of the inner mitochondrial membrane (IMM). MCU complex consists of the MCU itself, the essential MCU regulator (EMRE), MCU regulator 1 (MICUR1), MCUB, MICU1, and MICU2 (MICUs). The Ca^{2+} uptake into the mitochondria through the MCU is facilitated by its membrane potential (~ -180 mV) and is finely regulated by the protein MICU1. The latter presents EF-hands Ca^{2+} binding domains, which prevent Ca^{2+} entry unless the EF-hands are occupied by Ca^{2+} , resulting in Ca^{2+} entry facilitation. The matrix Ca^{2+} removal systems are mainly the $\text{Ca}^{2+}/\text{H}^+$ and $\text{Na}^+/\text{Ca}^{2+}$ exchangers (NCLX)¹. Previous studies have shown that MCU knockout (KO) mice are not lethal despite the absence of this Ca^{2+} transporter. In contrast, NCLX-KO is lethal, indicating that NCLX is an indispensable protein for mitochondrial survival^{2,3}. Therefore, this work aims to study the mechanisms of MCU regulation to understand the evolutionary pressure to express this protein. To determine whether mitochondrial $[\text{Ca}^{2+}]$ is in a steady state or not, we carried out simultaneous measurements of the $[\text{Ca}^{2+}]_i$ and mitochondrial Ca^{2+} ($[\text{Ca}^{2+}]_{mt}$) in HeLa cells with fura-2 and either CEPIA-2mt or mito4x-GCaMP6f targeted to the mitochondria matrix. Histamine (His) was applied to increase the $[\text{Ca}^{2+}]_i$ by activating IP_3Rs and thus increasing $[\text{Ca}^{2+}]_{mt}$. The data showed that His increased $[\text{Ca}^{2+}]_{mt}$ even with $[\text{Ca}^{2+}]_i$ below 500 nM and without external Ca^{2+} . To determine whether the $[\text{Ca}^{2+}]_{mt}$ is in a steady state, cells were exposed to CGP-37157, an NCLX inhibitor, resulting in no basal increase in $[\text{Ca}^{2+}]_{mt}$. To test this hypothesis further, we used $p\text{-Br-2APB}$, a new ER Ca^{2+} -releasing agent that is also a SOCE inhibitor. Unexpectedly, this compound induced a strong reduction of the $[\text{Ca}^{2+}]_{mt}$ despite transiently elevated the $[\text{Ca}^{2+}]_i$, suggesting a possible IP_3R -mediated basal Ca^{2+} entry into mitochondria.

References

1. R Bagur, G Hajnóczky (2017). *Molecular Cell*, **66**, 780–788.
2. X Pan, J Liu, T Nguyen, C Liu, J Sun, Y Teng, M Fergusson, I, M Allen, D Springer, A Aponte, M Gucek, R Balaban, E Murphy and T Finkel (2013). *Nature Cell Biology*, **15**, 1464–1472.
3. SM Emrich, RE Yeast, AJ Fike, KN Bricker, P Xin, X Zhang, ZSM Rahman, M Trebak, (2022). *Cell Calcium*, **108**

MOLECULAR INTERACTION BETWEEN P53, P21 AND BIOMOLECULES PRESENT AT MANILKARA ZAPOTA SEEDS.

García Alcudia Vanessa, Alfredo Téllez Valencia, Claudia I. Avitia Domínguez, Oswaldo I. Hernández Abreu, José A. González Garrido, Carlos J. López Victorio, Edgar Zenteno Galindo[‡], Carlos Josué Solórzano Mata[§], Vanessa Dehennaut[‡], Adelma Escobar Ramírez^{*}

^{*}Universidad Juárez Autónoma de Tabasco, División Académica de Ciencia Básicas, Grupo de investigación de bioquímica y biología molecular. Carretera Cunduacán-Jalpa Km1. Col. La Esmeralda, CP 86690, Cunduacán, Tabasco, México, adelma.escobar@ujat.mx, 9933892285, [‡] Instituto del cáncer ONCOLille, Cancer Heterogeneity, Plasticity and Resistance to Therapies UMR 9020 CNRS, Bd du professeur Jules leclercq 59000 Lille, France. [‡] Universidad Nacional Autónoma de México, Facultad de Medicina, Dpto. Bioquímica, Av. Universidad 3000, C.U., 04510 Ciudad de México. [§] Universidad Autonoma Benito Juárez de Oaxaca, Facultad de odontología, Oaxaca, Oaxaca.

Manilkara zapota, (Chicozapote), is a tree of the sapotacea family. The cultivation of *Manilkara zapota* has been limited to the production of fruits as food, as well as to the obtaining of the latex. The cultivation of *Manilkara zapota* has been limited mainly to the production of the fruit as food, as well as to the obtaining of the latex, the utilization of this species has been neglected, considering that all parts of the tree present a great amount of chemical-biological compounds of interest In this sense our work is directed to the identification and evaluation of the interaction of biomolecules of the seed of “*Manilkara zapota*” versus the crystallographic structures of the transcription factor, p53 and p21; by means analytical techniques LC/MS and molecular docking. Considering previous antiproliferative results on cancer cell lines HCT116 and Du145, we hypothesized that *Manilkara zapota* contains secondary metabolites, with antiproliferative properties, which can interact with transcription factors and have effect on sumoylation pathway having consequences on cell cycle regulation. The p53 protein is involved in cell cycle regulation, acts at the level of the G1 to S step and when activated induces apoptosis in response to DNA alteration. p53 mediates its effect in part through p21 and therefore they are usually studied together. p21 has both p53-dependent and p53-independent effects, and the latter in turn inhibits cyclin-dependent kinases (CDKs), arresting the cell cycle by inhibiting DNA replication. In the present work, 9 secondary metabolites and 6 peptides were selected, using the mass HPLC equipment, after a defragmentation analysis was readed using the “ChemDraw®” program and subsequently molecular docking of the selected molecular structures from *Manilkara zapota* versus the p53 and p21 crystallographic structures was assayed. The results of this work will let to evaluate the use of metabolites identified as resveratrol, dihidrocordoina, curmadiona by others, at the modulation of the activity of P53, and p21 in the cancer processes and evaluated their uses as potentials therapeutic phytomedicament in the future.

SPECIFICITY AND MECHANISMS OF c-TYPE CYTOCHROME BIOGENESIS IN MALARIA PARASITES

Aldo E. García Guerrero, Rebecca Marvin, Amanda Mixon-Blackwell and Paul Sigala

Department of Biochemistry, School of Medicine, University of Utah
E-mail: aldo.garcia@biochem.utah.edu

Malaria remains a devastating disease affecting millions of people around the world, especially in sub-Saharan Africa and south-east Asia. *Plasmodium falciparum* is the causative agent of severe malaria, including cerebral malaria. The lack of an effective vaccine and the emergence of resistance against frontline therapies highlight the urgent need to understand the basic molecular mechanisms of the parasite to discover novel therapeutic targets. *P. falciparum* infects two different hosts; mosquitoes, in which sexual reproduction occurs, and humans where it invades hepatocytes and red blood cells. All symptoms appear during the blood stage and during this phase the mitochondrial respiratory chain is essential to maintain the activity of the dihydroorotate dehydrogenase (DHOD), involved in pyrimidine biosynthesis. Specifically, the respiratory Complex III is a current target by the antimalarial atovaquone. The activity of Complex III depends on membrane-bound cytochrome c₁ (Cyt_{c1}) and soluble cytochrome c (Cyt_c), which differ from other subunits by covalently binding heme. In eukaryotes, all c-type cytochromes are hemylated by a single enzyme known as the holocytochrome c synthase (HCCS). In humans a single bifunctional HCCS hemylates both Cyt_c and Cyt_{c1}. However, most eukaryotes have two HCCS enzymes with different but partially overlapping specificities to hemylate Cyt_c (HCCS) or Cyt_{c1} (HCC1S). *P. falciparum* has two HCCS homologues (PF3D7_1224600/PF3D7_1203600) that are both refractory to disruption, suggesting non-redundant essential roles for hemylation of Cyt_c and Cyt_{c1}. We confirmed that both HCCS enzymes localize to the mitochondrion. To directly test essentiality for blood-stage parasites, we tagged both HCCS genes with the aptamer/TetR-DOZI system for conditional knockdown (KD). HCC1S KD caused a lethal growth defect due to diminished Cyt_{c1} levels and ETC dysfunction, and parasites were not rescued by overexpressing HCCS. KD of HCCS resulted in a more modest growth defect, but parasites were strongly sensitized to mitochondrial depolarization by proguanil, which targets a secondary pathway for maintaining transmembrane potential. These results and biochemical reconstitution of Cyt_c hemylation by HCCS in bacteria demonstrate a specific role for parasite HCCS in hemylating Cyt_c. This study provides fundamental insight into the key mechanisms evolved by malaria parasites to support essential ETC function. We have ongoing experiments to understand how parasites transport heme to the mitochondrion and to define the key interaction partners involved in c-type cytochrome hemylation.

CHARACTERIZATION OF THE VDAC FAMILY IN MAIZE AND THE PHYSIOLOGICAL ROLE OF THE ISOFORMS ZmVDAC1B AND ZmVDAC4B IN DROUGHT STRESS

Donají A. García-Ortiz¹, Andrés Burgos-Palacios¹ and Sobeida Sánchez Nieto¹

¹Laboratorio de Transporte y Percepción de Azúcares en Plantas, Departamento de Bioquímica, Facultad de Química, UNAM, 04510, Mexico City, Mexico.
E-mail: donaji.garciaortiz@gmail.com, sobeida@unam.mx

Voltage-dependent anion channels (VDACs) are the most abundant proteins in the mitochondrial outer membrane. They fulfill a variety of functions ranging from the exchange of high-energy molecules and metabolites to debatable functions such as cell death¹, having an important role between cytoplasmic and mitochondrial signaling events.

In plants, the study of VDAC is limited, what is known is that they belong to multigene families, which have been shown to participate in different types of biotic and abiotic stresses. However, to date there is scarce information about the VDAC family in important agronomic plants such as *Zea mays*. Here we present the identification and *in silico* characterization of the VDAC family in maize (ZmVDAC), the localization of ZmVDAC1b and ZmVDAC4b, the physiological effect of transient expression of these isoforms in *Nicotiana benthamiana* leaves under drought stress conditions, and the response with other proteins, such as ZmHXK4, for which there is evidence of interaction with VDAC through pull-down assays.

We identified 9 genes that comprise the ZmVDAC family through an *in silico* analysis to propose a nomenclature according to phylogenetic relations. In addition, by obtaining transcript levels along the germination time and in response to *Fusarium verticillioides* infection,² we chose ZmVDAC1b and ZmVDAC4b to conduct a deep study for its potential to have a key role in maize physiology. The recombinant proteins were detected in the mitochondria of *Nicotiana* leaves. The heterologous expression of ZmVDAC produced a notorious damage in the leaves, which is exacerbated under drought stress. We are still investigating the plant responses to the coexpression of ZmHXK4 with ZmVDAC to understand their role in cell death.

References

1. Colombini, M. (2016). The VDAC channel: Molecular basis for selectivity. *Biochimica Et Biophysica Acta. Molecular Cell Research*, 1863(10), 2498-2502
2. Rodríguez-Saavedra, C., García-Ortiz, D. A., Burgos-Palacios, A., Morgado-Martínez, L. E., King-Díaz, B., Guevara-García, Á. A., & Sánchez-Nieto, S. (2023). Identification and Characterization of VDAC Family in Maize. *Plants*, 12(13), 2542.

IMPACT OF DROUGHT STRESS AND RECOVERY IRRIGATION ON AMINO ACID ACCUMULATION AND ANTIOXIDANT ACTIVITY IN MEXICAN SOYBEAN

Julio César García Rodríguez¹, Roxana Silerio Espinoza², María Karina Manzo Valencia¹, Diego de Jesús Pantoja Gutiérrez², Víctor Olalde Portugal¹ and Silvia Valdés Rodríguez^{1*}

¹Centro de Investigación y de Estudios Avanzados del IPN-Unidad Irapuato, Carr. Irapuato-León Km 9.6, Libramiento Norte, Irapuato, Gto. C.P. 36821, ²Tecnológico Nacional de México-Celaya, Antonio García Cubas 600, Col. Fovissste, Celaya, Gto. C.P. 38010.

Corresponding author: silvia.valdez@cinvestav.mx

Drought is a significant abiotic factor that negatively impacts plant growth and productivity of soybean [*Glycine max* (L.) Merr]. To cope with drought stress, plants have developed biochemical mechanisms, such as accumulating specific amino acids and activating antioxidant enzymes. Drought-tolerant plants frequently amass particular amino acids to maintain cellular homeostasis and enhance antioxidant activity to mitigate reactive oxygen species. This study aimed to assess the free amino acids content and superoxide dismutase (SOD) activity in leaves of three Mexican soybean genotypes subjected to drought stress and subsequent recovery irrigation. One early (H02-2309) and two intermediate (H98-1240 and Huasteca 700) genotypes were evaluated in a randomized block design with three replicates under well-watered and water-deficit conditions. Water deficit was applied at the R2 stage of plants by reducing soil irrigation gradually (from 11% to 3% gravimetric soil moisture) over 17 days, followed by a re-watering phase. Accumulation profiles of amino acids and SOD activity were measured in the third leaf of plants at 17 days of water deficit (17 DWD) and 8 days after recovery irrigation (8 DRI). At 17 DWD, Pro content increased significantly in stressed plants of the H02-2309 and Huasteca 700 genotypes compared to irrigated plants. At 8 DRI, Huasteca 700 showed enhanced levels of Pro, Gly, Asn, and His content and increased SOD activity in the water deficit treatment. The H98-1240 genotype displayed the highest SOD activity at 17 DWD and elevated Pro, Gly, and Ala content at 8 DRI in the same condition. Results indicated that soybean genotypes responded differently to drought stress, suggesting that H02-2309 and Huasteca 700 appeared to utilize an osmotic adjustment response, while H98-1240 likely activated an antioxidant response. Interestingly, after recovery irrigation, plants that were subjected to drought increased the amino acid content in both Huasteca 700 and H98-1240 genotypes, while SOD activity was enhanced in Huasteca 700, possibly as part of a recovery mechanism.

CHALLENGES IN THE SOLUBILITY AND FOLDING OF RECOMBINANT LIPASES: THE CASE OF LIPGOM6

Elena Lizbeth Garcia-Villegas^{1,2}, Liliana Pardo-López² and Enrique Rudiño Piñera²

¹Centro de Investigación en Dinámica Celular, UAEM, Av. Universidad 1001, Cuernavaca, Morelos. ²Instituto de Biotecnología, UNAM, Av. Universidad 2001, Cuernavaca, Morelos.
E-mail: lizbeth.garcia@ibt.unam.mx

Lipases are enzymes that catalyze the hydrolysis of triacylglycerides and whose products are fatty acids and glycerol. These enzymes have a catalytic triad consisting of serine, an acid residue (glutamic acid or aspartic acid), and histidine. *Pseudomonas* lipases are used in many areas of biotechnology, nevertheless, the expression and soluble production of these enzymes become a challenge for their study and commercialization. This is because *Pseudomonas* lipases require a foldase protein, called lif, which is responsible for folding the lipase to a native state. In this study, we present a successful strategy for the soluble and active production of lipase 2 from *Pseudomonas sp. GOM6* (lipGOM6) through the dilution refolding method assisted by the foldase lifGOM6.

References

1. Azeem, F., Afzal, M., Javed, S., Riaz, M., Kouser, A., Hussain, S., Nadeem, H. (2017). Progress in Biophysics and Molecular Biology, 132, 23-34.
2. Nardini, M., Lang, D. A., Jaeger, E., Liebeton, K., & Dijkstra, B. W. (2002). Journal of Biological Chemistry, 275 (40), 31219-31225.
3. Tang, L., Su, M., Zhu, L., Chi, L., Zhang, J., & Zhou, Q. (2013). World Journal of Microbiology and Biotechnology, 29 (1), 145-151.

THE METHIONINE SYNTHASE IN PLANT DEVELOPMENT

Sara Margarita Garza Aguilar¹, Adriana M. Salazar Puente¹, Estefany Damaris Guerrero Molina⁴,
M. Guadalupe Frías Muñoz³, Rosa M. Bocanegra-García³, Hiram Hernández López³,
Rocío I Díaz de la Garza^{1,2}

¹Escuela de Ingeniería y Ciencias, Tecnológico de Monterrey, Ave. Eugenio Garza Sada 2501, Monterrey 64849, NL, México;

²Institute for Obesity Research, Tecnológico de Monterrey, Ave. Eugenio Garza Sada 2501, Monterrey 64849, NL, México;

³Unidad Académica de Ciencias Químicas, Universidad Autónoma de Zacatecas, Campus UAZ siglo XXI, Zacatecas, Zac;

⁴Facultad de Química, Departamento de Bioquímica, Universidad Nacional Autónoma de México,

Av. Universidad y Copilco, Ciudad de México 04510, México

Email: qfbsaramargaritaga@gmail.com

In plants, the Methionine Synthase (MS) catalyzes the biosynthesis of Methionine (Met) using a folate (5-CH₃-THF) as a cofactor. Folates may accept and donate 1C-units at different oxidation states for the biosynthesis of amino acids, purines, and other metabolites, such as S-adenosyl-methionine (SAM). Folate derivatives exist in nature in a variety of polyglutamyl forms (PG), and several folate-utilizing enzymes have higher affinity for the PG forms than for the monoglutamyl forms; one such enzyme is MS. Met donates 1C- units to SAM synthesis, a precursor of other metabolites involved in different plant development processes. For instance, ethylene is produced during postharvest ripening, and polyamines and nicotianamine are formed in nodules during symbiosis. Arabidopsis contains three *MS* genes: *MS1* and *MS2*, which produce Met through the salvage pathway in the cytosol, and *MS3*, which synthesizes Met *de novo* in plastids. Our previous analysis suggests that MS gene expression and protein activity could be regulated by metabolites produced downstream, such as ethylene in ripening and Met or SAM during symbiosis between *Rhizobium* and legumes. The role of the interaction between MS enzymes and substrates-products in the development process is not yet fully understood. This work aims to characterize the MS of plants. We retrieved and compared the deduced MS sequences from various plants, predicted their subcellular localization, and analyzed their phylogenetic relationship and primary sequence. Additionally, we conducted a docking analysis between the protein and folate-PG. Furthermore, we determined the free amino acid involved in Met synthesis during postharvest ripening of climacteric fruits (tomatoes and avocados), and in fixing and non-fixing nodules during symbiosis between *Rhizobium* and *Phaseolus vulgaris*. Additionally, an *in silico* transcriptomic analysis of *MS* genes was performed. We found that other plants encode at least two MS proteins, one cytoplasmic and one plastid isoform, similar to Arabidopsis. The *MS1* gene was primarily expressed during ripening, while the *MS3* gene was expressed in symbiosis. The plant MS protein sequences are highly conserved (>85%) and contained all essential domains, including the zinc binding domain, H₂Cy/Met binding domain, and folate binding domain, indicating the crucial role of this enzyme in plants development.

CLONING, EXPRESSION AND PURIFICATION OF RECOMBINANT HEMAGGLUTININS FROM AVIAN INFLUENZA VIRUS TYPE A

¹Margarita Fernanda Gómez Meza, ¹Adriana Castillo Villanueva, ¹Jocelyne Martínez Martínez, ¹Gabriel Isaías Torres Vernet, ²M. Escorcía Martínez, ¹Jesús Antonio Oria Hernández*

¹Laboratorio de Bioquímica Genética, Instituto Nacional de Pediatría. ²Departamento de Medicina y Zootecnia de las Aves, FMVZ-UNAM. *Av. Insurgentes Sur 3700-C Insurgentes Cuicuilco, Coyoacán, 04530, CDMX. 55-1084-0900 ext 1442. jesus.oria.inp@gmail.com.

The Influenza A Virus (IAV) can infect a wide range of hosts, mainly birds and humans, being Avian Influenza (AI) thus considered a zoonotic disease. The IAV family is subdivided based on their antigenic surface glycoproteins HA (hemagglutinin) and NA (neuraminidase); to date, 16 HA and 9 NA influenza A virus subtypes are recognized. AI H5N1 is classified as highly pathogenic avian influenza (HPAI). Since the first report of human infection in 1997 with the H5N1 AIV, hundreds of cases have been reported, reaching a mortality rate of more than 50%. AI has the potential to cause global epidemics, therefore, the identification and control of circulating viruses is a global health priority. The aim of this work was to generate recombinant hemagglutinins from IAV as antigens for potential vaccine development. Genes coding for hemagglutinins of two H5 and one H7 strains were commercially synthesized and cloned into the pCribb plasmid. pCribb adds both 6X-His and MBP tags and a cleavage site for TEV protease at the NH-terminal end. The recombinant proteins were expressed in *Escherichia coli* BL21(DE3) pLysS strain. Purification was accomplished by immobilized metal affinity chromatography (IMAC). Results indicate that each gene was successfully cloned in pCribb, as indicated by 0.8% agarose gel showing a band of the expected size (~1700 bp) after digestion with NdeI and BamHI. Full sequencing was performed to ensure proper construction. Expression conditions were OD₆₀₀ 0.6, IPTG 1 mM, 37°C for 3 hours and 20°C overnight. Purification after IMAC was verified on 12% SDS-PAGE showing a band of ~111 kDa as expected. Preliminary results indicate that proteins were hemagglutinins and immunogenic. In conclusion, recombinant hemagglutinins from AIV with zoonotic potential were obtained through expression in a heterologous system. We hypothesize that these proteins will provide a safe and viable option for the development of vaccines and serological tools for AI surveillance, prevention and control, safeguarding public health.

RELATIONSHIP BETWEEN MAIZE CELL CYCLE-RELATED REGULATORS AND THE REPAIR PROTEIN RAD51A

M. en C. Estefany Damaris Guerrero Molina, Dr. Jorge Manuel Vázquez Ramos*

Facultad de Química, Departamento de Bioquímica, Universidad Nacional Autónoma de México
Ciudad de México, 04510, México.
E-mail: jorman@unam.mx

Genome stability of seed cells is crucial for crop productivity and genotoxic agents and metabolic processes can cause DNA damage. To counteract this damage, plant cells have DNA repair mechanisms similar to mammals.

Cyc/CDK protein complexes are essential for the cell cycle but are inhibited in response to DNA damage, however, CycB1/CDKB1 complexes play a specific role in repairing double-strand breaks through homologous recombination. Also, the Proliferation Cell Nuclear Antigen (PCNA) is important for DNA replication and cell cycle regulation, but importantly participates in DNA repair, especially through homologous recombination (HR) involving RAD51 recombinase.

In this project, studies on maize demonstrated the effect of gamma radiation-induced DNA damage on germination and seedling establishment. Additionally, we observed differential changes in the expression and protein abundance of PCNA, CycB1;2, CDKB1;1, and RAD51 after radiation. Furthermore, interaction simulations were developed between PCNA and the aforementioned proteins. Predictions indicate a potential macro-complex formation between these proteins and DNA, suggesting their involvement in the regulation of DNA damage repair.

EXPLORING THE STRUCTURAL AND FUNCTIONAL CHANGES OF VIBRIO CHOLERAE PYRUVATE KINASE

Gloria Hernández-Alcántara¹, Alicia Vega-Segura¹, Martín González Andrade¹, José de Jesús García-Trejo², Lilian González-Segura², Itzhel García-Torres³, Sergio Enríquez-Flores³ and Leticia Ramírez-Silva¹

¹Departamento de Bioquímica, Facultad de medicina, UNAM, ²Departamento de Bioquímica, Facultad de Química, UNAM,

³Laboratorio de Biomoléculas y Salud Infantil, Instituto Nacional de Pediatría.

E-mail: ghernandez@bq.unam.mx

Pyruvate kinase (PK) isoform I, is a four-domain enzyme that is active as a homo-tetramer, it exhibits hyperbolic steady-state kinetic, although it is allosterically regulated in the bacteria. A previous phylogenetic analysis of the PK family showed a dichotomic tree, separating the family into two clusters. Cluster I is K⁺-dependent activity and cluster II is K⁺-independent¹. Prokaryotic bacterium as *Vibrio cholera* encoded three distinct PKs, one is in cluster I (VcIPK) and two are in cluster II (VcIIPK and VcIIIPK) this characteristic is unique for gamma-proteobacteria². Previously data showed that VcIPK and VcIIPK possess different allosteric effectors, whereas VcIIIPK did not show allosteric activation. This study was conducted with the goal of continuing to study the structural and functional properties between the three isoforms of pyruvate kinases from *Vibrio cholerae*. Our results indicate that when the three isoforms are incubated with reducing agents, only VcIIPK increases its enzymatic activity and in native gels it presents different oligomeric states such as dimer, tetramer, and other oligomers. A particular characteristic is that we have not been able to crystallize this isoform, which we believe is due to this property. In this regard, it is possible that S-S bridge are forming through cysteine residues. In an effort to understand these results, we analyzed the five Cys residues on VcIIPK and constructed single mutants where Cys was substituted by Ser, the results are in progress. In order to approach the biological meaning of these oligomers, we are analyzing the protein obtained directly from *Vibrio cholerae* by western blot. The first results indicated that the tetramer was the predominant oligomer found in the cell, as well as other lower molecular weight forms.

Acknowledgments. PPAPIIT-UNAM:IN205821 (GH-A).

References

1. Oria-Hernandez J, Riveros-Rosas H, Ramírez-Silva L. (2006) Dichotomic phylogenetic tree of the pyruvate kinase family: K⁺-dependent and -independent enzymes. *J Biol Chem.* 281(41):30717-24. Doi:10.1074/jbc.M605310200.
2. Guerrero-Mendiola C, García-Trejo JJ, Encalada R, Saavedra E, Ramírez-Silva L. (2017) The contribution of two isozymes to the pyruvate kinase activity of *Vibrio cholerae*: one K⁺-dependent constitutively active and another K⁺-independent with essential allosteric activation. *PLoS One* 12(7):e0178673. Doi:10.1371/journal.pone.0178673.

EVALUATION OF THE DNA GYRASE INHIBITORY EFFECT OF NATURAL PRODUCTS AND DERIVATIVES WITH KNOWN ANTI-*HELICOBACTER PYLORI* ACTIVITY

Erick Hernández-Hipólito¹, Erika Gomez-Chang¹, William Meza-Morales², Yair Álvarez-Ricardo², Raúl G. Enríquez² and Irma Romero¹

¹Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México, Cd. Universitaria, C.P. 04510, CDMX, México. ²Instituto de Química, Universidad Nacional Autónoma de México, Cd. Universitaria, C.P. 04510, CDMX, México.
E-mail: irma@bq.unam.mx

Helicobacter pylori is the etiological agent of gastritis, ulcers, and gastric cancer; the worldwide prevalence of the infection is ~44% and for Mexico it has been estimated of about 52%, thus being considered a national and international public health problem¹. Eradication treatments consist of a proton pump inhibitor and the combination of two or three antibiotics. Currently, the effectiveness of these therapies has declined due to the emergence of antibiotic-resistant strains, making it necessary to search for alternative treatments².

As part of these alternatives, various compounds and extracts derived from plants used in traditional medicine, with good anti-*H. pylori* activity, have been identified in the laboratory^{3,4}. In an attempt to explain the mechanism by which these products inhibit bacterial growth, the action of one extract and 17 compounds (including curcuminoids, cadalenes, eupatilin, and anacardic acid) were tested on the DNA gyrase activity.

This enzyme, which is essential and unique to bacteria, regulates DNA topology during replication processes by introducing negative supercoiling, so it constitutes an important pharmacological target⁵.

The results showed that two products, anacardic acid and the methanolic extract of *Cyrtocarpa procera* (CpMet), inhibited 100% the DNA gyrase supercoiling activity, with IC₅₀ values of 13.16 µM and 4.94 µg/mL, respectively.

In conclusion, we propose that the anti-*H. pylori* activity of these products is due to the inhibition of the DNA gyrase activity; so, they have the potential to be used in *H. pylori* eradication therapies, considering that they are novel drugs, with not known resistance.

Acknowledgments: To DGAPA, PAPIIT, UNAM IN207622, and CONAHCyT, FOINS PRONACES 307152.

Referencias:

1. Zamani *et al.*, (2018). *AP&T*, 47, 868-876.
2. O'connor *et al.*, (2014). *Helicobacter*, 19, 38-45.
3. Castillo *et al.*, (2007). *J. Ethnopharmacol.*, 114, 72-77.
4. Escobedo *et al.*, (2014). *J. Ethnopharmacol.*, 143, 363-371.
5. Bush *et al.*, (2015). *EcoSal Plus*, 6.

IN VITRO STUDY OF PHYSICOCHEMICAL PROPERTIES OF NOVEL VOLTAGE-GATED POTASSIUM ION CHANNEL BLOCKERS BASED ON 4-AMINOPYRIDINE

Karen Sofia Hernández-Medina¹, Sofía Rodríguez-Rangel¹, Jair A. Madera-Hurtado², Mario U. Delgado-Jaime², Aikaterini Diamanti² & Jorge E. Sánchez Rodríguez¹

¹Departamento de Física y ²Departamento de Química, CUCEI, Universidad de Guadalajara, Guadalajara, Jalisco 44430, México.

*These authors contributed equally

E-mail: jorge.srodriguez@academicos.udg.mx

4-aminopyridine (4AP) is a voltage-gated potassium ion channel (K_v) blocker used in patients diagnosed with multiple sclerosis (MS) for improving walk. Its mechanism of action involves a series of sequential processes that depend on lipophilicity, pH and voltage membrane. We implemented *in vitro* (titration curves, shake-flask method, and UV-vis spectroscopy) and *in silico* (computational chemistry) procedures to determine some physicochemical (,) properties of 4AP and the new K_v blockers 4AP analogs: 3-Fluoro-4-aminopyridine (3F4AP) and 3-Methyl-4-aminopyridine (3Me4AP). of 3F4AP (7.22) was units lower than produced by 4AP and 3Me4AP (9.58 and 9.82). At =7.4, of 4AP, 3Me4AP and 3F4AP was, and , respectively. This result was consistent with the previously measurements reported by Rodríguez-Rangel, *et al.*¹. Next, we determined at values for which of protonated () or deprotonated () species of these 4AP analogs exist. At , of these 4AP analogs was while at , of 3F4AP and 3Me4AP was 2-fold higher than the one produced by 4AP indicating that conjugation of F and CH₃ at position 3 of 4AP makes molecules more lipophilic. To explain this behavior, we performed a *in silico* assays by Density Functional Theory employing a wide range of functionals, spanning from the simplest local and gradient-corrected to more robust range-separated hybrid and combined with the cc-pVTZ basis set² and using the CPCM and SMD solvation models to calculate Gibbs free energies in liquid phase. The computed and values closely resembled to the measured ones in terms of absolute values and relative trends. We conclude that the best predictive levels of theory for 3Me4AP involved the use of LC-PBE and TPSS0 functionals, with absolute percentage errors (APE) of 2.03%, and 3.36%, respectively, whereas for 3F4AP the best predictive level of theory was achieved by using the functional BHANDHLYP with APE of 3.13%. This study begins to delineate a methodology to predict and determine 4AP based molecules with better lipophilicity.

References

1. Rodríguez-Rangel, S., Bravin, A. D., Ramos-Torres, K. M., Brugarolas, P., & Sánchez-Rodríguez, J. E. (2020). Scientific reports, 10(1), 52.
2. Pezzola, S., Venanzi, M., Galloni, P., Conte, V., & Sabuzi, F. (2024). Molecules, 29, 1255.

STRUCTURE AND PHYLOGENY OF CROTOXIN SUBUNITS A AND B

David Hernández¹, Andrés Reyes Chaparro^{*2}

National Polytechnic Institute, National School of Biological Sciences, México City (11350),
Extension Manuel Carpio and Plan de Ayala, Col. Santo Tomás, Miguel Hidalgo.
E-mail: areyesch@ipn.mx*

Crototoxin is a protein complex found in the venom of snakes from the Viperidae family, primarily composed of an acidic and a basic subunit. This protein functions as a phospholipase A₂, with several known and studied variants identified in ten species of snakes, including the genera *Crotalus*, *Bothrops*, and *Bothriechis*. The extracted sequences pertain to the species *C. durissus terrificus* (model organism), *C. scutulatus*, *C. tigris*, *C. tzabcan*, *C. viridis viridis*, *C. atrox*, *C. basiliscus*, *B. neuwiedi*, *B. jararacussu*, and *B. schlegelii*. The objective of this study is to elucidate the structure and phylogeny of crototoxin to explore its potential pharmacological applications, particularly in parasitology and as an anticancer agent. The methodology employed involved the utilization of the MEGA and CHIMERA programs, as well as the AlphaFold 2 platform and the GenBank and Protein Data Bank databases. The results include the similarity percentages of sequences compared to *C. durissus terrificus*, as well as the construction of four cladograms based on the nucleotide and amino acid sequences of both subunits of the protein. Furthermore, alignments of the conserved regions of each subunit, including both nucleotides and amino acids, were performed, culminating in the modeling of the preliminary structures of the ten crototoxin complexes.

It has been observed that there is greater amino acid variation at specific positions in the A and B subunits of crototoxin. In subunit A, these positions are 2, 5, 11, and 17, while in subunit B, the most notable regions are positions 1 to 23 and 51 to 99. This variability is reflected in the three-dimensional structure of protein models. Cladograms illustrate differences in amino acid and nucleotide sequences, indicating that the genus *Crotalus* forms a group in subunit A, whereas studied species of *Bothrops* and *Botriechis* share certain relationships in another clade.

On the other hand, in crototoxin B sequences, the genus *Crotalus* exhibits differences in cladograms, with *Bothrops jararacussu*, *Bothrops neuwiedi*, and *Botriechis schlegelii* showing structural relationships with *Crotalus viridis viridis* and *Crotalus atrox*. This may be related to the neurotoxicity of subunit B, although other factors are involved. It is known that the functional domain of subunit B includes specific functional domains, leading to functional selection in amino acid sequences. Thus, they are conserved to fulfill a specific function related to enzymatic activity and interaction with other proteins.

References

1. Hernández, I. (2015). Crototoxina en el veneno de la serpiente de cascabel *Crotalus simus* en México: su relación con la toxicidad y distribución geográfica. [Proyecto de tesis]. UNAM. México. Enero, 2015. 1-65
2. Neri, E., Bénard, M. y Alagón, C. (2014). RDU. 15(11). 1-8
3. Sampaio, S., Hyslop, S. Fontes, M., Prado, J., Zambelli, V. Magro, A., Brigatte, P. Gutierrez, V. y Cury, Y. (2010). ELSEVIER. 55. 1045-1060

RED AND INFRARED LIGHT AS AN ENHANCER FOR MITOCHONDRIAL RESPIRATION OF KERATINOCYTES CELLS

Manuel Alejandro Herrera¹, Camille Caldeira¹, Alicia Kowaltowski¹ and Maurício S. Baptista¹

¹Universidade de São Paulo, Instituto de Química, Av. Prof. Lineu Prestes, 748–Butantã, São Paulo.

Dirección Postal: 05508-900; E-mail: maherreral@usp.br; Telefono: +55 11 91054-9573

The capacity of cells and organisms to perceive and respond to environmental signals is fundamental to life. Several factors induce responses odors, temperature, and photons. Light is an important factor that influence our daily activities and, in fact, our metabolism. The red and infrared light in the electromagnetic spectrum emitted by the sun comprises the region of 600 to 2500 nm. These types of light has gained lots of attention for to the modulatory responses in cellular metabolism and its possible connection with photobiomodulation, a procedure increasingly important in photomedicine. The results we have obtained studying LEDs with emission in red light (λ_{max} 657 nm) and infrared light (λ_{max} 805 and λ_{max} 980 nm) are truly promising by photobiomodulatory effects on. Through respiration analysis of the different mitochondrial states (using different modulators such as Oligomycin, CCCP, Antimycin, and Rotenone) in human keratinocyte cells (HaCat cell line), using SeaHorse XF24, we definitively demonstrated that red and infrared light significantly increases mitochondrial respiration, especially in the basal respiration, ATP production and maximal respiration. Effects on mitochondrial respiration are related to ATP synthesis in keratinocytes, for that reason, we thought that a metabolic demand like proliferation or biosynthesis pathways are activated by light, for now, we demonstrated that red light increase proliferation analyzed 48 and 72 hours post treatment. We need to validate, but we hypothesize mediate literature that those effects are not relative to opsins, GPCRs proteins sensitive to light, HaCaT cells has no Opsins long-wave sensitive. Activation of electron transport chain and oxidative phosphorylation may be related to another type of photo-induced mechanism. These results have the potential to allow the understanding of how skin cells interact with red light, facilitating the development of photobiomodulation protocols.

STUDY OF PERIPHERAL DOMAINS IN STRUCTURE-FUNCTION OF ISOCITRATE LYASE (ICL) FROM *PSEUDOMONAS AERUGINOSA*

Mildred Jiménez Báez, Alma Laura Díaz Pérez, César Díaz Pérez, Jesús Campos García

Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo
Francisco J. Múgica S/N, Ciudad Universitaria, 58030 Morelia, Mich.
+52 443 367 7256, mildredjbaez@gmail.com

Pseudomonas aeruginosa metabolizes organic compounds such as carbohydrates, organic acids, fatty acids, and amino acids, but also a wide range of pollutants [1]. This quality is essential for niche colonization and contributes to its pathogenicity. The catabolic pathways involved in the assimilation of these compounds include the glyoxylate cycle. Isocitrate lyase (ICL) is the first glyoxylate cycle enzyme, breaking down isocitrate to produce glyoxylate and succinate. *P. aeruginosa* ICL (PaICL) is a virulence factor belonging to the AceA/ICL protein family characterized by the catalytic domain I, whose structure contains a triosephosphate isomerase barrel (TIM-barrel) [1]. Additionally, the PaICL contains a domain II inserted at the periphery of domain I, which, by X-ray crystallography, could participate in enzyme oligomerization. Another characteristic of the PaICL is the α 13-loop- α 14 motif (extended motif), protruding from the enzyme core, whose function is unknown [2]. The objective of this work was to determine the function of domain II, extended motif, and the carboxyl-terminus (C-ICL) and amino-terminus (N-ICL) regions in PaICL and their roles that play in the *P. aeruginosa* PAO1 virulence [3]. Deleting domain II and the extended motif resulted in structurally unstable enzymes without activity. His₆-tag fusion on the C-ICL protein produced a less efficient enzyme than fusion on the N-ICL without affecting acetate assimilation or virulence. The homotetramer of the PaICL enzyme was more stable in N-His₆-ICL than in C-His₆-ICL, suggesting that the carboxyl terminus is essential for quaternary structure conformation. The ICL-mutant A39 complemented with recombinant proteins N-His₆-ICL or C-His₆-ICL was more virulent than the wild-type PAO1 strain. The results indicate that domain II and the extended motif are essential for ICL function, while the carboxyl terminus was involved in quaternary structure conformation, concluding that the ICL is essential for acetate assimilation and virulence in *P. aeruginosa*.

References

1. Campos-García J, Díaz-Pérez C, Díaz-Pérez AL (2013). World J Microbiol Biotechnol. 29:991-999.
2. Crousilles A, Dolan SK, Brear P, Chirgadze DY, Welch M (2018). J Biol Chem 293:14260-14269.
3. Díaz-Pérez AL, et al., World J Microbiol Biotechnol. (2023) 39(12):339.

PURIFICATION AND CHARACTERIZATION OF CHIMERIC RECOMBINANT VARIANTS OF HUMAN DEUBIQUITINASE USP2 IN RELATION TO ITS SPECIFICITY FOR UBIQUITIN K63 CHAINS

Josahandy Jiménez-Peralta¹, Fidel O. Ramírez-Amador¹, Luis E. Pérez-Córdova¹, Leticia Olvera-Rodríguez¹, Alexey Llopiz¹, Emma Arévalo-Salina¹, Manuel S. Rodríguez², Gloria Saab-Rincón¹, Wendy Xolalpa-Villanueva¹

¹Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, UNAM, 62250. Cuernavaca, Morelos, México.
²Laboratoire de Chimie de Coordination (LCC) - UPR 8241 CNRS, and UMR 152 Pharma-Dev, Université de Toulouse, IRD, UPS, 31400 Toulouse, France.

E-mail: josahandy.jimenez@ibt.unam.mx

Ubiquitination involves the covalent binding of ubiquitin (Ub) to lysine (K) residues on proteins. The bound Ub can be modified by the addition of more Ub, forming polyUb chains which is a tag that directs the protein to specific destinations in the cell. K48-linked chains lead to degradation by the proteasome, whereas K63-linked chains are involved in cell signaling. Deubiquitinases (DUBs) reverse ubiquitination, most DUBs recognize the ubiquitinated substrate independently of chain topology, such as USP2. However, some DUBs such as CYLD are specific for certain ligands showing preference for K63- and M1-linked chains. In CYLD, it has been identified structural regions of interaction with Ub chains (BL1, BL2 and L3), which contribute to specificity. Based on these findings, in our working group, the construction of chimeras with the insertion of the CYLD structural regions in USP2 was performed. The objective of this work was to evaluate the role of the BL1 and L3 structural regions of CYLD by characterizing recombinant USP2-CYLD chimeras. Our human chimeric enzymes were expressed in *E. coli* cells (Rossetta Star II) and purified by glutathione affinity chromatography using a GST tag. The activity of the purified fractions was evaluated by fluorescence assays with the substrate Ub-Rhodamine110mp and by SDS-PAGE and diUb or tetraUb chains. Purified chimeric USP2-CYLD-L3, USP2-CYLD-BL1 and wild-type USP2 enzymes were obtained with a purity percentage of 92.8, 60.9, and 94.8 % respectively. The insertion of the BL1 and L3 structural regions of CYLD into the DUB USP2 influenced the solubility of the protein decreasing the purification yield with respect to the wild-type enzyme: USP2 37.8 mg/L, USP2-CYLD-L3 18 mg/L and USP2-CYLD-BL1 3.6 mg/L. The chimeric enzymes showed lower activity compared to USP2, which was successful even though sequence level modifications included sequence change and 6 additional aa in USP2-CYLD-L3, whereas for USP2-CYLD-BL1 5 aa were removed in addition to the structural loop sequence change. Project supported by the Fondo Sectorial de Investigación para la Educación, convocatoria 2018, CONAHCyT AI-S-22895.

References

1. Gonzalez-Santamarta, M., Bouvier, C., Rodriguez, M., Xolalpa, W. (2022). Seminars in Cell & Developmental Biology, **132**, 155-170.
2. Clague, M., Urbé, S., Komander, D. (2019). *Molecular cell biology*, **20**, 338–352.

EFFECT OF GROWTH TEMPERATURE ON CATALASE ACTIVITY OF *RHODOCOCCUS EQUI*

Francisco Javier Juárez Castañeda¹, Mónica Andrea Valdez Solana¹, Erick Sierra Campos¹

¹Universidad Juárez del Estado de Durango, Av. Artículo 123 s/n Fracc. Filadelfia,
35019 Gómez Palacio, Dgo. México.
E-mail: ericksier@ujed.mx

Catalases play a key role in defense against oxidative stress in bacteria by catalyzing the decomposition of hydrogen peroxide (H₂O₂). They are involved in multiple cellular processes, such as development and differentiation, as well as in the production of some metabolites¹. *Rhodococcus equi* possesses 4 genes encoding catalase isoforms, with *katA* being the one that is overexpressed 367.9 (±122.6) times when the bacteria is exposed to 50 mM H₂O₂ during the stationary phase². However, this work didn't perform the kinetic characterization of this enzyme. Therefore, in this study, the effect of temperature on catalase activity in *R. equi* was analyzed. For this purpose, *R. equi* were cultured at 30, 37, and 44 °C under aerobic conditions in YPD medium. In their three growth phases, their optical density at 625 nm (OD₆₂₅) and the presence of catalase were monitored with rapid tests on slides with H₂O₂. In the stationary phase, catalase-positive was detected under the conditions of 30 and 37 °C, while at 44 °C there was no reaction, even though the three growing conditions reached the stationary phase with a similar OD₆₂₅ of 1.4 to 1.7 AU. It is important to mention that the catalase activity of these cells requires high concentrations of H₂O₂ (5-11 M). Therefore, to determine the kinetic parameters, *in vivo* assays were performed by displacement of H₂O volume in a bioreactor³. Unlike other methods, this allows for precise measurement of catalase activity at high concentrations of H₂O₂, without interference from the detection limit of other equipment. For the one at 37 °C, a Km of 103.9 mM and a Vmax of 80.8 mol/min×mg were obtained. At 30 °C, the Km increased to 319.9 mM, while the Vmax increased by 74.38% (140.9 mol/min×mg). Using the same method, inhibition kinetics were performed with aminotriazole (specific for catalases) and KCN (for hemoproteins). The Ki with KCN was 0.76 mM at 37 °C and 1.05 mM at 30 °C. For aminotriazole, the Ki was 31.87 mM at 37 °C and 627.48 mM at 30 °C. Finally, a gradient zymogram (4-12%) was prepared, in which it was observed that in the ones at 30 and 37°C, the activity is given by a protein doublet, where each one contributes ~50% to the total activity. Kinetic parameters suggest 2 different enzymes, but zymogram revealed an identical doublet, indicating one enzyme with distinct properties at 30 and 37°C. These findings suggest that *R. equi* possesses a "supercatalase" that is expressed during the stationary phase at an optimal temperature of ~30 °C, which could be used in various biotechnological works for the elimination of H₂O₂.

THE GLUCOSE SENSOR HXK1 IS INVOLVED IN THE COLD ACCLIMATION AND FREEZING SURVIVAL OF *ARABIDOPSIS THALIANA* PLANTS

Beatriz King-Díaz, Andrés Burgos-Palacios y Sobeida Sánchez Nieto

¹Dpto. de Bioquímica, Fac. De Química, UNAM. Ciudad Universitaria, Coyoacán, Cd.Mex. México, C.P. 04510

Email: kingbeat@unam.mx , sobeida@unam.mx

Cold and freezing temperatures affect the development and yield of agronomic plants. However, many plant species have developed a set of mechanisms to cope with the negative effects of cold stress, called acclimation¹. In the acclimation, a protein kinase signaling is activated inducing three main cold-responsive genes: inducer of CBF Expression (*ICE*), C-repeat Binding Factors (*CBFs*), and the Cold-Regulated genes (*COR*). These *ICE-CBF-COR* genes are a central pathway responsible for initiating the cold response in plants². Low temperatures induce Reactive Oxygen Species (ROS), alteration of cell membrane permeability, ice formation, and solute leakage². In *A. thaliana*, anthocyanin biosynthetic genes are upregulated at low temperatures, their accumulation in plants protects from the damage effects of the ROS³. In apple, the hexokinase 1 (HXK1) phosphorylates the transcription factor (TF) MdbHLH3 enhancing the transcription of the anthocyanin biosynthesis genes⁴. The HXKs have a wide range of functions from glucose sensing to regulating programmed cell death (PCD)⁴. AtHXK1 is a moonlighting mitochondrial enzyme and a glucose sensor⁶. To determine the importance of AtHXK1 in the mechanism of cold acclimation-freezing challenge, *A. thaliana* plants, ecotype Ler, mutant plants of HXK1 enzyme glucose insensitive (*gin*) mutants, *gin2-1*⁶, and *gin2-1* complemented plants with the *Zea mays* HXK4 were subjected to cold and freezing stress. The results suggest that HXK1 is important for acclimation of *A. thaliana* plants and for anthocyanin biosynthesis. Our findings indicate that *gin2-1* plants complemented with ZmHXK4 response to cold acclimation involved a different ICE-CBF-COR pathway.

References

1. Ponce-Pineda et al., 2021. *Int J Mol Sci* 22.
2. Mukhopadhyay and Roychoudhury, 2018, In, Wani and Herath eds.
3. Xu and Rostein, 2018, *Plant signaling and behavior* 3, e1451708
4. Hu et al., 2016. *Plos Genet* 12(8), e1006273.
5. Rodríguez-Saavedra et al., 2021. *Front Mol Biosci*.
6. Moore et al. 2003, *Science* 300: 332-336.

A NOVEL CENTRAL METABOLISM CONTROL BY CELL CYCLE

Aurora Lara-Núñez*, Roberto Labastida-Cuevas, Karla G. Serrano-Dávila, Miranda M. Mojica-Prisciliano, Kevin E. Lugo García, P. Sarahy Soto-Juárez, Diego Valdez-Tapia

Facultad de Química-UNAM. Edificio E, Lab. 114, Ciudad Universitaria, CDMX C.P. 04510. Tel. 55 5622 5284

*E-mail: auroraln@unam.mx

It is widely acknowledged that central carbon metabolism undergoes intricate regulatory controls, encompassing various processes, including transcription, translation, protein folding, allosteric enzyme regulation, and post-translational modification. Post-translational modifications, such as ubiquitination, phosphorylation, or glycosylation, possess the capability to modify enzyme structure, consequently altering enzyme activity swiftly. Each metabolic pathway comprises one or more enzymes that modulate the metabolic flux to meet the cell's requirements. These enzymes are recognized as regulatory enzymes and are subjected to diverse levels of control mediated by assorted mechanisms.

Cell division, being a demanding process, necessitates precise coordination with central carbon metabolism to ensure the adequate supply of DNA synthesis precursors, formation of new organelles, *de novo* cell wall synthesis, among other functions. Therefore, the cell cycle must regulate central carbon metabolism in maize, as demonstrated in human cancer cell lines. We postulate that this regulation occurs through cyclin/CDK phosphorylation, considering that these complexes are the principal regulators of the cell cycle.

This study aimed to investigate the interaction of Cyclins or CDKs with regulatory enzymes that govern at least five different pathways within central carbon metabolism (glycolysis, tricarboxylic acid cycle, oxidative pentose phosphate pathway (OPPP), gluconeogenesis, and the anaplerotic reaction mediated by PEPC). The interaction was assessed using *in silico* methods and pull-down assays. Furthermore, the phosphorylation of the target enzymes by Cyclin/CDK complexes was validated *in vitro* and, in some instances, through semi-*in vivo* assays. Finally, the impact of post-translational modification on enzyme activity was evaluated. The target enzymes in this study were as follows: Glyceraldehyde 3 phosphate dehydrogenase, hexokinase 7 (cytosolic) pyruvate kinase, phosphofructokinase, malate dehydrogenase, glucose-6-phosphate, fructose-1,6-bisphosphatase, and phosphoenolpyruvate carboxylase.

Acknowledges: PAPIIT-DGAPA IN211423

STUDY OF THE CELL DEATH INDUCED BY THE IZTLI PEPTIDE 1 IN SACCHAROMYCES CEREVISIAE REVEALS AN UNKNOWN GENETIC CONNECTION BETWEEN ARREST, METABOLISM AND MATING

Ma. Teresa Lara Ortiz, Vladimir Juarez Arellano, Roberto Galindo Ramirez, Susana Castro-Obregon y Gabriel Del Río Guerra

Departamento de Bioquímica y Bilogía Estructural. Instituto de Fisiología Celular, UNAM. CD MX. C.P. 04510. gdelrio@ifc.unam.mx, Telf. 55 56 24 46 00

A current concern is the resistance of microorganisms to multiple drugs. In addition, most or these drugs act in cells that are actively dividing, but there is still no known antibiotic that acts specifically during cells cycle arrest. Developing antibiotics against non-diving cells is becoming a need to treat some diseases. In the laboratory of Dr. Gabriel Del Río, the antimicrobial peptide Iztli 1 (IP-1) derived from the α pheromone was developed. Including the α pheromone in IP-1 makes it specific target *Saccharomyces cerevisiae* cells that express the receptor for said pheromone (Mat a cells). We have reported that IP-1 only kills cells that are not dividing, either as a result of pheromone-mediated arrest or other cell cycle inhibitors, making it the first selective drug that kills cells only when they are arrested, providing an experimental model to study the mechanism for killing yeast cells arrested on the cell cycle. This work reports pharmacogenomic studies of yeast and human cells. We also identified that the null mutants of six genes (KAR5, TSA2, MNN4, ADH1, SPT20 and TDH1) that encode for IP-1-interacting proteins are not arrested by the α pheromone, although they present the Shmoo phenotype associated with mating and are required for the cell death induced by IP-1. Mouse Embryonic Fibroblasts (MEF) cells are not killed by IP-1 even when MEF were induced to arrest. Consistent with this finding is our observation that the set of mammalian proteins that interact with IP-1 differ from those identified in yeast. Therefore, we describe the genetic components of a programmed cell-death that is not conserved between yeast and mammalian cells providing new potential targets for the development of a new class of selective antibiotics against non-diving microbes. It is relevant to note that we are discovering an unexpected role for two genes coding for fermentative (ADH1) and respiratory (TDH1) metabolism that are involved in the cell-cycle arrest related to the pheromone pathway, suggesting a unknown connection between mating, cell-cycle and metabolism.

TIPS TO INCREASE THE THERMAL STABILITY OF A RECOMBINANT PROTEIN: THE PRACTICAL CASE OF CGI-58

Miriam L. Llamas-García¹, Edgar D. Páez-Pérez², Claudia G. Benítez-Cardoza³, Gabriela Montero-Morán², Samuel Lara-González^{1*}

¹IPICYT, ²Universidad Autónoma de San Luis Potosí, ³Escuela Nacional de Medicina y Homeopatía, IPN.

*Autor de correspondencia: Samuel Lara González,

Camino a la Presa San José 2055, Col. Lomas 4ta, CP 78216, San Luis Potosí, SLP.

Tel: 01(444)834200 ext 6239, Correo electrónico: samuel.lara@ipicyt.edu.mx

Working with recombinant human proteins in *E. coli* systems presents several technical challenges, including issues such as incorrect folding, formation of inclusion bodies, and low solubility, which complicate their functional analysis and the study of interaction mechanisms. In this research, we focus on the practical case of the human protein CGI-58, considered a novel therapeutic target for diseases such as cancer, diabetes, and metabolic syndrome. The primary function of CGI-58 is in adipose tissue lipolysis, where it has been reported to interact with proteins ATGL, PLIN1, and FABP4, playing a crucial role in the regulation of this process. Our research group aims to elucidate the protein-protein interaction mechanisms and obtain the structure of these complexes using X-ray diffraction. Initially, it is necessary to establish conditions that maintain the protein in a stable state. Specifically, we investigate the role of three tryptophan residues at the N-terminus of CGI-58 (W19, W23, and W27) involved in lipid droplet (LD) attachment and ATGL activation by generating a triple tryptophan-to-alanine mutant (3WA). Additionally, we examined the effect of phosphomimetic mutation at residue S237 by creating the 3WA/S237E mutant. Our experimental results indicate that the tryptophan residues at the N-terminus modulate the oligomerization state of the protein, with the CGI-58 3WA and 3WA/S237E mutants predominantly existing as monomers compared to the wild-type (WT) protein, which tends to aggregate. Furthermore, these mutations enhance thermal stability and increase the secondary structure content compared to the 3WA mutant alone, suggesting that phosphorylation plays a significant role in maintaining protein integrity.

STUDIES ON THE PHYSIOLOGICAL ROLE OF THE PUTATIVE GENTISALDEHYDE DEHYDROGENASE FROM *PSEUDOMONAS AERUGINOSA*

Alexey Llopiz, Carlos Mújica-Jiménez, Arline Fernández-Silva,
Gustavo A. Valero-Pibaral, Rosario A. Muñoz-Clares

Facultad de Química, Universidad Nacional Autónoma de México.
Ciudad Universitaria 04510, Ciudad de México, México.

E-mail: clares@unam.mx

In addition to be an important animal and plant pathogen, *Pseudomonas aeruginosa* is a soil and water bacterium with a versatile metabolism, allowing its growth on a wide variety of substrates. Despite *P. aeruginosa* relevance, much of its enzymatic machinery remains unstudied, including the aldehyde dehydrogenase product of the *PA2125* gene. Recent studies in our laboratory suggest that its physiological substrate is 2,5-dihydroxybenzaldehyde (gentisaldehyde), but a pathway involving the degradation of aromatic compounds via gentisaldehyde has not been described in *P. aeruginosa*. To address this, we investigated the growth of the PAO1 wildtype and a mutant strain devoid of the *PA2125* gene in different potential carbon and energy sources: (1) aromatic compounds that may yield gentisaldehyde when degraded (m-cresol, anthracene, phenanthrene, naphthalene, 1-naphthol, phenylalanine, homogentisate); (2) aldehydes that *in vitro* are substrates of the *PA2125* enzyme (gentisaldehyde, salicylaldehyde, benzaldehyde, 3-hydroxybenzaldehyde, 3,5-dihydroxybenzaldehyde, (hydroxymethyl)furfural) and their corresponding alcohols; and (3) products of these aldehydes oxidation. Additionally, since the *PA2124* gene encodes a probable dehydrogenase that could oxidize gentisyl alcohol to gentisaldehyde, we evaluated *in silico* the feasibility of them being substrates for *PA2124* and *PA2125*, respectively. The catalytically competent complexes, simulated by molecular docking were validated through molecular dynamics simulations. The results support the idea that gentisyl alcohol and gentisaldehyde may be the physiological substrates of *PA2124* and *PA2125*, respectively, and that these enzymes may participate in the degradation of aromatic compounds in *P. aeruginosa*. The discovery of the novel metabolic pathway in *P. aeruginosa* could open the possibility of diverse applications in bioremediation and/or biotechnological applications.

Acknowledgements: This work was financed by PAPIIT UNAM (grant IN203423) and PAIP Facultad de Química UNAM (grant 5000-9119) to RAMC. AL was supported by SNII, CONAHCYT (contract I1200/051/2023), AFS by a DGAPA, UNAM, postdoctoral fellowship, and GAVP by a CONAHCYT graduate scholarship.

BIOCHEMICAL CHARACTERIZATION OF OROTATE PHOSPHORIBOSYLTRANSFERASE FROM THE PHYTOPATHOGEN *PSEUDOMONAS CICHORII*

Amelia López-Albores¹ and Lilian González-Segura^{2*}

¹Instituto Tecnológico de Tuxtla Gutiérrez, Chiapas. C.P. 29050. ²Universidad Nacional Autónoma de México, Ciudad de México, C.P. 04510, México.
E-mail: *lilgonzalezseg@gmail.com

The enzyme orotate phosphoribosyltransferase (OPRTase) catalyzes the condensation of orotate (OA) with 5- α -D-phosphoribosyl 1-diphosphate (PRPP) in the presence of Mg²⁺ ion to produce pyrophosphate (PPi) and orotidine 5'-monophosphate (OMP) which is converted by OMP decarboxylase to uridine 5'-monophosphate (UMP), the main precursor to de novo pathway of all the pyrimidine nucleotides.

Previous studies in bacteria, protozoans and fungi have demonstrated the importance of de novo synthesis pathway of pyrimidine nucleotides and its influence on their pathogenicity. Studies in *Pseudomonas aeruginosa* and *Magnaporthe oryzae* have reported a significant reduction in virulence factors and pathogenicity as a result of the lack of the gene encoding OPRTase.

Pseudomonas cichorii is a Gram-negative phytopathogen that causes serious diseases in all the regions with warm temperatures and high humidity levels. Signs of its infection are necrotic lesions in leaves, stems and petioles. *P. cichorii* has a wide host range; some of its hosts include a huge diversity of vegetables, ornate flowers and economically important species such as tobacco (*Nicotiana tabacum*) and coffee (*Coffea arabica*).

In the present project, the biochemical characterization of the OPRTase from *P. cichorii* was carried out with the aim that this enzyme would be an innovative target for the design of drugs against the infection of coffee plants caused by *P. cichorii*. The recombinant enzyme was purified by using affinity chromatography and exclusion molecular chromatography. Thermal stability was measured by thermal shift assays. Finally, as a part of kinetic studies, values of Km for PRPP and Vmax were obtained. The kinetics results were compared with the values reported for other OPRTases.

Supported by FQ-5000-9129, PAPIIT-UNAM (IN211223)

A SEASONAL PROTEOMIC STUDY OF *CENTRUROIDES EXILICAUDA* VENOM FROM BAJA CALIFORNIA, MÉXICO

Joaquín López-Carrillo¹, Alan Hernández-Melgar¹, Roberto Arreguín-Espinosa²,
Martín Pallares-Perez³, Nadia Caram-Salas^{1,4}

¹Departamento de Innovación Biomédica, CICESE, Carretera Tijuana-Ensenada 3918, Fracc. Zona Playitas, 22860 Ensenada, B.C. México. ²Departamento de Bioquímica, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Ciudad Universitaria, A. P. 70242, México, D.F. 04510, México. ³Facultad de Medicina, Xochicalco Campus Ensenada, San Francisco 1139, Fracc. Misión, CP 22830, Ensenada, B.C., México. ⁴CONAHCYT, Avenida Insurgentes Sur 1582, Crédito Constructor, Benito Juárez, Ciudad de México, 03940, México
E-mail: ncaram@cicese.edu.mx

Extensive research has been conducted on scorpion venom for its potential applications in biotechnology. The specificity and potency of the venom components offer a unique advantage in developing new therapeutic strategies. However, most studies on scorpion venom provide only partial characterizations and do not fully elucidate all protein components from specific species. This study aims to uncover endemic Baja California scorpion *Centruroides exilicauda* venom's protein components at three seasonal periods. Venom was extracted using established electrical stimulation techniques from live specimens in the spring, summer, and fall seasons. The collected venoms were analyzed for protein components using matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS). MALDI-TOF-MS analysis revealed that the components of *C. exilicauda* venom mainly consist of molecules with a molecular weight below 15 kDa, including peptides as the major components, enzymes, and other bioactive molecules. The components of *C. exilicauda* venom exhibited distinct temporal variations; however, three peptides were consistently present in all venom samples regardless of the time. Our findings underscore the dynamic nature of scorpion venom and emphasize the importance of studying its components in different temporalities.

CHARACTERIZATION OF THE Cdc2 SUBUNIT REGULATORS TvCKS1 AND TvCKS2 IN *TRICHOMONAS VAGINALIS*

Karla Concepción López Pacheco, Nataly Morales Galeana,
Mónica Sarzo Gómez and María Imelda López Villaseñor

Instituto de Investigaciones Biomédicas, UNAM. Ciudad Universitaria, Cd. Mx.,
Tel. +52 55 5622 8952, e-mail: imelda@iibiomedicas.unam.mx

The cell cycle is the process by which a cell divides, producing two genetically identical cells. This process involves several phases that ensure the accurate duplication and cell division. Cell cycle progression is tightly regulated by components of a control system, specifically cyclins and cyclin-dependent kinases (Cdks). The essential activity of Cdks involves catalyzing the transfer of a gamma phosphate group from an ATP molecule to a Ser/Thr residue on substrates required for cell cycle progression, and this activity is primarily activated by cyclin binding. While Cdks and cyclins regulate the cell division cycle, some Cdks also form complexes with a small protein known as Cks (Cdc2 kinase subunit). Cks proteins are small proteins (8-12 kDa) that are essential for Cdk function and cell division in metazoans, although their precise roles are not yet fully understood. Although there has been extensive research on the cell cycle and its regulatory components in metazoans, the understanding of cell cycle regulation and regulators in protists, such as *Trichomonas vaginalis*, remains limited.

T. vaginalis is a microaerophilic protozoan of early evolutionary divergence that cause trichomoniasis, the most common non-viral sexually transmitted infection worldwide. Our group has identified and characterize several Cdks and cyclins in *T. vaginalis*, highlighting the importance of studying Cks proteins. Two Cks proteins in *T. vaginalis*, referred to as TvCKS1 and TvCKS2, have been identified. Previous work using yeast two-hybrid assays showed that both TvCKS1 and TvCKS2 can interact with three TvCRKs (*T. vaginalis* Cdks), suggesting that these proteins play a role in the cell cycle regulation in *T. vaginalis*. Additionally, preliminary data demonstrated that only TvCKS1 or TvCKS2 fused to the DNA-binding domain activate the transcription of both *lacZ* and *LEU2* gene reporters, indicating their potential activity as transcriptional activators.

On the other hand, yeast Cks1 is essential for the cell viability as it is required for G1/S and G2/M phase transitions and budding. To perform complementation assays, a *S. cerevisiae* CKS1/*cks1*Δ mutant strain was generated. Preliminary results from random spore assays suggest that both TvCKS1 and TvCKS2 can complement this mutant strain.

Our results indicate that there are two Cks proteins in *T. vaginalis* that share features with canonical Cks and need to be further characterized. Additionally, these molecules may play an important role as subunit regulators of TvCRKs, presumably involved in the cell cycle regulation in *T. vaginalis*.

ALDOSTERONE EFFECTS ON THE EXPRESSION OF RYANODINE RECEPTORS AND THEIR PARTICIPATION IN THE CALCIUM DYNAMICS OF RAT MESENTERIC ARTERIES

Hiram Lozano-Ruiz¹, Cristian J. Arriero-Carrillo¹, Agustín Guerrero-Hernández¹ and Angélica Rueda¹

¹Centro de Investigación y de Estudios Avanzados del IPN, Departamento de Bioquímica, Av. IPN 2508, Col. San Pedro Zacatenco, GAM. CP. 07360, México.

E-mail: arueda@cinvestav.mx

Intracellular calcium (Ca^{2+}) signals play a key role in the contraction and relaxation of vascular smooth muscle cells (VSMC), regulating the diameter of resistance mesenteric arteries (MA), hence blood flow. Phenylephrine (PE), an agonist of α_{1A} adrenergic receptors, produces intracellular Ca^{2+} oscillations (Ca^{2+} osc) which are periodic and uniform Ca^{2+} release events from the Sarcoplasmic Reticulum (SR) of VSMCs. The contribution of the intracellular Ca^{2+} channel/ryanodine receptors (RyR) to the PE-induced Ca^{2+} osc of Aldo-treated MA has not been studied.

The mineralocorticoid hormone Aldosterone (Aldo) increases the expression of both L-type voltage-dependent Ca^{2+} channels and SERCA2 pump in rat MA, increasing the frequency of Ca^{2+} sparks. These which are local Ca^{2+} release events produced by the activity of RyRs^[2]. However, whether Aldo modifies the expression of vascular RyR isoforms (RyR1, RyR2, and RyR3) or their subcellular distribution in VSMCs of Mas is unknown. Therefore, in this work, we evaluated the effect of Aldo treatment (10 nM, 24 h) in the expression and subcellular distribution of RyR isoforms, and the contribution of RyRs in PE-induced Ca^{2+} osc.

Our findings reveal a novel aspect of Aldo-treated VSMCs of MAs. The results show that the amplitude of PE-induced Ca^{2+} osc (PE 1 μM) was significantly increased in Aldo-treated VSMCs compared to control cells ($\Delta F/F_0 = 2.77 \pm 0.18$, n=17 control cells vs 3.72 ± 0.35 , n=18 Aldo-treated cells, $P < 0.05$), without changes in the Ca^{2+} osc frequency (Ca^{2+} osc/min = 0.44 ± 0.08 , n=17 control cells vs 0.31 ± 0.04 , n=18 Aldo-treated cells, $P = 0.14$). The pre-incubation of MA with ryanodine (100 μM , 30 min) and the application of caffeine (20 mM) at the end of the pre-incubation period (to block RyRs), inhibited the PE-induced Ca^{2+} oscs in 100% of VSMCs of both conditions (n=18 cells for each experimental group), arguing for a key role of RyRs in the generation of the intracellular Ca^{2+} osc independently of the Aldo treatment. Interestingly, Aldo had these effects without modifying the protein expression or the mRNA levels of RyR isoforms (RyR1, RyR2, and RyR3). Further work is needed to determine whether Aldo treatment alters the subcellular distribution of each RyR isoform.

This research holds significant implications for understanding the role of RyRs in the PE-induced Ca^{2+} osc of Aldo-treated MAs. By shedding light on the complex interplay between Aldo and RyRs, our study paves the way for potential therapeutic interventions targeting these pathways.

Work partially supported by Fondo Sectorial de Investigación para la Educación de Conahcyt (Basic Sciences project #A1-S-9082).

References

1. Gutiérrez A, Contreras C, Sánchez A, & Prieto D. (2019). *Frontiers in Physiology*, **10**: 55.
2. Salazar-Enciso R, Guerrero-Hernández A, Gómez AM, Benitah JP, & Rueda A. (2022). *Frontiers in Physiology*, **13**: 834220.

MITOCHONDRIAL IRON METABOLISM IS DISRUPTED IN A MURINE MODEL OF LIVER STEATOSIS

Luévano Martínez, L.A.^{1*}, A.C. Cepeda¹, Pérez-Cabeza de Vaca, R.¹, García Rivas G. de J.¹

Institute for Obesity Research. Tecnológico de Monterrey. Hospital Zambrano-Hellion. Av. Batallón de San Patricio 112, Real San Agustín, 66278 San Pedro Garza García, N.L.
Tel: +52.81.8888.2161. E-mail: aluevano@tec.mx.

Objective. Iron is an essential metal found in several metalloproteins involved in respiration, oxygen transport, control of oxidants and DNA maintenance. Accumulating evidence shows that obesity predisposes individuals to alteration in iron homeostasis in non-hematopoietic tissues. Mitochondria plays a key role in iron homeostasis since iron-sulfur clusters and hemes are synthesized within this organelle. However, despite the evidence showing iron homeostasis alterations in obesity, the information concerning the role that mitochondria play in iron homeostasis in obesity is scarce.

Methods. Livers from mice fed with a high fat diet (HFD, 60% cal from fat) were excised to isolate mitochondria and cytosol by differential centrifugation. Both fractions were analyzed for heme, non-heme, and labile iron. Mitochondrial respiratory function was determined by high resolution oximetry. Expression of target proteins was determined by western blot.

Results. Livers from HFD-animals present early signs of steatosis as observed by the presence of lipid bodies. Although fibrosis is still not profoundly settled. We observed a compromised respiratory activity in these mitochondria. Although the protein content of respiratory complex II, IV and cytochrome c were not affected. Preliminarily, we found that HFD induces a dysregulation in iron metabolism mainly at a mitochondrial level where we found an increase in the labile iron pool accompanied with oxidative damage in lipids and proteins. Moreover, two enzymes involved in heme metabolism, heme oxygenase-1 (degradation) and ferrochelatase (biosynthesis) were upregulated regulated in livers from HFD-animals at early stages of NAFLD. But interestingly, as time goes the expression of ferrochelatase decreases while the activity of heme oxygenase-1 increases. Additionally, we found that frataxin, an important mitochondrial iron chaperone involved in iron-sulfur cluster and heme biosynthesis, was downregulated in NAFL both effects could explain the increase in the labile iron pool in this fraction.

Conclusions. HFD alters the iron metabolism in livers but only at mitochondrial level. We proposed that the parallel increase in heme oxygenase and ferrochelatase could be a mechanism to avoid iron overload into mitochondria at early stages of liver steatosis. But as the diseases progresses hepatocytes lost the ability to manage iron leading to mitochondrial accumulation.

Keywords: *Iron, liver, mitochondria*

PEPTIDE VSAK DERIVED FROM THE C-TERMINAL REGION OF CETPI, ATTENUATES CELLULAR RESPONSES ASSOCIATED WITH LPS

Ismael Luna-Reyes¹, Erendira Perez-Hernandez¹, Blanca A. Delgado-Coello¹, Alejandro Zentella Dehesa², Miguel A. Avila-Rodriguez³ and Jaime Mas-Oliva¹

¹Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, 04510 Ciudad de México. ²Instituto Nacional de Ciencias Médicas y Nutrición "Salvador Zubirán", 14080 Ciudad de México. ³Unidad Radiofarmacia-Ciclotrón UNAM, Universidad Nacional Autónoma de México, 04360 Ciudad de México.
JM-O; e-mail: jmas@ifc.unam.mx ; phone: (+52) 55 5622 5584

Globally, sepsis and septic shock are among the most important health issues and leading causes of death in Intensive Care Units. Although both conditions are extremely complex, their development is clearly related to an unregulated response due to an infection. Mainly, Gram-negative bacteria are recognized as causal agents of sepsis and septic shock. Given many molecules present in bacteria, such as LPS (lipopolysaccharides), are potent immune stimulators, they trigger an excessive immune response that eventually could progress to sepsis and septic shock^{1,2}. In this regard, there is an urgent need for new strategies to tackle these conditions. Many years ago, our research group described the Cholesteryl-Ester Transfer Protein Isoform (CETPI), an isoform of CETP, which lacks of cholesterol transfer activity due to the substitution of the last 18 amino acids of the CETPI C-terminal region³. Afterwards, we found that VSAK-peptide, comprising those 18 residues, binds LPS with high affinity. *In vivo*, VSAK has demonstrated its ability to reduce the systemic response induced by LPS administration, including inflammatory markers and metabolic dysfunction⁴. Recently, we have tested the effects of VSAK treatment on macrophages and endothelial cell lines. These cells were treated with LPS and VSAK-peptide, to study the effect on the expression of inflammation markers, adhesion molecules, and endothelial activation markers. The expression of all marker molecules was reduced when cells were treated with LPS and VSAK-peptide in comparison with cells treated only with LPS. This effect could be mainly attributable to a reduced activation of TLR4 by LPS, due in turn to the competitive binding of VSAK to LPS. Interestingly, VSAK was also able to reduce the effect of Neoseptin-3, used along the assays as a positive control for TLR4 activation. The effects observed *in vitro* support the role of VSAK to mitigate the effects of LPS, which alongside traditional antibiotic therapy has shown to reduce the exaggerated activation of the immune system that could lead to unregulated host responses. Our observations collectively suggest that VSAK holds promise as a potential therapeutic agent for mitigating the harmful effects of LPS.

References

1. Luna-Reyes, I, Pérez-Hernández, EG, Delgado-Coello, B, Mas-Oliva, J.(2021). Arch Med Res 52, 798–807.
2. Pérez-Hernández, EG, De la Puente-Díaz de León, V, Luna-Reyes, I et al.(2022). Mol Med 28, 157.
3. Pérez-Hernández, EG, Delgado-Coello B, Luna-Reyes I, Mas-Oliva J.(2021) Biomed Pharmacother. 141:111890.
4. Luna-Reyes, I, Pérez-Hernández, EG, Delgado-Coello, B, Ávila-Rodríguez, MA, Mas-Oliva, J. (2021). Sci Rep 11,1-11.

A COMPUTATIONAL AND EXPERIMENTAL APPROACH TO DEVELOP A FRET-BASED SUBSTRATE FOR ASSESSING THE PROTEOLITIC ACTIVITY OF MYCOBACTERIUM TUBERCULOSIS MARP

Pablo A. Madero-Ayala¹, Rosa E. Mares-Alejandre¹, Patricia L.A. Muñoz-Muñoz¹, Víctor García González², Samuel G. Meléndez-López¹, Marco A. Ramos-Ibarra¹

¹Facultad de Ciencias Químicas e Ingeniería, Universidad Autónoma de Baja California, Tijuana, BC 22390.

²Facultad de Medicina-Mexicali, Universidad Autónoma de Baja California, Mexicali, BC 21000, México.

E-mail: pablo.madero@uabc.edu.mx; mramos@uabc.edu.mx.

Mycobacterium tuberculosis (Mtb), the causative agent of tuberculosis (TB) in humans, is an obligate intracellular bacterium. TB is one of the deadliest diseases worldwide, affecting approximately one-quarter of the global population.¹ Mtb infection begins when aerosol particles containing the pathogen are generated by individuals with active disease and deposited in the lungs of another (new, naïve) host. Once in the alveoli, mycobacteria are phagocytosed by macrophages and sequestered within a phagosome, where they are usually eliminated owing to their acquired bactericidal properties. However, Mtb possesses several evasion and resistance mechanisms against the immune system, allowing it to persist and reproduce in the host.² The Mtb survival mechanism involves inhibition of phagolysosome formation. However, macrophages activated by IFN- γ can overcome this inhibition and promote acidification, which is an important anti-tuberculosis mechanism. Several reports have indicated that Mtb is capable of persisting in low intracellular pH conditions, suggesting that it has specific mechanisms of resistance to acidic environment.³ One protein involved in pH homeostasis is MarP, a membrane serine protease encoded by the Rv3671c gene². Mtb mutants lacking MarP are non-dividing cells hypersensitive to acidic pH, indicating that the protein is required for the optimal separation of dividing cells at low pH.³ This inability of mycobacteria results in attenuated survival and viability. Despite the importance of MarP in Mtb pathobiology, full characterization of its enzymatic activity remains pending because of the lack of a bioassay that allows the assessment of its catalytic properties and susceptibility to inhibition by specific molecules. Therefore, we developed a FRET-based bioassay as a methodological alternative for the biochemical characterization of *M. tuberculosis* MarP and as a technological platform for the search for specific inhibitors.

References

1. Combating the global TB epidemic. (2024). Centers for Disease Control and Prevention. <https://www.cdc.gov/globalhivtb/who-we-are/about-us/globaltb/globaltb.html>
2. Biswas T, Small J, Vandal O, Odaira T, Deng H, Ehrt S, Tsodikov OV (2010). *Structure*, 18(10), 1353–1363.
3. Botella H, Vaubourgeix J, Lee MH, Song N, Xu W, Makinoshima H, Glickman MS, Ehrt S (2017). *The EMBO Journal*, 36(4), 536–548.
4. Vandal OH, Pierini LM, Schnappinger D, Nathan CF, Ehrt S (2008). *Nature Medicine*, 14(8), 849–854.

EFFECT OF A TRIAZASPIRAN-TYPE MOLECULE ON MIGRATION AND INVASION OF MDA-MB-231 TUMOR CELLS

Jesús Omar Medina Cruz¹, Javier de Jesús Vasconcelos-Ulloa¹, Georgina Quintana-González¹, Bryan Alexis Rivera-Suárez¹, Héctor Frayde-Gómez¹, Octavio Galindo-Hernández¹

¹Facultad de Medicina Mexicali, Universidad Autónoma de Baja California, Mexicali 21000, Baja California, México.
E-mail: octavio.galindo@uabc.edu.mx

Breast cancer is the most common carcinoma in women worldwide, being the first cause of death from neoplasia in women, exceeding 500,000 cases annually. In Mexico, an incidence of 29,929 cases was reported in 2020¹. Several therapeutic options are currently available, however, in patients with triple-negative breast cancer (TNBC) these are limited and have multiple side effects. Furthermore, this type of cancer is very aggressive, with a poor prognosis and survival². Therefore, the design and generation of novel treatments that improve the prognosis of patients with TNBC are necessary. Aim. Characterize the effect of the 8-Benzyl-1,3,8-triazaspiro-[4.5]-decane-2,4-dione (triazaspirane) on the migration and invasion processes in MDA-MB-231 tumor cells. Material and methods. By using MDA-MB-231 cancer cells as a TNBC model, the potential inhibitory effect of triazaspirane on migratory capacity will be evaluated by wound healing assay; the invasion will be evaluated through Boyden chambers assays. The data will be statistically analyzed through the GraphPad Prism 8 program (version 8.0.2) using the one-way ANOVA test compared by Dunette's multiple test. The statistical probability $P < 0.05$ will be considered significant. Results. The biological activity assays demonstrated an inhibitory effect on cell migration and invasion. Therefore, Triazaspirane-type molecules have important inhibitory effects on the mechanisms associated with metastasis in MDA-MB-231 breast cancer cells.

References

1. Sung, H.; Ferlay, J.; Siegel, R.L.; Laversanne, M.; Soerjomataram, I.; Jemal, A.; Bray, F (2020). Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. CA. Cancer J. Clin. n/a, doi: <https://doi.org/10.3322/caac.21660>.
2. Pareja F, Geyer FC, Marchiò C, Burke KA, Weigelt B, Reis-Filho JS (2016). Triple-negative breast cancer: the importance of molecular and histologic subtyping, and recognition of low-grade variants. Npj Breast Cancer. 2(1):16036.

PHENOLICS PROFILING OF NATIVE CHROMATIC CORN VARIETIES

Liliana Hortencia Méndez Barredo¹, Juan Luis Monribot Villanueva¹,
José Antonio Guerrero Analco¹, Eliel Ruiz-May¹

¹Instituto de Ecología A. C. Carretera antigua a Coatepec 351, Col. El Haya, Xalapa, Veracruz. C.P. 91073.
E-mail: liliana.mendez@inecol.mx, eliel.ruiz@inecol.mx, Telephone number: +52 5577108822

Maize (*Zea mays* L.) is a human and animal critical crop that supports food ¹. Pigmented maize contains many secondary metabolites, such as phenolic compounds, carotenoids, and tocopherols^{2,3}. The present study aims to perform a phenolics profiling in native maize of three different colours (red, white, and purple) from the states of Chiapas and Michoacán, highlighting the added value of coloured grains. The phenolics profiling was performed on an ultra-high-performance liquid chromatography coupled with tandem mass spectrometry (UPLC-MS/MS) using a dynamic multiple reaction monitoring method.

This study provides valuable insights into the phenolics diversity of native maize varieties. The high phenolic compounds content in purple maize highlights its potential as a functional food with important health benefits. These findings support the strategic importance of promoting and preserving colored native maize varieties due to their bioactive properties and contributions to food security and agricultural sustainability. Our results offer a comprehensive insight into the analyzed samples, highlighting patterns, similarities, and differences among native maize, as well as the presence of phenolic subgroups within. Twenty-six phenolic compounds were identified and quantified in the samples. Compounds such as protocatechuic acid, rutin, quercetin, vanillic acid, quercetin-3-glucoside, kuromanin, 4-hydroxybenzoic acid, naringenin, and (+)- catechin emerge as pivotal for distinguishing purple maize from Chiapas and Michoacán states compared to other maize varieties. The phenolic diversity observed in red maize from Chiapas suggests the presence of unique compounds.

References

1. Mejía, D. Maize Post-Harvest Operations. Food and Agriculture Organization of the United Nation 2003, 1-100.
2. Hu, Q.P.; Xu, J.G. J Agric Food Chem 2011, 59, 2026–2033, doi:10.1021/jf104149q.
3. Žilić, S.; Serpen, A.; Akillioğlu, G.; Gökmen, V.; Vančetović, J. J Agric Food Chem 2012, 60, 1224–1231.

WOLBACHIA SPP. INFECTION IN DROSOPHILA MELANOGASTER

Ofelia Méndez-Romero¹, Edson Mosqueda-Martínez¹, Carolina Ricardez-García¹,
Natalia Chiquete-Félix¹, Salvador Uribe-Carvajal¹

¹Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Mexico City, Mexico.
E-mail: suribe@ifc.unam.mx

Wolbachia pipientis is a Rickettsial bacterium that infects a wide variety of insects, including mosquitoes, fruit flies, and butterflies. It is transmitted maternally. In mutualistic associations, *Wolbachia* can provide the host with metabolites or assist in maintaining the chemical homeostasis of the host cell¹. The main species of *Wolbachia* that infect *Drosophila spp.* are *Wb_{pop}* (*Drosophila simulans*) and *Wb_{Mel}* (*Drosophila melanogaster*)². In the fly, *Wb_{pop}* has higher bacterial density, cytoplasmic incompatibility, vertical transmission, and invasion capacity than *Wb_{Mel}*. Additionally, it significantly enhances fertility and causes cytological defects. These differences can impact the interaction between *Wolbachia* and *Drosophila*, potentially leading to bioenergetic changes in the host³. In this study, mitochondria were isolated from Wt, *Wb_{pop}* and *Wb_{Mel}*, and it was found that they were functionally coupled, with a respiratory control ratio (RCR) of 1.99 (Wt), 1.62 (*Wb_{pop}*), and 1.79 (*Wb_{Mel}*), indicating efficient oxidative phosphorylation and adequate energy metabolism in isolated mitochondria. In *Drosophila* mitochondria infected with *Wb_{pop}* oxygen consumption was enhanced to a higher degree than that infected with *Wb_{Mel}*. However, the transmembrane potential was higher in Wt than in infected mitochondria. Additionally, significant differences in the activity of canonical complexes were observed. In *Wb_{pop}* and *Wb_{Mel}* activity of Complex I and V were lower than Wt, but the activity of CIV was higher in infected mitochondria. The mechanism for these effects is currently under study.

References

1. Bishop, C. and Asgari, S. (2021). *Molecular Microbiology*, 115, 1229-1243.
2. Bubnell, J. E., Fernandez-Begne, P., Ulbing, C. K. S. and Aquadro, C. F. (2021). *G3*, 11, 1-12.
3. Zhukova, M. V. and Kiseleva, E. (2012). *BMC Microbiol*, 12, 1-12.

PITAVASTATIN: ITS POTENTIAL ADVERSE EFFECTS EVALUATED IN HYPERCHOLESTEROLEMIC CD-1 MALE MICE

Rodrigo Miranda Zamora¹, Isela Esther Juárez Rojop², Andrés Eliu Castell Rodríguez³,
Jorge Luis Blé Castillo², Juan Cuauhtémoc Díaz Zagoya^{1*}

¹Departamento de Bioquímica, División de investigación, Facultad de Medicina, Universidad Nacional Autónoma de México, México. ²División Académica de Ciencias de la Salud, Universidad Juárez Autónoma de Tabasco, Villahermosa, Tabasco, México.

³Departamento de Biología Celular y Tisular, División de investigación, Facultad de Medicina, Universidad Nacional Autónoma de México, México.

*Juan Cuauhtémoc Díaz Zagoya, Av. Universidad 3000, Ciudad Universitaria, Ciudad de México, CP 04510, Tel. 5556232174, zagoya@unam.mx

Statins remain as the cornerstone for the treatment of hypercholesterolemia, a major risk factor for cardiovascular disease. According to INEGI's 2022 report the first cause of death in the Mexican population was cardiovascular disease.

Pitavastatin (PTV) lowers endogenous cholesterol synthesis but it also has potential adverse effects that were evaluated in hepatocyte mitochondria from hypercholesterolemic mice. The animals received a daily dose of 0.03, 0.06, 0.09 or 0.4 mg/Kg, along with a control diet or hypercholesterolemic diet, during 15 or 50 days. The animals were euthanized and the morphology and respiratory function of hepatocyte mitochondria was evaluated. After the 50 day treatment with 0.09 mg/Kg/day PTV or lower doses plus a control diet or a cholesterol-rich diet, the oxygen consumption was similar to that of the control group (42.36 nmol/min/mg protein). The body weight gain after the 15 or 50 day treatment was lower in all experimental groups. There were no deaths in all groups, except in the 0.4 mg/Kg/day PTV plus hypercholesterolemic diet (42.9%). This dose is six times of that used in humans (0.066 mg/Kg/day if we consider 4 mg ATV a day for a 60 Kg person) PTV is the most active statin of those available on the market; according to our preliminary results it is safe at therapeutic doses. We are still missing the results of the mitochondria microscopic observation.

CHARACTERIZATION OF SUCROSE SYNTHASE GENE FAMILY IN COMMON BEAN (*PHASEOLUS VULGARIS* L.) SUBJECTED TO MOISTURE RESTRICTION DURING POD FILLING

Norma Cecilia Morales-Elias¹, Eleazar Martínez-Barajas³, Monserrat Vázquez-Sánchez¹, Antonio García-Esteva¹, Iris Grisel Galván-Escobedo² and Daniel Padilla-Chacón^{2*}

¹Programa de Posgrado en Botánica, Colegio de Postgraduados. Carretera México-Texcoco, km 36.5, Montecillo, 56264, México.

²Programa de Posgrado en Botánica, CONAHACYT-Colegio de Postgraduados. Carretera México-Texcoco, km 36.5, Montecillo, 56264, México. daniel.padilla@colpos.mx. ³Departamento de Bioquímica, Facultad de Química. Universidad Nacional Autónoma de México. Ciudad Universitaria. Coyoacán. Ciudad de Mexico, 03000, México.

Sucrose synthase (SUS, EC 2.4.1.13) is considered as a key enzyme involved in plant sucrose metabolism which gene family has been identified and characterized in several plant species. However, to date scant information about the SUS genes is available in common bean (*Phaseolus vulgaris* L.). Here, we identified seven PvSUSs genes in common bean located on six different chromosomes. The proteins sequences showed conserved catalytic domains with molecular weights and lengths ranging from 90.5 to 105.1 kDa from 799 to 929 amino acids, respectively. Gene structure analysis indicated that the PvSUSs have similar exon-intron structures. Phylogenetic analysis revealed that the seven members could be classified into three groups (PvSUS1 and PvSUS2 in I, PvSUS5, PvSUS6 and PvSUS7 in II, and PvSUS3 and PvSUS4 in III), demonstrating evolutionary conservation in the SUS family across common bean and other plant species. According to results performed using simulation of molecular docking, the catalytic residues are conserved in all seven PvSUS isoforms, except for PvSUS3 where Arg-580 is substituted by Lys. The binding energy of UDP-glucose and fructose interaction values range among -6.45 to -7.7 Kcal/mol. In contrast, the dissociation constant (Kd) which reflects the ligand's affinity for all docked complexes revealed values of 2.01 μ M, 2.5 μ M and 1.54 μ M for PvSUS3, PvSUS7, PvSUS4 respectively. The 3D shape of the sequences allows us to visualize the supposed biological form and function. According to the functions of cis-elements all the PvSUSs promoters possessed at least one for abiotic/biotic elements such as drought-responsive elements MYB and MYC. Although significant progress in improving drought resistance of beans has been registered, little information exists on the role of pod wall as important contributions during pod filling, especially under terminal drought. In this study, we use *Phaseolus vulgaris* cv. OTI under drought stress to explore the metabolic responses of pod walls after 5, 10, 15, and 20 days of moisture restriction (MR) after reproductive stage eight (R8). The expression of PvSUSs on pods wall subjected the effect of terminal drought were investigated via real-time PCR. The results showed that PvSUS1, PvSUS3, and PvSUS4 were remarkably expressed during seed development under MR. Biochemical characterization showed that under MR, SUS activity increase associated with high levels of fructose while sucrose increased, hexoses decreased reciprocally. Fluorescent esculin were used, which is recognized by sucrose transporters. Cross sections of pods at 50 % field capacity (FC) revealed a much weaker esculin signal in the phloem of pods at 100 % FC. Our results provide new insights into physiological functions of SUS genes in common bean, especially roles in regulating sugar accumulation in common bean. These findings open new opportunities to research the sucrose distribution mechanism under moisture restriction.

ANALYSIS OF FATTY ACID TRANSPORT MEDIATED BY THE MCT11 TRANSPORTER.

Nicole Justine Moreno Licona¹, Zuleima Natali Domínguez Velazquez^{2,3},
Edgar Morales Ríos¹, Viridiana Olin Sandoval²

¹Department of Biochemistry-CINVESTAV, Zacatenco. ²Department of Biotechnology and Bioengineering-CINVESTAV, Zacatenco. ³Biomedical Sciences-UNAM
E-mail: nicole.moreno@cinvestav.mx, edgar.morales@cinvestav.mx, viridiana.olin@cinvestav.mx

The Monocarboxylate transporter (MCT) family is comprised of 14 members, classified in 2 categories according to transport mechanism. Nevertheless, not all members of this family have been fully characterized. That is the case of the MCT11 transporter, a diabetes association study identified a new risk haplotype (consisting of 5 SNPs) in the coding region of the *SLC16A11* gene that encodes for this transporter¹. Currently, the ligand of this transporter has not been identified and its relevance in the pathophysiology of diabetes is not yet understood. Metabolic studies have associated the phenotype of this haplotype with alterations in lipid metabolism^{2,3}. Thus, the aim of this project is to identify candidate ligands of the MCT11 transporter using an *in silico* approach and further experimentally verifying the results. First, with an AlphaFold structural model of MCT11, we proposed a transport site with the highest probability of appearance residues inside the pocket in conserved models and tested 200 substrates with multiple physicochemical characteristics that could be candidates for transport in docking molecular. Our results show that the proposed pore has hydrophobic characteristics and great affinity for high molecular weight monocarboxylate^{2,3}. To enrich our results, we have implemented molecular dynamics analysis with 6 target ligands chosen by the docking experiments, which provides us with information on the possible interactions of the ligand with residues of the transport pore. These results give us support in the search for the MCT11 transporter ligand. Besides, to corroborate the *in silico* analysis results, we are performing binding experiments using whole cell thermostability assays⁴ in Hek293 cells expressing the MCT11 transporter. Additionally, we will perform transport experiments using the selected ligands.

References

1. The SIGMA Type 2 Diabetes Consortium. (2014). Sequence variants in *SLC16A11* are a common risk factor for type 2 diabetes in Mexico. *Nature*, 506 97–101.
2. Rusu, V., Hoch, E., Mercader, J. M., Tenen, D. E., Gymrek, M., Hartigan, C. R., . . . Guzman, G. (2017). Type 2 Diabetes Variants Disrupt Function of *SLC16A11* through two distinct mechanisms. *Cell Press*, 170 199–212.
3. Zhao, Y., Feng, Z., Zhang, Y., Sun, Y., Chen, Y., Liu, X., . . . Wei, Y. (2019). Gain-of-Function Mutations of *SLC16A11* Contribute to the Pathogenesis of Type 2 Diabetes. *Cell Reports*, 26 884–892.
4. Hashimoto M, Girardi E, Eichner R, Superti-Furga G. (2018) Detection of Chemical Engagement of Solute Carrier Proteins by a Cellular Thermal Shift Assay. *ACS Chem Biol*. 15 1480-1486.

CHARACTERIZATION OF THE ROLES PRIMPOL IN DNA INTEGRITY MAINTAINING IN *ARABIDOPSIS THALIANA*

Johan Olalde-Hernández^{1,2}, Daniela Camacho-Manriquez², Karem Sánchez Martínez^{1,2},
Fernando Guzmán-Chávez³, Luis Briebe², José Antonio Pedroza-García^{1*}

¹Departamento de Bioquímica, Facultad de Química, UNAM, ²Langebio-Cinvestav Irapuato,

³CONAHCYT-Instituto de Investigaciones Biomédicas, UNAM.

e-mail: johan040701@gmail.com; *joped@quimica.unam.mx

PrimPol, short for “DNA Primase-Polymerase,” in human this enzyme was recently discovered, it can perform the roles of both primase and DNA polymerase on its own. Their functions are crucial to the DNA damage tolerance and repair as nucleus as mitochondria, it initiates the synthesis RNA or DNA primers de novo and extending from them while acting as a DNA polymerase. Moreover, it has been reported that its primase function is primordial for other DNA polymerases involved in repair mechanisms¹. In contrast to mammalian cells, PRIMPOL has not been extensively studied in plants, we previously have demonstrated that *Arabidopsis* PRIMPOL has activities of Primase and DNA polymerase in *in vitro* assays, and it showed triple subcellular location in nucleus, mitochondria, and chloroplast². Therefore, the aim of our work is to characterize *in vivo* the functions of *Arabidopsis thaliana* PRIMPOL in nucleus, mitochondria, and chloroplast. Firstly, three T-DNA mutant lines (*pp1*, *pp6*, and *pp7*) were isolated which were knockdown mutants, such plants did not display differences on phenotype in comparison to wild-type plants in normal grow conditions. However, lack of the DNA polymerase POLIA, one out of two DNA polymerases that carry out the DNA replication and repair in mitochondria and chloroplast lead to loss viability of *pp6* and *pp7* mutants, being that sesquimutants plants *pollA* (-/-) X *pp6* (+/-) showed around 25% of aborted embryos. Similar results were observed in crosses with a mutant of the DNA polymerase ϵ , whose function is the synthesis of leading strand during replication of nuclear DNA. Additionally, we generated PRIMPOL overexpressing lines and CRISPR-Cas9 mutants, which will contribute to our studies. Until now, our results have demonstrated the importance of plant PRIMPOL in response to DNA damage in nucleus and beyond.

References

1. Stephanie Tirman, Emily Cybulla, Annabel Quinet, Alice Meroni, Alessandro Vindigni. (2021). Critical Reviews in Biochemistry and Molecular Biology, 56, 17-30.
2. Paola L. García-Medel, Antolín Peralta-Castro, Noe Baruch-Torres, Alma Fuentes-Pascacio, José A. Pedroza-García, Alfredo Cruz-Ramirez, Luis G. Briebe. (2021). scientific reports, 11, 1-14.

METABOLIC FACTORS ASSOCIATED WITH METABOLICALLY HEALTHY OBESITY

Marisol Olivares-Arévalo¹, Samuel Canizales-Quinteros¹, Paola V. León-Mimila¹, Hugo Villamil-Ramírez¹

¹Unidad de Genómica de Poblaciones Aplicada a la Salud (UNAM-INMEGEN),
Periférico Sur 4809, Arenal Tepepan, Talpan. C.P. 14610. Mexico City, Mexico
E-mail: solmar.0413@gmail.com

Author corresponding: Canizales-Quinteros, Samuel. E-mail: cani@unam.mx

Abstract. Increased visceral adipose tissue is considered a major risk factor for insulin resistance and metabolic syndrome, probably induced by elevated free fatty acid levels^{1,2}. Some subjects with obesity do not have metabolic co-morbidities, that is, approximately 10% are metabolically healthy (MHO)³. The factors associated with the absence of metabolic complications and the presence of visceral adiposity in obese individuals is poorly understood. The aim of this study to compare the levels of FFA and adipokines in two groups of individuals with obesity and visceral adiposity: individuals with metabolic complications (metabolically unhealthy obesity, MUO), and individuals without complications (MHO). MHO was defined as obesity and visceral adiposity with fasting blood glucose level <100mg/dL, systolic blood pressure <130mmHg and/or diastolic <85mm Hg, high-density lipoprotein cholesterol > 50 mg/dL for women and > 40 mg/dL for men. Ninety-eight subjects with MHO and 189 with MUO were included. Visceral adiposity was measured using the Metabolic Score for Visceral Fat (METS-VF)⁴. The MHO group showed 34% lower HOMA-IR (P<0.001) and 25.6% lower insulin secretion measured by C-peptide levels (P<0.001). Interestingly, serum free fatty acids concentrations were not significantly different at MHO and MUO subjects. These results suggest that others factors independent of free fatty acid serum concentrations, could mediate the insulin sensitivity and metabolic complications in obesity. For this reason, we evaluated the serum concentrations of resistin, leptin and adiponectin. Only adiponectin levels showed significant differences, as this adipokine was increased 30% in the MHO (median 6.0, interquartile range 4.5-8.8 µg/mL) as compared to the MUO group (4.2, interquartile range 3.1-6.4 µg/mL; P<0.001). These results are consistent with studies that suggest that adiponectin improves insulin sensitivity and maintains healthy adipose tissue expansion⁵. However, the mechanisms regulating adiponectin levels in the presence of visceral adiposity have not been described. Genetics variants may contribute to modulate the concentrations of this hormone and its association with metabolically healthy obesity.

References

1. Smith, Gordon I., et al. (2019). "Metabolically Healthy Obesity: Facts and Fantasies." *Journal of Clinical Investigation*, **vol. 129**, pp. 3978–89.
2. Hagberg, C., Spalding, K. (2024). "White Adipocyte Dysfunction and Obesity-Associated Pathologies in Humans." *Nature Reviews Molecular Cell Biology*, **vol. 25**, pp. 270–89.
3. Wang, Jiang-Shui., et al. (2023). "Trends in the Prevalence of Metabolically Healthy Obesity Among US Adults, 1999-2018". *JAMA Network Open*, **vol. 6**, pp. 2-15.
4. Bello-Chavolla, O., et al. (2020). "Metabolic Score for Visceral Fat (METS-VF), a Novel Estimator of Intra-Abdominal Fat Content and Cardio-Metabolic Health." *Clinical Nutrition*, **vol. 39**, pp. 1613–21.
5. Aguilar-Salinas CA, et al. (2008). "High adiponectin concentrations are associated with the metabolically healthy obese phenotype". *J Clin Endocrinol Metab.* **Vol. 93**, pp. 4075-9

A NEW ER Ca^{2+} RELEASE AGENT SYNTHESIZED FROM 2-APB TESTED IN HELA CELLS

David Olvera¹, Marco González¹, María Teresa Percino², Manuel Arias³, Agustín Guerrero-Hernández¹

¹Biochemistry Dept. ²Chemistry Dept. Cinvestav, IPN Av 2508, Mexico City, 07360. ³Facultad de Estudios Superiores de Iztacala-UNAM, Tlalnepantla de Baz 54090, EdoMex, México.

E-mail: ol318497@uaeh.edu.mx

Ca^{2+} ion is essential in regulating various cellular processes such as neurotransmitter release, excitation-contraction coupling in muscle, proliferation, cell migration, and apoptosis, among others. The endoplasmic reticulum (ER) is the main intracellular Ca^{2+} reservoir, which possesses a Ca^{2+} toolkit for regulating this ion, such as Ca^{2+} release channels (i.e., IP_3R and RyR) and the SERCA pump¹.

Mikoshiha initially introduced 2-APB as a cell-permeable IP_3R inhibitor. However, it became an interesting modulator of Ca^{2+} entry through Orai1 channels, the so-called store-operated Ca^{2+} entry (SOCE); low concentrations stimulate while high concentrations inhibit SOCE. 2-APB has other targets besides SOCE and IP_3R ; for this reason, developing analogs of 2-APB with specific effects on SOCE would be desirable. One initial approach is adding halogens in different phenol ring positions. One such molecule is the *p*-Br-2APB that turned out to increase the $[\text{Ca}^{2+}]_i$ in the absence of external $[\text{Ca}^{2+}]$ while at the same time inhibiting the effect on $[\text{Ca}^{2+}]_i$ observed with thapsigargin (Tg), a SERCA pump inhibitor, in addition to being a better inhibitor of SOCE compared to 2-APB².

Therefore, to identify the ER ion channel targeted by *p*-Br-2-APB, we decided to characterize the intracellular Ca^{2+} dynamics in HeLa cells. We measured simultaneously the $[\text{Ca}^{2+}]_i$ with Fura-2 and the ER $[\text{Ca}^{2+}]$ with a GECI. The effect of 10 μM *p*-Br-2-APB induces an increase in $[\text{Ca}^{2+}]_i$ with a simultaneous reduction in $[\text{Ca}^{2+}]_{\text{ER}}$; however, a 100 times lower concentration (100 nM) slowly reduces the $[\text{Ca}^{2+}]_{\text{ER}}$ without any change in the $[\text{Ca}^{2+}]_i$. The data obtained with 100 nM *p*-Br-2-APB suggest that the ER Ca^{2+} leak is increased more at this concentration than the SERCA pump is inhibited. To determine whether the IP_3R was activated or not by *p*-Br-2-APB, HeLa cells were incubated with 20 mM of caffeine, which has been reported as an IP_3R inhibitor³, and the $[\text{Ca}^{2+}]_i$ and $[\text{Ca}^{2+}]_{\text{ER}}$ responses induced by 50 μM *p*-Br-2-APB was evaluated. The results suggest that *p*-Br-2-APB does not activate IP_3R . Since 2-APB modulates Orai channels, we investigated whether *p*-Br-2-APB activates the Orai1 or Orai3 channels to induce ER Ca^{2+} release. The dominant negative clones of these channels did not alter the increase of the $[\text{Ca}^{2+}]_i$ triggered by *p*-Br-2-APB, arguing against these channels being the target of *p*-Br-2-APB. In conclusion, *p*-Br-2-APB is a novel ER Ca^{2+} release agent in HeLa cells.

References

1. M.J. Berridge, M.D. Bootman, H.L. Roderick, *Nat Rev Mol Cell Biol* (2003), 4 517–529.
2. Schild A, Bhardwaj R, Wenger N, Tscherrig D, Kandasamy P, Dergiç J, Baur R, Peinelt C, Hediger MA, Lochner M. *International Journal of Molecular Sciences*. (2020); 21(16):5604.
3. Kang, SS, Han, KS, Ku, BM, Lee, YK, Hong, J, Shin, HY, Almonte, AG, Woo, DH, Brat, DJ, Hwang, EM, Yoo, SH, Chung, CK, Park, SH, Paek, SH, Roh, EJ, Lee, SJ, Park, JY, Traynelis, SF & Lee, *Cancer Research* (2010), vol. 70, no. 3, 1173-1183.

INDUCTION OF SYSTEMIC RESISTANCE IN ARABIDOPSIS AGAINST PHYTOPATHOGENS BY THE BENEFICIAL FUNGUS *TRICHODERMA ATROVIRIDE* THROUGH MODULATION OF SERINE BIOSYNTHESIS

Daniel David Pacheco Rodríguez¹, Saúl Jijón Moreno¹ and J. Sergio Casas-Flores¹

¹Instituto Potosino de Investigación Científica y Tecnológica (IPICYT). Camino a la Presa San José
No. 2055 Col. Lomas 4a. sección, C. P. 78216. San Luis Potosí, SLP.
E-mail: scasas@ipicyt.edu.mx

In their natural environment, plants are exposed to various types of stresses, both biotic and abiotic. Biotic stress includes attacks by pathogenic microorganisms, which plants counteract by activating their innate immune responses. The plant's immunity is further enhanced both locally and systemically by root-associated symbiont microorganisms, including beneficial fungi of the genus *Trichoderma*. The mechanisms by which *Trichoderma* spp. induce systemic resistance against pathogens have not yet been thoroughly described. During plant-microorganism interactions, alterations occur in the metabolism of amino acids related to plant defense. This study focused on analyzing whether the amino acid serine (Ser) induces systemic resistance in *Arabidopsis* against the necrotrophic pathogenic fungus *Botrytis cinerea* and the hemibiotrophic bacterium *Pseudomonas syringae* pv. *tomato* DC3000 (*Pst* DC3000). Additionally, it was evaluated the modulation of genes related to Ser biosynthesis in the plant induced by *Trichoderma atroviride*.

The results demonstrated that Ser induces systemic resistance in *Arabidopsis* against both *B. cinerea* and *Pst* DC3000. This resistance was correlated with the induction of genes associated with systemic acquired (SAR) and induced systemic resistance (ISR). When applied to the roots, Ser triggers hydrogen peroxide production in the leaves of *Arabidopsis*. The beneficial fungus *T. atroviride* as well as the phytopathogens *B. cinerea* and *Pst* DC3000 modulated the expression of *EDA9* and *GGAT1*, genes related to Ser biosynthesis in *Arabidopsis*. We hypothesized that the plant responds to the presence of beneficial microorganisms or pathogens through Ser biosynthesis, which contributes to the induction of systemic resistance. Finally, we found that the GGAT1 protein potentially acts as a positive regulator of plant immunity against *Pst* DC3000 and plays a minor role in the induction of systemic resistance mediated by *T. atroviride*.

Keywords: Systemic resistance; *Trichoderma*; *Arabidopsis*; serine; amino acids.

ALPHA AMYLASE INHIBITION BY SILVER NANOPARTICLES BIOSYNTHESIZED WITH *CNIDOSCOLUS ACONITIFOLIUS*

Eduardo Padilla Camberos¹, Angelica Sofia González Garibay¹, Ivan Sánchez Hernández¹
Biotecnología Médica y Farmacéutica, CIATEJ, A.C. Normalistas 800, Guadalajara, Jal., 44270, México
epadilla@ciatej.mx

Silver nanoparticles (AgNPs) have been studied for a long time for their various applications, one of which is their possible hypoglycemic effect which may be due to the inhibition of enzymes involved in carbohydrate metabolism that prevent the accumulation of glucose in the blood. Therefore, it has been proposed as a therapeutic approach in diabetic people¹.

Biosynthesis of AgNPs uses microorganisms or plant extracts which are used as a reducing and stabilizing agent. *C. aconitifolius* extracts can be used in the synthesis of AgNPs because it presents compounds that act as reducing agents such as flavonoids and phenolic compounds. Therefore, the objective of the present work was to evaluate the inhibition of the alpha amylase enzyme with AgNPS biosynthesized with an extract of leaves of *C. aconitifolius*.

The synthesis of AgNPS was carried out by reducing a 1.0 mM Silver Nitrate solution with the *C. aconitifolius* extract. The reaction was carried out at room temperature with continuous stirring. To estimate the particle size, a scan was performed from 300 to 650 nm in a spectrophotometer.

The enzyme inhibition assay was carried out with pancreatic alpha amylase enzyme dissolved in PBS. The mixture was incubated at 25 °C for 10 min, then a starch solution (0.5% w/v) was added, and the chromogen DNS (dinitrosalicylic acid) was added to the reaction, followed by incubation at 95-100 °C for 5 minutes and cooling to room temperature².

The sample absorbance was read at the wavelength of 540 nm in a plate reader. The results obtained in this work show that the aqueous extract of *C. aconitifolius* can react with Silver Nitrate forming nanoparticles, which is evident showing the highest absorbance value at a wavelength 430 nanometers which is indicative of a particle size between 10 and 15 nanometers because at that wavelength the plasmon resonance of the silver ion occurs.

The AgNPS synthesized with *C. aconitifolius* showed inhibition in the enzymatic activity of alpha amylase with values of 26 to 94% and an IC₅₀ value of 0.9 uL/mL. These results are like those reported in other investigations but with the advantage of using an environmentally friendly method.

In conclusion, the synthesis of AgNPS using a safe method without toxic solvents is possible with extracts from *C. aconitifolius* leaves. The inhibitory activity of alpha amylase may have an application as a regulator of high glucose levels in diabetic people.

References

1. Ashraf, J. M., Ansari, M. A., Khan, H. M., Alzohairy, M. A., & Choi, I. (2016). Green synthesis of silver nanoparticles and characterization of their inhibitory effects on AGEs formation using biophysical techniques. *Scientific reports*, 6, 20414.
2. Padilla-Camberos, E., Lazcano-Díaz, E., Flores-Fernandez, J. M., Owolabi, M. S., Allen, K., & Villanueva-Rodríguez, S. (2014). Evaluation of the inhibition of carbohydrate hydrolyzing enzymes, the antioxidant activity, and the polyphenolic content of *Citrus limetta* peel extract. *The Scientific World Journal*, 2014.

PROTEOMIC ANALYSIS OF THE RESPONSE TO GIBBERELIC ACID IN *CAPSICUM ANNUUM*

Diego de Jesús Pantoja Gutiérrez^{1,2}, Silvia Edith Valdés Rodríguez², Humberto R. Medina¹

¹Laboratory of Molecular Biology-LGO, Tecnológico Nacional de México. 600 Antonio García Cubas Av., Fovissste, Celaya, Gto., 38010 Mexico. ²Laboratory of Biochemistry and Molecular Biology of Proteins, Cinvestav-IPN. Km 9.6 Irapuato-León Rd, Irapuato, Gto., 36821 Mexico.
E-mail: djpantoja.gtz@gmail.com

Capsicum is a genus that groups at least 42 species within the Solanaceae family, native to the tropical regions of America. Most of these species synthesize capsaicinoids, such as capsaicin, an alkaloid that accumulates in the fruit and provides a pungent flavour. The anthocyanin and carotenoid pigments give the fruit its colour. Chili peppers are used to give food a spicy flavour and as a source of capsaicin, which is an active compound in some medicines. As an agriculturally relevant crop, the genome of representative species and cultivars has been sequenced and analysed, providing genetic resources for studies at the molecular level. Gibberellins are a large group of tetracyclic diterpenoid carboxylic acids synthesized from the intermediate *trans*-geranylgeranyl diphosphate by diverse organisms. Bioactive gibberellins, including gibberellic acid, are growth inducers and regulate several development processes in plants¹. In *C. annuum*, gibberellic acid increases leaf width, flowering, plant height, fruit production, and yield². Some of these effects, including the expression of a gene involved in response to gibberellins, have recently been corroborated in plants of *C. annuum* var. jalapeño in our laboratory. However, the molecular mechanism that governs the physiological effects of gibberellins has not been widely studied in this solanacea. This work aimed to explore the response to gibberellic acid in *C. annuum* at a molecular level. *C. annuum* var. jalapeño plants were treated with exogenous gibberellic acid. Samples of flower-buds (at the pre-anthesis stage) were collected, and an extract of total protein was prepared. The proteins were fractionated by two-dimensional electrophoresis. Gel image (proteome maps) and statistical analysis resulted in the identification of eight protein spots, differentially expressed in flower-buds of plants treated with gibberellic acid. This study describes a novel approach to gain insights into the molecular mechanism governing the response to gibberellic acid in *C. annuum*. Moreover, it represents a first instance of the standardization of a method to 2D proteomics application in *C. annuum* var. jalapeño. Progress in the characterization of the proteins identified and gibberellins signalling and effects will be discussed.

References

1. Hedden, P. (2020) The Current Status of Research on Gibberellin Biosynthesis. *Plant Cell Physiol.* 61:1832-1849. doi 10.1093/pcp/pcaa092.
2. Sahu, S., Kumar, V. and Sharma, H.G. (2019) Effect of plant growth regulators on growth and yield of chilli: A review. *Journal of Pharmacognosy and Phytochemistry* 8: 2122-2132. E-ISSN: 2278-4136 P-ISSN: 2349-8234.

SUBCELLULAR LOCALIZATION PLAYS A DETERMINANT ROLE ON THE FUNCTIONAL DIVERSIFICATION OF THE PARALOGOUS PROTEINS *BAT1* AND *BAT2*

Yamile Paredes-Chiquini¹, Jose Carlos Campero-Basaldua¹,
Edgar Adrian Ramirez-Gonzalez¹ and Alicia Gonzalez-Manjarrez¹

¹Instituto de Fisiología Celular, Universidad Nacional Autónoma de México.
Circuito Exterior s/n Ciudad Universitaria, Coyoacán, 04510 México D.F.
E-mail: yamile@ifc.unam.mx

BAT1 and *BAT2* are paralogous genes codifying Bat1 and Bat2 branched-chain aminotransferases. Utilization of branched chain amino acids (valine, isoleucine and leucine VIL) as nitrogen sources, and the biosynthesis of these amino acids are exclusively carried out by these two enzymes. *BAT1* and *BAT2* have opposite expression profiles. *BAT1* is preferentially expressed when ammonium is present as sole nitrogen source (biosynthetic conditions), while *BAT2* expression is prominent on VIL as sole nitrogen sources (catabolic conditions). Under biosynthetic conditions, a *bat1Δ* mutant shows reduced growth rate compared to that of a wild type strain or a *bat2Δ* mutant. Under catabolic conditions a *bat2Δ* mutant has deficient growth rate as compared to either the wild type strain or the *bat1Δ* mutant. Opposite phenotypes of the *bat1Δ* and *bat2Δ* mutants agree with the expression profiles of *BAT1* or *BAT2*, indicating that although Bat1 and Bat2 are both capable of synthesizing and degrading VIL, they cannot fully substitute lack of either one of the genes.

These enzymes have also diversified their subcellular localization: Bat1 is a mitochondrial enzyme while Bat2 is cytosolic. We have relocalized the enzymes to the opposite compartment in the presence or absence of their paralogue. Results show that the prominent biosynthetic role of Bat1 in the mitochondria is notably diminished in the cytoplasm. Bat2 catabolic role displayed in the cytoplasm, is lost in the mitochondria. Results will be presented showing *BAT1* and *BAT2* expression patterns and enzymatic activity, in order to discuss the effect of relocalization on Bat1 and Bat2 biosynthetic or catabolic roles.

A more important finding showed that when Bat2 is localized in the mitochondria, strains acquire a petite phenotype and are unable to grow on ethanol. Expression of mitochondrial genes is extremely decreased and the concentration of mitDNA VS nucDNA is notably reduced. These results suggest that Bat2 aminotransferase besides playing a role in VIL metabolism, could have an additional negative role in the maintenance of mitochondrial DNA integrity, and selection has thus favored Bat2 cytosolic localization.

HINGES, SPRINGS, AND STICKY SURFACES DETERMINE THE PREFERRED CONFORMATIONS OF CRE IN THE ABSENCE OF LOXP AND PRIME IT FOR BINDING

Nina Pastor¹, Marco A. Ramírez-Martínez¹

¹Centro de Investigación en Dinámica Celular, UAEM, Av. Universidad 1001, Col. Chamilpa, Morelos, México.

E-mail: nina@uaem.mx, marco.ramirez@uaem.edu.mx

Cre is a site-specific tyrosine recombinase that binds to a 34-base pair DNA segment known as *loxP*¹. The Cre/*loxP* system forms a complex (intasome) that is stabilized by protein-protein and protein-DNA interactions². The structure for Cre in the absence of DNA is still unknown, due to its intrinsic flexibility. Our aim is to characterize the conformational landscape of the Cre monomer in the absence of *loxP* to determine the availability of Cre-Cre and Cre-*loxP* interaction surfaces. Starting from five different seeds^{3,4}, we ran 5 μ s-long molecular dynamics simulations using the charmm36m⁵ forcefield and the GROMACS2020⁶ software. Our results show that Cre can extend and compact as shown by its radius of gyration; these motions result from the flexible linker between CBD and CAT domains, and a large positively charged surface that promotes different orientations of the two domains. To characterize these conformations, we measured the distance and angle formed between two alpha helices near the center of mass of each domain; Cre samples many different states but is biased to specific angle-distance combinations with weak interactions between domains. To determine whether the interaction between domains might be hiding or exposing *loxP*-interaction surfaces, leading to an autoinhibited structure or priming one of the domains for binding, we evaluated the accessible surface area that is in contact with *loxP* for each domain. CAT exposes more area compared to the CBD domain, suggesting that CAT could be primed for *loxP* binding. Finally, when we looked at the domains individually, we found that each domain has a hinge that describes an opening-closing movement that could be associated with the interaction with an adjacent subunit, allowing Cre to not only interact with *loxP* but also with another monomer.

We thank LNS (grants 202101023N and 202301027N) and LANCAD (2020 to 2024 grants) for supercomputing time and technical support. MARM thanks CONAHCyT (scholarship 858905).

References

1. Sternberg, N. et al (1981). *Journal of Molecular Biology*, **4**, 467-486.
2. Van Duyne, G. (2015). *Microbiology Spectrum*, **3**, 25-32.
3. Stachowski, K. et al (2022). *Nucleic Acids Research*. **50**, 1753-1769.
4. Ennifar, E. et al (2003). *Nucleic Acids Research*. **31**, 5449-5460.
5. Huang, J. et al (2016). *Nature Methods*. **14**, 71-74.
6. Abraham, M. et al (2015). *SoftwareX*. **1-2**, 19-25.

REDESIGN OF SHIKIMATE DEHYDROGENASE TOWARD COFACTOR SPECIFICITY EXCHANGE USING ARTIFICIAL INTELLIGENCE

Raúl Iván Pérez Bermúdez, Lorenzo Segovia

Instituto de Biotecnología, Universidad Nacional Autónoma de México; address: Avenida Universidad 2001, Chamilpa, 62210, Cuernavaca, Morelos; phone: 5556225675 |E-mail: lorenzo.segovia@ibt.unam.mx

The *Escherichia coli* shikimate dehydrogenase, an enzyme belonging to the aromatic amino acid synthesis pathway, will be redesigned for the exchange of NADP⁺ to NAD⁺ specificity. For this, the use of the deep learning-based protein sequence design method, called LigandMPNN, is proposed, due to its ability to explicitly model all non-protein components and protein-ligand interactions¹. The first step is to select those residues in which a redesign is allowed for optimization of cofactor binding. We have chosen residues based on patterns related to cofactor interaction. Subsequently, refinement of the sequences is proposed by obtaining other bioinformatically determined parameters, such as the fast relaxation of the models, the redesign of second shell mutations and finally side chain packing. Models are scored under several schemes including recall, pLDDT, rmsd, to the initial structures. We will use commercially synthesized genes of the selected sequences for their subsequent expression and kinetic characterization (activity and affinity for NAD⁺, NADP⁺, and shikimate); subsequently, these parameters will be associated physicochemically, structurally, and interactively with the mutations introduced in each case.

Similarly, for this project we also propose the redesign of another variant of shikimate dehydrogenase (S131A, L135A, N149D, V152F) obtained by García Guevara (2018)², which loses more than a thousand-fold activity with NADP⁺ and increases by an order of magnitude its activity with NAD⁺, compared to the wild-type enzyme. These approaches will allow us to understand the different contributions involved in ligand affinity and thus, its manipulation.

References

1. Dauparas, J., Lee, G. R., Pecoraro, R., An, L., Anishchenko, I., Glasscock, C., & Baker, D. (2023). Atomic context-conditioned protein sequence design using LigandMPNN. *BioRxiv*, 2023.12.22.573103. <https://www.biorxiv.org/content/10.1101/2023.12.22.573103v1.abstract>
2. García Guevara, J. F. (2018). Rediseño del dominio Rossmann de la enzima Shikimate Deshidrogenasa de *E. coli*: Intercambio de la actividad de NADP hacia NAD. Tesis de Doctorado. Instituto de Biotecnología, Universidad Nacional Autónoma de México.

CHOOSING SIDES IN *LOXP* AND THE IMPORTANCE OF THE INITIAL COLONIZATION BY CRE

Marco A. Perez C.¹, Adamary M. Hernandez P.¹, Marco A. Ramirez M.¹, Nina Pastor¹

¹Centro de Investigación en Dinámica Celular, ²UAEM. Av. Universidad 1001, Col. Chamilpa, 62209 Cuernavaca, México.
E-mail: marco.perezca@uaem.edu.mx; adamary.hernandezpin@uaem.edu.mx; marco.ramirez@uaem.edu.mx; nina@uaem.mx

Cre is a site-specific recombinase that recognizes the *loxP* sequence, composed of two palindromic RBEs of 13 bp flanking an 8 bp spacer sequence that makes *loxP* asymmetric. Each RBE site recruits a Cre monomer, forming a dimer. Cre cuts first on the right side of the spacer region and then on the left side¹. This is apparently due to the compression of the minor groove imposed by Cre, preventing the entry of K201, and therefore the cleavage of *loxP*; this suggests that the cut occurs at an undeformed site. However, a single Cre cannot cut *loxP*². The catalytic domain of Cre (CAT) binds to *loxP* but is not active³. Catalysis requires the simultaneous arrival of K201 (located in a long flexible loop) and Y324 (located at the end of helix M, close to an IDR), in addition to the proper conformation around the scissile phosphate. Models of Cre and CAT bound to half of a *loxP* site were constructed with variations in spacer sequence to investigate its influence on Cre or CAT binding due to its flexibility and composition. Each model was simulated with the charmm36m⁴ force field and GROMACS⁵ software for at least 1 μ s. We compared ribose shielding by Cre to the OH radical⁴ protection pattern as a positive control. To analyze DNA deformations, the roll, twist, tilt, and opening parameters, and the groove widths were measured, as well as the distances and angles between the IK, DG, and DJ helices of Cre. These pairs of helices monitor the response of the protein to the flexibility of the spacer. We calculated the contacts made by the catalytic residues towards the cleavable phosphate to study the integrity of the active site. Finally, we evaluated the stability of the complexes by calculating the contacts of Cre with *loxP*, and the contacts between the IDRs in Cre with the rest of Cre.

We thank LNS (grant 202301027N) and LANCAD (grant 2024) for supercomputing time and technical support. MARM thanks CONAHCyT (scholarship 858905).

References

1. G. Meinke, A. Bohm, J. Hauber et al. (2016). *Chemical Reviews*, **20**, 12785-12820.
2. K. Stachowski et al. (2022). *Nucleic Acids Research*, **50**, 1753-1769.
3. R. Hoess, K. Abremski, S. Irwin et al. (1990). *Journal of Molecular Biology*, **4**, 873-882.
4. J. Huang et al (2016). *Nature Methods*. **14**, 71-74.
5. M. Abraham, T. Murtola, R. Schulz et al. (2015). *SoftwareX*, **1-2**, 19-25.

MODULATION OF THE HEXOSAMINE BIOSYNTHETIC PATHWAY AND O-GLCNACYLATION INFLUENCES THE LOCALIZATION OF THE CD36 RECEPTOR IN MACROPHAGES

Yobana Pérez-Cervera^{1,2}, Edgar Zenteno Galindo³, Eleazar Ramírez-Hernández³, Roberta Salinas-Marin⁴, Adriana Moreno-Rodríguez⁵, Rafael Torres-Rosas¹, Liliana Argueta-Figueroa^{1,6}, Berenice Fernández-Rojas¹, Socorro Pina-Canseco², Alfonso E. Acevedo-Mascarúa¹, Alicia Hernández Antonio¹ y Karen Julissa Loeza-Reyes^{1,2*}

¹Centro de Estudios en Ciencias de la Salud y la Enfermedad, Facultad de Odontología, Universidad Autónoma Benito Juárez de Oaxaca, Oaxaca, México; ²Centro de Investigación Multidisciplinaria Facultad de Medicina-UNAM-UABJO, Universidad Autónoma Benito Juárez de Oaxaca, Oaxaca, México; ³Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México, Mexico City, Mexico; ⁴Laboratorio de Glicobiología Humana y Diagnóstico Molecular, Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Cuernavaca, Mexico; ⁵Facultad de Ciencias Químicas, Universidad Autónoma Benito Juárez de Oaxaca, Oaxaca, Mexico. ⁶CONAHCYT-Facultad de Odontología, Universidad Autónoma Benito Juárez de Oaxaca, Oaxaca, Mexico.
E-mail: yobanper@gmail.com.

CD36 is a type 2 cell surface scavenger receptor expressed in various tissues. In macrophages, CD36 recognizes oxidized low-density lipoprotein (ox-LDL), which promotes the formation of foam cells, the first step toward an atherosclerotic arterial lesion. CD36 possesses a variety of posttranslational modifications, among them N-glycosylation and O-GlcNAc modification. Some of the roles of these modifications on CD36 are known, such as N-linked glycosylation, which provides proper folding and trafficking to the plasma membrane in the human embryonic kidney. This study aimed to determine whether variations in the availability of UDP-GlcNAc could impact Rab-5-mediated endocytic trafficking and, therefore, the cellular localization of CD36. These preliminary results suggest that the availability of the substrate UDP-GlcNAc, modulated in response to treatment with Thiamet G (TMG), OSMI-1 (O-GlcNAcylation enzymes modulators) or Azaserine (HBP modulator), influences the localization of CD36 in J774 macrophages, and the endocytic trafficking as evidenced by the regulatory protein Rab-5, between the plasma membrane and the cytoplasm.

SCREENING OF A CHEMICAL FRAGMENT LIBRARY FOR THE SEARCH OF INHIBITORS AGAINST THE ACYL-HOMOSERINE LACTONE SYNTHASE OF *ACINETOBACTER BAUMANNII*

¹Jesús Ricardo Pérez Velázquez, ¹Gabriel Isaías Torres Vernet, ¹Jocelyne Martínez Martínez,
²Rodolfo García Contreras, ¹Jesús Antonio Oria Hernández, ¹Adriana Castillo Villanueva*

¹Laboratorio de Bioquímica-Genética, Instituto Nacional de Pediatría. ²Departamento de Microbiología y Parasitología, Facultad de Medicina, UNAM. *Av. Insurgentes Sur 3700-Letra C, Insurgentes Cuicuilco, Coyoacán, 04530 Ciudad de México, CDMX. Tel. 55 1084 0900, ext. 1442. acastilloinp@gmail.com

Antimicrobial resistance is one of the top 10 threats to global public health today. It has been reported that 4.95 million deaths were directly attributed to drug-resistant infections worldwide in 2019, and it is estimated that by 2050, up to 10 million deaths will be attributable to this cause. *Acinetobacter baumannii* is a multidrug-resistant gram-negative bacterium, classified as a critical priority pathogen by the World Health Organization. Multidrug resistance in *A. baumannii* is related to multiple factors, including quorum sensing. Quorum sensing regulates genes related to virulence and pathogenicity; therefore, it is hypothesized that quorum sensing inhibitors could serve as an alternative therapeutic approach to conventional antibiotics. The acyl-homoserine lactone synthase of *A. baumannii* (AbAHL) is a key enzyme in the production of signaling molecules known as N-acyl homoserine lactones that regulate quorum sensing in this bacterium. Therefore, AbAHL is considered a possible molecular target for the development of inhibitors with pharmacological potential. In this work, the AbAHL gene was cloned into the pCri1B and pET-HisTEV plasmids, expressed in *Escherichia coli* BL21(DE3)pLysS and purified by immobilized metal ion affinity chromatography. The recombinant protein was used to screen the effect of the 1,000 compounds from the Maybridge Ro3 Diversity Fragment Library on enzymatic activity. At a concentration of 100 μ M, 10 compounds were found to inhibit AbAHL activity by at least 70%. The inhibition effect of the molecules was specific to the enzyme and did not interfere with the enzymatic assay. The kinetic mechanism of inhibition of the best inhibitors will be explored. Overall, the identified compounds represent molecular scaffolds for the development of new AbAHL inhibitors with pharmacological potential.

CHANGES IN LIPID AND PROTEOME COMPOSITION OF THE INNER AND OUTER LIVER'S MITOCHONDRIAL MEMBRANES AND ITS CORRELATION WITH MITOCHONDRIAL RESPIRATION IN EXPERIMENTAL TYPE 1 DIABETES

Ramiro Perusquia-García, Ricardo Mejía-Zepeda†, Luis Arturo Baiza-Gutman

Universidad Nacional Autónoma de México, Facultad de Estudios Superiores Iztacala
Av. de los Barrios No. 1. Los Reyes Iztacala, CP: 54090, Tlalnepantla de Baz, Estado de México, México. +52 56 2131 5133.
Mail: ramiro.perusquia@gmail.com

It is known that Diabetes Mellitus (DM) modify cell dynamics by modulating lipid and protein metabolism at different levels, even so that affects at a to subcellular structures. As is known, the eukaryotic cells are full of membranous organelles, making them susceptible to the changes by pathologies like DM. One of the organelle that has been reported to change in its lipid and protein composition by DM is the mitochondria (MIT). MIT is composed of two membranes, the inner membrane (IMM) and the outer membrane (OMM); also, in its membranes takes places the electron transport chain (ETC) and the oxidative phosphorylation (OXPHOS), which make about 95% of the ATP. Since OXPHOS and ETC occur in the IMM, changes on the micro conditions of those membranes can alter those processes. In this study we evaluated the alterations of the Fatty Acid (FAc) and phospholipids composition (PLc) and protein profiles of the outer and inner membranes of the liver's mitochondria on Wistar rats, under the experimental model of Type 1 Diabetes Mellitus (T1DM). Methods: 2 months old Wistar rats were induced to T1DM using streptozotocin. The rats were euthanized after one moth and liver's mitochondria were isolated by differential centrifugation and mitochondrial respiration was evaluated. To obtain the OMM and IMM, we use the method of sucrose gradient ultracentrifugation. The FAc was determinated by gas chromatography, PLc by thin layer chromatography, lipid peroxidation by the thiobarbituric acid reaction and the protein profile by SDS-PAGE in both, the OMM and IMM. Results: we found that T1DM modify both the OMM and IMM FAc: In the OM, the saturated FA (SFA) have an increase in its percentage, and unsaturated FA (UFA) have a decrease proportion but; In the IMM, the FAc is more heterogeneous but change in the opposite way than OMM, in the IMM SFA decrease its percentage and UFA increases. The PLc of the IMM is more affected than de IMM. Lipid peroxidation increases in both membranes but is proportionally higher in the IMM. Also, the protein profile in both the OMM and IMM is also modified by the T1DM. Finally, we found that mitochondrial respiration is enhanced as a consequence of T1DM, respiration with and without ADP is almost one third higher in the DM1 group compared to the normoglycemic one. We infer that the lipid and protein modifications of the OMM and the IMM modulate the efficiency and/or the functionality of the ETC and OXPHOS, but more research is needed to prove this assumption.

PAPIIT: IN213421

CONACYT N° CVU: 1147028

UNAM Posgrado en Ciencias Biológicas

IDENTIFICATION OF *PARAMECIUM MULTIMICRONUCLEATUM* MITOCHONDRIAL ATP SYNTHASE

Maria Guadalupe Quintanar Solis¹, Anaiza Rico Luna¹, Héctor V. Miranda-Astudillo^{1*}

¹Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, México City, México.

*hmiranda@iibiomedicas.unam.mx

For decades, the study of mitochondrial bioenergetics has focused on a limited number of biological models, leaving out other organisms of ecological, medical, and biotechnological importance. In this way, photosynthesis has been mainly studied in the supergroup Archaeplastida, in organisms like *Arabidopsis thaliana* and *Chlamydomonas reinhardtii*, while oxidative phosphorylation in Opisthokonta, in organisms such as yeasts and animals. The study of new biological models has increased our knowledge about the megadiverse architecture of mitochondrial complexes and its mechanisms among organisms distributed in different Eukarya supergroups (1). Ciliates, which pertain to the supergroup SAR, are cosmopolitan unicellular microorganisms fundamental in the trophic chain of aquifer ecosystems, and in some cases human parasites. Mitochondrial complex V carries the synthesis of ATP from ADP and Pi from the conversion of a proton motive force generated by the electron transport chain. The aim of this project is to identify and characterize the ATP synthase from the ciliate *Paramecium multimicronucleatum*.

Cultivation of *P. multimicronucleatum* in minimum mineral Tris-phosphate medium supplemented with vitamins and yeasts was standardized. Growth curves showed an exponential phase between days three and seven. Cells were pelleted and cell lysis was carried out with a syringe. The lysate was further centrifugated to obtain total membranes. Membranes were solubilized with different non-ionic detergents, and mitochondrial complexes were separated by electrophoresis in blue polyacrylamide gels in native conditions (BN-PAGE). Mitochondrial ATP synthase was identified by *Western blot* after two-dimension electrophoresis (BN-PAGE followed by SDS-PAGE) and by zymography. Complex V polypeptide composition was further analyzed by two-dimension denaturing electrophoresis.

Acknowledgments. We thank the Program “Production of Biomolecules of Biomedical Interest in microorganisms” of the Institute of Biomedical Research (UNAM) and the PAPIIT program: IA204524 (DGAPA, UNAM) for the financing, as well as Conahcyt for the scholarship received by Anaiza Rico CVU 927622.

References

1. Miranda-Astudillo H. Vamos ., et al. (2022) *BBA-Bioenergetics* 1863: 148569

PARTIAL PURIFICATION AND CHARACTERIZATION OF THE NADH DEHYDROGENASE OF THE RESPIRATORY CHAIN OF *BACILLUS SUBTILIS*

Alexia Ramírez Ávila^{1,2}, Martín González Andrade³, Emma Berta Gutiérrez Cirlos Madrid^{2,*}

¹Posgrado en Ciencias Biológicas, Universidad Nacional Autónoma de México, Posgrado en Ciencias Biológicas, Unidad de Posgrado, Edificio D, 1° piso, Circuito de Posgrados, Ciudad Universitaria, Coyoacán, C.P. 04510, CDMX, México.

²Laboratorio 2 Bioquímica y Bioenergética, Unidad de Investigación en Biomedicina, Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México, Av. De los Barrios 1, Los Reyes Ixtacala, Tlalnepantla, C.P. 54090, Estado de México, México. ³Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México. Laboratorio de Biosensores y Modelaje Molecular, Torre de Investigación, 3° piso, Ciudad Universitaria, Coyoacán, C.P. 04510, CDMX, México.

*e-mail: ember@unam.mx

Bacillus subtilis has a branched respiratory chain, and the NADH dehydrogenase mainly feeds it. This complex belongs to the NDH-2 family: a group of peripheral enzymes composed of one polypeptide chain that does not translocate protons through the membrane. This complex has not been studied in detail in this bacterium, but it has been reported that the enzyme corresponds to the protein YjID.

We purified the NDH-2 from *B. subtilis* membranes by incubating them with 1.5 M NaCl to separate the transmembrane and peripheral proteins. The fraction containing the peripheral proteins was charged into an ion exchange chromatography, and a partially pure NDH-2 with minor contaminants was obtained.

The enzyme was obtained as a monomer of 42 kDa, which matches the predicted molecular mass of YjID. The enzyme did not lose its prosthetic group throughout the purification, and we detected unspecific activity with NADPH and O₂. The kinetic parameters for NADH and the quinones menadione and ubiquinone-1 were determined: a *km* of 3.32 μM for NADH, a *km* of 55.28 μM for menadione, and a *km* of 111.5 μM for ubiquinone-1. The IC₅₀ of 17.12 μM for the inhibitor HQNO was also obtained.

The tridimensional model of YjID was successfully docked in the membrane. The model was supplemented with the structure of NADH, FAD, and several quinones: menaquinone-7, menadione, ubiquinone-1 and HQNO. We also determined the catalytically important residues interacting ligands. We will show the complete characterization of this electron donor of the electron transfer chain of *B. subtilis*.

Grants: FICDTEM-2021-57 COMECYT, PAPCA 50-2022 and DGAPA UNAM, IN 204222.

THE PYRUVATE KINASE FROM *PYROBACULUM AEROPHILUM* SHARES WITH OTHER THERMOPROTEALES UNIQUE FEATURES OF THE CATALYTIC LID

Leticia Ramírez-Silva¹, Zuribet Baxin-Toto¹, Mitchel Daniela Berman-Flores¹, Alicia Vega-Segura¹, Jessica Díaz Salazar² and Gloria Hernández-Alcántara¹

¹Department of Biochemistry, School of Medicine, ²Department of Physical Chemistry, School of Chemistry, UNAM, Mexico City 04510. Phone 555623-2133.
iramirez@bq.unam.mx

Introduction. The phylogenetic analysis of the family of pyruvate kinase exhibits a dichotomic tree. One branch corresponds strictly, to the sequences of the K⁺-dependent pyruvate kinases and the other, to the sequences of the K⁺-independent enzymes. The branches of the K⁺-dependent and K⁺-independent enzymes present a very conserved signature: T113/K14/E117/T120 and L113/Q114/K117/(L,I,V) 120, respectively. The pyruvate kinase from *Pyrobaculum aerophilum* (*PaPK*) is a hyperthermophilic Thermoproteales found in the branch of the K⁺-independent enzymes. It has almost the complete signature of the branch, except for the corresponding position 117 (Ser 85). The *PaPK* has been purified, crystallized and few kinetic data reported. In a previous study of the pyruvate kinase from the Thermoproteales *Thermofilum pendens* (*TpPK*) it was described that the calorimetric and kinetic data indicated that the B domain or catalytic lid is more stable than the rest of the protein with a conformation that induces the catalytic readiness of the enzyme. Therefore, we were interested in studying the kinetic and thermal stability of *PaPK* and compare the results with *TpPK* to explore if they follow a general mechanism.

Methods. The gen of the pyruvate kinase from *Pyrobaculum aerophilum* in pET-15b was subcloned in pET3a and transformed in Rosetta (DE3) PLYS S. The enzyme was purified by ionic exchange (DEAE-Sepharose and CM-Sepharose). Activities were determined in a coupled assay with lactate dehydrogenase at 340 nm and 45° C. CD spectra were obtained in the far UV from 190 to 260 nm with 100 µg/ml in a 0.1 cm cell and in the near UV from 260 to 300 nm with 1 mg/ml in a 0.5 cm cell in a spectropolarimeter Chirascan (Japan) at 25°C and 3 repetitions. DSC experiments were performed in a capillary Differential Scanning Microcalorimeter from GE Health Science (USA) with 18 to 20 µM of protein concentration.

Results and Conclusions. *PaPK* is a K⁺-independent enzyme, not activated by Li⁺, Na⁺, K⁺, NH₄⁺, Rb⁺ or Cs⁺. Is the first PK described to be activated by 3PG [2] and is almost general for most Thermoproteales. It was a mixed allosteric effector in the presence of Mg²⁺ (*V*_{max} increased 2-fold; and *K*_{m_{PEP} decreased 4-fold). Catalytic efficiency is less than an order of magnitude higher with 3PG than without it. In the presence of Mn²⁺, *V*_{max} was the third part of that with Mg²⁺ and 20% less with 3PG. However, in the presence of Mn²⁺ compared to Mg²⁺, *K*_{m_{PEP} decreased 17-fold and 8-fold without and with 3PG, respectively; whereas *K*_{m_{Mn²⁺ free} were 286-fold and 137-fold smaller without and with 3PG, respectively than those for Mg²⁺_{free}. Catalytic efficiencies were more than 1 and 2 orders of magnitude higher for PEP and Mn²⁺_{free} in the manganese complexes than in the magnesium complexes without 3PG. According to secondary or tertiary structure, CD spectra with and without 3PG were alike either in the far as in the near UV. Finally, as expected we found a similar DSC as in *TpPK*, two calorimetric transitions indicative of an independent domain denaturation. This thermal denaturation has only been found in Thermoproteales; whereas mesophile like rabbit muscle pyruvate kinase (PK), *Vibrio cholerae* type I (PK) or the thermophile euryarchaeota *Thermoplasma acidophilum* PK exhibit one calorimetric transition or global protein denaturation. These results indicated that *PaPK* was slightly activated by 3PG. It was more efficient with Mn²⁺ complexes and exhibited an independent lid denaturation that induced the catalytic readiness of the enzyme as a general mechanism of these group of PKs.}}}

Pyruvate kinases from Crenarchaeota acquire their active conformation by a general mechanism.

Pyruvate kinase from *Pyrobaculum aerophilum* shares a general mechanism to acquire its active conformation with other Crenarchaeota.

GENERATION OF A QUANTIFICATION SYSTEM FOR *GALLERIA MELLONELLA* APOLIPOPHORIN-III

Uriel Ramírez Sotelo¹, Laura Cristina García Carnero¹, José Ascención Martínez Álvarez¹,
Manuela Gómez Gaviria¹, Héctor Manuel Mora Montes¹

¹Departamento de Biología, División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Guanajuato, Mexico.
Noria Alta S/N, Guanajuato, Gto; C.P. 36050, Tel 47320006 ext. 8153,
e-mail: hmora@ugto.mx

Research in animal models is essential to obtain more information about human infections and host-pathogen interactions, being murines the gold-standard. Invertebrate animals have emerged as alternative models to study the host-pathogen interaction with minimal ethical implications. *Galleria mellonella* is an alternative model that has proved useful in studying the interaction of the host with either bacteria or fungi, performing drug testing, and assessing the immunological response to different microorganisms. The *G. mellonella* immune response includes cellular and humoral components with structural and functional similarities to the immune effectors found in higher vertebrates, such as humans. An important humoral effector stimulated during infections is apolipoprotein III (apoLp-III), an opsonin characterized by its lipid and carbohydrate-binding properties that participate in lipid transport, as well as immunological activity. Despite some parameters, such as the measurement of phenoloxidase activity, melanin production, hemocytes counting, and expression of antimicrobial peptides genes are already used to assess the *G. mellonella* immune response to pathogens with different virulence degrees, the apoLp-III quantification remains to be a parameter to assess the immune response in this invertebrate. In this work, we develop an immunological tool based on an enzyme-linked immunosorbent assay that allows apoLp-III quantification in the hemolymph of larvae challenged with pathogenic agents. We tested the system with hemolymph coming from larvae infected with *Escherichia coli*, *Candida albicans*, *Sporothrix schenckii*, *Sporothrix globosa*, and *Sporothrix brasiliensis*. The results revealed significantly higher concentrations of apoLp-III when each microbial species was inoculated, in comparison with untouched larvae, or larvae inoculated with the vehicle. Moreover, our results demonstrate that the apoLp-III levels correlated with the strains' virulence, which was already reported. To our knowledge, this is one of the first attempts to quantify apoLp-III, using a quick and easy-to-use serological technique.

EVALUATION OF THE INTENSITY OF THE KILLER EFFECT IN *SACCHAROMYCES CEREVISIAE* USING DIFFERENT CARBON SOURCES AND ITS RELATIONSHIP WITH MITOCHONDRIAL BIOGENESIS

E. Ramos¹, V. Morales-Tlalpan^{1,2}, M. G. Martinez-Hernández¹, J. Trujillo-Barrientos¹ & Carlos Saldaña^{1,2*}

¹Laboratorio de Biofísica de Membranas y Nanotecnología, ²Laboratorio Nacional de Visualización Científica Avanzada UNAM-UAQ. Facultad de Ciencias Naturales, Universidad Autónoma de Querétaro. Juriquilla, Qro., México C.P. 76230. Phone +52 442 192 1200, ext. 65332. carlos.saldana@uaq.mx

Killer effect of *Saccharomyces cerevisiae* has attracted the attention of scientific community due to its interference competition dynamics. In this process, toxin-producer strains eliminate sensitive strains. The ability to produce *Killer* proteins is given by a persistent cytoplasmic coinfection of two double-stranded RNA (dsRNA) viral agents, belonging to the Totiviridae family. Four types of *Killer* toxins have been described in *Killer* system of *S. cerevisiae*: *K1*, *K2*, *K28* and *Klus*. This study focuses on *K1* protein, whose action mechanism is based on the interaction with sensitive cell's *TOK1* potassium channel, causing the opening of this channel and cell death due to loss of electrochemical gradient. This phenomenon is observed as a halo of inhibition between sensitive and toxin-producing yeast, where no yeast growth can be found; likewise, the magnitude of this effect is measured by the surface of the inhibition zone. The aim of the study is to determine if there are differences in the expression patterns of the *Killer* effect depending on the carbon source provided, as well as to evaluate its effect to promote the electron transport chain. Fermentable (succinate and glycerol) and nonfermentable (ethanol and lactate) substrates were used, obtaining partial results that indicate greater efficiency in *Killer* activity with fermentable substrates, particularly glycerol. Finally, mitochondrial biogenesis will be evaluated through PCR of two molecular markers, *Hrr25*, which positively regulates mitochondrial biogenesis, and *Puf3*, which negatively regulates it. These yeasts will be maintained under pH 7 conditions in aerobic, anaerobic, and fasting cell cultures reactivated with a carbon source, to evaluate whether the effectiveness of the *Killer* effect is correlated with the number of mitochondria per cell.

Acknowledgments. We extend our sincere gratitude to the funding institutions: FONDEC-UAQ, FONFIVE 2024 and CONAHcyT A1-S-26966 for their generous support and financial backing provided to Carlos Saldaña.

CHARACTERIZATION OF HUMAN EPIDERMAL GROWTH FACTOR (hEGF)-INDUCED Ca^{2+} RELEASE IN HeLa CELLS

Amanda Renté¹, Leonardo Gallegos¹, Lizeth Sandoval¹, Manuel Arias² and Agustín Guerrero-Hernández¹

¹Department of Biochemistry, CINVESTAV, CDMX, 07360. ²Programa de Neurociencias-UIICSE, Facultad de Estudios Superiores Iztacala, UNAM; Tlalnepantla de Baz, Estado de México, 54090, Mexico.

E-mail: amanda.rente@cinvestav.mx

Cells maintain a low cytoplasmic $[Ca^{2+}]_i$ (~100 nM) compared to the extracellular space (~1 mM) by utilizing ATP. They also possess various intracellular Ca^{2+} stores, such as the endoplasmic reticulum (ER), which produces localized and transient Ca^{2+} release events, thereby mitigating the Ca^{2+} toxic effect. The SERCA pump replenishes the ER, using the Ca^{2+} provided by different plasma membrane ion channels, via the store-operated Ca^{2+} entry (SOCE) that involves the activation of the Orai1 channel by STIM1, the ER- Ca^{2+} -binding protein. The ER also expresses Ca^{2+} release (RIP₃) and leak (Orai3) channels¹ and luminal proteins, enhancing its Ca^{2+} storage capacity. The Orai3 channel, expressed exclusively in mammals, is considered an oncogene due to its association with tumor growth. Notably, initial increases in the $[Ca^{2+}]_i$ induced by the VEGF require the expression of Orai3 channels². These findings suggest that the Orai3 channel might play a crucial role in $[Ca^{2+}]_i$ changes induced by growth factors, prompting our investigation into its role in Ca^{2+} signaling induced by growth factors other than VEGF, such as hEGF in HeLa cells. This study aims to determine whether Orai3 has a generalized role in Ca^{2+} signaling of all growth factors, underscoring its significance in cellular signaling pathways and potential implications for cancer biology.

This study delved into the hEGF-induced $[Ca^{2+}]_i$ response under different cytoplasmic Ca^{2+} buffering conditions (250 nM and 2 μ M of Fura-2 AM) in HeLa cells. The intriguing result was that the reduced Ca^{2+} buffering (low Fura-2) did not increase the transient elevation of the hEGF $[Ca^{2+}]_i$ response, challenging the notion of a positive Ca^{2+} feedback loop. Depleting the ER Ca^{2+} store with an inhibitor of the SERCA pump (thapsigargin) without external Ca^{2+} fully inhibited the hEGF-induced Ca^{2+} response, providing a novel insight into the role of the ER in this process. The amount of Ca^{2+} released by histamine was similar to that of hEGF. However, only the previous application of histamine decreased the amount of Ca^{2+} released by hEGF, but not vice versa, suggesting a unique interaction between these two factors. It has been proposed that hEGF activates PLC γ ; an inhibitor of this enzyme (U73122) and its inactive analog (U73343) were both used to verify this mechanism. U73122, whether chronically or acutely applied, inhibited the His $[Ca^{2+}]_i$ response but not the hEGF response, suggesting that hEGF releases Ca^{2+} from the ER without requiring an increase in IP₃. To evaluate the participation of the Orai3 channel in the hEGF response, we overexpressed both the WT Orai3, and its non-functional mutant Orai3 E81A (dominant negative channel). Our data suggest that RIP₃, but not the Orai3 channel, participates in the hEGF-induced $[Ca^{2+}]_i$ response, presenting a novel perspective on the role of Orai3 in this context.

References

1. D. Leon-Aparicio, J. Pacheco, J. Chavez-Reyes, J.M. Galindo, J. Valdes, L. Vaca, A. Guerrero-Hernandez (2017). *Cell Calcium*, 65, 91-101.
2. J. Li, A.F. Bruns, B. Hou, B. Rode, P.J. Webster, M.A. Bailey, D. O'Regan, K.E. Porter, R. Foster, M.T. Kearney, D.J. Beech (2015). *Arterioscler. Thromb. Vasc. Biol.*, 35,1987-1994.

KILL ME IF YOU CAN, THE LONG-LIVED YEAST RHODOTORULA MUCILAGINOSA

Carolina Ricardez-García¹, Ofelia Méndez-Romero¹, Natalia Chiquete-Félix¹ and Salvador Uribe-Carvajal^{1*}

¹Department of Genetics and Molecular Biology, Instituto de Fisiología Celular, UNAM, Mexico City, Mexico
suribe@ifc.unam.mx

Slowly aging organisms provide valuable insights into the mechanisms underlying the normal aging process. We now understand that aging is regulated by pathways conserved throughout evolution. Depending on the study organism, the aging process varies. There are organisms with an exceptionally long lifespan such as *Heterocephalus glaber* (naked mole-rat) or *Hydra vulgaris* (hydra). Experimentally studying these organisms is complex and expensive, so other model organisms have been considered. *Rhodotorula mucilaginosa* is a yeast from the Phylum Basidiomycota, that grows under various stress conditions. Additionally, it produces carotenes and lipids used in the pharmaceutical, cosmetic, and food industries. It has a versatile metabolism, featuring a branched respiratory chain and growth under oxidative stress conditions. Therefore, it was decided to evaluate the lifespan in *R. mucilaginosa* and comparing it with *Saccharomyces cerevisiae*, focusing on bioenergetic processes. Viability tests will be conducted, as well as assessments of mitochondrial function over time, measuring whole-cell oxygen consumption, respiratory complex activity, reactive oxygen species (ROS), ATP content, and carotene quantification.

KINETIC CHARACTERIZATION OF EUKARYOTIC MITOCHONDRIAL ATP SYNTHASES

Anaiza Rico Luna¹, Héctor V. Miranda-Astudillo^{1*}

¹Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, Mexico City, México.

*hmiranda@iibiomedicas.unam.mx

F_1F_0 ATP synthase is a multiproteic complex found in bacteria, thylakoid, and inner mitochondrial membranes. The core structure of the enzyme, corresponding to the *c* ring, the central stalk, and the catalytic region, is highly conserved among organisms. It uses a proton motive force to synthesize ATP from ADP and Pi through a rotational mechanism in which protons flow through the *c* ring triggering the anticlockwise rotation of the central stalk which in turn causes conformational changes in the catalytic region of the enzyme that favours ATP synthesis. When the enzyme rotates in a clockwise direction ATP hydrolysis takes place and the complex acts as a proton pump¹.

Unlike bacteria and thylakoids, the mitochondrial ATP synthase is found as a dimer which shows high structural divergence in the peripheral stalk and in the region embedded in the membrane among different eukaryote lineages. These differences have a direct impact on the stability of the dimer and contribute to the ultrastructure of the mitochondrial cristae². The above-mentioned lead us to ask if the structural differences observed in the mitochondrial complex V of phylogenetically separated lineages modify the overall catalytic activity of this enzyme. Mitochondrial preparations from *Polytomella parva* (Chloroplastida), and *Euglena gracilis* (Discoba) were obtained and solubilized with the non-ionic detergent n-dodecyl- β -D-maltoside. ATP synthases were purified by ion exchange liquid chromatography followed by exclusion chromatography. Complex V activity was further evaluated in a coupled assay of ATP hydrolysis.

Acknowledgments. This research was supported by grant IA204524 (PAPIIT, DGAPA, UNAM) and by the Institutional Program (IIBO-UNAM) "Production of biomolecules of biomedical interest in microorganisms". Anaiza Rico Luna has been awarded a scholarship by CONAHCYT (CVU 927622).

References

1. Miranda-Astudillo, H., Zarco-Zavala, M., García-Trejo, J. J. & González-Halphen, D. Regulation of bacterial ATP synthase activity: A gear-shifting or a pawl-ratchet mechanism? *The FEBS journal* vol. 288 3159–3163 (2021).
2. Miranda-Astudillo, H., Ostolga-Chavarría, M., Cardol, P. & González-Halphen, D. Beyond being an energy supplier, ATP synthase is a sculptor of mitochondrial cristae. *Biochim. Biophys. Acta Bioenerg.* 1863, 148569 (2022)

BIOCHEMICAL RESPONSE OF FOUR BREAD WHEAT GENOTYPES SUBJECTED TO HEAT STRESS

Edgar Robles-Rodriguez¹, Huizar L. Díaz-Ceniceros², Sergio G. Hernandez-Leon^{1*}

¹Centro de Investigación en Alimentación y Desarrollo, A.C. Carretera G.E. Astiazarán Rosas No. 46, Col. La Victoria, C.P. 83304, Hermosillo, Sonora, México ²Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias,

Carretera Norman E. Bourlaug SN, Cajeme 85000, Sonora, México.

Corresponding: sergio.hernandez@ciad.mx, (+52) 662 289-2400 ext 509 and 518

Text Wheat (*Triticum aestivum* L.) is a staple food crop globally, essential for human nutrition as it provides more calories and protein than any other cereal. Elevated temperatures can induce heat stress (HS), inhibiting wheat growth and metabolism, and adversely affecting its productivity worldwide. In response, wheat plants undergo physiological, biochemical, and metabolic changes to tolerate HS. The primary consequence of HS is the overproduction of reactive oxygen species (ROS), leading to oxidative stress. Therefore, investigating the biochemical responses of heat-stressed wheat genotypes is crucial for developing new crop strategies to ensure food security. In previous field experiments, conducted under optimal temperature conditions and HS during the reproductive stage, diverse wheat genotypes provided by CIMMYT and INIFAP were evaluated. These genotypes were classified based on reduced grain yield as tolerant or sensitive to HS. From these, four genotypes (two tolerant, one intermediate, and one sensitive to HS) were selected for detailed biochemical analysis. The study assessed the biochemical response to HS by measuring the concentrations of chlorophyll and carotenoids, performing ascorbate peroxidase (APX) and catalase (CAT) enzymatic assays, and analysing the accumulation of glycine betaine (GB) and proline. Results showed that all genotypes maintained photosynthetic pigment concentrations, with tolerant genotypes tending to increase while the sensitive ones to decrease, potentially impairing photosynthetic efficiency. Most genotypes exhibited a decrease in CAT activity under HS, while APX activity increased, suggesting that H₂O₂ scavenging under HS is predominantly managed by APX. GB content increased in tolerant genotypes, while proline levels decreased in most genotypes under HS. Both proline and GB act as critical osmoprotectants that accumulate under abiotic stress; however, proline tends to increase more under salinity, possibly explaining why wheat plants preferentially accumulate GB under HS. This research highlights the differential biochemical responses of wheat genotypes to HS, emphasizing the role of APX in ROS scavenging and the preferential accumulation of GB. These findings may contribute to developing wheat varieties with enhanced tolerance to heat stress to achieve sustained wheat production under changing climatic conditions.

EFFECT OF METABOLIC SYNDROME ON THE EXPRESSION OF THE RYANODINE RECEPTOR AND THE SERCA PUMP IN RAT CEREBRAL ARTERIES

Daniela Rodríguez-Benito¹, Daniel Hurtado-González¹, Rommel Sánchez-Hernández², Federico Castro-Muñozledo³, Angélica Rueda¹

¹Departamento de Bioquímica, ²Departamento de Fisiología, Biofísica y Neurociencias y ³Departamento de Biología Celular, Cinvestav-IPN. Unidad Zacatenco. Col. San Pedro Zacatenco, GAM. CP. 07360. Ciudad de México. México. arueda@cinvestav.mx

Metabolic syndrome (MS) is a pre-diabetic state characterized by insulin resistance, hypertension, visceral fat accumulation, and dyslipidemia, which increases the risk for developing cardiovascular diseases and stroke¹. Intracellular Ca²⁺ signals regulate contraction and relaxation of cerebral artery (CA) vascular smooth muscle cells (VSMC), hence blood flow². The functional coupling between Sarco/Endoplasmic Reticulum (SR) Ca²⁺ ATPases (SERCA pumps) and ryanodine receptors (RyRs) plays a key role in this regulation. The SERCA pump increases Ca²⁺ levels within the SR activating RyRs, which generates Ca²⁺ sparks in the cytoplasm promoting vascular relaxation². Previous data from our lab revealed a significant decrease in Ca²⁺ spark frequency of intact VSMC from the middle cerebral artery (MCA) of MS rats³. Therefore, we aimed to investigate how MS affects the expression of RyR and SERCA pump isoforms in rat CA.

Our MS experimental model (sucrose fed male *Wistar* rats) showed a significant increase in MCA wall thickness (in μm : 16 ± 1.04 in N=4 control rats vs 19.33 ± 0.95 in N = 6 MS rats, $P < 0.05$) but unchanged internal diameter (in μm : 70.24 ± 10.69 in N=4 control rats vs 68.98 ± 4.08 in N = 6 MS rats, $P = 0.9018$), unveiling structural alterations. Additional evidence of altered intracellular Ca²⁺ dynamics in VSMC of MS rats was further indicated by an increased incidence of intracellular Ca²⁺ waves (in %: 37.08 ± 5.29 in n=16 control cells vs 71.67 ± 8.17 in n=17 MS cells, $P < 0.05$), evidencing the abnormal regulation of intracellular Ca²⁺ cycling in MCA of MS rats. However, MS condition did not alter protein expression or mRNA levels of RyR isoforms (RyR1, RyR2, and RyR3) and SERCA2 pump isoforms (SERCA2a and SERCA2b). Therefore, the abnormal intracellular Ca²⁺ dynamic in cerebral artery VSMC of MS rats could be related to changes in the functional activity of these proteins, which deserves further investigation.

This research underscores the complex interplay between MS and intracellular Ca²⁺ dynamics, focused on SERCA pump and/or RyRs as potential targets for therapeutic intervention in vascular complications linked to MS.

References

1. Gaitán-González P., et al. (2021). *World Journal of Diabetes*, **12**(10), 1704.
2. Fernández-Velasco M., et al. (2014). *Cell Calcium*, **56**(5), 397-407.
3. León-Martínez, M. (2017). Tesis de Maestría. Departamento de Bioquímica. Cinvestav-IPN.

KINETICS OF TWO RECOMBINANT *CAPSICUM ANNUUM* L. SOLUBLE INORGANIC PYROPHOSPHATASES WITH DIFFERENT PHYLOGENETIC ORIGIN

Ximena Carranza-Trinidad¹, Lilian Gabriela Valencia-Turcotte¹ and Rogelio Rodríguez-Sotres^{1*}

¹Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México, Av. Universidad 3000, Col. Cd. Universitaria, Alc. Coyoacán, c.p. 04510. Cd. México.

*E-mail: sotres@unam.mx

Classic soluble inorganic pyrophosphatases (PPa, EC 3.6.1.1) are magnesium-dependent hydrolases, essential to cell survival and growth, because these hydrolyse the pyrophosphate formed as biosynthesis byproduct. These enzymes share a common fold (pfam PF00719) but have been subdivided into proteins with eukaryotic (PPa-ek) and prokaryotic (PPa-pk) origin. Their kinetic behaviour has been best characterized in baker's yeast (ScPPa-ek) and in *Escherichia coli* (EcPPa-pk), but plant genomes encode representatives of both groups. Plants have several cytoplasmic PPa-pk isoforms, but usually only one chloroplastic PPa-ek¹. Here we report the cloning and expression of two proteins from *Capsicum annuum* L. (chilli pepper), as recombinant fusion forms (MBP-CaPPa-ek, and MBP-CaPPa-pk4). This is the first report of a plant eukaryotic fully-active recombinant isoform (MBP-CaPPa-ek) expressed in bacteria. Their saturation kinetics was compared to previous kinetic data reported for two enzymes from *Arabidopsis thaliana* (AtPPa-pk1 & AtPPa-pk4)². While the CaPPa-pk4 isoform did conform well the kinetic model for the Arabidopsis enzymes, the CaPPa-ek enzyme initial velocity pattern could not be fitted to the same model. Theoretical molecular models, Molecular Dynamics simulations and semiempirical Quantum Mechanical calculations were used to propose a possible explanation for the differences, and these seem related to changes in the Mg²⁺ binding sites.

Funding: LANCAD-UNAM-DGTIC-215, CONAHCyT A1-S-8650, PAPIIT-DGAPA IN219518, PAIP-FQ 5000-9122

References

1. Gutiérrez-Luna, Navarro de la Sancha, Valencia-Turcotte, Vázquez-Santana, *et al.*, (2016) *Plant Science* 253, 229-242
2. Navarro de la Sancha, Coello-Coutiño, Valencia-Turcotte, Hernández-Domínguez, *et al.*, (2007) *Plant Science* 172, 796-807.

IDENTIFYING A HYDROPHOBIN CLASS I FROM *AGARICUS BISPORUS*: PRODUCTION OF DIFFERENT AMYLOID-LIKE FIBRILS

Jesús Rojas¹, Hugo Nájera¹

¹Laboratorio de Biofísicoquímica, Departamento de Ciencias Naturales, Universidad Autónoma Metropolitana-Cuajimalpa. Av. Vasco de Quiroga 4871. Col. Santa Fe Cuajimalpa, Mexico City. CP. 05348. México.
E-mail: jrojas@cua.uam.mx

Hydrophobins (Hfb) are small amphiphilic extracellular proteins produced by filamentous fungi; they are surface-active proteins, and their functions are mainly related to their ability to self-assemble into amphipathic monolayers at hydrophobic–hydrophilic interfaces. Their molecular weight is between 7-15 kDa; they are characterized by having eight conserved cysteine regions in their structure¹. Hfb are classified into classes I and II, depending on hydrophobicity patterns, purification requirements, and their ability to form stable structures called fibrils or rodlets, which are amyloid-like fibril structures. The fibrils generated by class I can be dissolved by treatment with trifluoroacetic acid. In contrast, the aggregates produced by class II can be dissolved by treatment with 60% ethanol and 2% sodium dodecyl sulfate. Hfb amphipathic nature is essential to change the nature of the surface from hydrophilic to hydrophobic or vice versa². Therefore, this work aimed to extract and purify hydrophobin class I from the fungus *Agaricus bisporus* to identify and predict its structure and the production of fibrils on different surfaces. The extraction, purification, and identification of hydrophobin class I (Hfb-I) were performed from the outer cap of *Agaricus bisporus*, as well as the prediction of the structure and production of amyloid-type-like fibers on different surfaces. Extraction and purification were efficient for Hfb-I, obtaining a band of ~12 kDa. Sequence identification showed eight cysteine residues; structure prediction exhibited α -helices and β -sheets. Hfb-I increased the contact angle in glass and mica. Fibrillogenesis decreased at basic pH, while fibril formation was favored at acidic and neutral pH. Scanning electron microscopy analysis showed that Hfb-I produced different amyloid-like structures in glass and mica.

References

1. Wösten, H. A. (2001). *Annu. Rev. Microbiol.* **55** (1), 625-646.
2. Kulkarni, S., Nene, S., Joshi, K. (2017). *Process Biochem.* **61**,1-11.

METABOLIC CHANGES AND ANTIOXIDANT RESPONSE IN *USTILAGO MAYDIS* GROWN IN ACETATE

Lucero Romero-Aguilar¹, Katia Daniela Hernández Morfin², Juan Pablo Pardo-Vázquez¹
and Guadalupe Guerra Sánchez²

¹Universidad Nacional Autónoma de México, Facultad de Medicina, Departamento de Bioquímica. Circuito Interior, Ciudad Universitaria, Coyoacán, Ciudad de México C.P. 04510, Mexico. ²Instituto Politécnico Nacional, Departamento de Microbiología.

¹. Plan de Carpio y Plan de Ayala S/N Santo Tomás, Miguel Hidalgo, Ciudad de México C.P. 11340, Mexico.

E-mail: lusromaguila@bq.unam.mx, pardov@bq.unam.mx

Abstract. In this research, we studied the metabolic adaptation of cells growing in acetate as a carbon source by measuring the activities of key enzymes involved in glycolysis, gluconeogenesis, and the pentose phosphate pathways. Since growth on acetate induces oxidative stress, the activities of some antioxidant enzymes were also assayed. The results show that cells grown in acetate plus nitrate did not increase the amount of lipid droplets, but increased the activities of glutathione reductase, glutathione peroxidase, catalase, and superoxide dismutase, suggesting a higher production of reactive oxygen species in cells growing in acetate. As expected, the phosphofructokinase-1 had the lowest specific activity in the glycolytic pathway. Moreover, the activity of the phosphoenolpyruvate carboxykinase, a gluconeogenic enzyme, was present only in the acetate condition. In the presence of acetate as the only carbon source, *U. maydis* synthesized fatty acids, which were directed into the production of phospholipids and neutral lipids for biomass generation, but without any excessive accumulation of lipid droplets.

References

1. Romero-Aguilar L, Hernández-Morfin KD, Guerra-Sánchez G, Pardo JP. Metabolic Changes and Antioxidant Response in *Ustilago maydis* Grown in Acetate. *J Fungi (Basel)*. 2023 Jul 14;9(7):749. doi: 10.3390/jof9070749. PMID: 37504737; PMCID: PMC10381545.

PHYLOGENETIC DISTRIBUTION AND RECONSTRUCTION OF ANCESTRAL PHYTASES

Rogelio Rosas¹, D. Alejandro Fernández Velasco¹

¹Laboratorio de Fisiología e Ingeniería de Proteínas. Departamento de Bioquímica. Facultad de Medicina, Universidad Nacional Autónoma de México, Universidad 3000, Coyoacán, Ciudad de México, México.

E-mail: rogelio.rosas@ciencias.unam.mx

Phosphorus (P) is an essential element for life, playing a crucial role in various biological processes including DNA and RNA synthesis. However, phosphorus is a non-renewable resource, and its availability in agricultural soils can be limited. Phytate is the primary storage form of phosphorus in plant seeds; however, it is not readily digestible by monogastric animals, limiting the availability of phosphorus for animal nutrition. In the agroecological context, phytate can have a negative impact on phosphorus use efficiency and animal health. Phytases are enzymes capable of degrading phytate and releasing the phosphorus it contains. Supplementation of animal diets with phytases can improve phosphorus digestibility and nutrient absorption, leading to better animal growth and performance. The known phytases are grouped into four structural categories: purple acid phytases (PAPhy), phosphotyrosin phytases (PTPhy), beta propeller phytases (BPPhy), and histidine acid phytases (HAPhy). Ancestral sequence reconstruction (ASR) is a powerful technique that allows the inference of amino acid sequences of enzymes that existed in the past. ASR has the potential to be used to develop new enzymes with improved properties, such as higher catalytic activity, substrate specificity, or thermostability. The generation of thermostable phytases using ASR could expand their range of application in industrial processes and improve their efficiency.

Homology searches were performed using model sequences of phytases crystallized with substrate to determine their phylogenetic distribution. After a cleaning process, the identified sequences were aligned, and phylogenetic trees were constructed.

HAPhys and PTPhy sequences were found in all three domains of life, while BPPhys sequences were only found in Firmicutes, particularly in the genus *Bacillus*, and in some fungoid protists. In the case of PAPhys, sequences were found in algae and in the major plant groups. High variability was found among BPPhys, even in organisms of the same genus. This same pattern of variability was also present in PTPhy, being extremely variable within Firmicutes and with very little similarity when compared to plants. PAPhys sequences are mainly from algae and are more present in plants than HAPhys, whose sequences are found in all domains, although with a higher presence in fungi and bacteria and to a lesser extent in plants. HAPhys sequences have the largest number of sequences with conserved segments, with the same pattern of variability and with limitations in sequence homology between eukaryotic and prokaryotic organisms.

References

1. Gen, X., et al. (2013). *Annual Review of Animal Biosciences*, **1**, 283-309.
2. Hochberg, G., Thornton, J. (2017). *The Annual Review of Biophysics*, **46**, 247-269.
3. Scott, B., Koh, K., Rix, G. (2024). *Current Research in Structural Biology*, **7**.

ANTIOXIDANT CAPACITY OF ASPARAGUS UNDER DIFFERENT GROWING METHODS

Adela Guadalupe Rosas Rosas¹; Juan Guillermo Cruz Castillo²; Diana Guerra Ramírez³;
Ana María Castillo González¹

¹Universidad Autónoma Chapingo, Departamento de Fitotecnia. Carretera México-Texcoco km 38.5, Chapingo, Estado de México, C. P. 56230, ²Centro Regional Universitario Oriente, Km. 6 Carretera Huatusco – Jalapa, Huatusco, Veracruz, ³Universidad Autónoma Chapingo, Departamento de Preparatoria Agrícola. Carretera México-Texcoco km 38.5, Chapingo, Estado de México, C. P. 56230.
E-mail: rosasag18@gmail.com

Asparagus is a perennial herbaceous plant formed by a subway part that has a main rhizome and this acts as a union between the root system and the aerial part, which is called fern and is constituted by erect stems and modified leaves where flowers and fruits develop and chemical substances are converted into organic matter in order to elaborate the necessary reserves¹. Last few years, asparagus production has had a remarkable rise, worldwide, one out of every one hundred tons harvested comes from the Mexican field ². The asparagus plant has certain nutrient demands without which it does not develop favorably, due to the growing demand to return to a sustainable agriculture, where the least possible damage to the environment, have been looking for alternative nutrition in an organic way, such is the case of bocashi, which is an organic fertilizer that is integrated into the soil to increase the amount of microorganisms, improve the physical characteristics and provide nutrients to plants, this will be reflected in its nutritional and nutraceutical quality³. For the above reasons, in this research, phenolic content, flavonoids, and antioxidant activity were determined by FRAP and ABTS methods in asparagus grown under different methods in Atenco, State of Mexico (AQ and ASO) and Huatusco, Veracruz (HF and HS). Data were analyzed under a completely randomized design, for total phenols, ASO and AQ were statistically equal with 6.0748 and 6.0748 mg EAG/g respectively, but different from HS and HF. For flavonoids, ASO was statistically superior to HS and HF with 3.3457 mg EAG/g. In the FRAP and ABTS trials, ASO was statistically superior to HS and HF, with a content of 25.75 $\mu\text{mol ET/g}$ and 201.77 $\mu\text{mol Trolox/g}$ respectively, indicating that the asparagus produced in Atenco with organic fertilizer had similar characteristics to those grown by the conventional method and that there is an area of opportunity in Huatusco to improve its production.

References

1. Del Pozo-L., A. (1999). Morfología y funcionamiento de la planta. En: El cultivo del espárrago (pp. 9-28). Boletín INIA N° 6. Chillán, Chile: Instituto de Investigaciones Agropecuarias (INIA)- Ministerio de Agricultura.
2. Sistema de Información Agroalimentaria y Pesquera (SIAP), 2020.
3. Boudet, A., Chinchilla, V., Boicet, T., González, G. (2015). Efectos de diferentes dosis de abono orgánico tipo bocashi en indicadores morfológicos y productivos del cultivo de pimiento (*Capsicum annum L.*) var. California Wonder. Centro agrícola, 42(4)

CANNABIDIOL ATTENUATES PALMITIC ACID-INDUCED INFLAMMATORY SIGNALING IN HUMAN MACROPHAGES BY CANNABIDIOL: INSIGHTS INTO METABOLIC STRESS MITIGATION

Rueda Munguía M.¹, Luévano Martínez, L.A.¹, García Rivas G. de J.¹, Lozano García O.*

¹Instituto Tecnológico y de Estudios Superiores de Monterrey, San Pedro Garza García, N.L.

*omar.lozano@tec.mx, OLG.

Introduction. Metabolic diseases like obesity are characterized by chronic low-grade inflammation, increasing the risk of type 2 diabetes, cardiometabolic diseases, hypertension, and mortality. Obese individuals present elevated circulating levels of palmitic acid (PA) which triggers inflammation responses in macrophages. PA is a pro inflammatory fatty acid, which acts as an agonist for toll-like receptors (TLRs), particularly TLR2 and TLR4, PA activates signaling cascades similar to those induced by lipopolysaccharide (LPS) inducing endoplasmic reticulum (ER) stress and increasing pro-inflammatory cytokines such as IL-1 β , TNF- α , and IL-6. Cannabidiol (CBD) from *Cannabis sativa*, protects against PA-induced inflammation by modulating tumor necrosis factor-alpha (TNF- α), reactive oxygen species (ROS), and nitric oxide (NO). CBD may interact with peroxisome proliferator-activated receptors (PPARs), particularly PPAR γ and PPAR α , which are involved in lipid metabolism and inflammation. Understanding macrophages behavior in response to PA and how CBD can modulate these responses is relevant for elucidating chronic inflammation and metabolic dysfunction mechanisms opening avenues for developing therapeutic strategies against metabolic conditions. The study was based on the need to understand the effects of palmitic acid and CBD on macrophage function given their relevance in metabolic diseases.

Methods. Macrophages were obtained by differentiating human monocytes derived from the U937 cell line, using 50 ng/mL of phorbol 12-myristate 13-acetate in 72 hours. Subsequently, they were treated at a concentration of 200 μ M PA in combination with 10 μ M CBD for 24 hours. Metabolic activity was evaluated by Alamar blue assays, triglyceride accumulation and cell viability were determined using kits and crystal violet assay, cytokines were measured by flow cytometry. The activity of PPAR γ and PPAR α was assessed using specific agonists and antagonists to determine their roles in the observed effects. Experiments were done in triplicate and data analyzed by ANOVA.

Results. The results suggest that treatment with palmitic acid and CBD modulates macrophage function. Changes in metabolic activity were observed upon exposure to PA, while CBD attenuates triglycerides (TG) accumulation without affecting cell viability. Notably the group treated with CBD showed TG levels similar to the control group, whereas the PA group exhibited 2.5-fold increase in TG, with a p-value of 0.001 indicating a statistically significant difference.

Conclusions. PA increased metabolic activity, while CBD reduced triglyceride accumulation. Future perspectives will investigate cellular respiration and the metabolic activity pathway through which CBD attenuates inflammation.

Keywords: *Cannabidiol, inflammation, macrophages.*

THE ROLES OF TRANSCRIPTION FACTOR SOG1 OF *ARABIDOPSIS THALIANA* BEYOND OF ITS NUCLEAR FUNCTION IN ROOT

Karem Sánchez Martínez¹, Fernando Guzmán-Chávez², José Antonio Pedroza-García^{1*}

¹Departamento de Bioquímica, Facultad de Química, UNAM, ²CONAHCYT-Instituto de Investigaciones Biomédicas, UNAM
e-mail: karem.sm10@gmail.com; *joped@quimica.unam.mx

Being sessile organisms, plants are exposed to adverse conditions in their environment, which can induce damage to their nuclear and organellar deoxyribonucleic acid (DNA), that decreases the stability of its genome, affecting their growth and survival. As a mechanism to cope with severe DNA damage, there are highly coordinated cellular networks, collectively called the DNA damage response (DDR). In yeast and mammals, the transcription factor p53 is a master regulator of the DDR, which controls the expression of genes associated with DNA repair mechanisms, cell cycle arrest and programmed cell death when the damage is severe. Moreover, p53 has functions beyond of its role as transcription factor in the nucleus, it has been reported that p53 is also located in mitochondria where interacts with other protein: 1) to triggers cell death mechanism and 2) with proteins of replication and repair machinery of mitochondria, indeed, p53 can bind directly to DNA mitochondria, therefore p53 participates actively to safeguard the mitochondria integrity. In plants, the transcription factor SOG1 has homologous functions to p53 activating the DDR in nucleus, although, both factors do not have conserved sequences. Recent studies have shown that SOG1 up-regulates hundreds of genes how response to DNA damage, several of them plant DDR specific genes. Until now, it is well known the functions of SOG1 in root meristems as transcription factor in nucleus. However, it has not explored if SOG1 is involved in maintaining the DNA integrity in leaf and whether SOG1 is in mitochondria and/or chloroplast. To respond to these questions we generated the *promSOG1::SOG1-GFP* construct, which was used to transform protoplasts from *Arabidopsis* rosette leaves, this analysis revealed that SOG1 has localization in the chloroplast and mitochondria, in addition to its nuclear localization. Additionally, stable lines of *promSOG1::SOG1-GFP* were generated and leaves were challenged to genotoxins revealing that SOG1 is accumulated in different leaf cell-type of treatment depend-manner. Therefore, these results shed light novel roles of SOG1 to DNA integrity maintaining in chloroplast and mitochondria and its potential functions in the division and differentiation of leaf cells in response to DNA damage.

THE USE OF THIOFLAVIN T TO ESTIMATE THE PLASMA MEMBRANE POTENTIAL (PMP) IN DIFFERENT YEAST STRAINS AND THE EFFECT OF pH

Norma Silvia Sánchez¹, Francisco Padilla-Garfias¹, Yazmín Ramiro-Cortés², Minerva Araiza-Villanueva¹, Martha Calahorra¹ and Antonio Peña¹

¹Departamento de Genética Molecular, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Circuito Exterior s/n, Ciudad Universitaria, México City 04510, México; ²Departamento de Neurodesarrollo y Fisiología, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Circuito Exterior s/n, Ciudad Universitaria, México City 04510, México.
E-mail: nsanchez@ifc.unam.mx

Previously, the application of the cationic dye thioflavin T (ThT) for assessing the electrical plasma membrane potential difference (PMP) in baker's yeast was described, using fluorescence changes, and considering correction factors for dye binding to internal cell components. It was deemed important to investigate the feasibility of this approach with other yeast strains, prompting an investigation into alternative methods such as flow cytometry and multi-well plate readers for PMP estimation. A pair of *Saccharomyces cerevisiae* strains (W303-1A and FY833) and various non-conventional yeasts including *Debaryomyces hansenii*, *Candida albicans*, *Meyerozyma guilliermondii*, and *Rhodotorula mucilaginosa* were included in the study. Across different conditions, fluorescence-based PMP estimation yielded consistent results with all strains, further validated by assessments on mutants of the main monovalent transporters, which confirmed the efficacy of ThT' in PMP monitoring.

Likewise, the assessment of yeast PMP responses to varying pH values and K⁺ concentrations involved evaluating fluorescence changes and ThT accumulation. Qualitative observations at pH 4.0 indicated a slightly lower PMP compared to pH 6.0 and 7.0. Applying the Nernst equation to ThT concentrations in and out of cells, allowed for PMP estimation at different pH levels, aligning with fluorescence-based observations. Given yeast's exposure to low pH environments during fermentation, maintaining a robust PMP for survival is crucial, so the results are trustable. The probable contribution of fermentation-derived bicarbonate to PMP establishment was also considered. These experiments reiterated the effectiveness of ThT-based methods for PMP analysis.

References:

1. Peña, A.; Sánchez, N.S.; Padilla-Garfias, F.; Ramiro-Cortés, Y.; Araiza-Villanueva, M.; Calahorra, M.
2. The Use of Thioflavin T for the Estimation and Measurement of the Plasma Membrane Electric Potential Difference in Different Yeast Strains. *J. Fungi* **2023**, *9*, 948. <https://doi.org/10.3390/jof9090948>
3. Antonio Peña, Norma Silvia Sánchez, Yazmín Ramiro-Cortés, and Martha Calahorra.
4. Effects of medium pH on the yeast plasma membrane potential. *Arch. Biochem. Biophys*, 2024. Under Review, May, 2024.

AKT AND PKC INHIBITORS ACTIVATE AN ER Ca^{2+} LEAK INVOLVING Sec61 TRANSLOCON IN HELA CELLS

Lizeth Sandoval¹, Juan Manuel Arias², Agutín Guerrero¹

¹Cinvestav, IPN Avenue 2508, Mexico City. ²Facultad de Instituto superior de Iztacala-UNAM, Tlalnepantla de Baz 54090, Mexico State
E-mail: lsandoval@cinvestav.mx

The endoplasmic reticulum (ER) participates in a wide variety of cellular processes, among them xenobiotic detoxification, lipid synthesis, protein translocation, folding, and modification; generation of lysosomes and secretion granules; and it is also the primary intracellular Ca^{2+} store. The ER membrane expresses the SERCA pump, which uses ATP to accumulate Ca^{2+} into the ER; also, there are Ca^{2+} release channels such as IP_3R and RyR, and Ca^{2+} leak channels including the translocon ($\text{sec61}\alpha$), presenilin, Orai2, and 3, and TRPVs. The $[\text{Ca}^{2+}]_{\text{ER}}$ homeostasis depends on the balanced action of SERCA pumps and leak channels, preventing ER Ca^{2+} overload. The phosphoprotein translocon complex allows co-translational and post-translational protein import into the ER. Additionally, puromycin, a protein synthesis inhibitor that induces the premature release of the nascent peptide, reduces the ER $[\text{Ca}^{2+}]_{\text{ER}}$ without the participation of the release channels but by leaving open Sec61 translocon. On the other hand, anisomycin, a protein synthesis elongation inhibitor, inhibits depletion of the ER Ca^{2+} store by leaving the nascent peptide in the translocon pore lumen, functioning as a plug. Ca^{2+} leak through the translocon is restricted by various conditions, including the nascent protein, the own plug of $\text{Sec61}\alpha$, accessory proteins (BiP, Calmodulin, and Sec62), and phosphorylation (PKC)¹⁻³. Staurosporine (Sts) is a general kinase inhibitor that can induce a transient increase in the $[\text{Ca}^{2+}]_i$ from the ER Ca^{2+} store, prompting us to identify the nature of the ion channel involved and the protein kinases that might be participating.

We have studied the effect of Sts on the internal Ca^{2+} stores with different ER luminal $[\text{Ca}^{2+}]_{\text{ER}}$ indicators (Mag-Fluo, erGAP3, and R-Cepia). Sts induced a rapid reduction in the $[\text{Ca}^{2+}]_{\text{ER}}$, which was blocked by emetine, an inhibitor of the Ca^{2+} leak activity of Sec61 translocon. The previous depletion of the ER Ca^{2+} stores via activation of IP_3Rs , together with blocking SERCA pumps, inhibited the effect of Sts on the $[\text{Ca}^{2+}]_{\text{ER}}$. However, inhibition of IP_3Rs did not modify the Ca^{2+} -releasing activity of Sts. Specific PKC inhibitors (Gö6983 and Gö6976) partially explained the effect of Sts, implying the participation of other kinases. The Akt inhibitor (Akti VIII) reduced the $[\text{Ca}^{2+}]_{\text{ER}}$ and was mutually exclusive with the effect of Sts. Indeed, Akti VIII did not affect the $[\text{Ca}^{2+}]_{\text{ER}}$ when cells were cultured in the absence of serum for 24 hours, a condition that deactivates Akt. In this condition, the inhibition of PKC still explained only half of the Sts effect on $[\text{Ca}^{2+}]_{\text{ER}}$ reduction. These data suggest that the activity of Akt, PKC, and another unidentified kinase redundantly inhibit the Sec61 translocon Ca^{2+} leak role in HeLa cells.

References

1. S. Lang, F. Erdmann, M. Jung, R. Wagner, A. Cavalie, R. Zimmermann, *Channels*, (2011) 5, 228–235
2. A. Dagnino-Acosta, A. Guerrero-Hernandez, *Front. Physiol.* (2022), 13 1–12.
3. C. Körbel, M. Linxweiler, Menger, R. Zimmermann, M. Greiner, M. Musterman, P. Placeholder, *BioMol Concepts*, (2018), 9, 53–63.

ZIKA VIRUS REGULATES CELL SURVIVAL THROUGH MODULATION OF MIR-125A EXPRESSION AND MITOCHONDRIAL FUNCTION

María E. Santana-Román, Gustavo Pedraza-Alva and Leonor Pérez Martínez

Laboratorio de Neuroinmunobiología, Departamento de Medicina Molecular y Bioprocesos,
Instituto de Biotecnología (IBt), Universidad Nacional Autónoma de México (UNAM),
Cuernavaca, Morelos 62210, Mexico.
Correspondence: leonor@ibt.unam.mx

Zika virus (ZIKV) is a single-stranded RNA virus belonging to the Flaviviridae family, and is the causative agent of ZIKV-associated congenital syndrome in newborns and Guillain-Barré syndrome in adults. ZIKV infects central nervous system (CNS) cells, causing reduced proliferation, inhibition of cell differentiation, and activation of cell death. Furthermore, cell survival is compromised during ZIKV infection as a result of changes in mitochondrial function and dynamics. ZIKV infection diminished mitochondrial fusion protein 2 (mitofusin 2), promoted cytochrome C release, reactive oxygen species generation, and aggregation of the pro-apoptotic factor BAX in the outer mitochondrial membrane.

Small non-coding RNAs (miRs) are regulators of gene expression at the post-transcriptional level by binding to their target messenger RNAs (mRNAs). Among miRs, miR-125a has been detected in the serum of individuals ZIKV-positive, and in neural stem cells ZIKV-infected. The miR-125a is expressed in the developing CNS and plays a role in neuronal differentiation. Additionally, miR-125a regulates cell survival by downregulating mRNAs encoding for proteins involved in mitochondrial function and dynamics, such as TP53-regulated inhibitor of apoptosis 1 (TRIA1), mitofusin 1, mitofusin 2, and hexokinase 2.

Research ongoing in our group shown that ZIKV induces miR-125a expression in embryonic neuronal cells. However, the role of miR-125a in modulating mitochondrial function during ZIKV infection in immature CNS cells and the signaling pathways mediating its role in cell survival, remains unknown. Therefore, we propose that ZIKV alters mitochondrial function and dynamics, compromising cell survival via upregulating miR-125a expression.

This study employs the SH-SY5Y neuronal cell line to evaluate miR-125a expression, mitochondrial dynamics, and cell survival in response to ZIKV.

This work was partially funded by DGAPA/UNAM projects IN217822 and IN216922. CONAHCYT supported PhD studies through the “Beca nacional para estudios de posgrado” CVU 856866.

Keywords: Zika virus, mitochondria, miR-125a, and SH-SY5Y cell line.

IN SILICO, IN VITRO AND IN VIVO ANALYZES DEMONSTRATE THAT ARSENIC AFFECTS CELLULAR RESPIRATION BY INTERACTING WITH CYTOCHROME C AND UBIQUINONE

Erick Sierra Campos¹, Grace X. Terrazas Quiñones¹, Carlos A. Rocha Martínez¹,
Erica K. Ventura García¹, Mónica A. Valdez Solana¹

¹Universidad Juárez del Estado de Durango, Av. Artículo 123 s/n Fracc, Filadelfia, 35019 Gómez Palacio, Dgo. Mexico.
E-mail: ericksier@ujed.mx

Mitochondria are organelles responsible for metabolism, they play an important role in hepatotoxicity, due to their abundance in hepatocytes. Arsenic is toxic to several organs, the liver being the most affected. A growing body of research demonstrates that arsenic is particularly hazardous to the mitochondria of cells, causing oxidative stress and disrupting a variety of signaling pathways and activities¹. Arsenic(III) in isolated mitochondria inhibits complexes I and II, increases mitochondrial ROS formation, lipid peroxidation, potential mitochondrial membrane collapse, cytochrome c release, and mitochondrial inflammation in a concentration-dependent manner². Therefore, we investigated how cytochrome c (cytc) and ubiquinone (Q) are affected during arsenic-induced mitochondrial dysfunction. Initially, an *in silico* analysis was self-performed with bovine heart cytc (PDB ID 6FF5) and the inorganic forms (arsenate ion, ART; and arsenite (3-), AST) with the LigBind server. The results show that ART interacts with several amino acids (H18, P30, N52, W59, Y67 and M80). While AST interacts only with H18, they are important for coordinating links with the heme group. Spectroscopic analysis (200-800 nm) revealed a significant decrease in the area under the cytc signal curve in the presence of AST. This decline was even more pronounced when cytc was pre-incubated with AST. In addition, changes in signal patterns, such as the decrease and widening of the Soret peak, indicated a change in the structure of the heme group. Similarly, the presence of AST affects the ability to reduce Q at a wavelength of 275 nm. On the other hand, oxygen consumption assays revealed a dose-dependent inhibitory effect of AST on respiration in mitochondria isolated from rat liver with arsenite. Complex I was the most affected, showing a 76% inhibition with the highest dose of AST (2.5 mg/kg). However, complex II showed an increase in oxygen consumption with succinate, suggesting a compensatory response to the dysfunction of the other complexes. Finally, inhibition assays between sodium arsenite and each complex (I and II) were measured spectrophotometrically. To our surprise, complexes I and II maintained their activity at low arsenite concentrations and only at concentrations <300 µM was inhibition greater than 50% in each of the mitochondrial complexes. Therefore, we conclude that arsenite does not interact directly with the complexes and that the inhibition of electron flow in respiration, but is due to the chemical interaction between arsenic and Q and/or cytc.

References

1. A Prakash C, Chhikara S, Kumar V.. Biol Trace Elem Res. 2022 Jan;200(1):261-270. doi: 10.1007/s12011-021-02624-2.
2. Hosseini MJ, et al., Iran J Pharm Res. 2013 Winter;12(Suppl):121-38.

EFFECT OF THE ETHANOLIC LEAVES EXTRACT OF *CALLISTEMON CITRINUS* ON THE ANTIOXIDANT ACTIVITY IN THE LIVER OF RATS ADMINISTERED WITH INDOMETHACIN

Uriel Noé Solano-Candia¹, Luis Alberto Ayala-Ruiz¹, Patricia Ríos-Chávez¹

¹Facultad de Biología, Universidad Michoacana de San Nicolás de Hidalgo,
edificio B-4. Cd Universitaria, 58030 Morelia, Michoacán.
1415049d@umich.mx

Nonsteroidal anti-inflammatory drugs (NSAIDs) have analgesic, antithrombotic, and antipyretic properties. Indomethacin (Indo) is mainly used as an analgesic, its mechanism of action is uncoupling oxidative phosphorylation and prevents platelet aggregation. The used of this drug promotes the generation of reactive oxygen species (ROS). Indo produce damage in the gastrointestinal, renal, hematologic and liver tissues. It is important to find new alternatives to reduce the damage cause by Indo. In this context, *Callistemon citrinus* has reported antioxidant properties. Hence, it's crucial to evaluate its effect on rats administered IndoM. The present stud was focused on investigating the antioxidant effect of ethanolic leaves extract of *Callistemon citrinus* in liver of rat's treatment with indomethacin. The rats were divided into three groups (n=5). Group 1: control, Group 2: indomethacin, Group 3: indomethacin and *C. citrinus*. All the groups were fed with a standard diet (Rodent diet®) and tap water *ad libitum* for 15 weeks. At the end of this period, Groups 2 and 3 were administered with a single dose of indomethacin before being sacrificed (30 mg/kg, orally); additionally, Group 3 was administered with a single dose of *C. citrinus* (250 mg/kg, orally). Animals were sacrificed and the liver was excised, the enzymatic activity of catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPx), and the levels of reduced glutathione (GSH) were determined. IndoM showed a significant reduction of antioxidant enzyme as SOD and CAT. Whereas the administration of ethanolic extract of *C. citrinus* significantly increase the SOD activity, meanwhile CAT and GPx were not improved. The protective effect of *Callistemon citrinus* could be attributed to its ability to boost the antioxidant system.

MORPHOLOGICAL AND GENETIC CHARACTERIZATION OF FRUIT DEHISCENCE IN *BIXA ORELLANA* L.

M.C. Rocío Tamayo-García, Dr. José Alberto Narváez-Zapata, M.C. Margarita de Lourdes Aguilar-Espinosa, Lic. Felipe Alonso Barredo-Pool, Dra. Renata Rivera-Madrid*

Unidad de Biología Integrativa. Centro de Investigación Científica de Yucatán, A.C. calle 43, No. 130, Col. Chuburná de Hidalgo. Mérida, Yucatán. C.P. 97200.
Dra. Renata Rivera-Madrid renata@cicy.mx Cel 999 993 5759

Bixa orellana L. is a plant of commercial importance that contains a pigment known as bixin in the aril of its seeds. This pigment is used in the textile, pharmaceutical, cosmetic, and food industries. The fruit of *Bixa orellana* L consists of a capsule with two valves joined at the dehiscence zone, a crucial mechanism that regulates the exposure of seeds to the environment. This study characterizes the genetic and morphological traits of dehiscence in contrasting types of fruits in *Bixa orellana* L. To achieve this, the fruit transcriptomes from contrasting accessions were assembled and analyzed using bioinformatic tools. Quality control was performed with FASTQC, preprocessing with Trimmomatic, read alignment with Bowtie, transcriptome assembly with Trinity, and assembly quality analysis with Nx50/BUSCO/CDHit. Additionally, ORF prediction was carried out with TransDecoder, contaminant search with Kyoto Encyclopedia, annotation with Trinotate (BLASTp/BLASTx), quantification expression with Cufflinks, and differential expression with DESeq/edgeR. For morphological characterization, tissues were fixed in 4% paraformaldehyde, embedded in an ethanol gradient, and included in JB-4® Resin. The 3 µm sections were stained with 0.5% toluidine blue and Periodic Acid-Schiff (PAS). Observations were made with a Zeiss Germany® Axioplan microscope. The dehiscent fruits were enriched in metabolic pathways related to pigment, sugars, and photosynthesis. Morphocellular differences were observed in the valves, exocarp, vascular bundle sheath, placenta, gland distribution, and exocarp appendages. Finally, it was concluded that the dehiscence mechanism in *Bixa orellana* L. is analogous to that reported in Brassicaceae.

References

1. Dardick, C., & Callahan, A. M. (2014). Evolution of the fruit endocarp: molecular mechanisms underlying adaptations in seed protection and dispersal strategies. *Frontiers in Plant Science*, 5, 284. <https://doi.org/10.3389/fpls.2014.00284>
2. Trujillo-Hdz, J. A., Cárdenas-Conejo, Y., Turriza, P. E., et al. (2016). Functional polymorphism in lycopene beta-cyclase gene as a molecular marker to predict bixin production in *Bixa orellana* L. (achiote). *Molecular Breeding*, 36(7), 135. <https://doi.org/10.1007/s11032-016-0555-y>

EXPRESSION, PURIFICATION, AND CHARACTERIZATION OF 3-DEHYDROQUINATE SYNTHASE (DHQS) FROM *ACINETOBACTER BAUMANNII*

Atl Emiliano Tarbuck Valle¹, Carol Siseth Martínez Caballero¹ and Eduardo Hernández Vázquez¹

¹Institute of Chemistry, UNAM, Circuito Exterior s/n Circuito de la Investigación Científica, C.U., 04510 Ciudad de México, México.
E-mail: a_tarbuck@outlook.com

The global rise of antimicrobial resistance is considered a silent pandemic that, according to the World Health Organization¹, is situated as one of the greatest concerns of health globally. It's estimated that by the year 2050, drug-resistant infections will cause up to 10 million deaths per year. Specifically, the bacterium *Acinetobacter baumannii* is one of the most important pathogens associated with opportunistic infections in nosocomial environments and is one of the most serious ESKAPE organisms, a group of six highly virulent pathogens that effectively escape the effects of antibacterial drugs². This organism commonly targets the most vulnerable hospitalized patients, particularly those who are critically ill or immunocompromised. Hospital-acquired pneumonia is the most common affectation, followed by infections involving the central nervous system, skin and soft tissue, and even bones³.

Consequently, the principal objective of this work is the expression, purification, and characterization of a vital enzyme of *A. baumannii* that has become the target of various research groups: the 3-dehydroquinate synthase (DHQS). This enzyme, expressed by the *aroB* gene, catalyses the second regulatory step of the shikimate pathway facilitating the conversion of DAH7P to DHQ, requiring NAD⁺ and metal cofactors Zn²⁺ or Co²⁺. The shikimate pathway plays an essential role in bacteria, fungi, algae and other microorganisms as it's necessary for the biosynthesis of chorismic acid, a precursor to aromatic amino acids and other metabolites. This pathway is an attractive target for drug development due to its absence in mammals and potential for low human toxicity⁴.

DHQS, as well as other enzymes within the shikimate pathway, has already been investigated as antibacterial targets, and compounds have been found that lead to the inhibition of cell growth in drug-resistant *Mycobacterium tuberculosis*⁵, thus paving the way for the development of new antimicrobial drugs to help combat this global health crisis.

References

1. Antimicrobial Resistance Collaborators (2022). *Lancet*, **399**, 629-655.
2. Lee C-R, Lee JH, Park M, Park KS, Bae IK, Kim YB, Cha C-J, Jeong BC and Lee SH (2017). *Front. Cell. Infect. Microbiol.*, **7:55**, 1-2.
3. Peleg, A. Y., Seifert, H., & Paterson, D. L. (2008). *Clinical microbiology reviews*, **21(3)**, 538-582.
4. Tillery LM, Barrett KF, Dranow DM, Craig J, Shek R, Chun I, Barrett LK, Phan IQ, Subramanian S, Abendroth J, Lorimer DD, Edwards TE, Van Voorhis WC (2020) *Protein Sci.* **29(3)**, 789-802.
5. Zhu N, Wang X, Li D, Lin Y, You X, Jiang J, Xu Y, Jiang W, Si S. (2018). *Sci Rep.*, **8(1)**, 1-3

CRYSTALLIZATION OF THE CATECHOL 1,2-DIOXYGENASE ENZYME FROM *STUTZERIMONAS STUTZERI* GOM2

Dalia Isabel Torres Aguirre¹, Arisbeth G. Almeida-Juárez² and Enrique Rudiño Piñera²

¹Centro de Investigación en Dinámica Celular, UAEM, Cuernavaca Morelos, ²Instituto de Biotecnología, UNAM, Cuernavaca Morelos.

E-mail: dalia.torres@uaem.edu.mx

The enzyme catechol 1,2-dioxygenase (C12DO) is an intradiol dioxygenase that catalyzes the cleavage of the aromatic ring of catechol to produce cis,cis-muconic acid, using Fe³⁺ as a cofactor¹. The *catA* gene, responsible for encoding this protein, was identified in the bacterium *Stutzerimonas stutzeri* GOM2, isolated from marine substrate in the Gulf of Mexico². The C12DO from *S. stutzeri* GOM2 has a primary structure consisting of 312 amino acid residues and has been heterologously expressed in *E. coli*, including a 6-histidine tag at its C-terminal to facilitate purification. Although enzyme oligomerization has been observed to vary depending on ionic strength², its three-dimensional structure has not yet been determined. In the Protein Data Bank (PDB), structures of C12DOs from 7 different bacterial species are registered, the enzyme from *Pseudomonas arvilla* being the closest homologue to C12DO from *S. stutzeri* GOM2. Protein crystallization is a technique used to determine three-dimensional structures through X-ray diffraction³. Therefore, the aim of this work is to express, purify, and crystallize C12DO to determine its three-dimensional structure. Crystallization is a critical point within this process, as it is not a trivial step to achieve. To crystallize a protein, a high degree of purity is required. In this work we obtain C12DO with a >90 % of purity, achieved through two purification steps: nickel affinity chromatography followed by size exclusion chromatography. Monodispersity of sample was evaluated by dynamic light scattering. We obtain crystals of C12DO enzyme under various conditions, including 0.1 M magnesium acetate, 15% PEG 4000 with HFIP as an additive, with two different buffers, 0.1 M Tris-HCl pH 8.5 and 0.1 M glycine-NaOH buffer pH 9 and 9.5. The C12DO crystals were diffracted at LANEM, IQ -UNAM.

References

1. Cathleen A. Earhart, Matthew W. Vetting, Ramachandraiah Gosu, Isabelle Michaud-Soret, Lawrence Que, Douglas H. Ohlendorf (2005) Structure of catechol 1,2-dioxygenase from *Pseudomonas arvilla*. *Biochemical and Biophysical Research Communications*. Volume 338, Issue 1
2. Rodríguez-Salazar, J., Almeida-Juarez, A. G., Ornelas-Ocampo, K., Millán-López, S., Raga-Carbajal, E., Rodríguez-Mejía, J. L., Muriel-Millán, L. F., Godoy-Lozano, E. E., Rivera-Gómez, N., Rudiño-Piñera, E., & Pardo-López, L. (2020). Characterization of a Novel Functional Trimeric Catechol 1,2-Dioxygenase From a *Pseudomonas stutzeri* Isolated From the Gulf of Mexico. *Frontiers in microbiology*, 11, 1100. <https://doi.org/10.3389/fmicb.2020.01100>
3. Jan Drenth (2007) *Principles of protein x-ray crystallography*. Third edition. Springer. Chapter 1 Crystallizing a Protein

FERMENTATION OF CACAO TO MODULATE ITS CONTENT OF ACTIVE COMPOUNDS

Tatiana Torres Loera¹, Roberto Ruiz Medrano^{1,2}, América Padilla Viveros^{1,2},
Juan Carlos Estrada Mora³, Beatriz Xoconostle Cázares^{1,2}

¹Department of Biotechnology and Bioengineering, ²Doctorado Transdisciplinario para la Sociedad,
³Colección Nacional de Cepas Microbianas y Cultivo Celular. Centro de Investigación y de Estudios Avanzados del IPN.,
Av. Instituto Politécnico Nacional 2508, 07360 Mexico City, Mexico.
E-mail: bxoconos@cinvestav.mx

Raw cacao beans are used as the main component in chocolate production. Several studies have shown the potential beneficial health effects related to cocoa and chocolate consumption, attributed to different bioactivities such as antioxidant and anti-inflammatory¹.

Raw cacao beans are inedible, bitter, and do not possess its characteristic chocolaty flavour, therefore, a fermentation process is needed, where the action of microorganisms in a lactic and then alcoholic fermentation produce the chocolate. Cocoa production may have inconsistent quality between batches, likely due to the uncontrolled presence of microorganisms, including pathogenic bacteria and fungi during fermentation. Theobromine and caffeine are the main methylxanthines found in cacao beans, and their content dictates the chocolate quality. At biochemical level, theobromine is methylated to produce caffeine and the enzyme has inhibition by product. For this purpose, fermentation was carried out including yeasts or coffee beans. The obtained chocolate was then analyzed for its theobromine and caffeine content. The results of fermentation kinetics and metabolite content will be presented, including recommendations for good fermentation practices, that would allow producers to obtain chocolate with better quality and innocuity for the benefit of consumers.

References

1. Grassi, D., Desideri, G., & Ferri, C. (2010). Blood pressure and cardiovascular risk: Whatabout cocoa and chocolate? *Archives of Biochemistry and Biophysics*, 501:112–115.
2. Latif, R. (2013). Health benefits of cocoa. *Current Opinion in Clinical Nutrition and Metabolic Care*, 16:669–674.
3. Martínez-Pinilla, E., Oñatibia-Astibia, A., & Franco, R. (2015). The relevance of theobromine for the beneficial effects of cocoa consumption. *Frontiers in Pharmacology*, 6:30.

FUNCTIONAL ANALYSIS OF THE CATALASE-PEROXIDASE OF PHYTOPATHOGENIC FUNGUS *USTILAGO MAYDIS*

¹Marisol Ubaldo-Aguilar, ²Juan Pablo Pardo-Vázquez, ²Lucero Romero-Aguilar, ¹Aranzazú López-Jaime, ¹Rodrigo Martínez-Acosta, ³Miguel A. Rosas-Paz, ⁵

¹Benemérita Universidad Autónoma de Puebla. ²Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México. Edificio D, primer piso en la Facultad de Medicina Circuito Interior Ciudad Universitaria Avenida Universidad 3000 CP. 04510 CDMX. ³Laboratorio de Biología Molecular y Genómica, Facultad de Ciencias, Universidad Nacional Autónoma de México.
Correspondencia: Dra. Lucero Romero-Aguilar. lusromaguila@bq.unam.mx

Ustilago maydis is a promising model organism for biotechnology and biochemistry due to its high similarity to higher eukaryotes. Its genome has been fully sequenced, and it is not pathogenic to humans. This yeast has a catalase-peroxidase (cat-px) that confers protection against oxidative stress, contributing to preventing lipid, protein, and DNA oxidation. This research aimed to characterize the cat-px of *U. maydis* and physiological responses under oxidative stress induced by chemical agents. Cell viability, specific activity, and expression of the cat-px gene were analyzed under different stress conditions induced by hydrogen peroxide (H₂O₂) for 24 h. The *U. maydis* cat-px has a higher specific activity at low H₂O₂ concentrations than control cells. The enzyme was sensitive to high temperatures (40 °C) and showed no activity at pH 6. The expression of the cat-px gene was dependent on H₂O₂ concentration, with lower expression at higher concentrations. Cell viability was affected and reduced by 60% at high H₂O₂ concentrations. Moreover, the presence of H₂O₂ induces morphological changes in *U. maydis*, with thinner cells observed compared to the control.

¿WHY *BACILLUS LICHENIFORMIS* CAN MAKE AN AEROBIC RESPIRATORY WHIT CYANIDE IN THE MEDIA?

Daniel Uribe-Ramírez¹, Héctor Vázquez-Meza², Eliseo Cristiani-Urbina¹ and Juan Pablo Pardo-Vázquez²

¹Escuela Nacional de Ciencias Biológicas-IPN, Av. Wilfrido Massieu 699. Unidad Profesional Adolfo López Mateos, G.A.Madero C.P. 07738, CDMX, México, ²Facultad de Medicina-UNAM, Ciudad Universitaria, Av. Universidad 3000, Coyoacán, C.P. 04510, CDMX, México.
E-mail: pardov@bq.unam.mx

The main harmful effect of cyanide is the inhibition of the mitochondrial cytochrome c oxidase, which prevents aerobic respiration. Unlike the electron transport chain of eukaryotic mitochondria, bacteria have branched respiratory chains, and depending on the growth conditions, bacteria can use different electron transfer pathways. The ability of energy-generating machinery to be flexible may play a significant role in enabling free-living bacteria, such those in the genus *Bacillus*, to adapt to the frequent variations in oxygen and nutrient availability seen in their natural habitat. As a cyanide-degrading bacterium, *B. licheniformis* has been employed to minimize pollutants found in wastewater. The aim of this study was to investigate potential changes in *B. licheniformis* respiratory chain that could result from the medium's cyanide content. Different growth conditions (Nut and MM-CN) were used to cultivate *B. licheniformis*, and the cells' oxygen consumption was assessed. In addition, three inhibitors were used to test the presence of different respiratory complexes, flavone (960 μM) to inhibit alternative NAD(P)H dehydrogenases, antimycin A (28.8 μM) for complex III and KCN (1 mM) for cytochrome c oxidase. It was found that the flavone exhibited a 25% inhibition, but antimycin A showed no inhibition at all. The cyanide-induced inhibition values varied based on the culture media used for the microorganism. For example, in MM-CN-grown cells, the addition of cyanide increased oxygen consumption by the cells. These findings suggest that antimycin A-sensitive complex III is not being utilized, leaving the quinol oxidases aa₃ and bd. We found that there is not a 100% inhibition when cyanide was present in the respiratory medium, indicating that cyanide-sensitive cytochrome aa₃ predominates in the respiratory chain of cells grown in nutritive medium. Furthermore, the residual respiratory activity in these cells might be due to the presence of cytochrome bd, which is insensitive to cyanide.

CHARACTERIZATION OF MITOCHONDRIAL COMPLEX I FROM THE DIATOM *PHAEODACTYLUM TRICORNUTUM*

Gloria Vargas-Romero¹, Zhaida Aguilar-Gonzalez¹, Girian Jaguer-García¹, Mónica Rodríguez-Bolaños¹, Marco I. Valencia-Sánchez², Pierre Morsomme³, Hervé Degand³, Pierre Cardol⁴, Héctor V. Miranda-Astudillo^{1,*}

¹Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, Mexico, ²Department of Biochemistry and Molecular Pharmacology, New York University, New York, USA. ³Institut des Sciences de la Vie, Université Catholique de Louvain, Louvain-la-Neuve, Belgium. ⁴Genetics and Physiology of microalgae, InBioS/Phytosystems, University of Liège, Belgium. *hmiranda@iibiomedicas.unam.mx

Mitochondrial NADH:ubiquinol oxidoreductase is the larger membrane complex involved in the respiratory chain. Structurally, this enzyme can be described as two large domains, one embedded in the membrane, involved in proton translocation, while the second domain protrudes from the membrane and it is involved in the electron transfer process. Historically, the mitochondrial complex I from two model organisms, *Bos taurus* and *Yarrowia lipolytica* has been studied and characterized in detail giving a slightly similar shape, however, with the recent inclusion of other organisms like *Polytomella parva*¹, and *Euglena gracilis*², divergent structures with extra domains have been described.

Diatoms are an important group of organisms with a major role in ecological and biotechnological fields. An important cooperation between the two major energetic routes, i.e. mitochondrial respiration and photosynthesis, has been characterized³, nevertheless, no detail description of the composition of the any complex involved in the oxidative phosphorylation system has been described so far. In the present work we showed purification and characterization of the mitochondrial complex I from this species.

P. tricornutum was cultured in an Antares I photobioreactor using green-enriched light⁴, and total membranes were prepared. Mitochondrial complex I was extracted with N-dodecyl-beta maltoside and purified using two steps of liquid chromatography followed by blue native electrophoresis. We identified two bands of different molecular weight (1100 and 800 kDa) through NADH/NBT activity. Their polypeptide composition was solved using a 3D gel system, followed by the identification of the different subunits by LC-ESI-Q-TOF-MS quantitative analysis. Our results showed two functional complexes with the carbonic anhydrase domain, but no evidence of a supercomplex. We observed the presence of the canonical and several lineage-specific complex I subunits, together our results lead us to propose a structural adaptation of this complex due to the gas availability in this species.

Acknowledgments. This research was supported by grant IA204524 (PAPIIT, DGAPA, UNAM) and by the Institutional Program (IIBO-UNAM) “The production of biomolecules of biomedical interest in microorganisms”.

References

1. Klusch, N., et al. *Plant Cell* **33**, 2072–2091 (2021).
2. Miranda-Astudillo, H. et al. *Sci. Rep.* **8**, 9698 (2018).
3. Bailleul, B. et al.. *Nature* **524**, 366–369 (2015).
4. Rodríguez-Bolaños, M. et al.. *Appl. Biochem. Biotechnol.* (2023).

DETERMINATION OF LCM AND β -GLUCOSIDASE IN NALTEL MAIZE SPROUTS FROM THE SIERRA NORTE, OAXACA

Pedro Ronel Vásquez-Díaz¹, Carlos Alberto Matias-Cervantes², Margarito Martínez Cruz¹,
María Teresa Hernández Huerta², Eduardo Lorenzo Pérez-Campos¹,
Iván Antonio García Montalvo¹, Laura Pérez-Campos Mayoral²

¹Division of Graduate Studies and Research, National Technology of Mexico/IT Oaxaca. Oaxaca, Mexico.

²CONAHACYT – Faculty of Medicine and Surgery, Autonomous University “Benito Juárez” of Oaxaca. Oaxaca, Mexico.

*pedro.ronel.diaz@gmail.com, martinezcu9@hotmail.com, carloscervantes.ox@outlook.com, marte-hh28@hotmail.com

Naltel maize is one of the oldest in Mexico and is considered an important reservoir of resistance genes to pests, diseases and abiotic factors. The expression of lectin (LCM) and β -glucosidase (Glu) has been associated with disease resistance in hybrid maize SB-3021. The purpose of the present work was to determine the expression of LCM and Glu in varieties of maize Naltel de Altura of the Sierra Norte de Oaxaca (SNO). Naltel de Altura varieties were collected in the SNO: Ixtlán de Juárez (IJ), La Luz (LL) and San Juan Chicomezuchil (SJC). A kinetic assay of lectin and β -glucosidase activity was performed on 0-8 day sprouts of SNO maize Naltel varieties using the hybrid breeds Asgrow and Berentsen SB-302 as control. Lectin activity was determined by hemagglutination with type O blood, and by D-inhibition of hemagglutination (IHA) D-galactose². Glu enzyme activity was performed using p-nitrophenyl- β -D-glucopyranosis substrate (Sigma-Aldrich Co.)³. The protein concentration was determined using the method of Bradford⁴. Naltel of SJC had a higher total protein concentration of 68.2 mg/ml, in contrast to the other native varieties. Naltel of IJ presented greater lectin expression on day 3, with 8787.8 UHA/mg and SB-302, 2002.6 UHA/mg, on day 6, the three varieties had IHA by D-galactose. All three varieties of Naltel showed maximum enzyme activity on day 3; IJ with highest activity, 599.6 U/mg, and SB-corn302 on day 4, with 689.8 U/mg. Results show high expression of lectin and enzyme from germinated native varieties on days 3 and 4 compared to reported in maize Berentsen SB-302². Therefore, the high activity of LCM and Glu in Naltel of the SNO, gives guidance to consider them as an alternative for the evaluation of genes of biotechnological interest associated with mechanisms of resistance to abiotic factors.

References

1. Gómez-Arduro G., E. Cenicerós-Ojeda, L. Casados-Velázquez, C. Bencivenni, A. Sierra-Beltrán, B. Murillo-Amador, A. Tiessen. 2011. Genome-wide analysis of the B-Glucosidase gene family in maize (*Zea Mays* L. var B73). *Plant Mol. Biol.* 77:159-183.
2. Molina J., Landa A., Bautista G., Martínez M. y Córdoba F. 2004. Molecular association of lectine and β -glucosidase in corn coleoptile. *Biochimica et Biophysica Acta* 1674 (2004) 200 – 304.
3. Esen, Asim, 1992. Purification and Partial Characterization of Maize (*Zea mays* L.) beta-Glucosidase. *Plant physiology*. 174, 82, Vol. 98. pp.174-182.
4. Bradford, M.M. 1976. A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytic Biochemistry* 72, 248-254.

PHYSICOCHEMICAL PROPERTIES OF THE B DOMAIN (CATALYTIC LID) OF THE PYRUVATE KINASE FROM *THERMOPILUM PENDENS*

Alicia Vega-Segura, Guillermo Salcedo-Barrientos, Jessica Díaz Salazar*,
Gloria Hernández-Alcántara and Leticia Ramírez-Silva

Departamento de Bioquímica, Facultad de Medicina, UNAM. *Departamento de Físicoquímica,
Facultad de Química, UNAM. Ciudad de México 045510. malivega@bq.unam.mx

Introduction. In a previous study of the pyruvate kinase from *Thermophilum pendens* (*TpPK*) it was proposed that the B domain exhibits a low mobility of the hinge and a high stability of the domain. It was suggested that it was due to the interactions π - π of the conserved aromatic residues present in the B domain of the pyruvate kinases (PKs) from Crenarchaeota. The presence of the hydrophobic core formed by the phenylalanines could explain the independent domain denaturation, i.e. the presence of two calorimetric transitions in the DSC observed in the *TpPK*. To understand how does the enzyme work and what mechanism does it use to achieve its active conformation, the aromatic residues from B domain of *TpPK* were substituted for the residues present in the B domain of rabbit muscle PK (RMPK).

Methods. Four *TpPK* mutants were built, expressed and purified (F89I, F109V, F89I/F109V, F108I/F109V). Mutants were kinetically characterized. To explore integrity of the mutants CD spectra in the far and near UV were obtained. Thermal stability was performed by DSC.

Results. *TpPK* is a K^+ -independent enzyme, not activated by Li^+ , Na^+ , K^+ , NH_4^+ , Rb^+ or Cs^+ . In order to confirm mutants maintained this property. The activation by monovalent cations was explored and showed no activation. It was also studied if the mutants were activated by allosteric effectors (AMP, Fru 1,6 BP, Glu 6P, Rib 5P, 3PG). The mutants exhibited no effect, even at 3PG that is almost a general effector for most Thermoproteales. In the presence of Mg^{2+} , the V_{max} for F89I, F89I/F109V, F108I/F109V mutants were 67%, 52% and 60%, respectively of that of the wild type enzyme; whereas for F109V it was 230%. Km_{PEP} increased 9-fold for F109V with no significant changes for the other mutants or substrates. In the presence of Mn^{2+} , the V_{max} for F108I/F109V mutant was 48%; whereas for F109V mutant was 141%. The Km_{PEP} for F89I, F109V, F108I/F109V decreased 28, 19, 77, respectively. The $Km_{Mn^{2+}}$ for all the mutants increased from 6 to 7- fold. According to the CD data, far UV spectra were alike the wild type enzyme, however significant differences were observed in the near UV spectra of all but one mutant (F108I/F109V). This result might indicate differences in conformations. In concert with the DSC data at 90°/h, the two calorimetric transitions were observed for all mutants and similar to those of the wild type enzyme. However, it was observed that Tm_1 and Tm_2 decreased 1 and 2 ° for F109V mutant; and from 3.2 to 4.6° for the double mutants. Only F89I increased 2° for both Tms . In the presence of saturating concentrations of Mn^{2+} , a single transition was induced in the wild type enzyme with a significant increase in the Tm . This experiment was not able to be performed due to the low affinity of Mn^{2+} for the mutants. Higher concentrations than 1 mM Mn^{2+} induced precipitation. In order to explore if the denaturation was kinetically controlled, different scanning rates were performed 30, 60, 90 and 120°/hr. Nearly no changes in the Tms were observed.

Conclusions. Up to now, we found that some kinetic differences and thermal stability properties of the mutants were observed. The physicochemical properties of these 4 mutants did not allow us to conclude that the aromatic residues were involved in the two calorimetric transitions. We will continue studying the single and double mutants F108I and F89I/F109V and will try to express the triple mutant F89I/F108I/F109V that has not been able yet.

Aknowledgments. PPAPIIT-UNAM: IN201021 (LRS)

THE SUBSTRATE OF GLUCOSE-6-PHOSPHATE DEHYDROGENASE OF *PSEUDOMONAS AERUGINOSA* INDUCES A CHANGE IN ITS QUATERNARY STRUCTURE

Roberto Velasco-García*, Edaena Benitez-Rangel* and Carlos Mújica-Jiménez**

Facultad de Estudios Superiores Iztacala* y Facultad de Química** (UNAM).

Tlalnepantla, Estado de México, CP 54090. Teléfono 55 59 64 76 80.

E-mail: robertvela2001@yahoo.com.mx

Glucose-6-phosphate dehydrogenase from *P. aeruginosa* bacteria (PaG6PDH) participates in the pentose phosphate pathway, generating NADPH and NADH. This enzyme is a homotetramer that cooperatively binds the substrate glucose-6-phosphate (G6P)¹. Unlike other G6PDHs structurally stabilized by NADP⁺ binding, G6P stabilizes PaG6PDH². This binding causes a change in the enzyme, detected in fluorometry experiments in which 8-anilino-1-naphthalenesulfonic acid (ANS) was used as a hydrophobic fluorescent probe. The enzyme resulting from this modification shows increased tolerance to physical and chemical inactivating factors and an increased affinity for G6P².

In the search for inhibitors of PaG6PDH, we have found that pyridoxal-5'-phosphate (PLP), a cofactor present in many enzyme systems, inactivates it. Our experiments suggest that PLP phosphate binds to lysine residue 179, essential in binding glucose-6-phosphate phosphate (G6P) at the active site. In addition, PLP also forms a Schiff base with another lysine at this site (probably K145). In this study, using fluorometric techniques and the ANS dye, we demonstrate that PLP binding induces a change in the enzyme similar to that produced by G6P. However, unlike the latter, PLP fails to maintain enzyme stability upon temperature increase. Dynamic light scattering (DLS) and high-performance liquid chromatography (FPLC) analyses of PaG6PDH modified with PLP or G6P reveal that, while the former remains a tetramer, the latter has a molecular mass corresponding to an octamer. These results suggest that, although both PLP and G6P induce conformational changes in the protein, only the binding of G6P modifies its quaternary structure. The latter change appears essential to transform it into a structurally more stable form.

References

1. Acero-Navarro, K., Jiménez-Ramírez, M., Villalobos, M., Vargas-Martínez, R., Perales-Vela, H., Velasco-García, R. (2018). *Protein Expr Purif* 142: 53-61.
2. Benítez-Rangel E., Rodríguez-Hernández, A., Velasco-García R. (2020). *BBA-PaP* 1868 (2): 140331.
3. Acknowledgments: This research received support from PAPIIT (project IN221623).

STRUCTURAL KEYS BEHIND UNUSUAL PRODUCT DIVERSIFICATION OF A NON-CLASSICAL CGTase

Beatriz Velazquez-Cruz^{1,2}, Laura Espinosa-Barrera², Hugo Serrano-Posada^{3*},
Juan A. Osuna-Castro^{4*} and Sara Centeno-Leija^{3*}

¹Facultad de Ciencias Químicas, Universidad de Colima, Carretera Colima-Coquimatlán km 9, 28400, Coquimatlán, Colima, México; ²Laboratorio de Biología Sintética, Estructural y Molecular (LaBioSEM), Universidad de Colima, Carretera Los Limones-Loma de Juárez, 28627 Colima, Colima, México; ³CONAHCyT-LaBioSEM, Universidad de Colima, Carretera Los Limones-Loma de Juárez, 28627 Colima, Colima, México; ⁴Facultad de Ciencias Biológicas y Agropecuarias, Universidad de Colima, Autopista Colima-Manzanillo km 40, 28100 Tecoman, Colima, México.

*Corresponding authors: hserrano0@uacol.mx; josuna@uacol.mx; scenteno0@uacol.mx

CldA is an exceptional bifunctional thermoacidophilic three-domain cycloglucanotransferase/ amylase (CGTase/amylase) from Thermoanaerobacterales. The structural determinants by which CldA exhibits significant competition between the disproportionation reaction ($k_{cat}/K_m = 1.3$) and the cyclization reaction ($k_{cat}/K_m = 0.2$) remain unknown. Here, a crystallographic structure analysis suggests two key residues influence this non-classical CGTase profile: Ser²⁰⁰ (subsite -6) and Met²⁸¹ (subsite +2). This work presents the comparative functional characterization between native CldA and the mutants; CldAS200G, and CldAM281F, aiming to optimize the cyclization activity profile. Hence, while CldA_{S200G} exhibits an exclusive α -amylase profile under acidic conditions (pH 4.0-5.0) and decreases the cyclodextrins (CDs) production yield 4-fold at pH 7.0, CldA_{M281F} increases CDs production yield 3-fold at both acidic and neutral pH compared to native CldA. The results revealed that the hydrogen bond between Ser200 and Phe216 (central aromatic), is necessary to conserve CGTase profile in CldA. Also, strengthening the “hydrophobic clamp” Trp²⁰⁴/Phe²⁸¹ located at subsite +2 enhances the CGTase activity regardless of pH. Together, functional and structural studies of native CldA, combined with rational protein engineering, offered an opportunity to gain insights into the structural keys affecting the cyclization activity profile of thermoacidophilic CGTases. Future CldA mutants are promising high-added value CDs production in the starch industry.

BVC is supported by a CONAHCyT student fellowship No. 994206. LEB acknowledges the postdoctoral fellowship CONAHCyT No. 218862. HSP, JAOC, and SCL acknowledge financial support from CONAHCyT grants CB-2018-AI-S-18011, CF-2019-33-549477, and INFR-2021-17-316456.

ON THE ROLE OF PRO16 IN THE STABILITY, FOLDING KINETICS AND FUNCTION OF THE LAO BINDING PROTEIN FROM *SALMONELLA TYPHIMURIUM*

D. Alejandro Fernández Velasco¹, Isabel Velázquez-López¹, Renan Vergara-Gutiérrez¹, Sergio Romero-Romero², Rogelio Rodríguez-Sotres³, Alejandro Sosa Peinado¹

¹Laboratorio de Físicoquímica e Ingeniería de Proteínas. Departamento de Bioquímica. Facultad de Medicina. Universidad Nacional Autónoma de México, Ciudad de México 04510.

Tel: +525556232259 Email: fdaniel@unam.mx.

²Instituto de Fisiología Celular Universidad Nacional Autónoma de México. ³Departamento de Bioquímica. Facultad de Química. Universidad Nacional Autónoma de México.

LAO is a periplasmic protein (PBP) which binds with high affinity the basic amino acids lysine, arginine and histidine. PBPs are composed of two lobes joined by a hinge region. Upon ligand binding the two lobes change from an open conformation towards a closed one, resembling a Venus flytrap mechanism.

PBPs share a Proline residue in the hinge region (P16 in the LAO from *S. typhimurium*). Molecular dynamics simulations (Cortés & Domínguez *PLOS one* 2017) suggest that this residue modulates the trap mechanism. In this work, the structure, unfolding/refolding and ligand binding of the P16A mutant of LAO were studied. The mutant is properly folded and in the closed form displays a 3D structure very similar to that of LAOwt. No crystals could be obtained in the absence of ligands. P16A unfolding by urea or temperature is reversible. Both the thermodynamic stability at room temperature and the T_m are lower in P16A than in LAOwt. The urea-induced unfolding/refolding of P16A shows only one of the two refolding limbs observed in LAOwt, confirming that the second limb is related to proline isomerization. The curvature observed in the refolding branch indicates the presence of an intermediate folding pathway. Isothermal Titration Calorimetry experiments are in process to determine the effect of the P16A mutation in the thermodynamic signature for ligand binding.

EFFECT OF *PLEUROTUS DJAMOR* ON MITOCHONDRIAL FUNCTION AND EXPRESSION OF GRP-75 AND UNCOUPLING PROTEINS IN AN INSULIN-RESISTANT MODEL

Erica Karime Ventura García¹, Mónica Andrea Valdez Solana², Nagamani Balagurusamy¹, Guadalupe García Arenas³, Atahualpa Guzmán de Casa⁴, Erick Sierra Campos²

¹Facultad de Ciencias Biológicas, Universidad Autónoma de Coahuila. Torreón, Coahuila, México.

²Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango. Gómez Palacio, Dgo, México.

³Facultad de Ciencias de la Salud, Universidad Juárez del Estado de Durango. Gómez Palacio, México.

⁴Centro de Investigación y Estudios Avanzados del IPN. Irapuato, México

E-mail: ericksier@ujed.mx

Recently, the glucose-regulated protein (GRP75) has gained relevance for its involvement in the assembly and stabilization of mitochondrial supercomplexes; prevention of mitochondrial fragmentation and release of mtDNA [1]; as well as negatively influencing the activity of uncoupling proteins (UCPs), a key component in lipid metabolism, and negatively regulation of insulin secretion. Bioactive compounds from *Pleurotus* have shown antidiabetic effects. However, most of these studies focus on polysaccharides and terpenoids, leaving aside other components such as proteins and pigments predominant in species like *Pleurotus djamor* [2]. Therefore, the present study evaluated the effects of *P. djamor* on the expression of GRP-75 and several UCPs isoforms and its influence on the antioxidant and mitochondrial control system in a type 2 diabetes model. The pathology was induced with streptozotocin and nicotinamide, and treated with crude extract for 4 weeks. The area under the curve (AUC) values obtained from glucose tolerance curves showed a post-treatment decrease in the treated diabetic group (DT); unlike the control (C) and diabetic (D) groups, which remained stable. Similarly, the effect on the enzymatic activity of catalase (C= 39 $\mu\text{M}/\text{min}$; D= 15 $\mu\text{M}/\text{min}$; DT=35 $\mu\text{M}/\text{min}$) and paraoxonase (C= 289 $\mu\text{M}/\text{min}$; D= 96 $\mu\text{M}/\text{min}$; DT=223 $\mu\text{M}/\text{min}$) demonstrated modulation of antioxidant response in the DT group. The observed changes at the mitochondrial level showed significant changes in oxygen consumption by respiratory complexes. In the case of Complex I (C= 15 nAtoms/min; D= 23 nAtoms/min; DT= 13.5 nAtoms/min), the activity in the D group was increased, an effect that was reversed in DT. Unfortunately, the activity of Complex II (C= 27 nAtoms/min; D= 23.4 nAtoms/min; DT= 13.6 nAtoms/min) and Complex IV (C= 132.1 nAtoms/min; D= 68.26 nAtoms/min; DT= 28.79 nAtoms/min) decreased considerably in the DT group. In addition, Western Blot analyses revealed a distinct pattern of UCPs in D and DT groups, showing a 3.8 times higher signal in the D group compared to the DT group. On the other hand, GRP-75 was expressed in control extract (CE), D, and DT; with an increase observed in CE. From this, we concluded that *P. djamor* is capable of internalizing GRP-75 and inducing changes on UCPs; as well as having an effect on antioxidant capacity and mitochondrial complex activity.

References

1. Zhao Q, Luo T, Gao F, Fu Y, Li B, Shao X (2022). Diabetes, 71(2), 233–48.
2. Shamim MZ, Mishra AK, Kausar T, Mahanta S, Sarma B, Kumar V. (2023). Molecules, 28(6), 283.

EFL1 INTERDOMAIN COMMUNICATION IS DISRUPTED AS A CONSEQUENCE OF THE R1086Q MUTATION

Jonathan Alejandro Zúñiga-Domínguez¹, Abril Gijbsbers¹, Rohit Jain², Erik R Farquhar³,
Mark R. Chance², Nuria Sanchez-Puig¹

¹Instituto de Química, Universidad Nacional Autónoma de México, Circuito Exterior s/n, Ciudad Universitaria, CDMX 04510, México. ²Centre for Synchrotron Biosciences, Case Western Reserve University, School of Medicine, 10900 Euclid Avenue, Cleveland, OH, 44106, USA. ³ Brookhaven National Laboratory, NSLS II, 11973-5000 Upton, NY, USA
E-mail: nuriasp@unam.mx

Shwachman-Diamond syndrome (SDS) is a multisystem autosomal-recessive disorder, characterized by exocrine pancreatic dysfunction and neutropenia. About 90% is caused by mutations in the SBDS gene but it has also been found that mutations in the GTPase EFL1 (Elongation Factor type 1), the latter, is responsible, together with SBDS, for removing the anti-association factor eIF6 from the Sarcin/Ricin Loop site (SRL), eIF6 is part of the 60S ribosomal subunit maturation system, which is responsible for protecting and preventing premature binding of the 40S subunit. One of the mutations that has been described as causing SDS is the R1095Q mutation (R1086Q in yeast) in EFL1 (1), which is located within domain 4. An attempt has been made to elucidate how this mutation affects EFL1 and prevents effective removal of eIF6. The X-ray footprint technique has been proposed to clarify the conformational changes, which consists of the evaluation of the zones accessible to the solvent through the free -OH radicals generated by radiation.

The most relevant conclusion of this study, is that the conformational change suffered by the native Efl1 is totally different from that suffered by the mutant, although the mutation is found in domain IV, it also affects domain I, where it is seen that the areas that were previously exposed to the solvent now, they hide, this also affects, their thermodynamic parameters where it changes from being an exothermic reaction, demonstrated by Perez 2022 in ITC experiments.

References

1. Stepensky P, Chacón-Flores M, Kim KH, Abuzaitoun O, Bautista-Santos A, Simanovsky N, Siliqi D, Altamura D, Méndez-Godoy A, Gijbsbers A, Naser Eddin A, Dor T, Charrow J, Sánchez-Puig N, Elpeleg O. (2017) *J. Med. Genet.* **vol. 54,8**: 558-566
2. Gijbsbers A, Nishigaki T, Sánchez-Puig N. (2016) *JoVE*, **vol 116**.
3. Pérez-Juárez J, Tapia-Vieyra JV, Gutiérrez-Magdaleno G, Sánchez-Puig N. (2022) *Biomolecules* **vol. 12,8** 1141.

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex molecule, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

BIOTECHNOLOGY

BACTERIA ISOLATED FROM AN ASPHALTENE ROCK FROM THE GULF OF MEXICO ARE CAPABLE OF DEGRADING OIL AT LOW TEMPERATURES

Libertad Adaya García¹, Ziomara Ramos-Jiménez¹, Jorge Rojas-Vargas^{1,5}, Elizabeth Ernestina Godoy-Lozano², Armando Hernández-Mendoza³, Adolfo Gracia⁴, José Luis Rodríguez Mejía^{1,6}, Alejandro Sánchez-Flores⁵ and Liliana Pardo-López^{1*}

¹Department of Molecular Microbiology, Institute of Biotechnology, UNAM, Cuernavaca, Morelos, Mexico. ²Center for Research on Infectious Diseases, National Institute of Public Health, Cuernavaca, Morelos, Mexico. ³Center for Research in Cellular Dynamics, Autonomous University of the State of Morelos, Cuernavaca, Morelos, Mexico. ⁴Institute of Marine Sciences and Limnology, UNAM, Ciudad Universitaria, Mexico City, Mexico. ⁵University Unit of Mass Sequencing and Bioinformatics, Institute of Biotechnology, UNAM, Cuernavaca, Morelos, Mexico. ⁶University Center for Biomedical Research, University of Colima, Central Campus Colima. ^{*}Department of Biology, University of Western Ontario, London, Ontario, Canada. Av. Universidad 2001. Cuernavaca, Morelos 62210. Mexico. 777-3291600. liliana.pardo@ibt.unam.mx

The Gulf of Mexico is a basin characterized by natural hydrocarbon (HC) emissions, which makes it an ecosystem whose bacterial diversity is constantly exposed to HC¹. An asphalt rock (called A43) collected at a depth of 850 meters during the COBERPES 09 campaign in 2017, was used to isolate bacteria capable of degrading hydrocarbons under extremophilic conditions. The bacterial population was enriched by storing the sample at 4°C in seawater for 4 months and subsequently by sequencing the DNA using the Illumina platform, the bacterial diversity capable of growing on the surface of the asphalt rock and surrounding water was analyzed. We found a decrease in bacterial diversity after enrichment, and an increase in bacterial genera reported as hydrocarbon degraders was also observed, which allowed us to isolate four bacterial strains: *Idiomarina loihiensis* GOM17, *Pseudomonas xanthomarina* GOM18, *Rhodococcus jialingiae* GOM19, and *Rhodococcus jialingiae* GOM20. Capacity degradation of the strains was analyzed considering the origin and treatment of the sample, using growth kinetics with light and medium oil as the only carbon source. Samples were incubated at 4°C for 3 months. The results obtained by GC-MS showed that both strains of the *Rhodococcus* genus can degrade HC from C12 to C40, while the *Idiomarina loihiensis* GOM17 strain can consume aromatic HC. *Pseudomonas xanthomarina* GOM18 did not show significant degradation. These strains were sequenced, and their genomes were analyzed using the HADEG² database, which allowed us to identify enzymes related to the degradation of aliphatic and aromatic HCs that correlate with HC consumption results. Additionally, the growth temperature range of the strains was analyzed, and it was determined that they can grow in a range of 4°C to 42°C, suggesting their metabolic potential for HC degradation under different conditions, which is an important advantage for future biotechnological applications.

References

1. Godoy-Lozano, E. E., Escobar-Zepeda, A., Raggi, L., Merino, E., Gutierrez-Rios, R. M., Juarez, K., ... & Pardo-Lopez, L. (2018). Bacterial diversity and the geochemical landscape in the southwestern Gulf of Mexico. *Frontiers in microbiology*, 9, 2528.
2. Rojas-Vargas, J., Castelán-Sánchez, H.G., Pardo-López, L. (2023) HADEG: A curated hydrocarbon aerobic degradation enzymes and genes database. *Computational Biology and Chemistry*, 107, 107966.

CO-CULTURE OF *DALDINIA ESCHSCHOLZII* AND *HUMPHREYA COFFEATA*

Alma Rosa Agapito-Ocampo¹, Edgar Martínez-Fernández¹, Porfirio Juárez-López¹, Anabel Ortiz Caltempa³,
Irán Alia Tejacal¹, Ma. de Lourdes Acosta Urdapilleta², Peña-Chora Guadalupe², Téllez-Téllez Maura^{2*}

¹Facultad de Ciencias Agropecuarias-Universidad Autónoma del Estado de Morelos. Av. Universidad 1001, Cuernavaca, Morelos. CP. 62210. Tel. 777 1345402. ²Centro de Investigaciones Biológicas, Av. Universidad Autónoma del Estado de Morelos, Morelos, México. Universidad No. 1001, Col. Chamilpa, CP. 62209. Cuernavaca, Morelos, México. ³Centro de Investigación en Biotecnología, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México.*maura.tellez@uaem.mx

Fungi have biologically active compounds, such as polysaccharides, proteins, phenolic compounds, and antioxidants¹ which can be found in the fruiting bodies, mycelium, and culture medium. *Daldinia eschscholzii* is an endophytic fungus that secretes specialized metabolites and emits volatile organic compounds. *Humphreya coffeata* is recognized for its medicinal properties, in addition to producing polysaccharides and has been reported to have an antioxidant effect. This work aimed to evaluate the effect of *D. eschscholzii* inoculation time on the production of biomass, extracellular compounds, and antioxidant activity of the co-culture of *H. coffeata* and *D. eschscholzii*. Liquid culture of *H. coffeata* was carried out and subsequently inoculated with *D. eschscholzii* at three different times (96, 144 and 196 h) and grown for 336 h. The biomass was 5.08 g/L at 96 h, 7.48 g/L at 144 h and 3.78 g/L at 192 h. Regarding the precipitate, 0.05 g/L was obtained at 96 h, 0.06 g/L at 144 h and 0.08 g/L at 192 h. The content of total polyphenols was 0.083 mg/AGE, 0.079 mg/AGE and 0.082 mg/AGE at 96, 144, and 192 h, respectively, and the antioxidant activity was 71.28%, 56.8% and 57.41% inhibition of the ABTS radical, at 96, 144, 192 h, respectively. This indicates that it is advisable to establish co-culture at 96 h since it allows more interaction time between the organisms and a differential response in the presence of secondary metabolites. The co-culture of fungi with biologically active compounds can enhance their biotechnological applications.

References

1. Gao, Y., Tang, W., Dai, X., Gao, H., Chen, G., Ye, J., Chan, E., Koh, H., L. Li, X., y Zhou, S. (2005). J Med Food, 8,159–68.

OPTIMIZATION OF PARAMYLON PRODUCTION IN *EUGLENA GRACILIS* USING DIFFERENT LIGHT SPECTRA

Zhaida I. Aguilar Gonzalez, Héctor V. Miranda-Astudillo

Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas,
Universidad Autónoma de México.
hmiranda@iibiomedicas.unam.mx

The carbohydrate β -1,3-glucan, also called paramylon, is the reserve carbohydrate of euglenoids. It is a water-insoluble polymer of great importance in pharmaceutical and biotechnological industries. A wide applications spectrum has been described so far, from anti-inflammatory and immunostimulatory action to wound healing [1] to diet supplement [2]. *Euglena gracilis* has a great metabolic capacity to grow under autotrophic, heterotrophic, or photoheterotrophic conditions. It has been described that its growth can be carried out with various organic substrates, such as acetate, ethanol, lactate, or glucose, in the presence of light or darkness. In photoheterotrophic conditions, it has been cultivated with red and white light, showing that the cells adapted in far red generated a greater amount of paramylon [3]. Paramylon production in this species can represent up to 80% of its dry weight [4].

In this work we sought to optimize the production of *E. gracilis* paramylon by adapting its photosynthetic machinery, using different wavelengths: near ultraviolet (UV), royal blue (RB), blue (B), green (G), yellow (Y), red (R), far red (FR) and infrared (IR) and 4 full spectra: 3 000 K, 10 000 K, 30 000 K, and full spectrum (Full). *E. gracilis* was cultured in Tris-minimal phosphate (TMP) medium supplemented with vitamins. It was observed that the cells adapted to full spectrum achieved a higher production of paramylon, and that *E. gracilis* suffered a modification in its photosynthetic machinery with respect of wavelengths used during the cultivation.

Acknowledgments. This research was supported by grant IA204524 (PAPIIT, DGAPA, UNAM) and the Institutional Program (IIBO-UNAM) "The production of biomolecules of biomedical interest in microorganisms".

References

1. Nakashima A. et. al., Biochem Biophys Res Commun 2017
2. Kawano T, et. al., Nutrients. (2020)
3. Rodríguez-Bolaños, M. et. al., Appl. Biochem. Biotechnol. (2023)
4. Gissibl, A. et. al., Front Bioeng Biotechnol. (2019)

EXPRESSION OF CHAGAS IN CHIMERAS WITH 4 EPITOPES FROM TRYPANOSOMA CRUZI ANTIGEN, TSA-1, IN PICHIA PASTORIS

Paola Eugenia Agustin Vélez¹, Marco Ibrim Rodríguez Sánchez¹,
Claudia Ivonne Flores Pucheta¹, Octavio Montes-Flores¹, Rossana Arroyo², Jaime Ortega López¹

¹Departamento de Biotecnología y Bioingeniería y ²Departamento de Infectómica y Patogénesis Molecular. Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN), Ciudad de México, México. C.P 07360
E-mail: paola.agustin@cinvestav.mx

Chagas disease, also known as American trypanosomiasis, is a neglected disease affecting marginated regions in Latin America.¹ This disease is caused by the intracellular parasite *Trypanosoma cruzi*, transmitted mainly by the bite of infected insects belonging to the Triatomine family. Trypomastigote Surface Antigen (TSA-1) is a protein of *T. cruzi* involved in the infective process that has been demonstrated as a promising antigen for developing a therapeutic vaccine against Chagas disease. Previous studies have shown that TSA-1 has five conserved epitopes among the *T. cruzi* DTUs that can induce an immunological response of cytotoxic T-lymphocytes.² However, the recombinant production of TSA-1 in *E. coli* is as inclusion bodies, requiring an extra refolding step.³ Also, the use of a soluble molecular scaffold containing two epitopes from TSA-1 has been proven in previous studies.⁴ This work aims to express chimeric proteins containing up to four TSA-1 epitopes in the loops L2, L4, and L6 of the endogenous cysteine-protease inhibitor chagasin of *T. cruzi*.

Using bioinformatic tools, chimeric proteins with up to four TSA-1 epitopes were designed, and their solubility was predicted *in silico*. The two most promising chimeras were selected for recombinant expression in the yeast *Pichia pastoris*. The expression of the recombinant protein of nine clones in flask cultures was analyzed using a Dot-Blot assay, and one clone was chosen for scale-up to the 2L bioreactor.

References

1. Haberland, S., Munoz, G., Wallukat, et al. (2013) Chronic Chagas disease: from basic to laboratory medicine. Clin Chem Lab Med, 51, pp. 271-294
2. Wrightsman RA, Dawson BD, Fouts DL, Manning JE (1994). Identification of immunodominant epitopes in Trypanosoma cruzi trypomastigote surface antigen-1 protein that mask protective epitopes. J Immunol. 1;153(7):3148-54. PMID: 7522250
3. Dela Cruz, J.J., Villanueva-Lizama, L., Dzul-Huchim, V., Ortega-Lopez, J., Flores-Pucheta, C. I., Gillespie, P., Zhan, B., Bottazzi, M. E., Hotez, P. J., and Dumonteil, E. (2019) Production of recombinant TSA-1 and evaluation of its potential for the immunotherapeutic control of Trypanosoma cruzi infection in mice, Hum Vaccin Immunother 15, 210-219
4. Cárdenas-Guerra RE, Montes-Flores O, Nava-Pintor EE, Reséndiz-Cardiel G, Flores-Pucheta CI, Rodríguez-Gavaldón YI, Arroyo R, Bottazzi ME, Hotez PJ, Ortega-López J (2024) Chagasin from Trypanosoma cruzi as a molecular scaffold to express epitopes of TSA-1 as soluble recombinant chimeras. Protein Expression and Purification. PMID: 38423156

HETEROLOGOUS EXPRESSION OF SCORPION TOXIN FROM CHIHUAHUA

Carolina Alvarado-González^{1,2}, Herlinda Clement³, Lourdes Ballinas-Casarrubias², Rosario Peralta-Pérez², Gerardo Corzo³, Gerardo Pável Espino Solís^{1,4}

¹Laboratorio de Investigación Traslacional, Facultad de Medicina y Ciencias Biomédicas, Universidad Autónoma de Chihuahua, Circuito Universitario s/n, Campus II, Chihuahua 31125, México. ²Facultad de Ciencias Químicas, Universidad Autónoma de Chihuahua, Circuito Universitario s/n, Campus II, Chihuahua 31125, México. ³Instituto de Biotecnología-UNAM, Universidad Nacional Autónoma de México, Av. Universidad 2001, Col. Chamilpa, Cuernavaca 62210, México. ⁴Laboratorio Nacional de Citometría de Flujo, Facultad de Medicina y Ciencias Biomédicas, Universidad Autónoma de Chihuahua, Circuito Universitario s/n, Campus II, Chihuahua 31125, México. C.P. 31125, Tel: 6141843467
E-mail: p274265@uach.mx, gepsinos@uach.mx

The heterologous expression of proteins is a fundamental process in biotechnology, involving the production of proteins in host organisms that are different from their natural origin. This technique has revolutionized various fields, from medicine to industry. It also holds critical importance in enhancing our quality of life, as it significantly contributes to the development of more effective medical treatments. In the case of recombinant proteins derived from venomous animals, it provides the opportunity to produce much larger quantities than can be obtained from the animals themselves. A toxin from the venom of the scorpion *Chihuahuanus coahuilae* was expressed based on the partial N-terminal sequence of this species toxin, oligonucleotides were designed for PCR amplification and cloned into the pQE-30 expression vector. Expression was achieved using the Origami strain of *E. coli*, with the toxin predominantly found in inclusion bodies. It was recovered by Nickel affinity chromatography, reduced with 1,4-dithiothreitol (DTT), and purified by reverse-phase high-performance liquid chromatography (RP-HPLC). Folding was performed using the cystine-cysteine redox pair, followed by another round of purification via RP-HPLC. The fractions collected from this process were analyzed by mass spectrometry, and their biological activity was subsequently tested in mice and crickets.

THE NATIONAL LABORATORY FOR ANALYSIS OF BIOTECHNOLOGICAL MOLECULES AND DRUGS (LAMMB) CHALLENGES AND PERSPECTIVES

Elianeth Amaro, Irlanda Camacho, Maricela Olvera, Laura Palomares, Tonatiuh Ramírez and Mabel Rodríguez*

National Laboratory for Production and Analysis of Biotechnological Molecules and Drugs (LAMMB).

National Autonomous University of México.

E-mail: analitico@lammb.unam.mx

E-mail: operaciones@lammb.unam.mx*

The increasing international demand for biotechnological products has led to a rise in their regulation by health authorities. As a result, laboratories with highly specialized scientific, technical, and enabling infrastructure capabilities are needed to provide services for the characterization of recombinant products. The Laboratory for Analysis of Biotechnological Molecules and Drugs (LAMMB) is a laboratory established in 2015 at the Institute of Biotechnology of UNAM, which has analytical capabilities to characterize pharmaceutical proteins. In December 2022, LAMMB obtained the Health License from COFEPRIS as an Auxiliary Testing Laboratory in Health Regulation for the biological, chemical, and physicochemical characterization of biotechnological drugs, implying a strong commitment to Mexico. Additionally, since 2021, LAMMB has been part of the CEPI (Coalition for Epidemic Preparedness and Innovations) Laboratory Network to support the evaluation of vaccines against Sars-Cov-2 and other pandemic diseases. One of the distinctive features of LAMMB is its self-sustainability, which constitutes a major challenge due to the current economic situation facing Mexico and the world.

It is of vital importance that both entrepreneurs and the governmental sector, along with the scientific community, join forces so that laboratories like LAMMB can expand in the country. This would help lower the barrier of entry for biotechnological medicines in Mexico, which are so necessary to combat chronic and high-impact health conditions such as diabetes, cancer, and autoimmune diseases. Otherwise, Mexico would be left behind in pharmaceutical biotechnology, and we would be at the mercy of imported medications and international demand, a situation that was starkly evident during the recent pandemic.

References

1. <https://www.webmd.com/breast-cancer/guide/her2-positive-breast-cancer-survival-rates>
2. <https://www.gob.mx/cofepris/documentos/listado-de-medicamentos-bioteconomicos-innovadores>
3. https://www.gob.mx/cms/uploads/attachment/file/468862/Listado_de_Medicamentos_Bioteconomicos_Biocomparables_Versión_6_11-06-2019.pdf
4. <https://www.mirusbio.com/blog/fda-approved-gene-cell-therapies>

POLYURONIDE SYNTHASES IN FILAMENTOUS FUNGI

Mariandrea Aranda-Barba¹, Marina Franco-Herrera¹, Paul Montaña-Silva¹,
Rosa María Camacho Ruiz¹, Eddy Sánchez-León² & Jorge Verdín^{1*}

¹Biología Industrial, Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, Camino Arenero 1227, El Bajío, Zapopan, Jalisco, 45019, Mexico. ²Michael Smith Laboratories, Department of Microbiology and Immunology, University of British Columbia, Vancouver, British Columbia, Canada.

*Tel: 0133455200 ext 2103; jverdin@ciatej.edu.mx

Hyaluronic acid (HA) is a heteropolysaccharide composed of repeated dimers of N-acetyl D-glucosamine and D-glucuronic acid. HA is an important molecule that has been found in vertebrates, bacteria, viruses, and the pathogenic yeast *Cryptococcus neoformans*, the only reported fungus that synthesizes HA so far. In those organisms, the enzyme that synthesizes HA is the Hyaluronic Acid Synthase (HAS). Therefore, by Hidden Markov Models (HMM), conservation of catalytical motifs and phylogenetic reconstruction, we found protein sequences in some filamentous fungi that could potentially encode to a HAS. These proteins share high sequence identity with the HAS of *C. neoformans* (Cps1p). Complementary to that, we found in these fungi the enzymatic machinery needed for the synthesis of HA precursors, as well using HMM, phylogenetic reconstruction and, additionally, the function prediction was performed with CLEAN.

One of these fungi was *Mucor circinelloides*, whose putative HAS showed 66% identity to Cps1p. We hypothesize that this putative HAS could be involved in the pathogenicity of *M. circinelloides*, as has been demonstrated for *C. neoformans* HAS. To characterize it, it was heterologously expressed in *Saccharomyces cerevisiae*, an ideal model because it does not synthesize hyaluronic acid. The expression was confirmed by Western blot and immunofluorescence. Afterwards, the activity was determined by an *in vitro* assay coupled with HPLC/Fluorescence detector. Under the reaction conditions used, we detected an enzymatic activity associated with the plasma membrane that was dependent on glucuronic acid. We still do not know the molecular identity of the product synthesized, but it could be any polyuronide. We need to further characterize it, as well the evaluation of different reaction conditions and performing complementation assays in a *C. neoformans* deletion mutant for Cps1p. This work is being supported by CONACYT-Ciencia de Frontera 2019, grant 552259.

FUNCTIONAL AND STRUCTURAL ANALYSIS OF BASIDIOMYCETE FUNGAL LIGNOCELLULOLYTIC ENZYMES FOR BIOTECHNOLOGICAL APPLICATIONS

Silvia Armenta¹, Sofía Tamara Gutiérrez Trejo¹, Oscar Arce-Cervantes¹

¹Instituto de Ciencias Agropecuarias, Universidad Km. 1 s/n Ex. Aquetzalpa, 43600 Tulancingo, Hidalgo.

E-mail: silvia_armenta@uaeh.edu.mx

Lignocellulose is the most abundant renewable raw material and has a complex composition, mainly containing lignin, hemicellulose, and cellulose. Basidiomycetes specialize in degrading lignocellulose by producing various enzymes such as laccases, cellulases, and xylanases.¹ Despite their potential for biotechnological applications, there are limited reports describing the enzymatic complexes they produce. This work aims to understand the diversity and structural characteristics of the lignocellulosic enzymes produced by Basidiomycetes, especially, those involved in lignin degradation, cellulose hydrolysis, and hemicellulose breakdown. The findings from this study suggest valuable insights into the functional and structural properties of Basidiomycete fungal lignocellulolytic enzymes and their potential applications in biotechnology.² Due to there is insufficient information about the lignocellulolytic enzyme complex from basidiomycetes such as *Hericium erinaceus* and *Pleurotus agave*. Despite indications in the literature that basidiomycete fungi produce a significant variety of enzymes capable of breaking down lignocellulose, evidence only identified five potential enzymes with cellulase and xylanase activity in *H. erinaceus*. Besides, no genetic sequences have been documented for *P. agave*, and only one exists for *H. erinaceus*. Hence, further investigation into this species is crucial. As for other basidiomycetes like *Pleurotus ostreatus* and *Trametes versicolor*, they seem to exhibit an enzyme profile featuring a higher concentration of laccases with distinct structural features that could serve as platforms for future evolutionary studies.³ Therefore, it is essential to continue research in this area to tap into the full potential of these fungi and their lignocellulosic enzymes for biotechnological purposes.

References

1. Téllez, A. Arana, A, et. al.. (2016) UPP Capitulo 5, 64-83J
2. Zhong, R. (2019, March 1). Secondary cell wall biosynthesis.
3. Kumar, A y Chandra, R. (2019). Science Direct.

INTRACELLULAR OXIDATIVE STRESS AT LOW TEMPERATURES IN THE PHYTOPATHOGEN BACTERIUM *PSEUDOMONAS SAVASTANOI PV. PHASEOLICOLA* NPS3121

Jackeline Lizzeta Arvizu-Gómez^{1*}, Alejandro Hernández-Morales², Kevin Daniel Llanos-Vargas³, Gabriela Olmedo-Álvarez⁴, Juan Campos-Guillén⁵, Alba Adriana Vallejo-Cardona⁶, José Luis Hernández-Flores⁴, Christian González-Reyes⁷ and Angélica Nallehly Rodríguez-Ocampo⁷

¹Secretaría de Investigación y Posgrado. CENITT.Universidad Autónoma de Nayarit.²Facultad de Estudios Profesionales Zona Huasteca. Universidad Autónoma de San Luis Potosí. ³Maestría en Ciencias Biológico Agropecuarias. Universidad Autónoma de Nayarit. ⁴Centro de Investigación y de Estudios Avanzados del IPN. Irapuato, Guanajuato.⁵Facultad de Química. Universidad Autónoma de Querétaro. ⁶Unidad de Biotecnología Médica y Farmacéutica, Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A.C. (CIATEJ). ⁷Unidad Académica de Ciencias Químico Biológico y Farmacéuticas. Universidad Autónoma de Nayarit.
E-mail: lizzeta28@gmail.com; jackeline.arvizu@uan.edu.mx

Temperature, particularly low temperatures conditions (16-20 °C) is one of the most important environmental factors influencing the occurrence and development of various plants diseases caused by phytopathogenic bacteria. The production and expression of diverse pathogenic and/or virulent determinants by these microorganisms are mostly induced under this condition¹. So far, the know about molecular bases or signal transduction pathway related with low temperatures in phytopathogenic bacteria is still scarce. Previous transcriptome works in the *P. savastanoi pv. phaseolicola* NPS3121 model bacterium, suggested the intracellular oxidative stress as a molecular event related with the low temperatures exposition², without this having yet been demonstrated. In this study, generation of an oxidative-stress biosensor (pKLI) and fluorometry analyses with the *P. savastanoi pv. phaseolicola*-pKLI bioreporter strain were performed. Changes in the intracellular redox potential of *P. savastanoi pv. phaseolicola* during its growth at 28 °C and/or 18 °C were observed, which showed greater oxidation degree in cells grown at 18 °C. These results demonstrated that intracellular oxidative stress is part of the signaling pathway related with low temperatures in *P. savastanoi pv. phaseolicola* NPS3121.

References

1. Smirnova A, Li H, Weingart H, Aufhammer S, Burse A, Finis K, Schenk A, Ullrich MS (2001). Arch Microbiol, 176(6):393-9.
2. Arvizu-Gómez JL, Hernández-Morales A, Pacheco-Aguilar JR, Álvarez-Morales A (2013) BMC Microbiology 13, 81.

CLE14 PEPTIDE INHIBITS REGENERATION AND CALLOGENESIS IN ARABIDOPSIS

Adrián Ávalos-Rangel¹, Dolores Gutiérrez-Alanis²,
Luis Herrera-Estrella², Javier Raya-González³, José López-Bucio^{1*}

¹Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo. Edificio B3, Ciudad Universitaria; 58030, Morelia, Michoacán, México. ²Institute of Genomics for Crop Abiotic Stress Tolerance (IGCAST), Department of Plant and Soil Science, Texas Tech University, 1006 Canton Ave #3135, Lubbock, TX 79409. ³Facultad de Químico Farmacobiología, Universidad Michoacana de San Nicolás de Hidalgo, Avenida Tzintzunzan 173, Col. Matamoros, 58240, Morelia, Michoacán, México.
adrian.avalos.rangel@umich.mx; jbcucio@umich.mx*

CLE14 belongs into a family of plant secreted peptides that interact with leucine-rich repeat receptor-like kinase (LRR-RLK) receptors to orchestrate plant morphogenesis. Previous studies indicated its important function in cell division, phosphate homeostasis and senescence, but specific involvement in cell fate determination and organogenesis remains largely unexplored. Here, through pharmacological, genetic and cell biology approaches, we show the critical roles for *CLE14* in determining the balance between cell division and differentiation in root tip regeneration and callogenesis. *CLE14* application or its overexpression in *Arabidopsis* repressed primary root growth and triggered root branching and root hair formation. After resection of the primary root tip, *CLE14* expression was located specifically at the cell layer adjacent to the cutting and at the outermost external cell layer of the root cap as the newly root cap formed. *cle14* mutants had comparable root tip regeneration when compared to WT seedlings, whereas *35S:CLE14* seedlings failed to regenerate the missing root tip after resection. The de-differentiation of tissue into proliferative growth was analyzed in WT, *cle14*, and *35S:CLE14* stem explants grown in callus-inducing media. The results showed comparable callus-biomass production for WT and *cle14*, but a dramatically reduced callogenesis for *35S:CLE14* explants. Our data show that *CLE14* acts as a “brake” for root tip regeneration as well as callus formation.

FUNCTIONAL DRINK FROM WINE INDUSTRY WASTE

José Ramón Barraza Amaya^{1,4}, Nathiely Ramírez Guzmán^{1,4}, Josefina Rodríguez González²,
Erika Flores Loyola¹ and Cristian Torres León^{3,4}

¹Faculty of Biological Sciences, Universidad Autónoma de Coahuila, Carretera Torreón – Matamoros km 7.5. Ejido El Águila. Ciudad Universitaria. C.P. 27087. Torreón, Coahuila. ²Research Center for Interdisciplinary Studies Universidad Autónoma de Coahuila. ³Research Center and Ethnobiological Garden (CIJE), Universidad Autónoma de Coahuila, ⁴Agri-food and Agro-industrial Bioeconomy Research Group.
nathiely.ramirez@uadec.edu.mx

The Kombucha drink is a type of alcoholic ferment that is generated from a symbiotic culture of bacteria and yeast called SCOBY you need both to achieve fermentation since they depend on the process of the others, this drink uses *Camellia sinensis*, better known as green tea, being its main characteristic the large number of antioxidants it has [1]. The waste from the wine industry is considered a potential source for obtaining compounds with biological interest and representing an environmental problem.[2] The objective of this project is to carry out a probiotic drink with antioxidant capacity from waste from the wine industry. For the Methodology: The processing of the waste is carried out by drying, grinding, and sieving for the preparation of the probiotic drink using SCOBY and green tea and grape pomace as substrates from the wine industry of Parras Coahuila, of which kinetics were made and evaluated the presence of probiotics and antioxidant capacity of each of the drinks. The results were two drinks with the presence of probiotics as well as antioxidant capacity giving better results in the residue from grape. We concluded that the waste from the wine industry can be used as a substrate for preparing functional drinks with probiotic capacity and a high amount of antioxidant activity.

References

1. Mousavi Khaneghah, A.; Abhari, K.; Eş, I.; Soares, M.B.; Oliveira, R.B.A.; Hosseini, H.; Rezaei, M.; Balthazar, C.F.; Silva, R.; Cruz, A.G.; et. al.. Interactions between Probiotics and Pathogenic Microorganisms in Hosts and Foods: A Review. *Trends Food Sci. Technol.* 2020, 95, 205–218,
2. Torres-León, C.; Ramírez-Guzman, N.; Londoño-Hernandez, L.; Martínez-Medina, G.A.; Díaz-Herrera, R.; Navarro-Macias, V.; Alvarez-Pérez, O.B.; Picazo, B.; Villarreal-Vázquez, M.; Ascacio-Valdes, J.; et. al.. Food Waste and Byproducts: An Opportunity to Minimize Malnutrition and Hunger in Developing Countries. *Front. Sustain. Food Syst.* 2022, 2, 1–17,

IMPROVEMENT OF PIGMENT PRODUCTION BY FILAMENTOUS FUNGI.

Iliana Geraldine Barreto-Flores¹, Divanery Rodríguez-Gómez¹, Juan Vázquez-Martínez¹, Varinia López-Ramírez¹

¹Tecnológico Nacional de México/ITS de Irapuato. Carr. Irapuato – Silao km 12.5 Col. El copal. C.P.36821.
varinia.lr@irapuato.tecnm.mx

Filamentous fungi are eukaryotic organisms capable of surviving in extreme environmental conditions due to the development of mechanisms that allow them to adapt. A biochemical response to ecological and nutritional stress is the synthesis of secondary metabolites such as dyes or colorful pigments. Several studies show that changes in pH, temperature, salinity, or carbon and nitrogen sources, affects the type of metabolite and its quantity. This study centers on improving growth conditions of fungi to obtain an increase in the production of pigments. 12 strains of Fungi isolated from Sierra de Lobos in the state of Guanajuato and from the Pedregal reserve in Mexico City with the ability to produce colorful exudates were evaluate, they were inoculated in Petri dishes containing potato dextrose agar with pH variation (5.5 and 6.5) and incubated at 28 °C for 10 days. A qualitative and quantitative evaluation of the produce dye was carried out by, divided the Petri dish into 4 quadrants to compare the intensity of the color in the medium and on the mycelia, as well as the quantitation of the volume of exudate droplets. It was found that for eight of the isolates an increase in dye production was obtained at pH 6.5 and for the remaining four isolates it was at pH 5.5. Related to the type of release of the dye, 5 of the isolates spread the pigment within the medium, while 2 isolates spread the dye within the medium and produce droplets on top of the mycelium, 3 isolates generated exudate droplets on the mycelium and the mycelium took the color and in the other 2 isolates the mycelium was completely colored and the agar as well. The results show the influence of modifying one of the growth parameters on the production of colorful metabolites. This study presents evidence of the improvement of the synthesis of fungal dyes and raises the question about other abiotic variables that could increase their production, such as temperature, and carbon source, among others. The search of fungal growth conditions to increase the synthesis of dyes could be an alternative as colorful compounds with multiple applications and lower environmental impact.

OPTIMIZATION OF THE FERMENTATION PROCESS TO PRODUCE WHISKEY

John Carlos Botia Becerra¹, Héctor Hernández Escoto², Gloria Angélica González Hernández¹, Israel Enrique Padilla Guerrero¹, Juan Carlos Torres Guzmán¹

¹Departamento de Biología, ²Departamento de Ingeniería Química División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Col. Noria Alta, C.P. 36050, Guanajuato, Gto, México. Tel: 4737320006 ext. 8160
E-mail: torguz@ugto.mx

Whisky is a beverage produced by distillation of fermented cereals and matured in wooden casks¹; it is generally classified as Scotch, American, Canadian, Irish, and Japanese, which vary not only by their region of origin but also by the raw material used. Scotch whisky is made only with malted barley; the other whiskies use a mixture of grains, including corn, rye, and barley². The fermentation process for distilled beverages begins with malting and mashing, where most of the fermentable sugars are released from the cereals used, generating a mixture called wort, which is inoculated mainly with *Saccharomyces cerevisiae* yeast to convert sugars into ethanol and carbon dioxide. It is then distilled up to twice to remove the alcohol from the water and to separate the unwanted fractions, such as methanol or ethyl esters, from the desired ones, such as potable liquor or “new spirit.” Finally, the process ends with maturation in wooden casks that confers unique aromas and enhances the organoleptic characteristics of the whiskey.

This work aims to improve the ethanol yield obtained in the whiskey fermentation process by evaluating various strains of *S. cerevisiae* from tequila agave juices and whiskey must. Fermentations were carried out at an initial concentration of 120 g/L of direct reducing sugars, analyzing glucose, fructose, and mannose consumption, cell growth, and ethanol production over 24 hours. So far, the fermentation efficiencies of 3 different strains have been analysed, showing ethanol production that varies between 28 – 30 g/L. The results are compared with the data obtained from industrial-scale fermentation at the facilities of the Corralejo Tequila Company in a volume of 10,500 L.

References

1. J.R. Piggott, 15, Current Developments in Biotechnology and Bioengineering, Elsevier, 2017.
2. Wanikawa, A. (2020). Flavors in Malt Whisky: A Review. JASBC.

ENHANCING DENITRIFICATION WITH ELECTRIC FIELDS IN *PARACOCCLUS DENITRIFICANS*

Arturo Cadena-Ramirez¹*, Julio Cesar Ríos Guzman¹

¹Universidad Politécnica de Pachuca, Carr. Cdad. Sahagún-Pachuca Km. 20, Ex-Hacienda de Santa Bárbara,
43830 Zempoala, Hgo. Tel. 771712125

*E-mail: arturocadena@upp.edu.mx

The biological denitrification process carried out by *Paracoccus denitrificans* is essential for the removal of nitrates in wastewater treatment¹. The application of electric fields in microbial cultures can influence various metabolic activities and physiological states of microorganisms². These fields can alter membrane permeability, enzyme activities, and overall metabolic pathways, thus presenting a potential method for enhancing microbial processes³. This study investigates the effects of applying electric fields on the anaerobic denitrifying respiration of *P. denitrificans*. Using a rotating cylinder reactor, different C/N ratios and electric field intensities were tested to determine their impact on key parameters such as nitrate consumption, nitrite accumulation, pH, ORP, and biomass growth. Higher electric fields resulted in lower nitrite accumulation, suggesting a positive modification in the electron transport and biochemical reduction processes within the bacteria. For instance, at 200 mV, nitrite levels remained minimal throughout the process, while at 100 and 150 mV, nitrite accumulation occurred at different times. The pH and ORP values were closely monitored, showing significant changes correlating with the reduction stages of nitrate and nitrite. The ORP values reached more negative levels at higher electric fields, indicating enhanced reductive conditions conducive to denitrification. The specific nitrate consumption rates were observed to increase with the application of electric fields, with a notable rate of 75.47 mg N-NO₃⁻/L h at 200 mV, compared to 26.41 N-NO₃⁻/L h without an electric field. Additionally, nitrogen gas production was significantly enhanced, achieving a maximum rate of 14.25 mg N-N₂/L h at 200 mV, compared to 6.02 N-N₂/L h without an electric field. These findings demonstrate that electric fields can accelerate the metabolic processes involved in denitrification, leading to more efficient nitrate reduction and nitrogen gas production. The electric fields appeared to alter the cell membrane permeability, facilitating more efficient electron transfer and thereby improving denitrification rates. The study provides insights into the potential for optimizing wastewater treatment processes by controlling electric fields to enhance the biological activity of denitrifying bacteria. The research contributes to a better understanding of the mechanisms underlying electric field-enhanced denitrification and provides a foundation for future studies aiming to refine and implement this approach in real-world scenarios.

References

1. Hu, C., Wu, J., Wang, L., & Hong, Y. (2022). *Microbiology Resource Announcements*, 11(4), e01236-21.
2. Cadena R. A., Texier, A. C., González, I., & Gómez, J. (2018). *Environmental technology*, 41(21), 2747-2755
3. Alshawabkeh, A. N., & Maillacheruvu, K. (2002). *Physicochemical groundwater remediation*, Springer, Boston, 73-90.

GREEN TECHNOLOGIES FOR THE TREATMENT OF PESTICIDES IN WATER BASINS

David A. Camarena-Pozos, Oriana Landa-Cansigno, Javier A. Arcibar-Orozco, José O. Saucedo-Lucero, Norma C. Calderón-Rosas, Norma L. Cruz-Ortiz, Catalina De la Rosa-Juárez, Miroslava A. Silva-Goujon

Centro de Innovación Aplicada en Tecnologías Competitivas (CIATEC)
Omega 201, Fraccionamiento Industrial Delta. CP 37545. León, Guanajuato, México
dcamarena@ciatec.mx

In Guanajuato, the water quality of the Turbio River is widely recognized by the National Water Commission (CONAGUA) as polluted, due to a considerable contribution of industrial wastewater from the leather-footwear sector in Leon and the surrounding area. Agricultural activity in the basin is affected by the use of river water for irrigation, and on the other hand, it contributes to being a diffuse source of pesticide contamination, resulting in its dispersion, affecting biodiversity and human health.

The objective of this project was to develop a water treatment technology through constructed wetlands using *Typha domingensis* plants and tezontle as substrate, adding bioadsorption process. Additional activities included the characterization of pesticides in the water of the Turbio River (n=15) and the dissemination of the results to the agricultural communities.

Organochlorine pesticides such as aldrin, chlordane, p,p-DDT, dieldrin, endrin, heptachlor, hexachlorobenzene, and methoxychlor were found in water samples from Rio Turbio, which exceed the criteria recommended by the “Ley Federal de Derechos”. The presence of organophosphate pesticides such as chlorpyrifos, ethoprophos, fenclorophos, guthion and methyl parathion were also found. Based on this diagnosis, a water treatment system was designed and constructed consisting of a linear configuration composed of 8 constructed wetland modules with the addition of a biocarbon adsorption system. The system has the potential to reduce a wide range of pollutants, including organochlorine pesticides, organophosphates and herbicides, and to reduce water quality parameters such as chemical oxygen demand (COD) and total suspended solids (TSS) by up to 90%.

With all this information, workshops were developed to disseminate information, raise awareness of pesticides and facilitate the adoption of constructed wetland water treatment technology. This type of workshop had a very positive response in the transformation of knowledge of the farmers. We believe that it is necessary to continue promoting the use of constructed wetlands in order to make more users aware of this solution and to continue sensitizing more farmers on the benefits of its implementation.

PRODUCTION AND OPTIMIZATION OF INDUCED *SERRATIA MARCESCENS* CARBOXYLESTERASES FOR THE DEGRADATION OF POLYOLEFINS

Amador Campos-Valdez¹, María Sinisterra-Sierra¹, Iliana Barrera-Martínez²,
María Marcela Robles Machuca³, Leticia Casas-Godoy^{2*}

¹Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, Unidad de Biotecnología Industrial, Camino Arenero 1227, El Bajío del Arenal, 45019 Zapopan, Jalisco, México. ²CONAHCYT-Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, Unidad de Biotecnología Industrial, Camino Arenero 1227, El Bajío del Arenal, 45019 Zapopan, Jalisco, México. ³Universidad Autónoma de Nayarit, Secretaría de Investigación y Posgrado. Tepic, Nayarit, México.
E-mail: lcasas@ciatej.mx

Abstract. The accumulation of plastic waste, specifically Low-Density Polyethylene (LDPE) and Polypropylene (PP) poses a significant environmental threat due to their resistance to degradation. These materials are among the most widely used plastics, accounting for approximately 33% of global plastic production^{1,2}. Their chemical structure contributes to their durability and widespread use in packaging, textiles, and other industries. However, this stability makes them resistant to natural degradation, leading to persistent environmental pollution. The development of biological methods to accelerate the breakdown of these polymers could significantly mitigate their environmental impact.

This study explores the enzymatic degradation capabilities of a strain of *Serratia marcescens*, isolated from a plastic-polluted site in Mazamitla, Jalisco, focusing on producing and optimizing carboxylesterases induced by mineral oil. Optimal production conditions were identified using a Box-Behnken design, with 30°C, 150 rpm, and 15 g/L mineral oil emulsion yielding a specific activity of 0.504 U/mg. Further optimization revealed an optimal pH of 9 and a temperature of 37°C, enhancing activity by 1.9-fold. The cell-free extract was tested for its ability to degrade LDPE and PP over 96 hours, both with and without UV pretreatment. Enhanced degradation rates were observed with UV pretreatment, attributed to the initial breaking of plastic bonds. Weight loss measurements and Fourier Transform Infrared Spectroscopy (FTIR) analyses indicated significant plastic oxidation processes, with a 60% increase in the Carbonyl Index (CI) for UV-pretreated PP. The enzymatic extract showed a 30% reduction in activity after 144 hours. FTIR analysis confirmed the presence of functional groups associated with plastic degradation. These findings demonstrate the potential of *S. marcescens* carboxylesterases in the biodegradation of LDPE and PP, with UV pretreatment enhancing the degradation rate.

References

1. Al-Salem, S. M., Lettieri, P., & Baeyens, J. (2009). Recycling and recovery routes of plastic solid waste (PSW): A review. *Waste Management*, 29(10), 2625-2643.
2. Geyer, R., Jambeck, J. R., & Law, K. L. (2017). Production, use, and fate of all plastics ever made. *Science Advances*, 3(7), e1700782.

SOLUTIONS BASED ON COPPER IONS AS ANTIFUNGAL CONTROL IN SOIL AGAINST THE PHYTOPATHOGENIC FUNGUS *NEOPESTALOTIOPSIS* SPP.

Jesús Emiliano Cantador-Campos¹, Elizabeth Quintana-Rodríguez², Domancar Orona-Tamayo², Karla Liseth Macías-Sánchez¹, José de Jesús Esparza-Claudio², Catalina De la Rosa-Juárez² and Nancy Edith Lozoya- Pérez²

¹Instituto Politécnico Nacional (IPN). ²Soluciones tecnológicas y de Investigación, CIATEC A.C. Omega# 201, Col. Industrial Delta, C.P. 37545, León, Guanajuato., México.
Tel (477)710 0011, E-mail: eqintana@ciatec.mx.

Strawberry cultivation in Mexico has produced 639 thousand tons from 2016 to 2020, with Guanajuato being the third national producer with a contribution of 12% of total production¹. However, this crop has been affected by a new phytopathogen *Neopestalotiopsis* spp. This phytopathogen is the cause of leaf spots, diebacks, crown, root, and fruit rots². As this is an emerging threat, no specific methodologies have been developed for the pathogen³. Copper ions have algacide, nematicide, molluscicide, antibacterial and antiviral activities have been attributed. Previously, antimicrobial activity on strawberry fruit pathogens was described, extending shelf life. In addition, low cytotoxicity of these solutions has been demonstrated⁴. In the present study, the effect of copper ions on the in vitro growth of the fungus *N. rosae* at different concentrations was evaluated. In addition, the fungicidal activity of these solutions was analyzed in soils contaminated with spores of this fungus.

It was observed less fungal growth and irregular and melanized colonies at 0.2% copper ions solution in comparison with the control. In addition, conidia showed abnormalities in structure and size. At the 0.4% concentration, growth was totally inhibited. For the fungicidal activity tests on soil, it was observed that with 0.4% copper ions, fungal growth was reduced by 50% compared to the control. These results show the effectiveness of copper ions as an antifungal against the pathogenic fungus *N. rosae*, which presents a great potential for its use in the disinfection of agricultural soils. Future field studies will allow us to verify their effectiveness under agronomic conditions.

References

1. Secretaría de Agricultura y Desarrollo Rural (2022).
2. U. Acosta (2022). Caracterización de aislamiento de *Neopestalotiopsis* spp. Y efectos de fungicidas en pre y post-infección en cultivos de fresa. Universidad Autónoma de Chapingo, Chapingo, Edo. De Mex.
3. Dirección General de Sanidad Vegetal, Centro Nacional de Referencia Fitosanitaria (2022).
4. C. L. De la Rosa Juárez, E. Quintana Rodríguez, D. Orona-Tamayo, M.A. Silva Goujon and N.E. Lozoya-Pérez (2022). J. CIM. vol. 10, 1.

IMMOBILIZATION OF *BACILLUS SUBTILIS* ON MAGNETITE NANOPARTICLES FOR ARSENIC REMOVAL IN WATER

César Abdiel Cárdenas Villanueva¹, Dr. Juan Antonio Rojas Contreras¹, Dra. Perla Guadalupe Vázquez Ortega¹, Dra. María Adriana Martínez Prado¹, Dr. Víctor Manuel Ayala-García²

¹TecNM/I.T. Durango. Posgrado en Ingeniería Bioquímica Felipe Pescador 1803, Nueva Vizcaya, 34080 Durango, Dgo., México, ² Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango, Av. Veterinaria S/N, Col. Valle del Sur, 34120 Durango, Dgo., México.
22041572@itdurango.edu.mx

Arsenic is a toxic and carcinogenic element that poses a significant threat to human health and aquatic ecosystems. As water demands increase and natural sources face greater anthropogenic pressure, the need to develop effective and sustainable technologies for removing this contaminant becomes imperative. Developing sustainable technologies for removing inorganic contaminants from water is crucial, and applying microorganisms immobilized on magnetic nanomaterials presents a cost-effective and environmentally friendly alternative for water resource management. Magnetite nanoparticles (MNPs) have gained attention due to their unique capacity to act as magnetic supports in capturing and retaining bacteria. This study focuses on the immobilization of *Bacillus subtilis* on magnetite nanoparticles (MNPs) to address arsenic contamination in water. This approach offers significant advantages in system handling and recovery by allowing easy separation of fixed cells from the environment using an external magnetic field. The MNPs were synthesized by chemical co-precipitation from FeCl₂ and FeCl₃ solutions. *Bacillus subtilis* cells were incubated for 24 hours at 37 °C, suspended in saline solution, and incubated with MNPs for 8 minutes at 150 rpm at 37 °C. The cells immobilized on MNPs were separated using an external magnetic field, and the resulting supernatant, representing free, uncoated cells, was collected. Bacterial capture efficiency (BCE) was calculated using the formula $BCE (\%) = 1 - (CFU \text{ supernatant} / CFU \text{ initial})$. The results showed that MNPs have the capacity to precipitate in the presence of a permanent magnet, confirming the desired purity and magnetic properties. It was found that the efficiency of MNPs for immobilizing *Bacillus subtilis* increased with the concentration of MNPs, reaching an efficiency of 98%. In conclusion, MNPs were successfully obtained by the chemical co-precipitation method and respond to the application of an external magnetic field. The immobilization of *Bacillus subtilis* on MNPs showed a 98% efficiency, which is favorable for future work on arsenic removal in potable water.

NEW BIODEGRADABLE PLASTICS (POLY-3-HYDROXYALKANOATES) PRODUCED BY AZOTOBACTER VINELANDII OP RECOMBINANT STRAINS

Alma Luz Carmona¹, Josefina Guzmán Aparicio¹, Dayren Díaz Gómez¹, Ma. Soledad Moreno, Carlos Peña²,
Guadalupe Espín¹ and Daniel Segura González¹

¹Departamento de Microbiología Molecular. Instituto de Biotecnología, UNAM. ²Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, UNAM. Av. Universidad 2001, Col. Chamilpa, Cuernavaca, Mor. CP 62250.
E-mail: alma.carmona@ibt.unam.mx

Polyhydroxyalkanoates (PHAs) are polyesters produced by some bacteria as reserve of carbon and energy. These polymers are used in the industry to produce biodegradable plastics that replace petroleum-based plastics. PHAs are composed of hydroxyalkanoic monomers and depending on the type of monomers present, they show different thermomechanical properties and can be similar to plastics such as polyethylene or polypropylene, but unlike these, they are biodegradable.¹

There are two classes of PHAs: short-chain-length PHAs (sclPHAs), that have monomers containing 3-5 carbon atoms, and medium-chain-length PHAs (mclPHAs), containing C6 to C14 monomers. The sclPHAs are materials thermoplastic but hard and brittle; meanwhile, mclPHAs are elastomeric and more flexible.

The copolymers scl-mclPHAs are elastomers with low crystallinity, low tensile strength and high elongation to break, similar to low density polyethylene.² The type of PHAs synthesized depends on the carbon source, the metabolic routes present for the synthesis of the monomers, and the specificity of the polymerizing enzyme (PHA synthase) of the organism.³

Azotobacter vinelandii is a bacterium that produces PHAs to levels as high as 85% of its cell dry weight, but it produces only sclPHAs. In this work 3 recombinant strains of *A. vinelandii* were developed in order to produce new bioplastics with different thermomechanical properties. Genetic modifications on the biosynthetic pathways of *A. vinelandii*, including the introduction of the *phaG* gene, which encodes a 3-hydroxyacyl-ACP thioesterase, and PA3924, which encodes a 3-hydroxyacyl-CoA ligase enzyme both from *Pseudomonas spp*, resulted in the ability to generate a diversity of mcl-HA monomers. In addition to the introduction of this new metabolic route that synthesizes different monomeric units, the PHA polymerase gene (*phbC*) of *A. vinelandii* was replaced with a gene (*phaC_{AC}*) from *Aeromonas caviae*, that codes for an enzyme with broader monomeric substrate specificity. This allowed the polymerization of different hydroxyacyl units to produce PHA copolymers with new composition and different thermomechanical properties. In conclusion, the modification of metabolic pathways in combination with the introduction of a broad substrate range PHA synthase allowed us to obtain a diversity of scl-mclPHAs with improved physical properties.

References

1. Madison, L. and G. Huisman (1999). Microbiol. Mol. Biol. Rev. 63 (1):21-53.
2. Rai, R. et. al.. (2011). Materials Sci. and Eng. Rep. 72(3):29-47.
3. Peña C., et. al.. (2014). Microb. Biotechnol 7(4):278-293.

AUXIN SYMPLASTIC TRANSPORT MODULATES ROOT HAIRS DEVELOPMENT OF ARABIDOPSIS INDUCED BY AZOSPIRILLUM BALDANIORUM SP245

E. Carrillo-Flores¹, Aguilera-Méndez A²., Mellado-Rojas M.E.¹ and Beltrán-Peña E.¹

¹Laboratorio de Transducción de Señales, Edificio B3, Instituto de Investigaciones Químico-Biológicas (IIQB) de la Universidad Michoacana de San Nicolás de Hidalgo (UMSNH). Morelia Mich., México. ²Laboratorio de Bioquímica de la Nutrición, Edificio B3, IIQB de la UMSNH. Morelia Mich., México.
E-mail: *eldabelt@umich.mx.

The plant growth promoting rhizobacteria, *Azospirillum baldaniorum* Sp245 when interacting with *Arabidopsis*, arrests the growth of root primary (RP), increases the lateral root number and the development of the root hairs (RH), it phenotype that has been attributed to the bacterial auxins. Indole-3-acetic acid (IAA), the natural auxin synthesized in the aerial part of the plants and mobilized to the root by rapid transport through the phloem and another that is carried out cell-cell and it is known as polar auxin transport (PAT). The PAT allows the formation of an auxin maximum in the root tip, which regulate the development of this organ. RH are originating from of some epidermis cells, known as trichoblasts. The initiation and growth of RH is regulated by IAA, which is mobilized from the RP tip to the differentiation zone of RH by the transporters AUX1/LAX and PIN2. On the other hand, it has been reported that auxins can diffuse through plasmodesmata (PD) small intercellular channels that cross the cell wall and connect the cytoplasm of neighbouring cells by the symplastic transport (ST), which depends on the PD permeability. This permeability is regulated by accumulation callose (polymer of glucose linked by a β -1,3 bond), through of callose synthases (CAL/GSL) and two types of proteins that help at callose deposition on the PD neck: PLASMODESMATA CALLOSE-BINDING PROTEIN (PDCB) and PLASMODESMATA-LOCATED PROTEIN (PDLP). On the other hand, callose degradation is carried out by β -1,3-glucanases. The present study aims to analyze whether the ST is involved in growth and density of RH induced by *A. baldaniorum*. For this, *pdlp5-1 DR5:GUS* mutant and *PDLP5OE DR5:GUS* overexpression lines were used. The results showed an increase in RH development in *PDLP5OE DR5:GUS* seedlings inoculated with *Azospirillum*, phenotype that could be due to the restriction of auxin symplastic transport caused by the PD closure.

IDENTIFICATION OF ANTIGENS IN THE IMMUNOPROTEOME OF *LISTERIA MONOCYTOGENES*

Kimberly Arisbeth Casal-Rodríguez¹, Ulises Vega-Castillo, Eva Sosa-Sillas, Evangelina Beltrán-López and Héctor Samuel López-Moreno¹

¹Laboratory of Molecular Biomedicine, CAC-UAS-264; Graduate Program in Biomedical Sciences, School of Chemical and Biological Sciences, Universidad Autónoma de Sinaloa, Cd. Universitaria, Culiacán, CP.80010, Sinaloa, México.

hslmoreno@ms.uas.edu.mx; CP.80010; +526675747963

Listeria monocytogenes is a Gram-positive bacillus responsible for listeriosis^{1,2}, one of the most critical foodborne illnesses³, characterized by high rates of hospitalization and mortality^{4,5,6}. This bacterium has the ability to adapt, survive, and disseminate under variable conditions, both in the environment and within the human host^{7,8}, because of its virulence factors integrated into its proteome^{9,10,11}. To analyze and identify these and others virulence factors or antigens that play a critical role in the host-bacteria relationship, the immunoproteomic profile study is a powerful biomedical biotechnology tool. Our aim was to identify antigens in the immunoproteome of *L. monocytogenes*. In this context, a proteomic profile of *L. monocytogenes* (ATCC 7448) was obtained using a proteinic *Listeria* extract (PLE) resolved by 2-D minigels (using IPGs strips pH 3-10 and 12% SDS-PAGE and Coomassie stain). Afterward, the *L. monocytogenes* immunoproteomic profile was obtained by nitrocellulose electrotransfer of the PLE to develop a western blot using the sera of BALB/c mice PLE immunized, the antigen-antibody reaction was evidenced by peroxidase and diaminobenzidine. A total of 24 spots were detected in the *L. monocytogenes* proteomic profile, ranging from 11 to 120 kDa and pI between 3.8 and 5.6. In this sense, the majority of the spots was observed in the acidic pH region. On other hand, the immunoproteomic analysis of *L. monocytogenes* showed 16 antigenic spots equivalent to 67% reactivity compared with its proteomic profile, the antigens ranging from 12 to 120 kDa and pI between 3.6 and 7.9. Finally, our experimental data showed pI5 (15kDa and pI 4.5), an unreported antigen of the immunoproteome of *L. monocytogenes*. Nevertheless, a more detailed analysis is necessary for biochemical identification of this *Listeria* antigen and to resolve its role in the host-bacteria relationship.

References

- Zakaria, A. I., & Sabala, R. F. (2024). Potential public health hazards related to consumption of poultry contaminated with antibiotic resistant *Listeria monocytogenes* in Egypt. *BMC microbiology*, 24(1). <https://doi.org/10.1186/S12866-024-03183-X>
- Centro para el Control y la Prevención de Enfermedades. (2017, junio 28). *Listeria* (Listeriosis). <https://www.cdc.gov/spanish/listeria/index.html>
- Organización Mundial de la Salud. (2018, febrero 20). *Listeriosis*. <https://www.who.int/es/news-room/fact-sheets/detail/listeriosis>
- Disson, O., Moura, A., & Lecuit, M. (2021). Making Sense of the Biodiversity and Virulence of *Listeria monocytogenes*. *Trends in microbiology*, 29(9), 811–822. <https://doi.org/10.1016/J.TIM.2021.01.008>
- Drolia, R., & Bhunia, A. K. (2019). Crossing the Intestinal Barrier via *Listeria* Adhesion Protein and Internalin A. *Trends in microbiology*, 27(5), 408–425. <https://doi.org/10.1016/J.TIM.2018.12.007>
- Finn, L., Onyeaka, H., & O'Neill, S. (2023). *Listeria monocytogenes* Biofilms in Food-Associated Environments: A Persistent Enigma. *Foods* (Basel, Switzerland), 12(18). <https://doi.org/10.3390/FOODS12183339>
- Chávez-Arroyo, A., & Portnoy, D. A. (2020). Why is *Listeria monocytogenes* such a potent inducer of CD8+ T-cells? *Cellular microbiology*, 22(4). <https://doi.org/10.1111/CMI.13175>
- Tönz, A., Freimüller Leischfeld, S., Stevens, M. J. A., Glinski-Häfel, D., Ladner, V., Gantenbein-Demarchi, C., & Miescher Schwenninger, S. (2024). Growth Control of *Listeria monocytogenes* in Raw Sausage via Bacteriocin-Producing *Leuconostoc carnosum* DH25. *Foods*, 13(2), 298. <https://doi.org/10.3390/FOODS13020298/S1>
- Cong, Z., Xiong, Y., Lyu, L., Fu, B., Guo, D., Sha, Z., Yang, B., & Wu, H. (2024). The relationship between *Listeria* infections and host immune responses: Listeriolysin O as a potential target. *Biomedicine & Pharmacotherapy*, 171, 116129. <https://doi.org/10.1016/J.BIOPHA.2024.116129>
- Lopes-Luz, L., Mendonça, M., Fogaça, B., Kipnis, A., Bhunia, A. K., Bühner-Sékula, S., Mendonça, M., Bernardes Fogaça, M., Kipnis, A. E., & BÉ Uhrer-S Ekula, S. (2021). *Listeria monocytogenes*: review of pathogenesis and virulence determinants-targeted immunological assays. *Critical Reviews in Microbiology*, 47(5), 647–666. <https://doi.org/10.1080/1040841X.2021.1911930>
- Quereda, J. J., Morón-García, A., Palacios-Gorba, C., Dessaux, C., García-del Portillo, F., Pucciarelli, M. G., & Ortega, A. D. (2021). Pathogenicity and virulence of *Listeria monocytogenes*: A trip from environmental to medical microbiology. *Virulence*, 12(1), 2509–2545. <https://doi.org/10.1080/21505594.2021.1975526>

COMPREHENSIVE STRATEGIES FOR THE DISCOVERY OF PLASTIC-DEGRADING MICROORGANISMS AND ENZYMES

Leticia Casas-Godoy^{1*}, Andrés Méndez-Zamora², María Sinisterra-Sierra², Jesús Alberto Urbar-Ulloa², Amador Campos-Valdez², Iliana Barrera-Martínez¹, Marcela Robles-Machuca³, Alejandro Pereira-Santana⁴

¹CONAHCYT- CIATEJ, Camino Arenero 1227, El Bajío, 45019, Zapopan, Jalisco, ²Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, Unidad de Biotecnología Industrial,

Camino Arenero 1227, El Bajío del Arenal, 45019 Zapopan, Jalisco, México, ³Universidad Autónoma de Nayarit, Secretaría de Investigación y Posgrado. Ciudad de la cultura s/n centro, 63000, Tepic, Nayarit, México,

⁴CONAHCYT- CIATEJ, Parque Científico Tecnológico de Yucatán, 97302, Mérida, Yucatán. E-mail: lcasas@ciatej.mx

Over fifty years ago, plastics started substituting natural materials, and nowadays, they are essential to the global economy. Plastics are long-chain synthetic polymeric molecules composed of carbon, hydrogen, silicon, oxygen, chloride, and nitrogen. Today, these materials represent a worldwide environmental problem due to their indiscriminate use and insufficient efforts for proper recycling or waste management. Some of the most important plastics are polyurethane (PUR), polyethylene (PE), polyamide (PA), polyethylene terephthalate (PET), and polystyrene (PS).

Plastics are resistant to microbial attack since they have not been a part of nature long enough to allow the evolution of highly active plastic degrading enzymes. Environmental biodegradation of plastics is a combination of abiotic and biotic factors. UV light and weather disruption currently represent the route for plastic disruption, converting them into micro and nanoplastics. However, they need further degradation.

Recently, the use of microorganisms and enzymes for the biodegradation of plastic waste has gained attention. Enzymes capable of plastic degradation include hydrolases such as carboxylesterases (EC 3.1.1.1), lipases (EC 3.1.1.3), cutinases (EC 3.1.1.74), PETases (EC 3.1.1.101), MHETases (EC 3.1.1.102), proteases (EC 3.4), and oxidoreductases as laccases (EC 1.10.3.2). This project aimed to discover microorganisms and enzymes capable of degrading plastics. Two strategies were followed: a traditional approach and a computational biology strategy. Using the traditional approach, 600 strains with the desired enzymatic activity were isolated from 9 sampled plastic-contaminated sites. Among this collection, a strain of *Serratia marcescens* stands out for its ability to degrade LDPE. The computational biology strategy allowed the identification of 3 fungi and 3 bacterial enzymes for PET degradation. These sequences have not been previously reported for plastic degradation. This project will lay the groundwork for an efficient approach to the plastics pollution problem, ensuring that in the future it is solved in an appropriate way.

CHARACTERIZATION AND EVALUATION OF THE BIOLOGICAL ACTIVITY OF THE TA-PASP POLYMER

Ileana Castillo Tobias¹, Luis Enrique Cobos Puc¹, Oyervides Muñoz Ernesto¹, Jesús Alejandro Claudio Rizo¹, Dennis Cabrera Munguía¹, Aguayo Morales Hilda¹, Rodríguez Salazar María del Carmen¹ y Silva Belmares Sonia Yesenia¹

Facultad de Ciencias Químicas, Unidad Saltillo, Universidad Autónoma de Coahuila. Boulevard V. Carranza S/N esq con Ing. José Cárdenas Valdés, República Oriente, CP 25280, Saltillo, Coahuila.
E-mail: ileana.castillo@uadec.edu.mx

In recent decades, polymers have been the subject of extensive research due to their biodegradability, biocompatibility, and versatility in terms of physicochemical properties¹. Currently, great importance has been given to polymer derivatives obtained from modifications to optimize their properties for greater utility². In this work, the synthesis of two polymers with potential anticancer, antibacterial activity, and low hemolytic effects has been carried out³⁻⁴. These were obtained by the reaction of sodium polyaspartate (PAspNa) and modified with two quaternary ammonium salts with a carboxylic terminal group. 1-(Carboxypentyl)pyridinium bromide, 1-(carboxypentyl)trimethylammonium bromide were sintered from a quaternization reaction between 6-bromohexanoic acid and the corresponding tertiary amines (pyridine and trimethylamine). The ammonium salts were chemically grafted onto the macromolecular chains of sodium polyaspartate using an acid-base reaction.

The chemical structures of the two polymers were analyzed and confirmed by FT-IR and ¹H NMR. The corresponding thermal stability was analyzed by thermogravimetric analysis (TGA). Once synthesized and characterized, hemolysis tests were carried out on isolated human erythrocytes, confirming the low hemolytic effect of the PA-PAsp polymer (12-200 ppm). In the case of the TA-PAsp polymer, it was slightly hemolytic at concentrations of 12 and 25 ppm. This suggests that TA-PAsp has a different sensitivity than PA-PAsp in erythrocyte membranes. Subsequently, *in vitro* tests were carried out against a cancer cell line (MCF-7) and a healthy cell line (3T3) as well as four different bacterial strains (*Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*). The antibacterial activity of the TA-PAsp derivative was determined by measuring the minimum inhibitory concentration and the minimum bactericidal concentration. Finally, cell proliferation was evaluated by 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) analysis after 24, 48 and 72 h of treatment. It was observed that the TA-PAsp polymer only had a cytotoxic effect in MCF-7 cells only at a concentration of 1800 µg/mL at 24 h.

References

1. Castillo-Tobías, I.; Berlanga, L.; Poblano, J.; Rodríguez-Salazar, M.d.C.; Aguayo-Morales, H.; Cobos-Puc, L.E. Fundamental Considerations of Targeted Drug Therapies for Breast Cancer. *Future Pharmacology* 2023, 3, 686-707.
2. Dahman, Y. *Biomaterials science and technology: fundamentals and developments*; CRC Press: 2019.
3. Mukhtar, A.; Saqib, S.; Ullah, S.; Sagir, M.; Tahir, M.; Mahmood, A.; Al-Sehemi, A.G.; Assiri, M.A.; Ibrahim, M.; Zulfiqar, A. Applications of Polymeric Materials in Biomedical Engineering. *Sustainable Production and Applications of Waterborne Polyurethanes* 2021, 133-142.
4. Torres-Rodríguez, A.; Avérous, L.; Pollet, E.; de Jesús Sosa-Santillán, G.; Zugasti-Cruz, A.; Sierra-Rivera, C.A.; Pérez-Aguilar, N.V.; García-Lobato, M.A.; Oyervides-Muñoz, E. Antimicrobial and anticancer potential of novel polyaspartate derivatives synthesized via quaternary ammonium grafting. *Journal of Applied Polymer Science* 2022, 139, e52907.

ENZYMATIC SACCHARIFICATION OF AGAVE DURANGENSIS BAGASSE FOR THE PRODUCTION OF BIOHYDROGEN

Gerardo Javier Chaidez-Gurrola¹, Gerardo Alfonso Anguiano-Vega¹, Maria Guadalupe Nieto-Pescador¹, Carlos Antonio Alba-Fierro¹, Francisco Javier Rios-Fránquez^{1*}

¹Facultad de Ciencias Químicas Durango, Universidad Juárez del Estado de Durango Av. Veterinaria s/n Circuito Universitario Col. Valle del sur C.P. 34120. Durango, Dgo., México.

*E-mail: javier.rios@ujed.mx

Agave durangensis bagasse is an abundant waste from mezcal industry in the state of Durango, which is currently underutilized to obtain value-added products. Second generation biofuels obtained from agro-industrial waste are an alternative to the use of fossil fuels, including hydrogen, which has gained popularity in recent years and of which studies continue to be carried out to optimize its production from lignocellulose. In the present work, enzymatic saccharification tests of agave bagasse were carried out with cellulolytic extracts obtained from a native strain of *Bacillus subtilis*, previously isolated from a fresh sample of bagasse obtained from the company 618 mezcal of Nombre de Dios, Durango. In the enzymatic hydrolysis tests carried out in 250 mL Erlenmeyer flasks containing 2 and 3% of alkalinely pretreated agave bagasse, suspended in Citrate-Phosphate buffer pH 5.5 and adjusting the enzymatic activity to 2 U/mL, values of reducing sugars of 465 µg/mL after 2 hours of enzymatic hydrolysis at 60°C. These sugars will be used as a substrate for the production of cellulosic biohydrogen in a sequential process of saccharification and anaerobic fermentation with the bacteria *Clostridium butiricum*.

REGENERATION *IN VITRO* OF JIPI PALM (*CARLUDOVICA PALMATA* RUIZ & PAVÓN) MEDIATED BY THIDIAZURON

Samuel Alejandro Chan Poot¹, Elidé Avilés Berzunza¹, Gregorio Godoy Hernández¹

¹Yucatan Scientific Research Center A. C., Street. 43 No. 130- for 32 and 34, Chuburna de Hidalgo Colony, Pinzón II, 97205 Mérida, Yucatan, Mexico.
E-mail: ggodoy@cicy.com.mx

Carludovica palmata, or *jipi palm*, is a terrestrial and stemless plant, with compound foliage made up of radical leaves and long petioles, native to the tropical and humid forests of America. The importance of *C. palmata* lies in its fibers, which are used to make the worldwide famous Panama hats (Harling, 1958, Ceballos, 1998). *Jipi palm crop* presents several limitations for the expansion of cultivation areas, including slow vegetative propagation and zero seed germination under traditional field cultivation conditions. Therefore, a viable biotechnological strategy is the *in vitro* plant tissue culture. In this study, the response of *C. palmata* explants to the growth regulator thidiazuron (TDZ) was evaluated to develop a mass multiplication protocol for this species. Seven different concentrations of TDZ and a control (0.00 μM TDZ) were evaluated. The tests were carried out in PC culture medium as a base, evaluating the size and number of shoots for each TDZ concentration. The results were compiled in an Excel matrix and processed in the MINITAB statistical program to obtain the ANOVAs and Tukey's tests for the evaluated parameters. The statistical analyses showed that the highest number of shoots was obtained with the 7.5 μM TDZ concentration, with an average of 85.2 ± 4.4 shoots per explant. However, the length of these shoots was the lowest compared to the other TDZ treatments, and the control (0.00 μM TDZ). Under *ex vitro* conditions, the micropropagated plants showed a phenotype similar to traditional way-propagated seedlings.

References.

1. Ceballos E., J. (1998). Elementos para la conservación y manejo de *Carludovica palmata* en Centroamérica. Tesis de maestría. Centro Agronómico Tropical de Investigación y Enseñanza (CATIE). pp. 80.
2. Harling, G. (1958). Monograph of the Cyclanthaceae. Acta de Horticultura. Berg, 18, pp. 428.

HSA-MIR-206 EFFECT ON TUMORIGENESIS AND CHEMORESISTANCE OF HCT116 COLORECTAL CANCER CELLS

Lizbeth Guadalupe Chávez-Ramos¹, Rosalío Ramos-Payán¹, Mariana Avendaño-Félix², Geovanni Romero Quintana¹, Alfredo Ayala-Ham², Jorge López-Gutiérrez¹, Maribel Aguilar-Medina^{1*}

¹Faculty of Biological and Chemical Sciences, Autonomous University of Sinaloa. Culiacán, Sinaloa, México. ²Faculty of Dentistry, Autonomous University of Sinaloa. Culiacán, Sinaloa, México.

*Correspondence: maribelaguilar@uas.edu.mx.

The significant impact of Colorectal cancer (CRC) worldwide has made necessary to search for new biomolecules with diagnostic and therapeutic potential that can prolong the life of people with this disease. Hsa-miR-206 has been identified as a miRNA with great therapeutic potential in CRC, since it has activity as a tumor suppressor. Objective: The aim of this study was to evaluate the effect of increase Hsa-miR-206 expression in HCT116 CRC cells on tumorigenesis and chemosensitization to 5-FU. Methodology: The Hsa-miR-206 expression was increased by pre-miR-206 transfection with lipofectamine. Tumorigenesis was assessed by a 3D spheroid formation assay in an ultra-low attachment plate. Chemosensitization to 5-FU was determined by MTT assay and morphological analysis. Cellular processes regulated by Hsa-miR-206 were identified by *in silico* analysis. Results: We found that overexpression of hsa-miR-206, decreased approximately 84.1% the size of spheroids ($p = 0.0012$) and reduced the half maximal inhibitory concentration (IC50) up to 72.7% ($p = 0.038$). *In silico* predictions suggested that Hsa-miR-206 could generate these significant effects by regulating a variety of genes, such as SOX9 and BCL2, which participate in cellular processes associated with CRC tumorigenesis and chemosensitization. Conclusion: Our results demonstrated that Hsa-miR-206 plays a pivotal role on decreasing the tumor formation capacity by potentiating cytotoxic damage of 5-FU in HCT116 CRC cells, showing potential as therapeutic biomolecule.

MOLECULAR AND PHYSIOLOGICAL CHARACTERIZATION OF YQCK LOCUS OF *BACILLUS SUBTILIS* AND ITS METABOLIC ROLE WITHIN THE ARS ARSENIC RESPONSE OPERON

Elizabeth Cisneros-Lozano¹, Luz Idalia Valenzuela-García², Maribel Cervantes-Flores¹,
Marcelo Barraza-Salas¹ and Víctor Manuel Ayala-García¹

¹Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur C.P. 34120., ²Centro de Investigación en Materiales Avanzados, Calle CIMAV n°110 Ejido Arroyo Seco Durango, Dgo. México CP.34147. Tel.6182727955, E-mail: elifonsi53@gmail.com

Exposure to arsenic (As) represents a serious risk to human health, being a global problem of considerable concern. This metalloid is highly toxic and carcinogenic, associated with diseases such as diabetes, cancer, and cardiovascular and neurological disorders¹.

In response to the constant exposure of As in the environment, bacteria have developed various genetic systems to counteract this toxicity. These systems include the *ars* operons, a set of genes that give them the ability to resist and metabolize As. This operon generally comprises genes that encode proteins involved in the detoxification and expulsion of As, which is regulated by a repressor that is activated in response to the presence of As in the environment².

The present study focuses on understanding the molecular mechanisms that allow *Bacillus subtilis* to resist and metabolize As with specifically interest in the characterization of the *yqck* gene within the *ars* operon of *B. subtilis*. This operon includes the genes *arsR*, *yqck*, *arsB* and *arsC*, transcriptionally regulated by ArsR³. The *yqck* gene encodes a putative arsenic lyase (*arsI*) responsible for the cleavage of C-As bonds in organ-arsenical compounds.

The methodology includes bioinformatics analyses to study the sequence homology and structure of YqcK, as well as the creation of a mutant strain to evaluate its response to As. Additionally, experiments will be performed to measure the expression of *yqck* and resistance to various forms of organic As.

References

1. Monteiro De Oliveira, E. C., Caixeta, E. S., Santos, V. S. V., & Pereira, B. B. (2021). Journal of Toxicology and Environmental Health, vol 24, 119–135.
2. Ben Fekih I, Zhang C, Li YP, Zhao Y, Alwathnani HA, Saquib Q, Rensing C, Cervantes C (2018). Frontiers in Microbiology, vol 9, 1-11.
3. Sato T, Kobayashi Y (1998). Journal of Bacteriology, vol 180, 1655–1661.

REMOVAL OF PHARMACEUTICALS COMPOUNDS (PCS) USED IN COVID-19 TREATMENT PRESENT IN WASTEWATER BY CONSTRUCTED WETLANDS

Jessica F. Contreras-Mojica, Víctor H. Robledo-Zacarias, David A. Camarena-Pozos

Centro de Innovación Aplicada en Tecnologías Competitivas (CIATEC)

Omega 201, Industrial Delta, 37545 León, Guanajuato, México.

E-mail:jcontreras.estudiantepicyt@ciatec.mx

Emerging contaminants (ECs), including pharmaceuticals compounds (PCs), are introduced into the environment through municipal discharges and impact water quality, ecosystems and human health¹. Undoubtedly, patient treatments during the COVID-19 pandemic increased the presence of PCs in wastewater. It has been reported that conventional water treatment plants remove less than 50% of PCs, requiring more expensive non-conventional technologies². Constructed wetlands, which combine plants, substrates and microorganisms, offer a cost-effective and sustainable solution³. This research aims to evaluate the elimination of PCs by constructed wetlands as a tertiary treatment.

The first objective was to characterize dexamethasone (DEX), diclofenac (DCF), ibuprofen (IBP), paracetamol (PCT) and azithromycin (AZT) in water samples from treated water discharges, from a COVID hospital and from a surface water body disturbed by wastewater in the city of León, Gto. The samples were analyzed by liquid chromatography coupled to mass spectrometry (LC-MS). Solid phase extraction (SPE) was performed using Strata cartridges using acetonitrile as eluent. The second objective was to evaluate the degradation of PCs using wetlands constructed at laboratory scale with a capacity of 25 L using tezontle as substrate and the *Typha domingensis* plant. Synthetic water containing 250 ng/L per PCs was used and three hydraulic retention times were evaluated (40, 60 and 80 hours).

Concentrations of PCs at the sampling points in the city were as follows: For treated water discharges, concentration ranges of 10.34-102 ng/L (PCT), 1-1113.33ng/L (DEX), 580-20263ng/L (AZT), 47.5-2863.5 ng/L (DCF) and 39.31-1741.33 ng/L (IBP) were found. Hospital raw water samples averaged 107.33ng/L (PCT), 176ng/L (DEX), 12566.7 ng/L (AZT), 166.6 ng/L (DCF), and 1563.8ng/L (IBP). For the disturbed surface water body the concentrations were 71.95ng/L (PCT), 32060ng/L (AZT), 563.33ng/L (DCF), and 216 ng/L (IBP). Treatment using constructed wetlands showed a removal above 72% for all PCs evaluated with a retention time of 60 hours.

References

1. Rivera-Utrilla et. al., (2013). Chemosphere 93(7): 1268-1287.
2. Yuvaraj-Ravikunmar et. al., (2022). Environmental Technology & Innovation 26: 2352-1864
3. Huma-Ilyas et. al., (2020). Environmental Science and Pollution Research 27:14342-14364

HOST-MICROBIOTA INTERACTIONS DRIVE THE PROBIOTICS ENRICHMENT IN THE MICROBIOTA OF *L. VANNAMEI*: A HOLOGENOME PERSPECTIVE

Fernanda Cornejo-Granados¹, Luigui Gallardo-Becerra¹, Sandra Romero-Hidalgo², Alonso Lopez-Zavala³, Rogerio Sotelo-Mundo⁴, Andrés Cota-Huizar⁵, Adrián Ochoa-Leyva^{1*}

¹Departamento de Microbiología Molecular, Instituto de Biotecnología, UNAM, México. ²Departamento de Genómica Computacional, Instituto Nacional de Medicina Genómica, S. de Salud, México. ³Departamento de Ciencias Químico Biológicas, Universidad de Sonora, México. ⁴Laboratorio de Estructura Biomolecular, Centro de Investigación en Alimentación y Desarrollo, México. ⁵Camarones El Renacimiento SPR de RI, Sinaloa, México
*adrian.ochoa@ibt.unam.mx

The holobiont is the sum of a host and its microbiota, forming a mutually beneficial ecological unit¹. In decapod shrimps, it is known that different species can carry different microbiota. However, it is still unknown how the genetic line affects the microbiota within organisms of the same species and whether genetic features can help “create” a microbiota that promotes health and growth. To address this, we analyzed the microbiota of the intestine and hepatopancreas associated with two genetic lines of *L. vannamei* (Gen1 and Gen2) confirmed by genotyping of 6,465 SNPs. Then, we analyzed the V3-V4 regions of the 16S rRNA gene and found that Gen1 had higher bacterial richness and significantly ($p < 0.05$) higher diversity than Gen2. Additionally, Gen1 had a significantly ($p < 0.0001$) wider niche breadth than Gen2, indicating that the microbiota of Gen1 could better exploit available resources. Further, an ANOSIM analysis showed that the genetic line variable had a more significant ($p < 0.05$) impact on modulating the microbiota of the hepatopancreas, explaining 30% ($r = 0.30$) of microbial differences between Gen1 and Gen2, while in the intestine, the effect was not significant. Finally, we performed a targeted search of taxa reported as beneficial for shrimp² and observed that Gen1 had a higher abundance of beneficial taxa than Gen2. Interestingly, the same analysis revealed a higher abundance of beneficial bacteria on healthy shrimps than on diseased ones. These results emphasize the importance of studying how specific genetic traits affect the shrimp microbiota and promote beneficial microbes. This information can be integrated into breeding programs and help producers to develop better larvae for shrimp production.

References

1. I Zilber-Rosenberg, E Rosenberg (2008) Role of microorganisms in the evolution of animals and plants: the hologenome theory of evolution. *FEMS Microbiol Rev* 32, 723-735.
2. JP Ochoa-Romo, F Cornejo-Granados, AA Lopez-Zavala, MT Viana, et. al.. (2022). Agavin induces beneficial microbes in the shrimp microbiota under farming conditions. *Sci Rep* 12, 6392.

COMPARATIVE ANALYSIS OF THE OSTEOGENIC EFFECT OF THE MIXTURE OF CHITOSAN HYDROGEL WITH BONE OR BLADDER ECM HYDROGELS AND BONE MINERAL

Juan Luis Cota Quintero¹, Rosalío Ramos-Payán², Geovanni Romero Quintana², Alfredo Ayala-Ham³, Jorge Armando López Gutiérrez², Mariana Avendaño-Félix³, Elsa Maribel Aguilar-Medina^{2*}

¹Faculty of Biology, Autonomous University of Sinaloa, Culiacán, Sinaloa, México; ²Faculty of Chemical Biological Sciences, Autonomous University of Sinaloa, Culiacán, Sinaloa, México; ³Faculty of Odontology, Autonomous University of Sinaloa, Culiacán, Sinaloa, México.

* Correspondence: maribelaguilar@uas.edu.mx.

Introduction. Bone is one of the hardest tissues in the human body, of a mineralized organic-inorganic nature and is formed by hydroxyapatite and type I collagen, cells and blood vessels. Tissue engineering through the implantation of artificial materials has become one of the most researched scientific fields and can be used in regenerative medicine. The main purposes of tissue engineering can be classified as restoring, replacing, maintaining, or improving the function of different types of biological tissues. For the success of tissue engineering, it is necessary to combine a scaffold with living cells and/or active molecules, which will serve as promoters of tissue regeneration. Chitosan is a natural cationic biopolymer derived from the deacetylation of chitin with different applications and characteristics that make it ideal for biomedical use, among which are: biocompatibility, degradation, zero toxicity, adhesion, as well as broad antibacterial agent activity. and antifungals. **Objective.** The present research proposes a comparative, longitudinal and prospective study with the objective of comparing the osteogenic effect of the mixture of chitosan hydrogel with bone or bladder ECM hydrogel, and bone mineral *in vivo* and *in vitro*. **Methodology.** For the *in vitro* tests, cultures of human peripheral blood macrophages and mesenchymal cells derived from human adipose tissue (hAd-MSCs) will be used to assess cytotoxicity by LDH release, expression of cytokines by CBAs, proliferation by MTT, life and death by fluorogenic stain, and osteoblast differentiation of hAd-MSCs in osteogenic medium. Finally, the *in vivo* bone regeneration potential of blended materials in 3-month-old male Wistar rats with a calvarium critical defect will be evaluated by cone beam computed tomography at 30, 60 and 90 days. **Statistical analysis.** Multiple comparison tests will be performed between groups using 2-way ANOVA. The results will be evaluated with the GraphPad Prism V8 software, the values will be taken as significant when there is a $p < 0.05$.

GENERATION AND PROPAGATION OF IMPROVED VARIETIES OF MALT AND FORAGE BARLEY: AN ALTERNATIVE CROP FOR VULNERABLE AREAS IN THE STATE OF JALISCO

Dianey Celeste Cruz Muñoz¹, Julio Armando Massange Sánchez¹

¹Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C. Camino Arenero 1227, El Bajío, Zapopan, Jalisco. ZIP code: 45019. Phone number: +523333455200 ext.1700
E-mail: jmassange@ciatej.mx

Barley (*Hordeum vulgare*) is a plant member of Poaceae family, mainly used as livestock feed and for malt production¹. It is the fourth most important worldwide cereal, and the fourth most produced cereal in Mexico after corn, sorghum and wheat^{2,3}. In Mexico, two of the main regions that produce barley are the Bajío region and Altiplano region⁴. In Bajío region, one of the most cultivated varieties with quality suitable for malt production is “Esperanza” variety while, for the Altiplano region, the attributes of fodder barley “Maravilla” variety stand out^{5,6}.

Currently, there is a focus on crop's yield stability to face adverse impacts from climate change and new diseases⁷. Therefore, in the case of barley, production and supply of forage and grain are at risk. Barley grain production in Mexico exhibits a stagnation with a downward trend (from 1,031,533 tons to 1,023,969 tons in 2012 and 2022 respectively³), this does not cover the requirements for malt production and then, beer manufacturing. To maintain the country's position as the main beer exporter, it is imperative to achieve self-sufficiency in barley production and grain procurement.

Barley tends to have low rates of outcrossing (<5%)⁸ so, their genetic variability is limited. Its improvement through chemical mutagenesis combined with plant tissue culture techniques is a viable way to generate genetic variability and then, develop new barley varieties⁹. The aim of this work is to generate and propagate improved varieties of malt and fodder barley through chemical mutagenesis with ethyl methanesulfonate (EMS) in seeds and immature embryos. Morphological traits of interest, such as plant size, spike expansion, among others, will be evaluated. In addition, genetic changes will be tracked through molecular markers.

The development of new barley varieties also provides an opportunity to promote the use and cultivation of this crop in the country, particularly in Jalisco, where some municipalities have high levels of marginalization and at the same time have suitable environmental conditions for barley growth.

References

1. Shewry P.R. & Ulrich S.E. (2014). Barley: Chemistry and Technology. Book (2nd ed.), 1-10.
2. FAO (2022) Agricultural production statistics. 2000-2021.
3. SIAP (2022) Anuario Estadístico de la Producción Agrícola. <https://nube.siap.gob.mx/cierreagricola/>
4. González-González, M., Zamora-Díaz, M.R., Solano-Hernández, S., Huerta-Zurita, R., Gómez-Mercado, R. & Rojas-Martínez, I. (2021). Mejoramiento genético de cebada en el INIFAP (1985-2020). Revista Mexicana de Ciencias Agrícolas, 25, 15-20.
5. González-González, M., Zamora-Díaz, M.R. & Solano-Hernández, S. (2016). Evaluación agronómica y física en líneas avanzadas de cebada maltera. Revista Mexicana de Ciencias Agrícolas, 7, 159-171.
6. Zamora-Díaz, M.R., Pérez-Ruíz, J.A., Huerta-Zurita, R., López-Cano, M.L., Gómez-Mercado, R., Rojas-Martínez, I. (2017). Maravilla: variedad de cebada forrajera para los Valles Altos de México. Revista Mexicana de Ciencias Agrícolas, 8(6), 1449-1454.
7. Lamichhane, S., & Thapa, S. B. (2022) Advances from conventional to modern plant breeding methodologies. Plant breeding and biotechnology, 10(1), 1-14.
8. Harwood, W.A. (2019) Barley: Methods and protocols. Book, 21-22.
9. Serrat, X., Esteban, R., Guibourt, N., Moysset, L., Nogués, S., & Lalanne, E. (2014). EMS mutagenesis in mature seed-derived rice calli as a new method for rapidly obtaining TILLING mutant populations. Plant Methods, 10.

IDENTIFICATION OF SIGNALING MOLECULES INVOLVED IN BACTERIAL COMMUNICATION ISOLATED FROM LICHENS

Miriam Joceline Cruz-Ramos¹, Varinia López-Ramírez¹, Juan Vázquez-Martínez¹
and Cynthia Paola Rangel-Chávez¹

¹Tecnológico Nacional de México/ Instituto Tecnológico Superior de Irapuato. Carretera Irapuato-Silao Km 12.5,
Col. El Copal. C.P.36821
varinia.lr@irapuato.tecnm.mx

Lichens are symbiotic associations between a mycobiont and one or more photobionts; recent research has revealed the presence of various bacterial genera, which play a crucial role in the physiology of lichens. Although bacteria play a very important role, understanding how they communicate within lichens is still an underexplored area. It is known that bacterial communication is mediated by the production, detection and response to molecules that act as chemical signals. These chemical signals make up complex communication systems that trigger the induction/repression of target genes, allowing the coordination of joint behaviors. Here we investigate the phenotypic change induced by bacterial interaction about the synthesis of signaling molecules in bacteria from lichen isolates collected in the state of Guanajuato, to characterize the specific molecules involved in the communication between lichen bacteria and determine whether these bacteria can coordinate joint behaviors that allow them to establish mutualistic or antagonistic interactions. To know the chemical signals involved in bacterial communication, interaction tests and gas chromatography were carried out. Through gas chromatography, it is expected to find *N*-acyl-homoserine lactone derivatives which are typical signaling molecules in Gram-negative bacteria. These molecules allow bacteria to produce biofilms and secrete secondary metabolites, which could influence bacterial dominance in a multi-species environment. Identifying signaling molecules lays the foundation for future research focused on evaluating the biotechnological potential of these molecules and their biological function in lichen symbiosis.

EVALUATION OF THE THERMOSTABILITY OF THE MUTANT PET HYDROLASE

Danna Cruz Rico¹, Jorge Marcos Viquez² and Silvia Armenta¹

¹Institute of Agricultural Sciences, Av. Universidad Km. 1. Exhacienda Aquetzalpa. C.P. 43600 Tulancingo, Hidalgo México,

²Open Lab, Cerro del Agua 274, Copilco Universidad, Coyoacán, Ciudad de México. C.P. 04360

E-mail: cr376434@uaeh.edu.mx, jorge@open-lab.com.mx, silvia_armenta@uaeh.edu.mx

Polyethylene Terephthalate (PET) is a commonly used plastic with important properties in the industry. It is non-biodegradable in the natural environment, making PET pollution one of Earth's most alarming problems today. Many researchers have discovered organisms such as *Thermobifida fusca* that excrete enzymes like cutinases capable of degrading PET¹. However, their catalysis time still takes too long, resulting in a slow degradation percentage. Similarly, *Ideonella sakaiensis* produces PET hydrolases, which may offer an effective solution for PET degradation². In this study, this PETase is being utilized as a platform for evolving mutants with enhanced catalytic properties through protein rational design and engineering techniques. Also, we aim to investigate the effects of several mutations on enhancing the thermostability of PETase. As a result of *in silico* analysis, we selected some specific amino acids (e.g., Asp, Glu, and Arg found in hyperthermophile proteins), which could increase the thermostability compared to the wild type. We obtained a mutant that could be more thermostable with PET degradation capability. Both of them were cloned into the plasmid PET22b+, obtaining the wild-type gene for PET hydrolase and another plasmid carrying the mutant gene, utilizing the T7 expression system. Both genes were equipped with a C-terminal His tag and an N-terminal pelB to facilitate their purification and secretion, respectively. IPTG at a concentration of 1mM was added to induce gene expression for 24 hours at 17°C in Luria-Bertani broth before harvesting. The purified enzymes will be characterized using various biochemical and biophysical techniques to assess their catalytic activity and thermostability. Finally, understanding the effects of these mutations on both, T_m and catalytic activity could accelerate the degradation process and lead to successful degradation because the efficient breakdown of Polyethylene Terephthalate is essential in tackling the increasing pollution problem.

References

1. Yoshida, S., Hiraga, K., Takehana, T., Taniguchi, I., Yamaji, H., Maeda, Y., Toyohara, K., Miyamoto, K., Kimura, Y., & Oda, K. (2016). A bacterium that degrades and assimilates poly(ethylene terephthalate). *Science*, 351(6278), 1196-1199. <https://doi.org/10.1126/science.aad6359>
2. Han, X., Liu, W., Huang, J., Ma, J., Zheng, Y., Ko, T., Xu, L., Cheng, Y., Chen, C., & Guo, R. (2017). Structural insight into catalytic mechanism of PET hydrolase. *Nature Communications*, 8(1). <https://doi.org/10.1038/s41467-017-02255-z>

CHARACTERIZATION OF WILD BLUEBERRY (*VACCINIUM LEUCANTHUM*) BY CONVENTIONAL AND FT-IR SPECTROSCOPIC TECHNIQUES

Acela Cuamatzi de Gante¹, Abdú Orduña Díaz² and Silvia Luna Suárez³

¹Centro de Investigación en Biotecnología Aplicada-IPN Unidad Tlaxcala, Ex-Hacienda San Juan Molino Carretera Estatal Tecuexcomac-Tepetitla Km 1.5, Tlaxcala C.P. 90700, México. Tels.: (55) 57296000, Ext. 87816.
E-mail: acuamatzig2300@alumno.ipn.mx

The consumption of blueberries has expanded significantly due to its nutraceutical ingredients¹, which offer numerous health benefits including the prevention of diabetes, cardiovascular, and neurological illnesses, among others². In the present study we characterized wild blueberry (*Vaccinium leucanthum*) at different stages of ripening from the state of Michoacán, using conventional techniques and FT-IR spectroscopy to obtain spectra for subsequent structural analysis. Total phenols were found to have representative bands between 1180 and 1260 cm⁻¹, ranging from 5.75±0.71 to 9.46±0.50 mg EGA/g; flavonoids were found to have representative bands between 1260 and 1280 cm⁻¹, 1025-1198 cm⁻¹, and 760-780 cm⁻¹, with a concentration between 1.75±0.07 and 2.29±0.16 mg EQ/g; and anthocyanins were found to have representative bands between 2910 and 2970 cm⁻¹, 1400-1700 cm⁻¹, and 900-1100 cm⁻¹, with a concentration between 2.45±0.26 and 126.24±13.74 mg C3G/100g

Keywords: *Vaccinium leucanthum*, characterization, FT-IR

References

1. Cosme, F., Pinto, T., Aires, A., Morais, M. C., Bacelar, E., Anjos, R., Ferreira-Cardoso, J., Oliveira, I., Vilela, A., & Gonçalves, B. (2022). Red Fruits Composition and Their Health Benefits—A Review. In *Foods* (Vol. 11, Issue 5). MDPI. <https://doi.org/10.3390/foods11050644>
2. Kalt, W., Cassidy, A., Howard, L. R., Krikorian, R., Stull, A. J., Tremblay, F., & Zamora-Ros, R. (2020). Recent Research on the Health Benefits of Blueberries and Their Anthocyanins. In *Advances in Nutrition* (Vol. 11, Issue 2, pp. 224–236). Oxford University Press. <https://doi.org/10.1093/advances/nmz065>

EXPLORING ESTERASE/LIPASE PRODUCTION IN *ASPERGILLUS FUMIGATUS* FOR PLASTIC DEGRADATION

Roberto-De la Rosa-Mora¹, Leticia Casas-Godoy³, Eduardo Trillo-Hernández^{1,2}, Marcela Robles-Machuca^{1*}

¹Universidad Autónoma de Nayarit, Secretaría de Investigación y Posgrado. Ciudad de la cultura s/n centro, 63000, Tepic, Nayarit, Mexico. *marcela.robles@uan.edu.mx; ²Estancias Posdoctorales-CONAHCYT, Coordinación de Apoyos a Becarios e Investigadores, Dirección de Posgrado, Ciudad de México; ³CONAHCYT-Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A.C., Unidad de Biotecnología Industrial, Camino Arenero 1227, El Bajío del Arenal, 45019 Zapopan, Jalisco, Mexico.

Abstract. The increase in plastic production and its excessive consumption by the population generates an environmental pollution problem because plastics are not rapidly degraded. The impact on ecosystems and humans is indirectly generated through microplastic particles. Alternatives for degrading plastics using fungi as a biological method are exemplified by Chien and Tsai (2022), who evaluated the degradation rates of plastic using lipolytic enzymes from the *Aspergillus* genus. Aim: To study the presence of plastic-degrading esterase/lipase secreted by *Aspergillus fumigatus* isolated from San Blas, Nayarit. Methodology: 45 fungal isolates from San Blas were analyzed for esterases/lipases (E/L) activity. The presence of extracellular E/L was analyzed by observing a degradation halo around the growth on a solid medium with tributyrin 10g/L + chloramphenicol 100mg/L at 30°C for 48 hours. Positive E/L strains were identified using MALDI-TOF MS. The production of the E/L enzyme was carried out in a liquid medium using tributyrin 10g/L as inducer at 30°C and 150 rpm for 9 days, and quantified using p-NPB 40mM at 405nm for 20 minutes. Results: 11.11% of the isolated fungi were identified as *A. fumigatus*. 51.11% of the total strains were positive for the E/L expression in a solid medium, with 13.04% corresponding to *A. fumigatus*. Two strains with higher E/L production were obtained at 72 and 144 hours, respectively. Conclusion: Two *A. fumigatus* strains isolated from a saline environment that produce extracellular E/L were obtained, which cleave the ester bonds of plastic polymers. *A. fumigatus* will be exposed to different plastics to determine their biodegradability.

References

1. Chien, HL, Tsai, YT, Tseng, WS, Wu, JA, Kuo, SL, Chang, SL, Huang, SL, & Liu, CT (2022). Biodegradation of PBSA films by elite isolates of *Aspergillus* and agricultural soil. *Polymers*, 14(7), 1320.

GENERATION OF MUTANT STRAIN DISRUPTED IN *ASER* GENE FOR STUDYING THE ARSENIC RESPONSE OF *ASE* OPERON IN *BACILLUS SUBTILIS*

Miguel Angel De La Torre-Arellano¹, María Teresa Alarcón-Herrera², Luz Idalia Valenzuela-García²,
Norma Urtiz Estrada¹ an Víctor Manuel Ayala-García¹

¹Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur C.P. 34120., ² Centro de Investigación en Materiales Avanzados, Calle CIMAV n°110 Ejido Arroyo Seco Durango, Dgo. México CP.34147.
E-mail: 1104144@alumnos.ujed.mx

Arsenic (As) is a toxic and carcinogenic metalloid that occurs naturally in soil, water, and air. It is known for its toxicity and has historically been used as a poison¹. In the environment, As exists in two main forms: inorganic and organic. Despite its toxicity, bacteria have developed mechanisms to metabolize As, allowing them to survive and proliferate in contaminated environments².

The *ase* operon in *Bacillus subtilis* is a key component in the response to As. Although information available about this operon is limited, it is known to be involved in As detoxification and resistance to this metalloid. In this study, a genetic construction was generated using the vector pMutin4 to disrupt *aseR* gene from *ase* operon from *B. subtilis* cells. By removing the function of AseR repressor, we would like to understand its specific function in this microorganism and its contribution to As detoxification process. To achieve this, an internal region of the *aseR* gene was cloned in the pMutin4 vector. Subsequently, *B. subtilis* cells were transformed with this construct to promote its integration by homologous recombination at the *aseR* locus to disrupt the gene and subsequently evaluate the As response in Δ *aseR* strain.

In conclusion, this study will provide valuable information about the function of the *aseR* gene in *ase* operon of *B. subtilis* and its role in the response to arsenic. These findings may have implications for biotechnology and environmental.

References

1. Páez-Espino, D., Tamames, J., de Lorenzo, V., & Cánovas, D. (2009). *Biology, Biochemistry, and Medicine*, vol 22, 117–130.
2. Huda, N., Khanom, A., Rahman, M. M., Huq, M. A., Rahman, M. M., & Banu, N. A. (2021). *International Journal Of Environmental Science And Technology*, vol 19, 9189-9208.

THE GENOTOXIC EFFECT OF FLUORIDE PRESENTS IN ENDEMIC SOIL ON ONION (*ALLIUM CEPA*) BULB CELLS

Angel R. Díaz Duarte¹, Rene Homero Lara-Castro¹, Roberto Briones Gallardo², Patricia Ponce-Peña¹, Gerardo A. Anguiano Vega¹, Estela Ruiz-Baca¹

¹Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N. Col. Valle del Sur C.P. 34120, ²Instituto de Metalurgia, Facultad de Ingeniería, Universidad Autónoma de San Luis Potosí, Av. Manuel Nava S/N, Lomas 4º sección, 78230.
E-mail: lcrh75jed.mx.

Fluoride (F⁻) is a highly reactive element in the earth's crust and the planet's aquifers. These fluoride ions can be in contact with plant species, inducing the constant absorption of this element. However, prolonged exposure to F⁻ can cause significant physiological, biochemical, and structural damage to cells, leading to cell and organism death. *Genotoxic damage* is the induced alteration of nucleic acid molecules carrying the cellular genome or other macromolecules that transmit genetic information. Several techniques, including the 'Comet Assay', can quantify this damage¹. This technique is a relatively simple and efficient method to determine DNA damage at the cellular level, assessing the integrity of the genome DNA molecule in each independent cell^{2,3}. After conducting a bioassay of laboratory exposure in soils contaminated with Fluorspar mineral (CaF₂) in the presence of onion bulbs

(*Allium cepa*) for 7, 15, and 25 days, was evaluated a genotoxic damage using the Comet

Assay. The findings revealed that genotoxic damage is significantly higher as a function of F⁻ concentration and with an increased exposure time between 7 and 15 days. However, a significant decrease in genotoxic damage was observed at 25 days of exposure. This research shows that once F⁻ is mobilized due to the erosion of fluorinated minerals in endemic soil, it can potentially bioaccumulate inside the cells in the first stages of exposure. Finally, F⁻ is recognized as a genotoxic agent in plant systems and can be considered a high-risk contaminant for plant species present in fluorspar ore endemic sites.

References

1. Santos, C. L. (2015). The use of comet assay in plant toxicology: recent advances. *Front. Genet.*, 6(216), 1-18.
2. Parvan, et. al.. (2020). Bioensaio com *Allium cepa* revela genotoxicidade de herbicida com flumioxazina. *RPAS*, 11, 10-10.
3. Peng, C. et. al.. (2021). Fluoride absorption, transportation and tolerance mechanism in *Camellia sinensis*, and its bioavailability and health risk assessment: a systematic review. *J. Sci. Food Agric.*, 101(2), 379-387.

IDENTIFICATION OF BACTERIAL PROTEASES ISOLATED FROM A MANGROVE IN NAYARIT

Fernando Díaz-Partida¹, Yuma-Citlalli Ávila-López¹, Leticia Casas-Godoy², Marcela Robles-Machuca*

¹Universidad Autónoma de Nayarit. Secretaría de Investigación y Posgrado. Ciudad de la cultura s/n centro, 63000, Tepic, Nayarit, México. ²CONAHCYT–Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A.C., Unidad de Biotecnología Industrial, Camino Arenero 1227, El Bajío del Arenal, 45019 Zapopan, Jalisco, México. Contacto: *marcela.robles@uan.edu.mx

Abstract. Plastics represent a colossal global impact in terms of sustainable development. Polyurethane (PUR) is a common contaminant found in the mangrove of San Blas, Nayarit, a saline environment where halophilic microorganisms thrive. Currently, bacteria have been associated with the degradation of plastics, serving as allies in preventing accumulation by secreting enzymes that cleave the bonds of these polymers without affecting the environment. Protease hydrolyzes the ester, amide, and urethane bonds of PUR. The aim of this work is to identify the protease activity of bacteria isolated from a mangrove in San Blas, Nayarit. Bacteria isolated from plastic samples collected from the mangrove and identified by MALDI-TOF MS were primarily screened in a solid medium to determine their capacity to produce protease, using 10 g/L whole milk powder as an inducer at 30°C for 24 hours. The formation of a degradation halo around the bacterial growth was analyzed. Protease production was then carried out in a liquid medium with peptone as an inducer at 30°C and 150 rpm. The analysis of protease activity was performed at 30 minutes using potassium caseinate 15 g/L at pH 8, and 10% TCA to stop the reaction, while ninhydrin was used to quantify free amino acids at 570 nm. A total of 35 isolated bacteria were analyzed, of which 51% tested positive for protease activity in a solid medium, with the genus *Bacillus* comprising 44%. Until now, 17% have demonstrated extracellular expression of protease enzymes in a liquid medium, with six bacteria of the genus *Serratia* showing high protease activity at 24 hours. A significant difference in protease production was observed when using whole milk powder and peptone as inducers in the screenings resulting in a 34% reduction in strains with protease activity.

DEVELOPMENT OF AN INOCULANT TO ACCELERATE THE COMPOSTING PROCESS OF BOVINE MANURE

Frida Sofía Doroteo Platas¹ and Antonino Báez Rogelio¹

¹Centro de Investigación en Ciencias Microbiológicas, Benemérita Universidad Autónoma de Puebla, Puebla, C.P. 72592
E-mail: frida.doroteo@alumno.buap.mx

Abstract –Livestock companies face daily challenges regarding the disposal of solid waste, especially the excreta generated by animals. The disposal of untreated or partially treated excreta poses a risk to the environment and public health. In Mexico, according to the National Agricultural Survey (ENA) conducted in 2019, there are approximately 32 million head of cattle, which could generate 672 tons of manure per day. The excessive production of bovine manure has led to inadequate management and indiscriminate application of manure on agricultural fields. The composting process allows the transformation of manure into a less polluting and stable material for use as a biofertilizer. This oxidative process includes four stages: mesophilic, thermophilic, cooling, and maturation. Each stage is characterized by microbial communities that adapt to the availability of nutrients, temperature, humidity, and pH. The objective of this study was to develop an inoculant to accelerate the composting process of bovine manure, in order to significantly reduce the process time and mitigate the environmental impact. To achieve this, 3 thermophilic and 4 mesophilic strains were isolated and identified from compost samples. The isolates were selected based on their ability to degrade cellulose and were then inoculated into compost piles at the beginning of the thermophilic phase and during the cooling phase. The results showed that the piles inoculated with microorganisms had a lower C/N ratio at the end of the composting process compared to the uninoculated pile. Additionally, the compost pile inoculated with microorganisms showed less nitrogen loss compared to the control pile. These results suggest that inoculation can mitigate the emission of polluting gases such as nitrous oxide (N₂O) inherent to composting.

References

1. Gómez-Brandon, M., Lazcano, C., & Domínguez, J. (2008b). The evaluation of stability and maturity during the composting of cattle manure. *Chemosphere*, 70(3), 436-444.
2. Hurtado, J. (2014). Evaluación del Efecto de Microorganismos Transformadores de Materia Orgánica (TMO) en el Proceso de compostaje de las Deyecciones de Bovinos, Porcinos y Conejos. [Título de magister, Universidad de Manizales.
3. Román, P., Martínez, M. M., & Pantoja, A. (2013). MANUAL DE COMPOSTAJE DEL AGRICULTOR. Oficina Regional de la FAO. <https://www.fao.org/3/i3388s/i3388S.pdf>

FUNCTIONAL CHARACTERIZATION OF HUB PROTEINS IN THE PHYTOHORMONE SIGNALING NETWORK IN *ARABIDOPSIS THALIANA*

Andrea Duarte Elías¹, Cinthya Paola Rangel-Chávez¹, Humberto Herrera-Ubaldo³, Paola Isabel Angulo Bejarano²

¹Departamento de Bioquímica. Instituto Tecnológico Superior de Irapuato, Irapuato 36 821, Guanajuato México. ²Departamento de Agronomía. Tecnológico de Monterrey campus Querétaro, Epignemio González 500, San Pablo Santiago de Querétaro, Qro.

³Department of Plant Sciences. University of Cambridge, Downing St., Cambridge CB2 3EA, United Kingdom.
E-mail: pangulobe@tec.mx

Phytohormones act at the site of synthesis or in different areas where they regulate specific physiological events. To accommodate the variety of metabolic signals involved in development, there are signaling pathways where phytohormones interact with each other through crosstalk, rendering the existence of a linear representation obsolete. Hub proteins, or central proteins, have high connectivity in a protein-protein interaction (PPI) network, with many interaction partners, connecting different network modules. Hormonal responses in plants involve sensory mechanisms, and PPIs regulate these processes.

Previous studies report the identification of nineteen hub proteins with potentially relevant involvement in hormonal signaling pathways in *A. thaliana* (Alzati Ramírez, 2022). However, these proteins have not been functionally characterized, so their role in the interconnection of these pathways is unknown. Characterizing these proteins is important to advance the understanding of plant functioning in their development and to understand how these proteins are involved in signal perception or transduction.

Therefore, the present work aims to functionally characterize hub proteins in the hormone signaling pathways in *A. thaliana*. To achieve this, we reconstructed the PPI networks for each reported hub, considering only their interacting proteins involved in the signaling pathways of seven hormones (auxins, cytokinins, ABA, ethylene, jasmonate, gibberellins, and brassinosteroids) and co-expression interactions. This allowed us to identify two hub proteins related to two and three hormonal signaling pathways. Subsequently, we obtained co-expression maps of each hub protein with their interactors, which allowed us to identify the tissues in which each hub and their interactors are co-expressed. Based on this information, we have proposed *in vitro* assays in *A. thaliana*, where the effect of different hormones on the plant phenotype will be evaluated. Keywords: phytohormones, protein-protein interactions, phytohormone signaling pathways, crosstalk, hub, interconnection.

EXPLORING ESTERASE/LIPASE PRODUCTION IN *ASPERGILLUS FUMIGATUS* FOR PLASTIC DEGRADATION

Roberto-De la Rosa-Mora¹, Leticia Casas-Godoy³, Eduardo Trillo-Hernández^{1,2}, Marcela Robles-Machuca^{1*}

¹Universidad Autónoma de Nayarit, Secretaría de Investigación y Posgrado. Ciudad de la cultura s/n centro, 63000, Tepic, Nayarit, Mexico. *marcela.robles@uan.edu.mx; ²Estancias Posdoctorales-CONAHCYT, Coordinación de Apoyos a Becarios e Investigadores, Dirección de Posgrado, Ciudad de México. ³CONAHCYT-Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A.C., Unidad de Biotecnología Industrial, Camino Arenero 1227, El Bajío del Arenal, 45019 Zapopan, Jalisco, Mexico.

Abstract. The increase in plastic production and its excessive consumption by the population generates an environmental pollution problem because plastics are not rapidly degraded. The impact on ecosystems and humans is indirectly generated through microplastic particles. Alternatives for degrading plastics using fungi as a biological method are exemplified by Chien and Tsai (2022), who evaluated the degradation rates of plastic using lipolytic enzymes from the *Aspergillus* genus. Aim: To study the presence of plastic-degrading esterase/lipase secreted by *Aspergillus fumigatus* isolated from San Blas, Nayarit. Methodology: 45 fungal isolates from San Blas were analyzed for esterases/lipases (E/L) activity. The presence of extracellular E/L was analyzed by observing a degradation halo around the growth on a solid medium with tributyrin 10g/L + chloramphenicol 100mg/L at 30°C for 48 hours. Positive E/L strains were identified using MALDI-TOF MS. The production of the E/L enzyme was carried out in a liquid medium using tributyrin 10g/L as inducer at 30°C and 150 rpm for 9 days, and quantified using p-NPB 40mM at 405nm for 20 minutes. Results: 11.11% of the isolated fungi were identified as *A. fumigatus*. 51.11% of the total strains were positive for the E/L expression in a solid medium, with 13.04% corresponding to *A. fumigatus*. Two strains with higher E/L production were obtained at 72 and 144 hours, respectively. Conclusion: Two *A. fumigatus* strains isolated from a saline environment that produce extracellular E/L were obtained, which cleave the ester bonds of plastic polymers. *A. fumigatus* will be exposed to different plastics to determine their biodegradability.

References

1. Chien, HL, Tsai, YT, Tseng, WS, Wu, JA, Kuo, SL, Chang, SL, Huang, SL, & Liu, CT (2022). Biodegradation of PBSA films by elite isolates of *Aspergillus* and agricultural soil. *Polymers*, 14(7), 1320.

GENERATION OF MUTANT STRAIN DISRUPTED IN *ASER* GENE FOR STUDYING THE ARSENIC RESPONSE OF *ASE* OPERON IN *BACILLUS SUBTILIS*

Miguel Angel De La Torre-Arellano¹, María Teresa Alarcón-Herrera², Luz Idalia Valenzuela-García², Norma Urtiz Estrada¹ and Víctor Manuel Ayala-García¹

¹Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur C.P. 34120., ² Centro de Investigación en Materiales Avanzados, Calle CIMAV n°110 Ejido Arroyo Seco Durango, Dgo. México CP.34147.
E-mail: 1104144@alumnos.ujed.mx

Arsenic (As) is a toxic and carcinogenic metalloid that occurs naturally in soil, water, and air. It is known for its toxicity and has historically been used as a poison¹. In the environment, As exists in two main forms: inorganic and organic. Despite its toxicity, bacteria have developed mechanisms to metabolize As, allowing them to survive and proliferate in contaminated environments².

The *ase* operon in *Bacillus subtilis* is a key component in the response to As. Although information available about this operon is limited, it is known to be involved in As detoxification and resistance to this metalloid. In this study, a genetic construction was generated using the vector pMutin4 to disrupt *aseR* gene from *ase* operon from *B. subtilis* cells. By removing the function of AseR repressor, we would like to understand its specific function in this microorganism and its contribution to As detoxification process. To achieve this, an internal region of the *aseR* gene was cloned in the pMutin4 vector. Subsequently, *B. subtilis* cells were transformed with this construct to promote its integration by homologous recombination at the *aseR* locus to disrupt the gene and subsequently evaluate the As response in Δ *aseR* strain.

In conclusion, this study will provide valuable information about the function of the *aseR* gene in *ase* operon of *B. subtilis* and its role in the response to arsenic. These findings may have implications for biotechnology and environmental.

References

1. Páez-Espino, D., Tamames, J., de Lorenzo, V., & Cánovas, D. (2009). *Biology, Biochemistry, and Medicine*, vol 22, 117-130.
2. Huda, N., Khanom, A., Rahman, M. M., Huq, M. A., Rahman, M. M., & Banu, N. A. (2021). *International Journal Of Environmental Science And Technology*, vol 19, 9189-9208.

THE GENOTOXIC EFFECT OF FLUORIDE PRESENTS IN ENDEMIC SOIL ON ONION (*ALLIUM CEPA*) BULB CELLS

Angel R. Díaz Duarte¹, Rene Homero Lara-Castro¹, Roberto Briones Gallardo², Patricia Ponce-Peña¹, Gerardo A. Anguiano Vega¹, Estela Ruiz-Baca¹

¹Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N. Col. Valle del Sur C.P. 34120, ²Instituto de Metalurgia, Facultad de Ingeniería, Universidad Autónoma de San Luis Potosí, Av. Manuel Nava S/N, Lomas 4° sección, 78230.
E-mail: lcrh75jed.mx.

Fluoride (F⁻) is a highly reactive element in the earth's crust and the planet's aquifers. These fluoride ions can be in contact with plant species, inducing the constant absorption of this element. However, prolonged exposure to F⁻ can cause significant physiological, biochemical, and structural damage to cells, leading to cell and organism death. *Genotoxic damage* is the induced alteration of nucleic acid molecules carrying the cellular genome or other macromolecules that transmit genetic information. Several techniques, including the 'Comet Assay', can quantify this damage¹. This technique is a relatively simple and efficient method to determine DNA damage at the cellular level, assessing the integrity of the genome DNA molecule in each independent cell^{2,3}. After conducting a bioassay of laboratory exposure in soils contaminated with Fluorspar mineral (CaF₂) in the presence of onion bulbs

(*Allium cepa*) for 7, 15, and 25 days, was evaluated a genotoxic damage using the Comet

Assay. The findings revealed that genotoxic damage is significantly higher as a function of F⁻ concentration and with an increased exposure time between 7 and 15 days. However, a significant decrease in genotoxic damage was observed at 25 days of exposure. This research shows that once F⁻ is mobilized due to the erosion of fluorinated minerals in endemic soil, it can potentially bioaccumulate inside the cells in the first stages of exposure. Finally, F⁻ is recognized as a genotoxic agent in plant systems and can be considered a high-risk contaminant for plant species present in fluorspar ore endemic sites.

References

1. Santos, C. L. (2015). The use of comet assay in plant toxicology: recent advances. *Front. Genet.*, 6(216), 1-18.
2. Parvan, et. al.. (2020). Bioensaio com *Allium cepa* revela genotoxicidade de herbicida com flumioxazina. *RPAS*, 11, 10-10.
3. Peng, C. et. al.. (2021). Fluoride absorption, transportation and tolerance mechanism in *Camellia sinensis*, and its bioavailability and health risk assessment: a systematic review. *J. Sci. Food Agric.*, 101(2), 379-387.

IDENTIFICATION OF BACTERIAL PROTEASES ISOLATED FROM A MANGROVE IN NAYARIT

Fernando Díaz-Partida¹, Yuma-Citlalli Ávila-López¹, Leticia Casas-Godoy², Marcela Robles-Machuca*

¹Universidad Autónoma de Nayarit. Secretaría de Investigación y Posgrado. Ciudad de la cultura s/n centro, 63000, Tepic, Nayarit, México. ²CONAHCYT–Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A.C., Unidad de Biotecnología Industrial, Camino Arenero 1227, El Bajío del Arenal, 45019 Zapopan, Jalisco, México.
Contacto: *marcela.robles@uan.edu.mx

Plastics represent a colossal global impact in terms of sustainable development. Polyurethane (PUR) is a common contaminant found in the mangrove of San Blas, Nayarit, a saline environment where halophilic microorganisms thrive. Currently, bacteria have been associated with the degradation of plastics, serving as allies in preventing accumulation by secreting enzymes that cleave the bonds of these polymers without affecting the environment. Protease hydrolyzes the ester, amide, and urethane bonds of PUR. The aim of this work is to identify the protease activity of bacteria isolated from a mangrove in San Blas, Nayarit. Bacteria isolated from plastic samples collected from the mangrove and identified by MALDI-TOF MS were primarily screened in a solid medium to determine their capacity to produce protease, using 10 g/L whole milk powder as an inducer at 30°C for 24 hours. The formation of a degradation halo around the bacterial growth was analyzed. Protease production was then carried out in a liquid medium with peptone as an inducer at 30°C and 150 rpm. The analysis of protease activity was performed at 30 minutes using potassium caseinate 15 g/L at pH 8, and 10% TCA to stop the reaction, while ninhydrin was used to quantify free amino acids at 570 nm. A total of 35 isolated bacteria were analyzed, of which 51% tested positive for protease activity in a solid medium, with the genus *Bacillus* comprising 44%. Until now, 17% have demonstrated extracellular expression of protease enzymes in a liquid medium, with six bacteria of the genus *Serratia* showing high protease activity at 24 hours. A significant difference in protease production was observed when using whole milk powder and peptone as inducers in the screenings resulting in a 34% reduction in strains with protease activity.

DEVELOPMENT OF AN INOCULANT TO ACCELERATE THE COMPOSTING PROCESS OF BOVINE MANURE

Frida Sofía Doroteo Platas¹ and Antonino Báez Rogelio¹

¹Centro de Investigación en Ciencias Microbiológicas, Benemérita Universidad Autónoma de Puebla, Puebla, C.P. 72592
E-mail: frida.doroteo@alumno.buap.mx

Abstract. Livestock companies face daily challenges regarding the disposal of solid waste, especially the excreta generated by animals. The disposal of untreated or partially treated excreta poses a risk to the environment and public health. In Mexico, according to the National Agricultural Survey (ENA) conducted in 2019, there are approximately 32 million head of cattle, which could generate 672 tons of manure per day. The excessive production of bovine manure has led to inadequate management and indiscriminate application of manure on agricultural fields. The composting process allows the transformation of manure into a less polluting and stable material for use as a biofertilizer. This oxidative process includes four stages: mesophilic, thermophilic, cooling, and maturation. Each stage is characterized by microbial communities that adapt to the availability of nutrients, temperature, humidity, and pH. The objective of this study was to develop an inoculant to accelerate the composting process of bovine manure, in order to significantly reduce the process time and mitigate the environmental impact. To achieve this, 3 thermophilic and 4 mesophilic strains were isolated and identified from compost samples. The isolates were selected based on their ability to degrade cellulose and were then inoculated into compost piles at the beginning of the thermophilic phase and during the cooling phase. The results showed that the piles inoculated with microorganisms had a lower C/N ratio at the end of the composting process compared to the uninoculated pile. Additionally, the compost pile inoculated with microorganisms showed less nitrogen loss compared to the control pile. These results suggest that inoculation can mitigate the emission of polluting gases such as nitrous oxide (N₂O) inherent to composting.

References

1. Gómez-Brandon, M., Lazcano, C., & Domínguez, J. (2008b). The evaluation of stability and maturity during the composting of cattle manure. *Chemosphere*, 70(3), 436-444.
2. Hurtado, J. (2014). Evaluación del Efecto de Microorganismos Transformadores de Materia Orgánica (TMO) en el Proceso de compostaje de las Deyecciones de Bovinos, Porcinos y Conejos. [Título de magister, Universidad de Manizales.
3. Román, P., Martínez, M. M., & Pantoja, A. (2013). MANUAL DE COMPOSTAJE DEL AGRICULTOR. Oficina Regional de la FAO. <https://www.fao.org/3/i3388s/i3388S.pdf>

FUNCTIONAL CHARACTERIZATION OF HUB PROTEINS IN THE PHYTOHORMONE SIGNALING NETWORK IN *ARABIDOPSIS THALIANA*

Andrea Duarte Elías¹, Cinthya Paola Rangel-Chávez¹, Humberto Herrera-Ubaldo³, Paola Isabel Angulo Bejarano²

¹Departamento de Bioquímica. Instituto Tecnológico Superior de Irapuato, Irapuato 36 821, Guanajuato México. ²Departamento de Agronomía. Tecnológico de Monterrey campus Querétaro, Epignemio González 500, San Pablo Santiago de Querétaro, Qro. ³Department of Plant Sciences. University of Cambridge, Downing St., Cambridge CB2 3EA, United Kingdom.
E-mail: pangulobe@tec.mx

Phytohormones act at the site of synthesis or in different areas where they regulate specific physiological events. To accommodate the variety of metabolic signals involved in development, there are signaling pathways where phytohormones interact with each other through crosstalk, rendering the existence of a linear representation obsolete. Hub proteins, or central proteins, have high connectivity in a protein-protein interaction (PPI) network, with many interaction partners, connecting different network modules. Hormonal responses in plants involve sensory mechanisms, and PPIs regulate these processes.

Previous studies report the identification of nineteen hub proteins with potentially relevant involvement in hormonal signaling pathways in *A. thaliana* (Alzati Ramírez, 2022). However, these proteins have not been functionally characterized, so their role in the interconnection of these pathways is unknown. Characterizing these proteins is important to advance the understanding of plant functioning in their development and to understand how these proteins are involved in signal perception or transduction.

Therefore, the present work aims to functionally characterize hub proteins in the hormone signaling pathways in *A. thaliana*. To achieve this, we reconstructed the PPI networks for each reported hub, considering only their interacting proteins involved in the signaling pathways of seven hormones (auxins, cytokinins, ABA, ethylene, jasmonate, gibberellins, and brassinosteroids) and co-expression interactions. This allowed us to identify two hub proteins related to two and three hormonal signaling pathways. Subsequently, we obtained co-expression maps of each hub protein with their interactors, which allowed us to identify the tissues in which each hub and their interactors are co-expressed. Based on this information, we have proposed *in vitro* assays in *A. thaliana*, where the effect of different hormones on the plant phenotype will be evaluated. Keywords: phytohormones, protein-protein interactions, phytohormone signaling pathways, crosstalk, hub, interconnection.

OBTAINING STARCH FROM *PACHYRHIZUS EROSUS* AS AN ALTERNATIVE TO CONVENTIONAL STARCHES

Jimena Carolina Enríquez Fonseca¹, Alfonso Jiménez Adón², Karla Lizbeth Macías Sánchez³,
Estefania Odemarís Juárez Hernández⁴

Unidad Profesional Interdisciplinaria de Ingeniería Campus Guanajuato Instituto Politécnico Nacional.
Silao de la Victoria Gto. México.

*E-mail: ejuarezh@ipn.mx

Starch is a significant polysaccharide in human history, as it has been a fundamental part of our diet and a versatile raw material. In the pharmaceutical industry, starch works as a dispersion agent and binding agent for active ingredients. In cosmetics, it acts as an absorbent, viscosity enhancer, and carrier for various substances. Starch can be extracted from diverse sources, including legumes, roots, tubers, cereals, and fruits. Among the conventional sources, corn, wheat, rice, sorghum, potatoes, yams, sweet potatoes, and sago are prominent. However, research on non-conventional starch sources has gained momentum. Exploring alternatives to conventional sources aims to discover starches with distinct physicochemical and functional properties. Sustainability, utilization of organic waste and byproducts, local availability, technological feasibility, and cultural significance are driving factors for investigating non-conventional starch sources. In this context, jicama (*Pachyrhizus erosus*) emerges as a potential non-conventional starch source. Native jicama starch was extracted and subsequently chemically modified to enhance its hydrophobicity. Positive results were obtained, with a yield of 2.22% from 4500 g of raw material (without peel), resulting in 100 g of native starch from *Pachyrhizus erosus*. The modified *P. erosus* starch exhibited acetyl group percentages and degrees of substitution of 0.1141 and 0.0043, respectively. Given that starch is a major constituent of roots, exploring jicama as a raw material opens new avenues for sustainable utilization. The viability of using *P. erosus* starch lies in its cost-effectiveness and positive market demand, contributing to the local economy.

ANTIOXIDANT ACTIVITY AND PHENOL CONTENT OF LIQUID-FERMENTATION OF *MORCHELLA ESCULENTA*

Martha Elena Espin-Reza¹, Maura Téllez-Téllez², Edgar Martínez-Fernández²,
Ma. de Lourdes Acosta Urdapilleta², Denis Castro-Bustos², Gerardo Díaz-Godínez³

¹Maestría en Manejo de Recursos Naturales, Centro de Investigaciones Biológicas, Universidad Autónoma del Estado de Morelos. Av. Universidad 1001, Chamilpa, 62209. Cuernavaca, Mor. ²Centro de Investigaciones Biológicas, Universidad Autónoma del Estado de Morelos. Av. Universidad 1001, Chamilpa, 62209. Cuernavaca, Mor.

³Centro de Investigación en Ciencias Biológicas, Universidad Autónoma de Tlaxcala. Av. Universidad Núm. 1, Col. La Loma Xicohténcatl, 90070. Tlaxcala, Tlax.

E-mail: maura.tellez@uaem.mx

Morchella spp. (morels) is an Ascomycota that includes an important group of mushrooms that are not very abundant in their natural environment; for centuries, it has been commercialized due to its flavor and nutritional value. However, it has been reported that these mushrooms are valuable not only for their nutritional content but also for their antioxidant activity, which gives them various medicinal qualities (Li *et. al.*, 2023). Furthermore, we can find these compounds not only in the fruiting body but also in the mycelium and culture broth (Yan *et. al.*, 2023). In the present work, the antioxidant activity and content of phenolic compounds of the culture broth of *Morchella esculenta* were quantified. It was grown in a medium with glucose and mineral salts, and incubated at 25 °C/130 rpm for 20 days; the antioxidant activity was determined: 2,2'-azino-bis-3-ethylbenzothiazolin-6-sulfonic acid (ABTS) and 1,1-diphenyl-2-picryl-hydrazyl (DPPH) and the content of total phenols by the Folin-Ciocalteu technique. The culture broth of *M. esculenta* presented a percentage of inhibition of the ABTS radical of 78.92% (360 h) and 82.72% (312 h) for the DPPH radical; the highest content of total phenols was 0.043 mg AGE/mL (360 h) and 3.36 g/L of biomass was obtained. In the present work, it is concluded that the culture broth of *M. esculenta* presents antioxidant activity similar to that which has been reported in other works, in which the fruiting body and mycelium of *Morchella* spp. were used, so the culture broth can be a viable alternative for obtaining compounds of medical importance, in addition, the cultivation of *Morchella* can be optimized to improve antioxidant activity.

References

1. Yan MQ, Feng J, Liu YF, Hu DM and Zhang JS. (2023). *Foods*, 12, 2086.
2. Li Y, Chen H and Zhang X (2023). *Frontiers Nutrition*, 10, 1159029.

INDUCTION OF THE TANNASE SYSTEM IN MARINE FUNGI FOR PET (POLYETHYLENE TEREPHTHALATE) DEGRADATION

Jossie Fernanda Espinoza Ávila; Noemí Marchena Echeverría;
Alfredo Herrera Estrella; Edgar Balcázar López

Universidad de Guadalajara. Centro Universitario de Ciencias Exactas e Ingenierías.
Blvd. Gral. Marcelino García Barragán 1421, Olímpica, 44430 Guadalajara, Jal.

Responsible author: Edgar Balcázar López; edgar.balcazar@academicos.udg.mx, 331706565646.

Given the significant environmental damage caused by various forms of pollution, including the overuse of PET (polyethylene terephthalate), this study delves into a potential solution. We explored using natural inducers in marine fungi to stimulate enzymatic systems capable of breaking ester bonds and degrading aromatic compounds commonly found in PET. This research is crucial in our ongoing efforts to combat environmental degradation.

Several enzymes of bacterial and fungal origin that can depolymerize and degrade PET have been reported in the literature. These enzymes are PETases, MHETases, Cutinases, Esterases, and Lipases. On the other hand, bacterial tannases with this same capacity have been reported. Tannases are enzymes capable of degrading tannins present in plant biomass. Given their nature of interacting with plants, fungi present a wide set of enzymes that are part of the feruloyl esterase family. The organic compound selected as an inducer for this project is a tannin present in plants as a protective barrier, and some enzymes in different microorganisms can degrade it. It also has a chemical structure similar to that of PET, which is the main reason to deduce that the mentioned enzymes can realize the degradation of both compounds and, therefore, can serve as an enzymatic inducer.

In our recent investigations, we made a significant discovery. We found a tannase of *Trichoderma asperellum* that bears a striking structural similarity to the MHETase of *Ideonella sakaiensis*, one of the most extensively studied enzymes in PET degradation. Moreover, there is evidence that *Trichoderma asperellum* can thrive on PET as its sole carbon source. These findings are novel and hold great promise for PET degradation.

During the study of five fungal strains, which were isolated from a plastic bag in an Estero in Mazatlan, Sinaloa, when the organic compound was added to them at a minimum concentration (1 mg/mL) in a medium with PET as the only carbon source, we observed a considerable increase in biomass (approximately 20 times) in two strains (*Aspergillus flavus* and strain HM2 in the process of genetic identification) compared to those in which this compound was not added.

In preliminary studies of *Aspergillus flavus*, we identified eleven tannases in its genome, four of which have a structural identity with the MHETase of *Ideonella sakaiensis*. These results suggest that the tannases in *Aspergillus flavus* may act on PET, allowing the fungus to utilize or bioassimilate this substrate as a carbon source. To corroborate this hypothesis, we are working with the heterologous expression of these enzymes in recombinant systems.

INSIGHTS IN THE ANTIBACTERIAL STRUCTURE-FUNCTION RELATIONSHIP OF THE PEPPER DEFENSIN J1-1

Estrada Georgina, Muñoz-Pérez Gilberto Andrés, Sánchez-Cach Lucila A., Guillén-Chablé Francisco A., Gerardo Corzo

¹Centro de Investigación Científica de Yucatán A. C., ²Institute of Biotechnology UNAM-Cuernavaca
Centro de Investigación Científica de Yucatán. Unidad de Biología Integrativa. Calle 43 No. 130. Col. Chuburná de Hidalgo,
CP 97200, Mérida, Yucatán, México. correspondencia: Georgina Estrada Tapia.
E-mail: georgina.estrada@cicy.mx

Plant defensins have been extensively characterized as antifungal peptides expressed in response to pathogen infection. In recent years some plant defensins have been characterized as antibacterial peptides *in vitro*. In this work we have studied the effects in structure and selectivity of a mutation K45E in the recombinant defensin J1-1 from Capsicum genus. The mutant J1-1_K45E was expressed in a previously reported bacterial expression system and purified in the same conditions as the recombinant parental defensin J1-1[1]. Comparative oligomerization profiles were observed in the presence and absence of their ligand lipid, phosphatidic acid (PA). In terms of activity of the mutant, two main differences are noticeable: it showed a gain in activity against *Staphylococcus aureus* and additionally to PA it binds to phosphatidyl serine lipid ligand. In conclusion, our work provides new insights into structure-function properties of plant defensin J1-1 that might be important to improve defensin properties against antibiotic resistant bacteria [2].

References

1. Guillén-Chablé, F., Arenas-Sosa, I., Islas-Flores, I., Corzo, G., Martínez-Liu, C., & Estrada, G. (2017). Antibacterial activity and phospholipid recognition of the recombinant defensin J1-1 from Capsicum genus. *Protein Expression and Purification*, 136, 45–51. <https://doi.org/10.1016/j.pep.2017.06.007>
2. Muñoz-Pérez Gilberto Andrés. Tesis doctoral (2024). repositorio CICY

OVERPRODUCTION OF MEDICALLY AND BIOTECHNOLOGICALLY RELEVANT PHENAZINES USING A MUTANT IN RSM A OF PSEUDOMONAS AERUGINOSA ID4365 WITH ATTENUATED VIRULENCE

¹Misael Josafat Fabian del Olmo, ¹Abigail González Valdez, ²Luis Fernando Montelongo-Martínez, ²Adelfo Escalante Lozada, ²Gloria Soberón-Chávez and ¹Miguel Cocotl Yañez

¹Departamento de Microbiología y Parasitología, Facultad de Medicina, Universidad Nacional Autónoma de México, C.P. 04510. ²Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, C.P. 04510.
E-mail: misaelfabian8@gmail.com

Phenazines are heterocyclic organic compounds produced by bacteria, characterized by properties such as redox potential and acid-base behavior. These compounds possess a range of biological activities, allowing them to be utilized as antimicrobials, pesticides, and antitumor agents. Enhancing phenazine production is therefore significant, and genetic manipulation of bacteria presents a promising strategy.

Pseudomonas aeruginosa, an opportunistic pathogen, holds significant biotechnological potential for phenazine production. This bacterium synthesizes four primary phenazines: phenazine-1-carboxylic acid (PCA), phenazine-1-carboxamide (PCN), 1-hydroxyphenazine (1-OHPHZ), and pyocyanin (PYO). Their biosynthesis involves two operons, *phzABCDE1* and *phzABCDE2*, which convert chorismic acid into PCA. The enzymes PhzS, PhzH, and PhzM further modify PCA to produce 1-OHPHZ, PCN, and PYO¹.

The *P. aeruginosa* strain ID4365, an environmental isolate from the Indian Ocean, overproduces pyocyanin compared to the reference strain PAO1. Additionally, the *rsmA* mutant strain (IDrsmA) shows a five-fold increase in pyocyanin production relative to the ID4365 strain². In this study, our goal was to use the IDrsmA strain to achieve overproduction of different phenazines. We found that *rsmA* inactivation inhibited the type III secretion system, and abolished cytotoxicity and pathogenicity in the *Galleria mellonella* model. Furthermore, we generated several mutations that boosted PCA production. Moreover, our results revealed a potential dependency of PhzM for the proper functionality of PhzS, which could, in the future, lead to the overproduction of 1-OH-PHZ. Consequently, IDrsmA strain shows reduced virulence, making it a safer candidate to produce pyocyanin and PCA.

References

1. Mavrodi DVBonsall RF, Delaney SM, Soule MJ, Phillips G, Thomashow LS2001.Functional Analysis of Genes for Biosynthesis of Pyocyanin and Phenazine-1-Carboxamide from *Pseudomonas aeruginosa* PAO1. *J Bacteriol*183
2. Cocotl-Yañez M, Soto-Aceves MP, González-Valdez A, Servín-González L, Soberón-Chávez G. Virulence factors regulation by the quorum-sensing and Rsm systems in the marine strain *Pseudomonas aeruginosa* ID4365, a natural mutant in *lasR*. *FEMS Microbiol Lett.* 2020 Jun 1;367.

IDENTIFICATION OF AN ANTIGENIC PROTEIN FROM *HELICOBACTER PYLORI* THROUGH PREDICTION OF T AND B EPITOPES AND PRODUCTION OF THE RECOMBINANT PROTEIN IN *ESCHERICHIA COLI*

Brenda Yarely Fernández Madrid¹, Juan Antonio Rojas Contreras¹, Jesús Páez Lerma¹,
Alma Karina Tamez Castrellón¹, Norma Urtiz Estrada²

¹Tecnológico Nacional de México/Instituto Tecnológico de Durango, Blvd. Felipe Pescador 1830 Ote. Col. Nueva Vizcaya, 34080 Durango, Dgo., México. ²Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango, Av. Veterinaria S/N, Col. Valle del Sur, 34120 Durango, Dgo., México.
E-mail: jrojas@itdurango.edu.mx

Helicobacter pylori is a spiral-shaped Gram-negative bacteria that is estimated to affect more than half of the world's population, in Mexico the reported seroprevalence is 66%.^{1,2} This microorganism has been associated with various gastric disorders that range from gastritis to cancer, of which approximately 37% of cases come from chronic infections due to *H. pylori*, which is why it is recognized by the World Health Organization as a class I carcinogen.^{3,4} The reason that this microorganism presents a high pathogenicity is due to the fact that it has multiple virulence factors and currently the appearance of multi-drug resistant strains have led to the failure of therapeutic regimens, placing its eradication below 80%.¹ The objective of this project is the Identification and production of an antigenic protein from the enteropathogenic bacteria *H. pylori* in an expression system for *Escherichia coli*. First, the identification of proteins and immunogenic epitopes was carried out *in silico* by searching for protein sequences in the reference strain *H. pylori* ATCC 26695 through the PSROTdb server, from which only proteins with cytoplasmic subcellular localization associated with virulence factors were chosen. These proteins were subjected to antigenicity evaluation using the Vaxijen 2.0 server (cutoff point of 0.4) and to the discarding of homology with human proteins using the NCBI BLAST tool. The accessory protein *UreE* was obtained as a selected candidate, with an antigenicity of 0.5011 and no homology with human proteins. This accessory protein is an essential metallochaperone enzyme involved in the maturation process of the urease enzyme produced by *H. pylori* as a virulence factor. Subsequently their main antigenic epitopes in the amino acid sequence were determined. For the B cells, the ABC-pred program was used, obtaining a peptide with an antigenicity value of 1.2691 given by the Vaxijen 2.0 server. For T cell epitopes prediction, the IEDB server was used, in which peptides for MHC class I and MHC class II were obtained, with antigenicity values of 2.053 and 1.5485 respectively. The tertiary structure of *UreE* was obtained, in this model was observed that the epitopes are located in external regions of the protein. The conclusion of this work is that the *UreE* protein presents epitopes with high antigenicity values and its structural location is favorable for immunogenic identification.

References

1. Katelaris, P., Hunt, R., Bazzoli, F., Cohen, H., Fock, K. M., Malfertheiner, P., Mégraud, F., Piscocoy, A., Quach, D., Vakil, N., Coelho, V., & LeMair, A. (2021). *H. pylori*. Directrices de la organización mundial de gastroenterología.
2. Ladrón de Guevara, L., Bornstein-Quevedo, L., González-Huezo, S., Castañeda-Romero, B., Costa, F. G., & di Silvio-López, M. (2019). Helicobacter pylori eradication in Mexico with a levofloxacin-based scheme versus standard triple therapy: Results from an open-label, randomized, noninferiority phase iiiib trial. Mexican Journal of Gastroenterology, 84(3), 274-283.
3. Bosques Padilla, F. J., J. M. Remes-Troche, M. S. González-Huezo, G. Pérez-Pérez, J. Torres-López, J. M. Abdo-Francis, M. V. Bielsa-Fernandez, M. C. Camargo, F. Esquivel-Ayanegui, E. Garza-González, A. I. Hernández-Guerrero, R. Herrera-Goepfert, F. M. Huerta-Iga, Y. Leal-Herrera, A. López-Colombo, N. X. Ortiz-Olvera, A. Riquelme-Pérez, C. L. Sampieri, L. F. Uscanga-Domínguez, y J. A. Velarde-Ruiz Velasco. 2018. «IV consenso mexicano sobre Helicobacter pylori.» Revista de Gastroenterología de México 83(3):325-41.
4. Malfertheiner, Peter, M. Constanza Camargo, Emad El-Omar, Jyh-Ming Liou, Richard Peek, Christian Schulz, Stella I. Smith, y Sebastian Suerbaum. 2023a. «Helicobacter Pylori Infection». Nature Reviews Disease Primers 9(1):19.

COLORIMETRIC EVALUATION AND PIGMENT CONTENT IN LEAVES OF DIFFERENT VARIETIES OF *A. HYPOCHONDRIACUS*, *A. HYBRIDUS* AND *A. CAUDATUS* IN VEGETATIVE STAGE

Guadalupe Abigail Flores Pérez, Vladimir Flores Benavides, Flores Benavides-Flor,
Flor de Fátima Rosas Cárdenas

Instituto Politécnico Nacional- Centro de Investigación en Biotecnología Aplicada
CP. 90700, Tel. 2461137296, Correo electrónico:gfloresp2300@alumno.ipn.mx

Amaranth is a dicotyledonous plant of the Amaranthaceae family, rich in nutrients, minerals and fiber that contribute to human health. Thanks to its phenotypic variability, it has a wide spectrum of attractive colors ranging from yellows, violets to intense reds, positioning it in the culinary and ornamental fields. In the specific case of food, color is a relevant attribute that allows us to recognize its quality and give us a perception of its flavor. This attribute is given naturally by the presence of pigments in the plant, varying its concentration according to the stage of development in which it is found. Characterization of amaranth color is important because it provides valuable information on crop management, it is indicative of the health and vigor of the plant, and it can be useful data for genetic improvement focused on the optimization of desired characteristics of amaranth plants. The objective of this work was to characterize the color of 15 amaranth varieties in vegetative stage of the species *A. hypochondriacus*, *A. hybridus* and *A. caudatus* with specific values within the CIE L* a* b* space and to quantify their chlorophyll and betalain content through spectrophotometry, providing an integral evaluation of the color and pigments present in the amaranth leaves of each variety. A portable colorimeter CHN Spec CS-10 was used for color measurement, with which measurements were taken at three different points on both the upper and lower sides of the amaranth leaves. Chlorophyll and betalains were quantified using extracts of 0.02 g of fresh plant tissue in 80% acetone and water at pH=5 (adjusted with 0.1M HCl) respectively. Spectrophotometric readings for chlorophyll were performed at wavelengths of 663 and 646 nm and for betalains at 538 and 480 nm, all tests were performed in triplicate. As a result the concentration of total chlorophyll was found in a range of 10 to 18 mg/g being Ab1 the one that obtained the highest values of total chlorophyll, *a* and *b* and at the same time adopting a color angle of 125.8°. The concentration of betacyanins obtained from the possible varieties ranged from 0.2 to 2.3 g/mg, being Ap5 and Ab4 the varieties with the highest concentration and showing a color angle of 344.8° and 344.5° respectively.

FINDING NOVEL ALKYL GLUCOSIDE-PRODUCING AND METHANOL-TOLERANT VARIANTS FROM AN AMYA ALPHA-AMYLASE LIBRARY

Kimberly Flores-Rivera¹, Josahandy Jiménez-Peralta¹, Wendy Xolalpa-Villanueva¹, Leticia Olvera-Rodríguez¹, Humberto Flores-Soto¹ and Gloria Saab-Rincón¹

¹Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, UNAM, Cuernavaca Morelos, 62210.
E-mail: kfloresr09@gmail.com

Alpha amylases are enzymes of great industrial importance hydrolysing the α 1-4 glycosidic linkage of starch to form glucose, maltose and maltotriose units as end products. Amylase from *Thermotoga maritima* AmyA can also catalyse transfer reactions using other carbohydrates as acceptor molecules (transglycosylation) or other molecules such as primary alcohols (alcoholysis). In the latter case, the final product obtained are alkyl glycosides, which have surfactant properties with important applications in the cosmetic, food, pharmaceutical and other industries. In the laboratory, variants of the AmyA gene of *T. maritima* have been obtained by error-prone PCR. To find a more alcoholytic variant and because there are approximately 2000 variants to evaluate, a 96-well plate method based on fluorescence with 1-Anilinonaphthalene-8-sulphonic acid (ANS) that binds to the hydrophobic regions of the micelles that form the alkyl glucosides has been developed. This method was applied to improve the alcoholytic properties of AmyA by directed evolution. On the other hand, the same variant library was screened to identify hydrolytic variants with increased tolerance to 40% and 50% methanol concentrations. *E. coli* C41 (DE3) cells were transformed with the variant library, culture and induction with IPTG were performed in 96-well plates, cells were heat lysed at 70°C. The alcoholysis reaction was carried out in the presence of 10% butanol or hexanol and starch. The production of alkyl glucosides from the improved variants was tested by thin layer chromatography. To evaluate the hydrolytic activity of the variants in the presence of methanol, the 3,5-dinitrosalicylic acid (DNS) reducing sugars method was used. So far, 2 variants with higher alcoholysis activity compared to the wild-type enzyme and 4 variants with higher tolerance to 40% methanol have been identified.

References

1. Damián-Almazo JY, Moreno A, López-Munguía A, Soberón X, González-Muñoz F, Saab-Rincón G (2008). *Appl Environ Microbiol*, 74 (16):5168-77.
2. Slavík, Jan (1982). *Biochim Biophys Acta*, 694 (1):1-25.

CELLULOLYTIC ACTIVITY OPTIMIZATION OF ENZYMATIC EXTRACTS FROM A NATIVE STRAIN OF *BACILLUS SUBTILIS* ISOLATED FROM *AGAVE DURANGENSIS* BAGASSE

Miguel Antonio Franco-Vázquez¹, Gerardo Alfonso Anguiano-Vega¹, Carlos Antonio Alba-Fierro¹,
Maria Guadalupe Nieto-Pescador¹, Francisco Javier Rios-Fránquez^{1*}

¹Facultad de Ciencias Químicas Durango, Universidad Juárez del Estado de Durango Av. Veterinaria s/n Circuito Universitario
Col. Valle del sur C.P. 34120. Durango, Dgo., México.

*E-mail: javier.rios@ujed.mx

Cellulases are enzymes widely used in industry, which are produced in nature mainly by microorganisms such as fungi and bacteria, which use them to release the monosaccharide contents in plant. In the state of Durango Mexico, *Agave durangensis* bagasse is an abundant and underused agroindustrial waste, which through enzymatic hydrolysis could be transformed into fermentable sugars to obtain value-added products. In our research group, a strain of *Bacillus subtilis* native from agave bagasse, was isolated and identified by biochemical and molecular techniques. Through cultures in a stirred tank bioreactor in a medium containing carboxymethylcellulose as the only carbon source, cellulolytic extracts were obtained from the culture supernatants. In 12-hour kinetics of the *B. subtilis* strain in mineral medium with chemically pretreated bagasse with 2% NaOH as a substrate, it has been determined that the maximum point of cellulase production is reached after 8 h of culture. To optimize the conditions under which the enzymatic activity is quantified, and using the enzymatic extracts obtained in the bioreactor, tests were performed varying the temperature, incubation time, type of buffer and pH values. These parameters establish the bases to carry out enzymatic degradation tests of *Agave durangensis* bagasse to obtain fermentable sugars.

SYNTHESIS OF GOLD NANOPARTICLES USING A LIPID-RICH EXTRACT FROM MEXICAN AVOCADO SEED AND EVALUATION OF ITS CYTOTOXICITY ON MURINE MELANOMA CELLS

Minerva Frutis-Murillo^{1,2}, Alejandra Ochoa-Zarzosa², G. Rosas¹ and Joel Edmundo López-Meza²

¹Instituto de Investigación en Metalurgia y Materiales, Universidad Michoacana de San Nicolás de Hidalgo, Ciudad Universitaria, Morelia, Mich., México. ²Centro Multidisciplinario de Estudios en Biotecnología-FMVZ, Universidad Michoacana de San Nicolás de Hidalgo, Tarímbaro, Mich., CP. 58893, México.
E-mail: 1505595k@umich.mx

Gold nanoparticles (AuNps) exhibit attractive properties due to their size-volume ratio, such as thermal stability, biocompatibility, low toxicity, and optical properties. AuNps can be synthesized by green methods using medicinal plant extracts rich in reducing and stabilizing agents to convert Au³⁺ ions to Au⁰. Green synthesis with extract plants has the advantage of coating the nanoparticles with plant biomolecules, which enhances their medicinal effects¹. This work combined a lipid-rich extract from Mexican avocado seed (LEAS) (*Persea americana* var. *drymifolia*)², whose chemical composition is long-chain fatty acids and acetogenins, with tetrachloric auric acid precursor salt solution to synthesize AuNps. The nanoparticles were characterized by UV-vis microscopy, showing a characteristic surface plasmon peak at 542 nm. By XRD pattern, an FCC cubic crystalline structure was determined, observing intense peaks located at 38.1°, 44.2°, 64.7°, and 77.6° which agree with the Miller indices (111), (200), (220), (311). Subsequently, by transmission electron microscopy, an average size of the AuNps (~10 nm) and a spherical morphology were determined. The cytotoxic effects analyzed by trypan blue assay of AuNps functionalized with LEAS (3-42.3 µg/ml) (AuNps: LEAS) were evaluated in murine melanoma cell line B16-FO for 24 h. The results showed that the AuNps: LEAS at 14.1 µg/ml decreased the cell viability to 34.49% compared with LEAS alone (14.1 µg/ml). Also, the death cell mechanism induced by AuNps:LEAS is being evaluated. In conclusion, the gold nanoparticles enhance LEAS cytotoxicity at low concentrations once they are functionalized.

References

1. H. R. Ghorbani. (2015). *Oriental Journal of Chemistry*, vol. 31, no. 1, pp. 303–305.
2. M. Báez-Magaña, A. Ochoa-Zarzosa, N. Alva-Murillo, R. Salgado-Garciglia, and J. E. López-Meza. (2019). *Journal of Immunology Research*, vol. 2019, pp 10.

CONSTRUCTION OF AN EXPRESSION VECTOR FOR THE SPSK_04019 GENE ENCODING AN HSP70 FROM *SPOROTHRIX SCHENCKII* AND EXPRESSION OF THE RHSP70 PROTEIN

Connie Gallegos-Rivera^{1,2}, Ana Lilia Martínez-Rocha³, Angélica López-Rodríguez¹, Iván Meneses-Morales¹, Maricela Aguilar-Duran², Estela Ruiz-Baca^{1*}

¹University Juárez of Durango State, Faculty of Chemistry Sciences. *Av. veterinaria s/n, Circuito Universitario, Col. Valles del Sur, C.P. 34120, Durango, Dgo. México. Tel: +52 6188271340, ²University Juárez of Durango State, Scientific Research Institute. ³ University of Guanajuato, Division of Natural and Exact Sciences, Department of Biology. *E-mail: eruiz@ujed.mx

Sporotrichosis is a subcutaneous mycosis caused by *Sporothrix* spp., a fungus whose initial point of contact with the host is the cell wall, where immune responses initiate through immune receptors [1]. In previous studies, we identified a 70 kDa heat shock protein (Hsp70) associated with the cell wall of *S. schenckii*, which modifies its expression in response to oxidative stress and could be involved in the adhesion mechanisms of the fungus [2]. Hsp70 is commonly considered a chaperone protein, but it has been shown to have potential as a virulence factor, motivating its study to understand its possible immunoprotective effect in sporotrichosis, an occupational mycosis [3]. The objective of this study was to clone the open reading frame (ORF) of the SPSK_04019 gene encoding Hsp70 from *S. schenckii* into the pJET1.2/blunt vector to subsequently subclone it into the pET28a expression vector. First, we synthesized cDNA by reverse transcription using total RNA from *S. schenckii*. Then, we amplified the ORF of Hsp70 by PCR and purified it. We cloned the purified product into the pJET1.2/blunt vector and then subcloned it into the pET28a vector. We confirmed the cloning and the subcloning products by sequencing them and using digestion with *Bam*HI and *Hind* III enzymes. Finally, the rHSP70 protein obtained from the construction of the pET28a vector was expressed by induction with isopropyl- β -D-1-thiogalactopyranoside (IPTG) in *E. coli* Rosetta (DE3), reaching an optical density of 0.5 Abs at a wavelength of 595 nm. This approach will allow further studies on the potential of Hsp70 as an immunoprotective agent in sporotrichosis.

References

1. Carrada-Bravo, T. (2012). Revista Latinoamericana de Patología Clínica, 59, 147-171.
2. Saucedo-Campa, D. O., Martínez-Rocha, A. L., Ríos-Castro, E., Alba-Fierro, C. A., Escobedo-Bretado, M. A., Cuéllar-Cruz, M., & Ruiz-Baca, E. (2022). Pathogens, 11(2).
3. Wang, J., Chen, J., Hu, Y., Ying, S.H., & Feng, M.G. (2020). Fungal Genetics and Biology, 144, 103437.

EVALUATION OF THE EFFECT OF ZINC OXIDE NANOPARTICLES ON JALAPEÑO PEPPER (*CAPSICUM ANNUUM*) CROP. BIOACCUMULATION, METABOLIC PROFILE AND BACTERIOME

Luis Alberto García Casillas¹, Gilberto Velázquez Juárez¹, Sandra Fabiola Velasco Ramírez¹

¹Department of Chemistry, Centro Universitario de Ciencias Exactas e Ingenierías, Universidad de Guadalajara, 1421 Marcelino García Barragán Blvd, Guadalajara, Jalisco.
E-mail: luis.garcia3988@alumnos.udg.mx

In the last decade, the production and application of metal nanoparticles have experienced exponential growth. Among the various nanoparticles, zinc oxide nanoparticles (ZnONPs) stand out for their wide use in disinfection, medicine, cosmetics, and catalysis. In 2020, global production of ZnONPs reached 58 thousand tons, placing them third in nanomaterial production worldwide. However, their growing production poses significant environmental challenges.

Agriculture, particularly crucial crops such as chili (*Capsicum annuum*), emerges as an essential field of study to understand the effects of ZnONPs in agricultural systems. Mexico, recognized as the point of origin of chili peppers, has a particular interest in this crop, both economically and culturally.

Most studies focus on evaluating how ZnONPs affect plants by measuring parameters such as germination, root growth, and enzyme activities linked to oxidative stress. However, less attention is paid to analyzing effects on secondary metabolites and the crop microbiome.

This work focuses on investigating the impact of zinc oxide nanoparticles on the jalapeño pepper crop as a model, addressing aspects such as zinc bioaccumulation in plant tissues, alterations in the production of secondary metabolites through non-targeted analysis, and the bacterial composition of the crop's phyllosphere and rhizosphere.

IDENTIFICATION AND ISOLATION OF STRAINS OF AGRONOMIC INTEREST IN AGRICULTURAL SOIL WITH CLAY TEXTURE

Jesús García Pereyra, Mónica García Montelongo, Sebastián E. González Villarreal, Ana María García Montelongo, Gabriel N. Aviña Martínez, Rosa Bertha Rubio Graciano

Tecnológico Nacional de México
jpereyra5@otmail.com

Summary. The purpose of this study is to identify different types of fungal organisms present in an agricultural soil with a clay texture within the agricultural area of the Guadiana Valley Technological Institute of Durango. A ground trapping process was conducted to capture fungi. For this, wet precooked rice was used as bait, the fungi were isolated and identified by color and growth form. The fungi were sown on potato dextrose agar with 2% lactic acid and a streptomycin-based antibiotic. Subsequently, an antibiosis was carried out employing a biological fungicide based on *Trichoderma harzianum*, as a growth inhibitor. The results indicate the presence in soil of *Trichoderma harzianum*, *Fusarium*, *oxisporum* *Metarhizium anisopliae*, *Aspergillus nigger* and *Glomus intraradices*. Bean seeds of the pinto Saltillo variety were placed with different doses of inoculation of *Trichoderma harzianum* and *Glomus intraradices* in a bioclimatic chamber in a controlled environment to observe the degree of root growth. The results indicate an inhibition of fungal growth in antibiosis using *Trichoderma harzianum* of 75%, both in germination and root growth *Glomus intraradices* surpassed *Trichoderma harzianum* by up to 40%.

References

1. Tapia, C. V., & Amaro, J. (2014). Género *Fusarium*. *Revista Chilena De Infectología*, 31(1), 85- 86. <https://doi.org/10.4067/s0716-10182014000100012>
2. Grageda-Cabrera, O. A., Díaz-Franco, A., Peña-Cabriales, J. J. y Vera-Nuñez, J. A. (2012). Impacto de los biofertilizantes en la agricultura. *Revista mexicana de ciencias agrícolas*, 3(6), 1261-1274. https://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S2007-09342012000600015.

GREEN SYNTHESIS OF SILVER NANOPARTICLES AND ITS EFFECTS ON PLANTS

Cristina Garcidueñas Piña, José Francisco Morales Domínguez, Sadoc Gamaliel Pérez Medina

Universidad Autónoma de Aguascalientes
Av. Universidad 940, Ciudad Universitaria, C.P. 20100, Aguascalientes.
Teléfono: 4499108420. Correo: cristina.garciduenas@edu.uaa.mx

Nanoparticles (NPs) are a type of material that has dimensions between 1 and 100 nm. Metallic NPs, especially silver NPs (AgNPs) have attracted attention for their variety of applications. Currently, they are used in areas as diverse as optoelectronics, catalysis, medicine, textiles, food, agriculture and as antimicrobials, among others. Studies have shown that AgNPs have beneficial effects on plant development. Therefore, the possibility of applying them to plants of agricultural interest in order to enhance their development has been raised. However, there are also reports in which the toxicity of AgNPs has been demonstrated. AgNPs can be obtained by chemical, physical and biological processes. Among the biological processes is the so-called green synthesis, in which the silver of a salt is reduced with the help of compounds present in the extract of plants or microorganisms, to produce AgNPs. This technique is less polluting than chemical synthesis and, in addition, the resulting NPs have a coating of organic compounds from the plant, which provide stability in suspension. In order to evaluate the benefit of AgNPs on plant development, in this work they were obtained by green synthesis, using aqueous extracts of leaves and fruit of *Psidium guajava* (guava), a species that is widely cultivated in the state of Aguascalientes. The obtained AgNPs showed different physical and chemical characteristics depending on the extract. However, no significant differences were found in the effects produced in plants between those exposed to AgNPs obtained with leaf or fruit extracts. The effect on the development of plants such as *Arabidopsis thaliana*, *Solanum lycopersicum* (tomato), *Raphanus sativus* (radish) and guava, exposed to these AgNPs, was analyzed. AgNPs were found to produce positive effects on germination at low concentrations (0.1, 0.01 and 0.001 mg/mL), while at higher concentrations (1 mg/mL) a lower percentage of germination or a delay was observed. At low concentrations of AgNPs, greater plant elongation was also observed than in the control group.

EFFECT OF DISSOLVED OXYGEN IN THE GROWTH OF A RECOMBINANT GLYCOPROTEIN-PRODUCING *PICHIA PASTORIS*

Rogelio D. Gaytan-Castro^{1,2}, Norma A. Valdez-Cruz¹, Mauricio A. Trujillo-Roldán¹

¹Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, Cd. Universitaria, Coyoacán, Ciudad de Mexico, México. AP. 70228, CP. 04510, México, D.F, Mexico. ²Posgrado en Ciencias Biológicas, Universidad Nacional Autónoma de México.
E-mail: maurotru@iibiomedicas.unam.mx

Pichia pastoris is a yeast capable of using methanol as a carbon and energy source ¹ and widely used to produce large quantities² of recombinant proteins. *P. pastoris* cultures are highly demandant of oxygen³ which must be constantly added ⁴. To maintain a constant value of dissolved oxygen, the specific rate of consumption (OUR) and the rate of transfer must be equal (OTR).

The yeast was grown in three controlled different conditions of dissolved oxygen tension (10%, 40% and 70%). To achieve this, duplicate batch reactors (0.8-1L) containing BMMY media were inoculated with the recombinant strain. pH control was set to 6 and temperature to 30°C. Dissolved oxygen tension was controlled manually with the addition of nitrogen and oxygen gas according to deviations from the setpoint. Agitation was set constant during the fermentation. The working strain was kinetically and stoichiometrically characterized. Samples were taken each 2.5 h, and optical density (600 nm) was measured.

Final biomass showed no difference between conditions. Natural logarithm plots have two slopes, the first between 0 and 12 h and the second for the last part of the culture. The specific growth rate has no differences between 10% and 40% in both cases. However, 70% is different. Thus, oxygen concentration affects kinetically but not stoichiometrically the growth of *P. pastoris*. Further protein characterization is recommended to assess if a similar trend is observed as in the case of growth.

Acknowledgments. RDG-C is a graduate student from Programa de Maestría en Ciencias Biológicas, UNAM, and received a fellowship from “Consejo Nacional de Humanidades, Ciencias y Tecnologías” CONAHCYT (CVU 869630). Project from Consejo Nacional de Humanidades, Ciencias y Tecnologías (CONAHCYT, CF-2023-I-1248 and CF-2023-I-1549).

References

1. David R Higgins y James M Cregg. (1998) *Pichia* protocols, 1-15.
2. Pan, Y., Yang, J., Wu, J., Yang, L., y Fang, H. (2022). *Frontiers in Microbiology*, **13**, 1059777.
3. Richard AJ Darby, Stephanie P Cartwright, Marvin V Dilworth, and Roslyn M Bill. (2012). Recombinant protein production in yeast: methods and protocols, 11-23
4. Felix Garcia-Ochoa y Emilio Gomez. (2009) *Biotechnology Advances* 27.2, 153-176

EXPRESSION OF MEMBRANE RECEPTORS IN CELL-FREE SYSTEMS FROM *ESCHERICHIA COLI* AND THEIR USE IN THE GENERATION OF BIOSENSORS

Karen Stephania González Ponce¹, Luz Edith Casados Vázquez¹, José Eleazar Barboza Corona¹

¹Division of Life Sciences (DICIVA), Irapuato-Salamanca Campus. University of Guanajuato. Ex Hacienda El Copal km. 9, Irapuato-Silao highway. Irapuato, Guanajuato. C.P. 36500. Mexico.
E-mail: edith.casados@ugto.mx

Biosensors based on cell-free extracts are detection systems that use the cellular machinery without the need for a living cell. For the generation of this type of biosensors it is required that the cell from which the components will be obtained, expresses the bioreceptor that will detect the analyte¹. The detection of pathogens is one of the applications that has generated interest in the development of biosensors. In particular, the detection of one of the main pathogens in the food industry, such as *Listeria monocytogenes*, establishes the need to generate tools capable of reducing the prevalence of food infections caused by this bacterium². The present work used the agr censoring system of *L. monocytogenes*, which consists of four genes: *agrACBD*, the first two (*agrA* and *agrC*) must be expressed in the cell that will generate the extracts (*Escherichia coli*). For this purpose, a biological circuit was generated with the *agrCA* genes, within a regulated system under the control of the T7 promoter (DHRF vector). Once the DHRF/*agrCA* loop was obtained, it was transformed into the BL21 C41pRARE strain and then the conditions for expression of the recombinant proteins were standardized, for this purpose *E. coli* BL21 C41 pRARE transformed with the DHRF/*agrCA* loop was cultured at 37 °C/200 rpm to an OD₆₀₀ of 0.5, then the culture was incubated for 15 min at 4 °C, after the incubation period, IPTG was added to the cultures (0.1 mM, 0.25 mM and 0.5 mM, final concentration of the inducer), in addition to the different concentrations of inducer, different temperatures were also tested for induction (18, 28 and 37 °C). The results indicate that the DHRF/*agrCA* circuit transformed into BL21-C41pRARE is only expressed at 18 °C/200 rpm with a final IPTG concentration of 0.5 mM, the rest of the conditions tested show no expression of the DHRF/*agrCA* circuit.

References

1. Voyvodic P & Bonnet J. (2020). Current Opinion in Biomedical Engineering. Vol.13, 9-15.
2. Banerji R, Karkee A., Kanojiya P., Patil A., Saroj S. (2022). LWT-Food Science and Technology, Vol. 154, Page: 112703.

IN SILICO ANALYSIS OF THE C8 CLONE AND TOXIC ACTIVITY OF THE ATP-DEPENDENT RNA HELICASE FROM THE ENTOMOPATHOGENIC BACTERIUM *SERRATIA ENTOMOPHILA* MOR 4.1 IN PHYLLOPHAGA BLANCHARDI LARVAE (COLEOPTERA: MELOLONTHIDAE)

Luz Areli Gorostieta-Nava¹, Hugo Castelán Sánchez² y María Eugenia Núñez-Valdez³

Centro de Investigación en Dinámica Celular³, Universidad Autónoma del Estado de Morelos, Av. Universidad 1001³, Col. Chamilpa, C. P. 62209, Cuernavaca, Mor.; México.C. P. 62209.
eugenia@uaem.mx

In the state of Morelos, the entomopathogenic strain *Serratia entomophila* Mor 4.1 (SeMor4.1) was isolated, which presents toxic activity towards larvae of *Phyllophaga blanchardi* (Coleoptera: Melolonthidae) and *Spodoptera frugiperda* (Lepidoptera: Noctuidae). Five insecticide clones were isolated from the SeMor4.1 gene library expressed in *Escherichia coli*. Clone C8 represents a 38845 pair base fraction of the SeMor 4.1 genetic sequence. This work presents the *in-silico* analysis of the sequence and the observed toxic activity associated with one of its genes. The sequence contains 37 possible ORFs distributed in 19 transcription units of which 7 correspond to operons. The GC content within the C8 fraction is of 62%, distant to the reported GC content in the whole genome of 52%. Furthermore, the presence of diverse virulence-associated genes is observed. The information suggests that the sequence could be part of a *Serratia* genus exclusive pathogenicity island. To identify the genes associated with toxicity, C8 subclones were constructed. C8-F3 subclone presents toxic activity against *Phyllophaga sp* larvae and contains genes codifying for the glycosyltransferase (GT) of a domain of a penicillin binding protein 1b (PBP) and a RNA Helicase (RNA-HEL). The RNA-HEL toxic activity in insects was characterized by a genetic cloning strategy to isolate the gene in the cloning vector pBluescript SK+ (pSK; along with its respective promoter regions). From the C8-F3 clone and the pSK vector digested with the EcoRI and SmaI restriction enzymes, the construction pSK-HEL (5.6 Kbp) was obtained with which the *E. coli* Epi 300™ strain was transformed. The toxic activity of the pSK-HEL construct was evaluated by injection bioassays in *Phyllophaga sp* with bacteria-free culture supernatants. In a first bioassay, a maximum mortality of 100% was observed on day 7 after starting the bioassay with the injection of 50 µg of protein/larva of culture supernatant ($P < 0.0001$ compared to the H₂O control and the empty vector). In order to compare the individual and collective activity of PBP and RNA-HEL, the Lethal Dose 50 (LD50) of F3 (construct carrying RNA-HEL and GT-PBP) and pSK-HEL was determined. It was observed that on day 7 of the bioassay the LD50 of pSK-HEL was 18,661 µg/larva and the LD50 of C8-F3 was 20,159 µg/larva. The results indicate that the RNA-HEL protein exhibits toxic activity on the larvae of *P. blanchardi*.

Acknowledgement. We thank CONACYT for the support granted to carry out this work, scholarship number 930323 by Dr. Maria Eugenia Nuñez Valdez.

METZAL AS A TEXTURIZING AGENT IN THE DEVELOPMENT OF BARLEY (*HORDEUM VULGARE*)

Neftali Esteban Guzmán-Mendoza¹, Emiliano Osorio-Osorio¹, Victor Manuel Hodgkin-Bastard¹, Carlos Getzael Vazquez-Osorno¹, Josefa Espitia-López¹, Benito Flores-Chávez¹, Paul Misael Garza-López¹

¹Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo. Tulancingo, Hgo. 43600
Número: 7753610223 E-mail: gu476253@uaeh.edu.mx

Metzal is a residue derived from the agave pulquero (*Agave salmiana*), which has woody characteristics due to its lignocellulose content, making it an ideal substrate for the development of some crops (1). The inefficient disposal of waste is also reported, including metzal. In addition, various studies highlight the physicochemical contributions of lignocellulosic waste as texturizing agents in plant germination and development (2).

Barley seeds (*Hordeum vulgare*) were sown in 7.85 L bags, 5 treatments were carried out in different concentrations of metzal (0%, 5%, 10%, 20% and 30%) each with five repetitions. At 120 days after transplanting, the following variables were measured: fresh weight (kg), height (cm), germination (%), number of ears, pH, EC (mS/cm), and concentrations of Na⁺, Ca²⁺, NO³⁻ and K⁺ (ppm). A completely randomized design was used and the data were analyzed using ANOVA, with significant differences determined by Tukey's multiple range test ($\alpha=0.95$). Additionally, a principal component analysis (PCA) was performed to identify behavioral trends between the different treatments and variables (XLSTAT by Lumivero® 2023.1).

It was observed that the treatment with 10% metzal was the most effective, with measurements of fresh weight of 0.33 kg, height of 47.29 cm and number of 6 ears, being significantly higher than the other treatments ($P<0.05$). On the other hand, the rest of the variables did not show significant differences between the treatments ($P>0.05$). In the PCA, the data presented a reliability of 83.64%, identifying three large clusters that relate the variables EC and Ca²⁺ with the observations of the treatments at 5% and 0%, while the observations of the treatments at 20% and 30 % showed a greater relationship with the variable K⁺ and the rest of the observations are correlated with the rest of the variables.

Metzal had an influence on barley growth, since the observed trends indicate that metzal concentrations caused differences in barley development, with the treatment with 10% being the most effective.

References

1. Figueredo-Urbina, C. J., Medina-Pérez, G., Juárez-Muñoz, J., & Arce-Cervantes, O. (2023). *Mexican Journal of technology and Engineering*, 2, 10-20.
2. Reyes-Medina, A. J., Fraile-Robayo, D., & Álvarez-Herrera, J. G. (2019). *Revista Temas Agrarios*, 24, 34-41.

PLANT GROWTH-PROMOTING BACTERIA ENHANCES THE BIOMASS OF *AGASTACHE MEXICANA* SUBSP *MEXICANA* AND IMPROVES THE PRODUCTION OF HIGH-VALUE SECONDARY METABOLITES IN GREENHOUSE CONDITIONS

Héctor Daniel Hermenegildo Rosas¹, Gabriela Carmona Castro², José de Jesús Arellano García¹, Susana Valencia Díaz¹, Ramón Suarez Rodríguez¹, José Augusto Ramírez Trujillo¹, Samuel Enoch Estrada Soto³, Mario Rodríguez Monroy², María Guadalupe López Rodríguez⁴, Irene Perea Arango^{1*}

¹Centro de Investigación en Biotecnología, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México.

²Centro de Desarrollo de Productos Bióticos-IPN, Yauatepec, Morelos, México. ³Facultad de Farmacia, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México. ⁴Facultad de Ciencias Agropecuarias, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México.

E-mail: iperea@uaem.mx

Agastache mexicana subsp. *mexicana*, commonly known as toronjil morado (purple or red hyssop), is a medicinal plant belonging to the Lamiaceae family. It exhibits a range of pharmacological properties, including antioxidant, anxiolytic and vasorelaxant effects. These pharmacological properties derive from the chemical composition of this plant, which is a source of secondary metabolites, predominantly terpenes and flavonoids. Of this last group, tilianin (acacetin-7-O-glycoside) stands out, a molecule that has been shown to possess pharmacological activities, including neuroprotective, anti-inflammatory, antidiabetic, antihyperlipidemic, cardioprotective, pro-apoptotic, and others. As research into the biological and pharmacological functions of this and other metabolites obtained from this medicinal plant advances, it becomes increasingly clear that improvements must be made to the production of biomass and the content of secondary metabolites. The yield of these bioactive compounds is limited, with yields of up to 1% of the dry weight of plant extracts from this medicinal specimen being typical. The use of sustainable, beneficial, and environmentally friendly biotechnological tools facilitates the resolution of this problem. In this research project, the beneficial relationships between bacteria and plants are being investigated. The bacterial strains used are *Azospirillum brasilense*, *Azospirillum baldaniorum* and *Curtobacterium* sp. (isolated strain of *Selaginella nothohybrida*). These bacteria can solubilize inorganic phosphate compounds and produce phytohormones, which has the potential to reduce the demand for phosphate fertilizers while promoting plant growth and productivity. These microorganisms are beneficial to toronjil plants grown in greenhouses, promoting plant growth. Additionally, the levels of bioactive compounds were influenced by biotic stress, resulting in quantitative modifications as determined by phytochemical analyses (HPTLC, phenolic compounds).

EXPRESSION OF THE RECOMBINANT BODEF1: A CLASS I DEFENSIN FROM *BIXA ORELLANA*

Hernández-Díaz Raquel Fabiola¹, Sánchez-Cach Lucila A.¹,
Rivera Madrid Renata¹, Estrada Georgina¹

¹Centro de Investigación Científica de Yucatán. Unidad de Biología Integrativa. Calle 43 No. 130. Col. Chuburná de Hidalgo, CP 97200, Mérida, Yucatán, México. ²Universidad de Colima. Laboratorio de Agrobiotecnología. Correspondencia: Georgina Estrada. E-mail: georgina.estrada@cicy.mx.

Bixa orellana is well known to be the source of the food additive “achiote”. Among its extended food and cultural uses by mayan culture, *Bixa orellana* has been used in traditional medicine. Plants are sessile organisms that are exposed to numerous types of stress, either abiotic or biotic. Plants have developed several immune responses to pathogen infection to achieve tolerance or to activate cell death mechanisms, one class of peptides appears to be conserved in response to infection among plants, invertebrates and vertebrates; these are defensins. In plants, defensins have shown antifungal and/or antibacterial activity, mature peptides are composed by 45 to 55 amino acids and contain eight cysteines that form four disulfides. Its *in vitro* antimicrobial activity depends on the binding of the peptide with membrane lipids, being important in this interaction the C-terminal region defined as the γ -core loop. BoDef1 was identified in the transcriptome of *Bixa orellana*, the defensin sequence was amplified from cDNA to obtain a construct to express the recombinant HisBoDef1. BoDef1 has a percentage of identity of 51% with J1-1 in its mature peptide sequence. Therefore, the objective of this work was to obtain a pure His-tagged BoDef1 peptide by recombinant DNA techniques, expressed in bacteria, to determine the efficiency of the system previously used to express the pepper defensin J1-1[1]. The best *E. coli* strain and purification methods to obtain the recombinant peptide defensin BoDef1 will be presented.

References

1. Guillén-Chable, F., Arenas-Sosa, I., Islas-Flores, I., Corzo, G., Martínez-Liu, C., & Estrada, G. (2017). Antibacterial activity and phospholipid recognition of the recombinant defensin J1-1 from *Capsicum* genus. *Protein Expression and Purification*, 136, 45–51. <https://doi.org/10.1016/j.pep.2017.06.007>

IN SILICO ANALYSIS OF HOTSPOTS IN REPLICATIVE POLYMERASES AND THEIR RELATIONSHIP WITH GYNECOLOGICAL CANCER

Kayley Aileen Hernández Ramírez¹, Silvia Armenta²

Instituto de Ciencias Agropecuarias Universidad Autónoma del Estado de Hidalgo,
Avenida Universidad Km. 1 s/n Exhacienda Aquetzalpa, 43600 Tulancingo de Bravo, Hgo.
he357227@uaeh.edu.mx¹, silvia_armenta@uaeh.edu.mx²

The Ministry of Health of the State of Hidalgo reports that breast and cervical cancer are the leading causes of death due to this type of tumor in women¹. The increasing number of cancer cases represents a public health challenge, therefore, in this work, we propose an *in silico* approach that provides relevant information on the molecular basis of this disease that can contribute to developing early diagnosis strategies and personalized medicine.

Different studies and databases have reported the relationship between these types of cancer and mutations (hotspots) in δ and ϵ polymerases, which are largely involved in genomic DNA replication and have a proofreading exonuclease activity, and pol ϵ additionally has the P-Domain which surrounds the DNA and has an impact on processivity².

Therefore, this work aims to perform an *in silico* study of the hotspots of polymerases δ and ϵ , to identify and predict the functional effect of the identified mutations using bioinformatics tools and to evaluate the potential impact of the mutations and their relationship with carcinogenesis.

The genomic studies platform of the cBioPortal³, which collects data related to different types of cancer, was used to search for mutations in δ and

polymerase genes. The most relevant hotspots were selected and visualized in a three-dimensional structure in ChimeraX⁴ obtained with the AlphaFold prediction tool. A structure-function analysis was performed on the selected mutations with the SIFT and PolyPhen-2 prediction tools. Finally, the geneious prime program was used to predict the changes in the secondary structure of the protein.

It was observed that unlike in POLE, where the overwhelming majority of cancer mutations are located in the exonuclease domain, POLD1 mutations occur in the exonuclease and polymerase domains, and those related to gynecological cancer are located in a region close to the site of interaction with the synthesized DNA chain. Cancer-related mutations alter exonuclease proofreading activity in addition to inducing significant alterations in the structure and function of polymerases, leading to polymerase instability. Therefore, it is proposed to perform *in vitro* and *in situ* studies for the characterization of the enzymatic and molecular activity of polymerases and to analyze the cellular interactions underlying the mutations in order to propose new treatments for gynecological cancer.

References

1. Secretaría de Salud de Hidalgo.(2019) Exhorto Ref. a Cáncer.
2. Hoitsma, N. M., Whitaker, A. M., Schaich, M. A., Smith, M. R., Fairlamb, M. S., & Freudenthal, B. D. (2019) *Cellular and Molecular Life Sciences*.
3. Cerami *et. al.*. The cBio Cancer Genomics Portal: An Open Platform for Exploring Multidimensional Cancer Genomics Data. *Cancer Discovery*. May 2012 2; 401. PubMed.
4. Pettersen EF, Goddard TD, Huang CC, Meng EC, Couch GS, Croll TI, Morris JH, Ferrin TE. (2021) *Protein Sci.* UCSF ChimeraX.

IDENTIFICATION OF VIRULENCE GEN *FIMH* IN CLINIC ISOLATES OF *KLEBSIELLA PNEUMONIAE*

Nubia Francelica Hernández-Sánchez¹, Ulises Vega-Castillo¹, Ana Karen Gómez-García¹,
Emiliano Bueno González¹, Evangelina Beltrán-López¹, Nanzhuan Napaketzalli Tiznado-Parra¹
and Héctor Samuel López-Moreno

Laboratory of Molecular Biomedicine, CAC-UAS-264; Graduate Program in Biomedical Science, School of Chemical and
Biological Sciences, Universidad Autónoma de Sinaloa. Cd. Universitaria, Culiacán, CP: 80010, Sinaloa, México.

*hslmoreno@uas.edu.mx; (52-6677137860)

Klebsiella pneumoniae is a Gram-negative bacillary, anaerobic enterobacterium, opportunistic pathogen responsible for a variety of community and hospital infections such as urinary tract infections, pneumonia, and liver abscesses. The emergence of hypervirulent (hvKP) multidrug-resistant (MDR) strains led the World Health Organization (WHO) to include this species in the list of primary pathogens for investigation to optimize treatments. In Mexico, an incidence of nosocomial infections ranging from 4 to 15.4 per 1000 live births and from 4.1 to 8.8 per 100 discharges in hospitals in the region has been recorded, representing a potential risk for all hospitalized patients. However, it is unknown how many of these infections are due to *K. pneumoniae* and how many of them are hvKP. Therefore, we consider it of a great importance to start with the identification of virulence genes such as *FimH* present in clinical isolates for conducting analyses that allow the design of specific subsequent treatments. For this purpose, specific primers were used to identify the presence of the gene *FimH*, using the polymerase chain reaction (PCR) technique, and 14 clinical samples obtained from patients hospitalized at the Hospital General de Culiacán were analyzed. The results showed a 900 bp product corresponding to the *FimH* gene, which was present in 10 of the 14 clinical isolates, these are *Kp1*, *Kp2*, *Kp3*, *Kp4*, *Kp5*, *Kp8*, *Kp9*, *Kp11*, *Kp12* and *Kp13*, indicating a high prevalence of hypervirulent strains in the patients studied. Therefore, these results suggest the need to implement strategies for the early diagnosis of *K. pneumoniae* strains that have the potential to cause severe diseases, as well as the control of their spread in the hospital environment.

DOING BIONANOTECHNOLOGY WITH DSDNA AND CRISPR-DCAS12A: DESIGNING LINEAR AND BRANCHED NANOFIBERS

Armando Hernández-García* & Jesús Ortiz-Saucedo

National Autonomous University of Mexico, Institute of Chemistry, Department of Chemistry of Biomacromolecules, Laboratory of Biomolecular Engineering and Bionanotechnology.
E-mail: armandohg@iquimica.unam.mx*

In nature DNA serves as the storage of genetic information, however, recently DNA has become a very useful material for building nanostructures which have multiple applications in biotechnology and biomedicine¹. Conventional DNA nanotechnology assembles nanostructures using single-stranded DNAs (ssDNA) and high-temperature annealing processes. The scope of DNA nanotechnology can be expanded by incorporating DNA-binding proteins that can help to self-assemble DNA into predictable nanostructures at room temperature in a programmable fashion². Also, hybrid -DNA-protein- nanostructures can exploit the advantages of both biomolecules: the predictability and specificity of DNA, along with the wide functional and structural diversity of proteins, thus mitigating their individual disadvantages. Particularly, CRISPR-Cas systems offer advantages since they can be programmed to bind to specific dsDNA sequences through a guide RNA, offering a way for creating hybrid nanostructures³. Here, we genetically fused to dCas12a protein the chemically induced heterodimerization domains FKBP and FRB. Two resultant proteins, dCas12a-FKBP and dCas12a-FRB, were positioned into short dsDNAs using gRNAs and polymerized into nanofibers upon the addition of rapamycin, which dimerizes FKBP and FRB domains. Also, branched nanofibers were obtained when three chimeric proteins were positioned into a dsDNA. The formation of single nanostructures was analysed in detail through biochemical assays and by Atomic Force Microscopy. This hybrid nanomaterials may find applications as nanorulers, nanoscaffolds and for biosensing.

References

1. Li, F., Li, J., Dong, B., Wang, F., Fan, C., & Zuo, X. (2021). *Chemical Society Reviews*, 50(9), 5650-5667.
2. Hernandez-Garcia, A. (2021). *Nanomaterials*, 11(5), 1332.
3. Calcines-Cruz, C., Finkelstein, I., Hernandez-Garcia, A. (2021). *Nano Letters*, 21(7), 2752-2757.

BIOTECHNOLOGICAL SYNTHESIS OF PRECURSORS OF INTEREST IN THE PRODUCTION OF SSRIS

Olga Livier Jasso Durón¹, Camila Barrio Santoyo¹, Ilse Araceli Careaga Rojas¹, Juan Cristóbal García García¹

¹Unidad Profesional Interdisciplinaria de Ingeniería Campus Guanajuato, Instituto Politécnico Nacional.
Av. Mineral de Valenciana 200 Col. Fraccionamiento Industrial Puerto Interior, 36275 Silao de la Victoria, Gto.
Tel: 4772727577. E-mail: jcgarciaga@ipn.mx

Huntington's disease (HD) is an inherited disorder that affects nerve cells in the brain, leading to their gradual deterioration and death. It manifests with involuntary movements, abnormal body postures, and behavioral issues; its progression is continuous and worsens over time¹. Symptoms may persist and include outbursts of anger, suicidal thoughts, profound depression, and psychosis². Reduced levels of serotonin have been found in studies of HD in both animals and humans, sparking interest in the study of treatment with selective serotonin reuptake inhibitors (SSRIs), particularly fluoxetine in adults with HD³. Among the newer second-generation antidepressant treatments are SSRIs, inhibitors, selective serotonin and noradrenaline reuptake inhibitors, and other medications⁴. Fluoxetine, or N-methyl-3-phenyl-3-[4-(trifluoromethyl)phenoxy]propan-1-amine, is a member of the (trifluoromethyl)benzenes, a second-generation antidepressant categorized as an SSRI. Nowadays, it is prescribed to manage depression and other pathologies⁵. Its safety and tolerability profile make it an attractive therapeutic option. A crucial step for the synthesis of fluoxetine is the reduction of ethyl benzoylacetate to obtain the intermediate ((S)-3-phenyl-3-hydroxypropionate ethyl). In this study, biotechnological obtaining of said intermediate will be carried out using the microorganisms *Geotrichum candidum*, *Kluyveromyces marxianus*, and *Saccharomyces cerevisiae*, as studies have reported their utility for the enantioselective reduction of ketones to alcohols^{6,7}. Additionally, the optimal operating conditions will be evaluated, the kinetic parameters of microbiological reduction will be determined, and the use of immobilized cells as an alternative method of the process will be assessed. Finally, the mechanism of fluoxetine synthesis from the intermediary obtained with bioreduction will be proposed.

References

1. National Institute Of Neurological Disorders And Stroke (2023). Huntington's Disease. *National Institutes of Health*.
2. National Human Genome Research Institute (2011). About Huntington's Disease. *National Institutes of Health*
3. Beglinger, L. J., Adams, W. H., Langbehn, D., Fiedorowicz, J. G., Jorge, R., Biglan, K., Caviness, J., Olson, B., Robinson, R. G., Kieburtz, K., & Paulsen, J. S. (2014). Results of the citalopram to enhance cognition in Huntington disease trial. *Movement disorders: official journal of the Movement Disorder Society*, 29(3), 401-405.
4. Gartlehner, Gerald (2008). Comparative Benefits and Harms of Second-Generation Antidepressants: Background Paper for the American College of Physicians. *Annals of Internal Medicine*, 149(10), 734.
5. National Center for Biotechnology Information (2024). PubChem Compound Summary for CID 3386, Fluoxetine. *National Library of Medicine*.
6. Ribeiro, J. B., Ramos, M. D. C. K. V., de Aquino Neto, F. R., Leite, S. G. F., & Antunes, O. A. C. (2005). New microbiological catalytic accesses to (S)-fluoxetine. *Catalysis Communications*, 6(2), 131-133.
7. Chênevert, R., Fortier, G., & Rhlid, R. B. (1992). Asymmetric synthesis of both enantiomers of fluoxetine via microbiological reduction of ethyl benzoylacetate. *Tetrahedron*, 48(33), 6769-6776.

MOLECULAR IDENTIFICATION OF A HIGHLY POLYHYDROXYBUTYRATE-PRODUCING STRAIN

Alfonso Jiménez Adón¹, Alma Rosa Corrales Escobosa², Andrea Margarita Rivas Castillo³, Norma Gabriela Rojas Avelizapa⁴, Karla Lizbeth Macías Sánchez^{1*}

^{1,1*}Unidad Profesional Interdisciplinaria de Ingeniería Campus Guanajuato Instituto Politécnico Nacional. Silao de la Victoria Gto., México. ²Departamento de Química, DCNE, Campus Guanajuato, Universidad de Guanajuato, Gto., México.

^{3,4}Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada del Instituto Politécnico Nacional. Querétaro, Qro., México.

*E-mail: amrivasc@ipn.mx, kmaciass@ipn.mx

Given the environmental impact generated by the production of plastics, there is an interest in obtaining such materials from alternative sources; one of these alternatives is the obtaining of plastics through bacteria; however, this source has shown limited feasibility due to low yields and high production costs. Previous studies report the isolation of a *Bacillus* strain from a mining site. Its relevance lies in its ability to produce 5 times more polyhydroxybutyrate (PHB) than most bacterial strains reported to date (55.32 ± 9.76 g/L). According to reports, the genus *Bacillus* belongs to class IV of microorganisms that accumulate polyhydroxyalkanoates (PHA) as lipid storage materials. In this genus, the PHB production pathway is mediated by the phaRBC operon, consisting of the genes phaR, phaB, and phaC. A molecular identification of the microorganism has begun, involving the amplification of the 16S ribosomal gene, followed by a purification process, and subsequent sequencing. Upon obtaining the sequencing results, the obtained sequences were compared with the NCBI database, revealing the highest similarity with *Bacillus cereus*. To confirm these results, a MALDI-Biotyper analysis was performed, obtaining mass spectra of the protein profile, which were compared with database. The results indicated a higher probability with *B. cereus*, which has been described as a promising model to produce PHA.

EX SITU FERMENTATION BIOMASS MONITORING BY IMPEDANCE SPECTROSCOPY

José Raúl Junco Carmona¹, Adrián Díaz Pacheco², Víctor Eric López y López²

¹Centro de Investigación en Biotecnología Aplicada, Instituto Politécnico Nacional, Tepetitla de Lardizábal, Tlaxcala, Mexico. ²Unidad Profesional Interdisciplinaria de Ingeniería Campus Tlaxcala del Instituto Politécnico Nacional, Tepetitla de Lardizábal, Tlaxcala, Mexico
E-mail: vlopezyl@ipn.mx

Biomass is the most important variable in bioprocesses¹, despite its importance, it is extremely difficult to measure and consuming so time², a technique that have been use with successful to monitoring biomass in fermentations is Dielectric Spectroscopy³ increasing its applicability as well to monitor cell cultures. During their growth, Bacillus species have different phases which involve the production and consumption of different metabolites, culminating in the cell differentiation process that allows the generation of bacterial spores. In order to use impedance spectroscopy as a tool to monitor industrial interest Bacillus cultures, we conducted batch fermentations of Bacillus species such as *B. subtilis*, *B. amyloliquefaciens*, and *B. licheniformis* coupled with this technique. Each fermentation was characterized by the scanning of 50 frequencies between 0.5 and 5 MHz every 30 min. Pearson's correlation between impedance and phase angle profiles (obtained from each frequency scanned). However, the aeration and agitation during *in situ* monitoring produces the presence of interference in the measurements⁴ where not only the quantity produced in a culture but also the behavior that is presented are important concerns. It is clear that conditions of operation in a bioreactor affect biomass production, but how operation conditions affect the measurement of biomass on-line is of special interest. We studied the effect of bioreactor operating condition variations on model parameters using impedance spectroscopy for biomass monitoring. The model parameters analyzed were capacitance, resistance, alpha (α , hence the proposal to perform *ex situ* monitoring to obtain stable measurements, using a device connected by bypass, which is continuously fed, within which are performed measurements of impedance from 42 to 5 MHz at 10 mV using a Hioki Hi Tester 3532-50 analyzer. It has been found that the presence of bubbles in the measurement chamber is not eliminated because its pass through the bypass and throughout the *ex situ* duct, causing interference in the measurements. Bypass pumping speed may be the key to reducing bubble effect and help to improve the monitoring of fermentations where biomass is more difficult to monitor, as it is for filamentous fungi.

References

1. Biechele P, Busse C, Solle D, Scheper T, Reardon K. Sensor systems for bioprocess monitoring. *Eng Life Sci.* 2015;15(5):469-488. doi:10.1002/elsc.201500014
2. Flores-Cosío G, Herrera-López EJ, Arellano-Plaza M, Gschaedler-Mathis A, Kirchmayr M, Amaya-Delgado L. Application of dielectric spectroscopy to unravel the physiological state of microorganisms: current state, prospects and limits. *Appl Microbiol Biotechnol.* 2020;104(14):6101-6113. doi:10.1007/s00253-020-10677-x
3. Díaz Pacheco A, Dinorín-Téllez-Girón J, Martínez Montes FJ, Martínez Tolibía SE, López y López VE. On-line monitoring of industrial interest Bacillus fermentations, using impedance spectroscopy. *J Biotechnol.* 2022;343(October 2021):52-61. doi:10.1016/j.jbiotec.2021.11.005
4. Díaz Pacheco A, Delgado-Macuil RJ, Díaz-Pacheco Á, Larralde-Corona CP, Dinorín-Téllez-Girón J, López-Y-López VE. Use of equivalent circuit analysis and Cole-Cole model in evaluation of bioreactor operating conditions for biomass monitoring by impedance spectroscopy. *Bioprocess Biosyst Eng.* 2021;44(9):1923-1934. doi:10.1007/s00449-021-02572-0

AMYLASE PRODUCTION FROM SOLID STATE FERMENTATION AND SUBMERGED LIQUID FERMENTATION BY THERMOTOLERANT FILAMENTOUS FUNGI OBTAINED FROM THE TOLANTONGO CAVES

Adriana Jazmín Legorreta-Castañeda¹, Karina García-Gutiérrez¹,
María Guadalupe Guerra Sánchez², Darío Rafael Olicón-Hernández¹

¹Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Departamento de Microbiología,
Laboratorio de Bioquímica y Biotecnología de hongos, México.
E-mail: doliconh@ipn.mx

The isolation of thermotolerant microorganisms present in natural environments has generated great interest in recent year, due to the extracellular enzymes or secondary metabolites that they can produce, among which are useful enzymes in the degradation of different substrates, as they are the amylases. These enzymes break down starch to dextrin, maltose, or free glucose and are used commercially in many industrial processes, such as food production, textile manufacturing, and detergents.

In the present work, thermotolerant filamentous fungi were isolated from sediment and water samples from the river, tunnel and caves of Tolantongo, Hidalgo, Mexico. The amylolytic activity in the isolated fungi were evaluated at a semiquantitative level, by measuring hydrolysis halos on agar plates with the respective substrate.

The fungi that presented the highest enzymatic index of amylolytic activity were selected to measure the production of amylases in solid and liquid fermentation states using the methods modified described by López-García *et. al.*, 2024 and Olagoke 2014, respectively.

Thirty thermotolerant filamentous fungi (38°C) were isolated from the Tolantongo river and caves, through the various isolation techniques used.

Of the 31 amylolytic fungi, 21 belong to the samples from the river (68%), 9 were isolated from caves (29%) and 1 corresponds to the tunnel (3%). The fungal isolates with the highest enzymatic index were H9 (2.51), H14 (2.61), H26 (2.34) and H31 (2.73). The strains H13 and H14 presented the highest levels of amylolytic activity by liquid fermentation (2.4U/mL) on day 6 at 28°C, and (6.35 U/mL) on day 4 at 45°C, respectively. The result of solid-state fermentation shows varied with respect to liquid state fermentation and depend on each strain, finding promising results for the biotechnological application for this fermentation.

References

1. Olagoke, O.A. (2014). *Am J Microbiol Biotechnol*, 1, 64-70.
2. López-García, C.L. *et. al.* (2024). *Preparative Biochemistry & Biotechnology*, 1, 1-11.

O/W EMULSIONS STABILIZED WITH MODIFIED CHIPILIN PROTEIN (*CROTALARIA LONGIROSTRATA*)

Diana Itzel López-Monterrubio¹, Consuelo Lobato-Calleros¹, Eleazar Aguirre-Mandujano¹

¹Departamento de Preparatoria Agrícola, Universidad Autónoma Chapingo,
Km. 38.5 Carretera México-Texcoco, 56230, Texcoco, México

E-mail: ilopezmonterrubio@gmail.com, consuelobato@yahoo.com, eleagman@yahoo.com

Chipilín (*Crotalaria longirostrata*) is a quelite native to southern Mexico and Central America, which is consumed in typical dishes of the region. Chipilín leaves are rich in vitamins, calcium minerals, and secondary metabolites (Mendez-Lopez *et. al.*, 2023). Likewise, the protein content of the leaves of this plant ranges between 26 % and 38 % depending on the collection site, which makes it a viable source of protein for use in the food industry (Martínez, 2012). However, chipilín protein has not been studied for use as an emulsifier. In this sense, the objective of this work was to evaluate the influence of the modification method on the emulsifying properties of the chipilín protein. In proximal chemical analysis of the chipilín leaf flour, we demonstrated its high protein content (32.53 ± 0.26 %), confirming its high potential as a source of vegetable protein. The chipilín protein was used by three methods: *i*) the pH of the protein was adjusted to 12 for 2 h and subsequently adjusted to 7.0 (PCh₁₂); *ii*) the high-pressure treatment (PCh_{MP}) was subjected to 3 cycles at high pressure (101 MPa) in a Microfluidics equipment (model M-110P, Newton, MA, USA); *iii*) the protein modified by dual method (PCh_{12MP}) was first subjected to alkaline treatment and subsequently to high pressure treatment. The ζ -potential of the chipilín protein was favored in the alkaline treatment and the high-pressure treatment, however, there were no significant differences between the control treatment (PCh_N) y PCh_{12MP}. O/W emulsions were prepared where the aqueous phase had a protein concentration of 0.5 % w/w and an oil phase equal to 0.05. The hydrodynamic diameter of the emulsions decreased with the treatment: PCh_N 1.65 ± 0.07 μm , PCh₁₂ 1.23 ± 0.05 μm , PCh_{MP} 1.39 ± 0.10 μm y PCh_{12MP} 1.02 ± 0.07 μm . This was corroborated with optical microscopy, where the formation of smaller droplets could be observed with the dual treatment. For its part, the polydispersity index of the emulsions was less than 0.2 for all emulsions. On the other hand, the highest creaming index was observed in the emulsion stabilized with PCh_N (9.26 ± 0.07 %), while the treatment with PCh_{12MP} presented the lowest value (8.27 ± 0.02 %). This demonstrates the good capacity of the chipilín protein for use as an emulsifier, which improves with the dual modification.

References

1. Mendez-Lopez, A. Y., del Carmen Lagunes-Espinoza, L., González-Esquínca, A. R., Hernández-Nataren, E., & Ortiz-García, C. F. (2023). *South African Journal of Botany*, 154, 140-148.
2. Martínez Muñoz, A. B. (2012). Consejo Superior Universitario Centroamericano.

IDENTIFICATION OF ANTIGENS IN THE IMMUNOPROTEOME OF PORPHYROMONAS GINGIVALIS: PROGRESS

Evangelina Beltrán-López¹, Fred Luque-Ortega¹, Eliakym Arámbula-Meraz¹, Karen Virginia Pineda-Hidalgo¹, Ulises Vega-Castillo¹, and Héctor Samuel López-Moreno^{1*}

¹Laboratory of Molecular Biomedicine, CAC-UAS-264; Graduate Program in Biomedical Sciences and Graduate Program in Biotechnology, Faculty of Chemical and Biological Sciences and Faculty of Odontology. Universidad Autónoma de Sinaloa, Cd. Universitaria, Culiacán, CP.80010, Sinaloa, México.

*hslmoreno@ms.uas.edu.mx; CP.80010; +526675747963

Porphyromonas gingivalis is a coccobacillus Gram-negative anaerobic bacteria that causes 20 to 50% of chronic periodontitis worldwide, making it a significant health concern. In Mexico, the Ministry of Health does not report its diagnosis, so its prevalence is unknown. Reports associate *P. gingivalis* with various systemic diseases, such as rheumatoid arthritis, Alzheimer's, colorectal cancer, and heart diseases, highlighting its biomedical significance. Currently, its genome sequence is available, and some proteins like gingipains have known functions and pathological roles. However, about 20% of its proteins have poorly studied or unknown functions. In this context, studying the immunoproteome provides a biomedical biotechnological tool to enhance our understanding of the host-bacteria relationship. This could lead to identifying unknown antigens, optimizing diagnostic methods, discovering therapeutic targets, or designing a vaccine to control *P. gingivalis* and mitigate its impact on public health and related systemic diseases. For this study, the biomass of a *P. gingivalis* ATCC 33277 culture maintained in anaerobiosis was collected by centrifugation and processed by sonication for proteomic analysis. Using strips of immobilized pH 3-10, the analysis was visualized in a 12.5% SDS-PAGE minigels stained with Coomassie Blue R-250. A total of 81 spots were counted, with molecular weights ranging from 18 to 200 kDa, 97.5% of which had a MW of 23 to 75 kDa. Although the pI covered the entire range, 80% of the spots were concentrated in a pI of 5 to 9, with a notable concentration in the range of 7 to 8. These findings align with literature reports but show a unique concentration in the pI range of 7 to 8. In conclusion, these results support further study of the *P. gingivalis* immunoproteome, focusing on characterizing previously unreported spots.

ANALYSIS OF THE EFFECT OF VASCULAR AND MESOPHYLL EXPRESSION OF A COMMON BEAN AQUAPORIN PVAQP1 ON *ARABIDOPSIS THALIANA* GROWTH

Valeria Alexandra López Toledo*, Brenda Yazmín Vargas Hernández*, Leandro Alberto Núñez Muñoz*,
María Eugenia Hidalgo Lara, Beatriz Xoconostle Cázares*, Roberto Ruiz Medrano*

Depto. de Biotecnología y Bioingeniería, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional.
Av. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, 07360 Ciudad de México, CDMX.
E-mail: rmedrano@cinvestav.mx

Drought stress has a negative impact on crop yields, thus affecting its biological cycle and productivity. This stress is the result of an imbalance between water uptake and loss in the plant and is currently caused by climate change and low water availability¹. To cope this stress at physiological level, plants have developed different mechanisms involving ABA-dependent and independent pathways². Among drought-induced transcripts, we have previously identified an RNA for an aquaporin in *Phaseolus vulgaris* cv Pinto Villa, a tolerant cultivar. Aquaporins belong to a family of tonoplast intrinsic proteins, allowing the selective water transport among adjacent cells. Interestingly, this RNA was expressed in the phloem only in response to water deficit³. In this work, we will determine the effect of expressing the corresponding mRNA, termed *PvAQPI*, in *Arabidopsis thaliana* under the control of vascular- and mesophyll-specific promoters. The effect on plant growth and development, as well as physiological traits such as photosynthesis, relative water, chlorophyll and anthocyanin contents in plants grown under fully irrigated conditions and water deficit will be shown. Transcriptomic analyses will be carried out to understand the mechanisms involved in drought tolerance mediated by *PvAQPI*.

References

1. dos Santos, T. B., Ribas, A. F., de Souza, S. G. H., Budzinski, I. G. F., Domingues, D. S. (2022). *Stresses*, 2, 113–135.
2. Xoconostle-Cázares, B., Ramírez-Ortega, F. A., Flores-Elenes, L., Ruiz-Medrano, R. (2010). *American Journal of Plant Physiology*, 5, 241–256.
3. Montalvo-Hernández, L., Piedra-Ibarra, E., Gómez-Silva, L., Lira-Carmona, R., Acosta-Gallegos, J. A., Vazquez-Medrano, J., Xoconostle-Cázares, B., Ruiz-Medrano, R. (2008). *New Phytologist*, 177, 102–113.

R-LUXHR: QUANTITATIVE HOMOLOGOUS RECOMBINATION REPORTER SYSTEM

Luis Brito-Elías, Verónica García Castillo,
Carlos Pérez-Plasencia and Eduardo López-Urrutia

Facultad de Estudios Superiores Iztacala, UNAM
Av. De los barrios 1, Los Reyes Iztacala, 54090, Tlalnepantla de Baz, México
E-mail: e_urrutia@unam.mx

DNA is constantly exposed to damage caused by both endogenous and exogenous agents. The cells possess several mechanisms to repair said damage, collectively known as DNA Damage Response (DDR), whose study is important due to its participation in cancer development and treatment choice. The scientific community has developed methods to evaluate each DDR pathway specifically. One of these pathways is Homologous recombination (HR), commonly measured using fluorescence-based methods such as single-cell electrophoresis, in-situ hybridization, and the direct repeat GFP assay (DR-GFP); however, while sensitive, these methods offer fundamentally qualitative measurements. The R-LuxHR reporter system (Luciferase repetition to measure HR) relies on the advantages of luminometry over fluorescence to quantitatively measure the repair of a reporter gene through HR. This method consists of the transfection of three plasmids: one expressing the I-SceI endonuclease, followed by the pLuc Δ 3 and pLuc Δ 5 plasmids 24 h later. The pLuc Δ 3 plasmid encodes the luciferase ORF interrupted by two I-SceI digestion sites and a spacer sequence; the pLuc Δ 5 plasmid contains the full-length luciferase ORF devoid of a promoter. The I-SceI endonuclease interrupts the luciferase ORF in the pLuc Δ 3, blocking its expression unless repaired by the endogenous HR machinery, which will repair it using the full sequence from the pLuc Δ 5 plasmid. Luciferase expression will be proportional to the HR rate in the cell; thus, measuring luciferase activity by luminometry yields a quantitative measurement of HR. The R-LuxHR reporter system can be used in most transfectable cell lines and has been submitted to the Instituto Mexicano de la Propiedad Industrial) under patent application MX/E/2024/024917.

PHENOLOGICAL EVALUATION IN *CAPSICUM ANNUM* PLANT BY THE APPLICATION OF CHITOSAN NANOPARTICLES

Eva Mariela Lunar-Mata¹, Enrique González Pérez², Angela María Chapa Oliver¹,
Jesús Rubén Rodríguez Núñez¹, Laura Mejía Teniente^{1*}

¹Programa de Ingeniería en Biotecnología, Departamento de Ingeniería Agroindustrial, División de Ciencias de la Salud e Ingenierías, Campus Celaya-Salvatierra, Universidad de Guanajuato, Av. Mutualismo Esq. Prolongación Río Lerma S/N, Celaya, Gto. C.P. 38060, México. ²Campo Experimenta Bajío. Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias. Carretera Celaya-San Miguel de Allende km 6.5, Colonia Roque, Celaya, Guanajuato, México. C. P. 38110.
E-mail: laura.mejia@ugto.mx

Chitosan nanoparticles (NP-QT) highlighting for their chemical-biological properties, including their tissue regenerative capacity, phenological stimulant, antimicrobial and antiviral activity, in addition to being biodegradable and highly biocompatible. In agriculture, chitosan can act as a biostimulant or elicitor, promoting growth and regulation, crop nutrition, increasing tolerance to biotic and abiotic stress, being a viable and sustainable alternative to increase crop yields with minimal environmental impact and improving their development conditions. Based on the above, the objective of this research was to evaluate and compare the effect of the application of different concentrations of chitosan nanoparticles on the phenological development of *Capsicum annum* plants. A completely randomized experimental design was used. Foliar sprays of 0%, 0.005%, 0.01%, 0.015% and 0.02% of chitosan nanoparticles showed that the 0.01% NP-QT treatment was statistically significant respect to the response variables of height, stem diameter, number of leaves and leaf area, compared to the other treatments and the control plants.

References

1. Chamnanmanoontham, N., Pongprayoon, W., Pichayangkura, R., Roytrakul, S., & Chadchawan, S. (2015). Plant Growth Regulation, 75, 101-114.
2. TENIENTE, L. M. (2018). Application of Elicitors AS, Chitosan, and H2O2 in *Capsicum annum* and their effect on the response to biotic stress.

CLONING OF FLIPPASE IA OF *GIARDIA INTESTINALIS*

Carmen Alejandra Luque-Camacho¹, María Cristina Villa-Medina¹, Evangelina Beltrán-López¹,
Ulises Vega-Castillo¹, Kimberly Arisbeth Casal-Rodríguez¹ and Héctor Samuel López-Moreno^{1*}

¹Laboratory of Molecular Biomedicine, CAC-UAS-264; Graduate Program in Biomedical Sciences and Graduate Program in Biotechnology, Faculty of Chemical and Biological Sciences. Universidad Autónoma de Sinaloa, Cd. Universitaria, Culiacán, CP.80010, Sinaloa, México.

*hslmoreno@ms.uas.edu.mx; CP.80010; +526675747963

The loss of plasma membrane asymmetry associated with cell death or apoptosis has been observed in *Giardia intestinalis* treated with different antiparasitic agents; however, proteases related to phosphatidylserine translocation have been poorly studied. In this scenario, the objective of the present project is the cloning of Flippase IA from *G. intestinalis* as the beginning of understanding the initial phases of apoptosis in this parasitic protozoan. Our *in silico* results show that Flippase IA contains important structural domains such as the transmembrane region, cytoplasmic region, caspase target motifs, and phosphorylation/dephosphorylation sites. Subsequently, specific primers were designed, and cloning of the 621 bp region consisting of domain A of *G. intestinalis* Flippase IA was performed. The cloned sequence is in the process of being registered in GenBank and will allow us to strengthen the molecular bases for a better understanding of the biology of *Giardia* that mediates its apoptosis.

EVALUATION OF MICROALGAE GROWTH PORPHYRIDIUM CRUENTUM IN RELATION TO LIGHT INTENSITY

Mildred Y. Manriquez-Zamora¹, Braulio Trujillo-Santoyo¹, JC Rodríguez-Sierra¹,
Ana L. Quintero-Ramirez¹ & Samuel Celaya- Herrera¹

¹UPIIG-IPN, Avenida Mineral de Valenciana 200
Col. Fraccionamiento Industrial Puerto Interior, 36275 Silao de la Victoria, Gto.
E-mail: alquinteror@ipn.mx

Summary. The study of microalgae has maintained great importance in recent decades. With microalgae and cyanobacteria being the most studied, the potential of red microalgae continues to be investigated. In the case of *Porphyridium cruentum*, a Rhodophyta microalga characterized by the production of exosulfated polysaccharides and phycoerythrin as a pigment, its analyses are affected by the low quantity of these metabolites in laboratory assays. In the present study, experiments were conducted at the laboratory level under different light intensity conditions, ranging from 15.41 to 92.48 PPF (Photosynthetic Photon Flux Density), within the RBW LED light spectrum. Photoinhibition was observed at the highest light intensity values, as well as a color change from red to green. The highest biomass productivity was obtained at a light intensity of 41.1 PPF, reaching a maximum concentration of 3.27×10^6 cells/mL and a specific growth rate of 0.0164 h^{-1} over a period of 7 days. This light intensity allowed for a higher growth rate compared to higher light intensities. The study demonstrates that the microalga *Porphyridium cruentum* grows better at low light intensities (15.41 and 41.1 PPF) compared to high light intensities (66.79 and 92.48 PPF). In a graph showing the specific growth rate as a function of light intensity, a peak is observed at low light intensities, followed by a decrease at high light intensities. Additionally, a pigment change from red to green was observed under high light intensities, probably due to inhibition. These findings provide specific data that can improve the cultivation of *P. cruentum*. The importance of controlling light intensity to maximize biomass productivity is highlighted. Future research could explore other factors that may improve the biomass production of *Porphyridium cruentum* at light intensities below 15.41 PPF.

SYSTEMATIC EVALUATION OF SELECTIVE ANTIMICROBIAL PEPTIDES

Gabriel Marcelino Pérez¹, María Teresa Lara Ortiz¹, Carlos Alberto Brizuela Rodríguez², Gabriel Del Río Guerra¹

¹Departamento de Bioquímica y Biología Estructural, Instituto de Fisiología Celular, UNAM; ²Departamento De Ciencias de La Computación División De Física Aplicada, Centro de Investigación Científica y de Educación Superior de Ensenada.
E-mail: gdelrio@ifc.unam.mx

Currently, the excessive use of antimicrobials in different areas has caused an increase in the resistance of pathogenic microorganisms [1]. This has caused infectious diseases in humans, animals, and plants to become more difficult to manage with the compounds present in the market, representing a significant challenge for public health, with millions of deaths attributable to multidrug-resistant (MDR) microorganisms each year, according to data from the United Nations (UN) [2]. Therefore, new strategies are required to control and treat MDR bacteria. Antimicrobial peptides (AMPs) are small peptides that protect their host from bacteria, fungi, viruses, and protozoa, in addition to having functions in immune regulation, inflammation, and wound healing [3]. Therefore, this work proposes to use an experimental method that allows to systematically evaluate the antimicrobial activity of recombinant AMPs against multidrug-resistant (MDR) bacteria, with the objective of finding new alternatives for the treatment and control of these bacteria.

References

1. Kon, Kateryna; Rai, Mahendra (ed.). *Antibiotic resistance: mechanisms and new antimicrobial approaches*. Academic Press, 2016.
2. Larsson, D.G.J.; Flach, C.F. (2022). *Nat. Rev. Microbiol.*, 20, 257–269.
3. Huan, Y.; et. al.. (2020). *Front. Microbiol.*, 11, 1–21.

PHYLOGENETIC ANALYSIS OF *ASPERGILLUS FLAVUS* ENZYMES WITH HIGH POTENTIAL FOR PET (POLYETHYLENE TEREPHTHALATE) DEGRADATION

Noemí Marchena Echeverría, Jossie Fernanda Espinoza Ávila,
Alfredo Herrera Estrella, Edgar Balcázar López

Universidad de Guadalajara. Centro Universitario de Ciencias Exactas e Ingenierías. Blvd. Gral. Marcelino García Barragán 1421, Olímpica, 44430 Guadalajara, Jal. Responsible author: Edgar Balcázar López; edgar.balcazar@academicos.udg.mx; Blvd. Gral. Marcelino García Barragán 1421, Olímpica, 44430 Guadalajara, Jal.

Our research offers a promising alternative to address the environmental and public health challenges posed by the extensive use of plastics. We can potentially mitigate these issues by harnessing the power of microorganisms such as fungi and bacteria. Recent studies have shown that bacteria like *Ideonella sakaiensis* and fungi such as *Humicola insolens*, *Fusarium oxisporum*, and certain fungi of the *Trichoderma* genus are capable of degrading PET (polyethylene terephthalate), a major component of plastic waste.

Interestingly, these fungi and bacteria produce Cutinases, Esterases, and MHETases, which act directly on PET depolymerization and degradation to simpler compounds. Likewise, a Tannase with the capacity to degrade PET was reported in a non-culturable marine bacterium. Preliminary studies in our research group showed that *T. asperillum* presents a tannase structurally similar to the MHETase of *Ideonella sakaiensis*. Within the search for new organisms capable of degrading PET, two marine fungi (*Aspergillus flavus* and HM2 in the process of molecular identification) were isolated and identified in our laboratory from a plastic bag from Estero de Urias in Mazatlan, Sinaloa. By analyzing the genome of one of them (*Aspergillus flavus*), we discovered that this fungus presents eleven tannases. Through phylogenetic analysis, we observed that five of them are grouped next to the clade of an MHETase from a non-culturable marine bacterium and the MHETase from *Ideonella sakaiensis* (which is one of the best-studied enzymes in PET degradation). Furthermore, by structure analysis using the I-TASSER program, we observed that four of the five tannases (ID:2166952, ID:2287271, ID:9945, and ID:2287564) showed high similarity to the crystallographic structure of *Ideonella sakaiensis* MHETase (6QZ1).

Analysis of this filter suggests that these tannases might be involved in degrading PET (polyethylene terephthalate), so their expression in bacterial or fungal recombinant systems is considered.

MECHANISTIC ASSESSMENT OF THE BIOSYNTHESIS OF CYANOBACTERIAL SECONDARY METABOLITES: TWO CASES

J. A. Martínez Yerena^{1,3}, L. Zdvoráková¹, J. Mareš^{1,3}, Z. Kameník¹,
P. Hrouzek^{1,3}, T. H. J. Niedermeyer⁴, M. Schwark⁴

¹Biology Centre, Czech Academy of Sciences, Branišovská 1160, 37005 České Budějovice, Czech Republic,

²Institute of Microbiology, Czech Academy of Sciences, Vídeňská 1083, 14200, Prague, Czech Republic,

³Faculty of Science, University of South Bohemia, Branišovská 1760, 37005 České Budějovice, Czech Republic,

⁴Institute of Pharmacy, Free University of Berlin, Königin-Luise-Straße 2-4, 14195 Berlin, Germany.

jose.yerena@hbu.cas.cz

Cyanobacteria is an ancient group of photosynthetic prokaryotes that is almost ubiquitous in the biosphere, that is notable for its resilience under extreme environmental conditions in part due to the rich diversity of unique secondary metabolites that they may produce as a direct response to environmental stressors, or just constitutively produce for yet unknown purposes.

Their abundant production of bioactive metabolites firmly sets them as a prime source of novel compounds with potential pharmaceutical applications, and as a hazard to be monitored, as many of their compounds are known to be harmful for humans and animals. Hence, why the structure, ecology and biosynthesis of their compounds is being intensively studied, to both assess their potential and reduce their impact.

Here we present our work on the biosynthesis of two cyanobacterial metabolites with antiproliferative properties. By heterologously expressing genes predicted to catalyze key steps of their synthesis, and reconstituting their reactions *in vitro*, we have confirmed their relationship to their producers and open the gate for further manipulation.

Our first metabolite is *Aetokthonostatin* (AEST), is a compound structurally related to dolastatin, which is produced by *Aetokthonos hydrillicola*, the eagle killing cyanobacteria. Here we expressed and purified the last enzyme of its biosynthesis gene cluster, a S-Adenosyl methionine (SAM) dependent methyltransferase, and demonstrated *in vitro* that it exclusively methylates stand-in analogs for the late stage of its biosynthesis, confirming the relationship between AEST and its producer.

Our second metabolite is Nostatin A, a Ribosomally Synthesized Post-translationally-modified Peptide produced by a terrestrial Nostoc strain. We have heterologously expressed the first enzyme of its biosynthetic gene cluster, and we are working towards a confirmation of its predicted step. This enzyme belongs to a different group of SAM dependent methyltransferases, one which cleaves SAM to produce a radical intermediary for their predicted reaction.

ADVANCEMENTS IN BARLEY VARIETY DEVELOPMENT: TECHNOLOGIES AND APPLICATIONS

Julio Armando Massange Sánchez¹, Samuel Madera-González¹, Claus Krogh Madsen²

¹Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C. Camino Arenero 1227, El Bajío, Zapopan, Jalisco. ²Department of Agroecology, Aarhus University, Slagelse, Denmark.
ZIP code: 45019 Phone number: +523333455200 ext.1700. E-mail: jmassange@ciatej.mx

Barley is the fourth most important cereal worldwide after maize, wheat, and rice in terms of tonnes produced. It is widely distributed across various geographical regions, including high latitudes, subarctic and arid zones, cold regions, and Mediterranean and subtropical climates. Barley is mainly used as animal feed and for malting in whisky and beer production, but it also serves human consumption, biofuel, and paper production. Its adaptability to extreme environments, along with the end use and local practices, has driven barley's evolution, resulting in varieties with traits such as two-six spike, spring-winter growth habits, and hulled-hull less grain. Similarly, barley's genetic improvement has been guided by its uses, demand, and available technologies. Since the establishment of genetic principles in the 19th century, hundreds of varieties (e.g., Triumph, Golden Promise, Clipper, Bowman, Flagship, and Sukai Golden) have been released to face adverse environmental conditions and/or improve yield and quality characters. This work presents, the advancements in barley variety development across different world regions and shows the implementation of conventional and modern technologies in creating new varieties. Notably, industrialized countries lead in using modern technologies, given the availability of resources, infrastructure, and trained personnel. However, developing varieties in non-industrialized countries proves fruitful using conventional technologies, resulting in relevant and impactful outcomes within their regions. This information provides orientation on the use of technologies in barley genetic improvement across various countries, contributing to address specific breeding goals.

References

1. Åberg, E., & Wiebe, G. A. (1946). Technical Bulletin of Department of Agriculture, 907, 1-190.
2. Angessa, T. T., & Li, C. (2016). In G. Zhang & C. Li (Eds.) Academic Press, 223-240.
3. Cíván, P., Drosou, K., Armisen-Gimenez, D., Duchemin, W., Salse, J., & Brown, T. A. (2021). BMC Genomics, 22(1), 1-17.
4. Cuesta-Marcos, A., Kling, J. G., Belcher, A. R., Filichkin, T., Fisk, S. P., Graebner, R., Helgerson, L., Herb, D., Meints, B., Ross, A. S., Hayes, P. M., & Ulrich, S. E. (2015). Elsevier Inc, 4, 287-295.
5. Dai, F., & Zhang, G. (2016) Academic Press, 1-26.
6. Friedt, W., Horsley, R. D., Harvey, B. L., Poulsen, D. M. E., Lance, R. C. M., Ceccarelli, S., Grando, S., & Capettini, F. (2010). John Wiley & Sons, Ltd, 160-220
7. Gonzalez Gonzalez, M., Zamora Díaz, M. R., Solano Hernández, S., Huerta Zurita, R., Gómez Mercado, R., & Rojas Martínez, I. (2021). Revista Mexicana de Ciencias Agrícolas, 25, 15-20.
8. Muñoz-Amatriaín, M., Cuesta-Marcos, A., Endelman, J. B., Comadran, J., Bonman, J. M., Bockelman, H. E., Chao, S., Russell, J., Waugh, R., Hayes, P. M., & Muehlbauer, G. J. (2014). PLoS ONE, 9(4), 1-13.
9. Ramage, R. T. (1987). In Plant Breeding Reviews, 95-138. Sato, K. (2020). DNA Research, 27(4).
10. Verma, R.P.S., Lal, C., Malik, R., Kharub, AS., Kumar, L., & Kumar, D. (2022) Springer Singapore, 93-134.

EFFECT OF DIFFERENT C:N AND MAGNESIUM RATIOS ON PHB ACCUMULATION IN *BACILLUS THURINGIENSIS*

Yazmin Melendez-Xicohtencatl¹, Adrián Díaz Pacheco² and Víctor Eric López y López¹

¹Centro de Investigación en Biotecnología Aplicada, Tepetitla de Lardizábal, Tlaxcala, México,

²Unidad Profesional Interdisciplinaria de Ingeniería Campus Tlaxcala, Guillermo Valle, Tlaxcala, México

E-mail: vlopezyl@ipn.mx

Plastic pollution is currently causing a disaster in the ecosystem. The focus of research has been on searching for alternatives through biodegradable products that have a high potential to replace conventional plastics. The family of polyhydroxyalkanoates (PHAs), more especially polyhydroxybutyrate (PHB), is one of the options. PHB is comparable to most common polymers in terms of both chemical and physical properties¹. Its limited production, mostly because of its high production cost, is one of its disadvantages nevertheless². Consequently, efforts are being made to optimize the PHB synthesis process. While there is a deficiency of phosphorus, magnesium, or nitrogen, several microbes can produce PHB. According to reports, *Bacillus* strains that accumulate higher PHB might have to be deficient in nutrients such as nitrogen, phosphorus, magnesium, and oxygen. Compared to other microorganisms that require substantial amounts to accumulate more PHB, like *pseudomonas*^{3,4}. A 2² factorial design was used to examine the impact of N and Mg on PHB accumulation. The response surface revealed that the ideal C:N ratio is 22.6 at a 0.05 Mg concentration. Thus, may conclude that while a N deficiency is necessary, it should not be excessive to get a higher PHB accumulation. At the same time, moderately higher Mg levels could potentially contribute to increased production. For that, *B. thuringiensis* is therefore a viable option for large-scale PHB production.

References

1. Hamdy, S., Danial, A., Gad, Shoreit, A., & Latif, A. (2022) BMC Microbiol, 22, 183
2. Javaid, A., Aslam, S., Qaisar, H. (2023) Jenrs, 2(12), 7-14.
3. Trakunjae, C., Boondaeng, A., Apiwatanapiwat, W., Kosugi, A., Arai, K., & Vaitthanamsat, P. (2021), Sci Rep, 11, 1896.
4. Thammasittirong, A., Saechow, S., & Thammasittirong, S. N. (2017) Turkish journal of biology, 41(6), 992-1002

CHARACTERIZATION OF A RECOMBINANT PROTEASE INHIBITOR FROM BROCCOLI (BRASSICA OLERACEA VAR. ITALICA)

María Fernanda Mendoza-Acosta¹, Luz Edith Casados Vázquez^{1,2} y José Eleazar Barboza Corona^{1,2*}

¹Posgrado en Biociencias, División de Ciencias de la Vida, Universidad de Guanajuato Campus Irapuato-Salamanca.

²Departamento de Alimentos División de Ciencias de la vida, Universidad de Guanajuato Campus Irapuato-Salamanca.

*josebar@ugto.mx

Protease inhibitors of plant origin are protein compounds present in various natural sources such as seeds, tubers, and legumes; they have emerged as versatile tools with potential applications in multiple fields. Its main function in plants is defense against insects, inhibiting the activity of essential digestive enzymes. However, its potential extends beyond plant protection, encompassing agriculture, nutrition, and medicine applications. In this work, a protease inhibitor (BraDef1) from broccoli (*Brassica oleracea* var. *italica*) was expressed and characterized. BraDef1 was amplified using oligonucleotides designed based on its sequence previously identified in broccoli extracts. The amplicon was cloned into the pET32α (+) vector and transformed into *Escherichia coli* BL21 Rosseta 2. The induction of the recombinant protein was carried out with 0.5 M IPTG and its purification by affinity chromatography with nickel columns and molecular exclusion, obtaining a yield of 29 µg/mL from a 1.5 L culture. It was characterized by evaluating its protease inhibitory activity and stability in different temperature and pH ranges. It was also determined that BraDef1 can permeabilize the *Fusarium oxysporum* membrane. The relevance of this work lies in the fact that we report for the first time the cloning and characterization of a protease inhibitor from broccoli (*Brassica oleracea* var. *italica*).

A NOVEL CIS-ELEMENT AS A POTENTIAL REGULATOR IN BEGOMOVIRUS COMPLEMENTARY STRAND SYNTHESIS

Daniel A. Mendoza-Magaña, Daniel A. Rodríguez-García, Yair Cárdenas-Conejo*

Laboratorio de Biología Sintética Estructural y Molecular de la Universidad de Colima.
Carretera los limones-Loma de Juárez CP. 28629, Colima, México.

*E-mail: ycardenasco@conahcyt.mx

Geminiviruses are circular single-stranded DNA plant viruses of significant economic interest, causing estimated global losses of \$30 billion annually. Within the *Geminiviridae* family, the *Begomovirus* genus is the most relevant, comprising over 445 reported species. Begomoviruses (BGVs) replicate via rolling circle replication facilitated by the replication protein Rep, encoded in their genome. For replication and transcription to occur, the complementary strand must be synthesized. Recently, it has been determined that the host proteins involved in initiating the complementary strand synthesis (CCS) of BGVs are DNA polymerase α^1 and the large subunit of DNA primase², which together with the small subunit of DNA primase form the DNA polymerase α complex. However, the exact origin of replication of the complementary strand (sso) in BGVs has not been precisely determined, although it is proposed to lie within the replication origin (ori) of BGVs². In contrast, in *Mastrevirus*, another genus within the same family, the sso was identified in a different region of the genome, where a set of molecules functioning as primers for CSS was found. These molecules are located downstream of two hairpins (a hairpin and a mini-hairpin 5'-GNA-3')³. Similarly, in nanoviruses, endogenous primers associated with BBTV virions were discovered⁴. As in the case of mastrevirus, this set of molecules was located downstream of a region capable of forming a mini-hairpin 5'-GNA-3'. Here, a conserved palindromic sequence (CPS) was discovered within BGVs, capable of forming a mini-hairpin of the type 5'-GNA-3'. The CPS is conserved across all BGV species and their associated satellites. We demonstrated that this CPS has a biological function, as mutating this sequence drastically reduces viral titers and symptom severity. Thus, we hypothesize that the role of CPS is to function as a signaling element involved in the generation of the Double-Stranded Intermediary, acting as the signal for the host primase to synthesize the primer required for the synthesis of the complementary strand. To determine if the CPS can recruit the DNA polymerase alpha complex, we are working on the expression of the two primase subunits in *Nicotiana benthamiana* for subsequent use in EMSA assays to determine if they bind to the CPS.

References

1. Nat Commun. 2021 May 13;12(1):2780. doi: 10.1038/s41467-021-23013-2
2. Front Plant Sci. 2023 Mar 17;14:1130723. doi: 10.3389/fpls.2023.1130723.
3. EMBO J. 1984 Dec 20;3(13):3069-73. doi: 10.1002/j.1460-2075.1984.tb02260.x.
4. J Gen Virol. 1997 Feb;78 (Pt 2):479-86. doi: 10.1099/0022-1317-78-2-479

FORMULATION AND CHARACTERIZATION OF RASPBERRY-BASED PRODUCTS PRODUCED IN HUEJOTZINGO, PUEBLA

Maricarmen Meneses-Juárez¹, Maricarmen Saavedra-López²,
José Hugo Castorena-García², Carolina Ramírez-López¹

¹Instituto Politécnico Nacional, CIBA – Tlaxcala. ²Instituto Tecnológico del Altiplano de Tlaxcala.
E-mail: mmenesesj2400@alumno.ipn.mx

The raspberry is a fruit with excellent nutritional properties, but it is also very perishable if it is not kept in suitable conditions for fresh consumption, in addition to the fact that its cultivation in Mexico is mainly carried out in season, so alternative products must be sought for its consumption and conservation while maintaining nutritional and functional value.

The aim of this project, involved the formulation of four raspberry waste products, such as jam, ferment, beer and liquor. The raspberry used is of the “Driscoll Maravilla” variety produced by the company Frambuesas Buenavista located in San Diego Buenavista Huejotzingo, Puebla, the analyses for characterization were carried out and the sensory evaluation was carried out by an untrained panel of 30 judges; men and women between 20 and 50 years of age.

The sensory analysis showed a preference for the batch of jam containing the proportions of raspberry 34%, tejocote 23%, sugar 43%, this formulation obtained the following characteristics: pH 3.08, °Bx 45.4, % titratable acidity 0.728, total sugars 18.413 g/ml. In the case of liquor, preference was given to the batch formulated with cane alcohol and filtered raspberry juice in a 1:1 ratio, obtaining the following characteristics: pH 3.65, °Bx 30, % titratable acidity 0.7455, Total phenols 4.4005 mg/ml. In the case of wine, the formulation was formulated with the characteristics of pH 2.47, °Bx 5.9 %, Acidity 0.8015, total phenols 2.9063 mg/ml. In the case of raspberry beer, the formulation was formulated with the following characteristics: pH 2.99, °Bx 7.6, % acidity 0.521, total phenols 3.6399 mg/ml.

By following up on this project, the raspberry producing company can reduce its waste and expand the variety of products, it is recommended to pay attention to mechanical cultivation, post-harvest management and biological control to avoid losses that impact economically.

References

1. Bascopé J., Andrés (2013-03-18) Productive reality of raspberry in the U.S. and Mexico. Berries. Expert Report. (Accessed: 03 November 2022).
2. Aguirre, L. P., & Cadenas, A. (2016). Analysis of raspberry competitiveness in Mexico. Mexican Journal of Agricultural Sciences, 37(1), 145-156.

RELATION BETWEEN ENVIRONMENTAL CONDITIONS AND ORGANIC FERTILIZERS IN CORN SEEDS QUALITY

Lucila Méndez Morán*, Celia Robles Murguía,
Héctor Gerardo Frías Ureña, Ignacio Patricio Cáceres Salazar

Centro Universitario de Ciencias Biológicas y Agropecuarias, Universidad de Guadalajara. Camino Ramón Padilla Sánchez
No. 2100 C.P. 45200, Nextipac, Zapopan, Jalisco, México.
E-mail: *lmendez@cucba.udg.mx.

Organic fertilizers have been used to improve the growth and development of crops, the vermicompost is a good fertilizer that increases soil quality, crop growth and yield. But, the environmental conditions can be decisive for its application in the field. In the present work, corn seeds obtained from crops grown in four different environmental conditions with and without vermicompost were used. For which a comparative analysis of the seeds was carried out in relation to the sugar and protein content, as well as their germination rate and seedling growth from said seeds. The results so far in relation to the bromatological and germination analysis demonstrate that there is a comparative relationship between environmental conditions and seed quality, as well as a relationship with the use of organic fertilizer. The results obtained have been decisive in characterizing the growing areas with the greatest capacity for growing corn under the different environmental conditions analyzed and the use of organic fertilizers.

ENCAPSULATION OF THE *BACILLUS SUBTILIS* PAD123-PARS::*GFPMUT3A* BIOSENSOR IN ALGINATE BEADS FOR THE DETECTION OF ARSENIC IN WATER

Noé de Jesús Miranda-Breceda¹, Luz Idalia Valenzuela-García², María Teresa Alarcón-Herrera², Patricia Ponce-Peña¹ and Víctor Manuel Ayala-García¹

¹Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur C.P. 34120., ²Centro de Investigación en Materiales Avanzados, Calle CIMAV n°110 Ejido Arroyo Seco Durango, Dgo. México CP.34147.
E-mail: nmiranda01mx@gmail.com

Arsenic is a naturally occurring element found in the terrestrial cortex. Water flows through certain rock formations allowing that arsenic (As) can dissolve and move to underground aquifers, streams or rivers that can be sources of drinking water. As is an extremely toxic element for humans and many living organisms not only in high concentrations, where exposure causes acute effects that can become lethal, but also exposure over a long period to low relative concentrations (e.g., in water intake) producing chronic negative health effects.

Whole-cell bacterial biosensors hold great promise as a practical complementary approach for in-field detection of As. Although there are various bacterial bioreporter systems for arsenic detection, fewer studies reported the immobilization of arsenic bioreporters in order to enable better detection strategies for this contaminant, as well as to make its use easier and more versatile. Previously, a whole cell biosensor for As detection constructed using *Bacillus subtilis* cells was reported by our group³. This biosensor termed PAD123-Pars::*gfpmut3a* can respond to concentrations of As(III) and As(V) below the minimum levels recommended by the WHO (10 µg/L) and emit green fluorescence. In this study we evaluated the immobilization of the PAD123-Pars::*gfpmut3a* biosensor in alginate biopolymer to immobilize cells and detect arsenite and/or arsenate in water. Parameters as alginate and CaCl₂ concentration as well as cell density were evaluated to establish the best conditions in which this biosensor can be used in an immobilized form.

References

1. Elcin, E., & Öktem, H. A. (2020). Journal of Environmental Health Science & Engineering, vol 18, 137-148.
2. Jang, Y.-C., Somanna, Y., & Kim, H. (2016). International Journal of Applied Environmental Sciences, vol 11, 559-581.
3. Valenzuela-García, L. I., Alarcón-Herrera, M. T., Ayala-García, V. M., Barraza-Salas, M., Salas-Pacheco, J. M., Díaz-Valles, J. F., & Pedraza-Reyes, M. (2023). Microbiology Spectrum, vol 11, e00432-23.

ANTIMICROBIAL AND GROWTH-PROMOTING PROPERTIES OF SILVER NANOPARTICLES FROM *BETA VULGARIS* L. LEAVES ON *M. BOMBYCINA* AND *S. UNDATUS* IN VITRO.

José Francisco Morales Domínguez, Cristina Garcidueñas Piña, Liliana Mercado Díaz de León

Departamento de Química. Universidad Autónoma de Aguascalientes

francisco.morales@edu.uaa.mx

Silver nanoparticles (AgNPs) are currently used in a wide range of products. In plants, they are mainly used for their antimicrobial and plant. AgNPs can be synthesized by physical, chemical and biological methods. Biological methods have the advantage of being cheaper, simpler and using less toxic reagents. In their biosynthesis, extracts of various plant species have been used and have demonstrated their usefulness as antimicrobials and in plant tissue culture as growth stimulators, promoting germination, sprouting and rooting. In this work, AgNPs were synthesized from *B. vulgaris* leaves (BvAgNP). Analysis of the phytochemical composition of plant showed that contain phenolic compounds as flavonoids that can participate in the synthesis reaction. Scanning electron microscopy and energy dispersive X-ray spectroscopy analysis showed that BvAgNPs are 78 nm diameter spheres with 39.76 % silver. The Z potential values found indicate that are stable in phosphate buffer. Twenty-five, 50, and 100 mg/L BvAgNP were used to study the effect on the growth of *M. bombycina* and *S. undatus* *in vitro*; rooting was inhibited in *M. bombycina* and root elongation was promoted in *S. undatus*. No sprouts were observed in *M. bombycina* and an average of 2.5 sprouts per L were observed in *S. undatus* at 25 mg/L. BvAgNPs showed antimicrobial activity against *Escherichia coli* and *Klebsiella oxytoca*, bacteria of clinical interest, and against *Agrobacterium tumefaciens*, a bacterium used in the genetic transformation of plants.

GENETIC TRANSFORMATION OF *MIMOSA TENUIFLORA* FOR THE ESTABLISHMENT OF HAIRY ROOT CULTURES

Jorge Humberto Mundo Ariza¹, Irene de la Concepción Perea Arango¹,
María del Carmen Gutiérrez Villafuerte¹, Maribel Lucila Herrera Ruiz², Manasés González Cortazar²,
Antonio Ruperto Jiménez Aparicio³, José de Jesús Arellano García¹

Centro de Investigación en Biotecnología (CEIB-UAEM), Av. Universidad 1001, Col. Chamilpa, Cuernavaca Morelos¹. Centro de Investigación Biomédica del Sur (CIBIS-IMSS), Argentina No. 1 Col. Centro, Xochitepec, Morelos². Centro de Desarrollo de Productos Bióticos (CEPROBI-IPN), Carretera Yautepec-Jojutla Km. 6, calle CEPROBI No. 8 Col. San Isidro, Yautepec, Morelos³.
E-mail: jorgemundo@uaem.mx

Mimosa tenuiflora, known as tepexcohuite, is traditionally used to treat infections, burns and as a hallucinogen. Its antimicrobial, cytotoxic and nervous system properties have been demonstrated, attributed to various secondary metabolites¹. The cultivation of transformed roots has been shown to promote metabolic production and have rapid growth rates, genetic and biochemical stability. In this project, a genetic transformation protocol mediated by *Agrobacterium rhizogenes* was implemented for the establishment of hairy root crops producing bioactive compounds. For infection with *A. rhizogenes* strains ATCC15834, A4 and K599, axenic seedlings were used as explants. The roots generated were individualized and disinfected. DNA extraction was carried out from the plant material and the genetic transformation of the established lines was corroborated by PCR *rolC* gene amplification. The biomass produced was macerated with the solvents n-hexane (C₆H₁₄), ethyl acetate (C₄H₈O₂) and ethanol (C₂H₆O) to obtain extracts, which were characterized by HPLC.

The formation of hairy roots was observed from the tenth day after infection, with a transformation efficiency of 64% for strain ATCC15834 and 53% for A4. From the PCR analysis, confirmation of genetic transformation was achieved by the expression of *rolC* gene from established lines. HPLC analysis of the extracts showed the presence of alkaloids, terpenes and flavonoids. Implementing a genetic transformation protocol mediated by *A. rhizogenes* infection allowed the establishment of hairy root cultures of *M. tenuiflora* producing molecules of pharmacological interest.

References

1. Octaviano R. Paulino U. Monteiro J. Cavalcanti E. *Mimosa tenuiflora* Poir: a Review of its Traditional Use, Phytochemistry and Pharmacology. Braz. Arch. Biol. Technol. 51(2008)937-947.

UNVEILING REMOTE HOMOLOGS OF FUNGAL CELL WALL PROTEINS USING AI-BASED STRUCTURAL PREDICTIONS

Pablo Valentín Navarro-Enguilo, Jesús Urbar Ulloa, Jorge Verdín*

Biología Industrial, Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco.
Camino Arenero 1227, El Bajío, Zapopan, JAL, México 4501
*jverdin@ciatej.mx

Cell wall resident proteins (CWPs) are the main interaction interface of the fungal cell with its surroundings. CWPs undergo rapid sequence changes that introduce variability, enabling the cell to adapt to diverse and changing environments. Consequently, CWP orthologs have typically been identified only among phylogenetically proximal species. Traditional methods for inferring protein function and phylogenetic relationships rely on sequence conservation, often failing to detect the intricate structural similarities shared by remote homologs.

We identified a collection of putative cell wall-resident proteins based on the presence of signal peptides, GPI signals, and the absence of transmembrane regions. This approach allowed us to track their conservation among various fungal species in our database. In parallel, we identified remote orthologs of cell wall proteins using AI-based structural prediction to challenge these results.

Traditional sequence-based searches often fail to detect these remote homologs due to low sequence identity. However, using structural analysis, we identified that the protein CCG-15 from *Neurospora crassa* exhibited structural homology with 15 remote proteins, all showing less than 30% sequence identity, but remarkably similar structures. Secondary structure analysis indicated that, despite low sequence conservation, a significant portion of the secondary structure was preserved. Additionally, repetitive motifs and logos were consistently observed among these remote homologs, highlighting the effectiveness of structural analysis in uncovering evolutionary relationships missed by sequence-based methods.

Comparing the structural dendrogram with sequence based phylogenetic reconstruction of AI-identified CCG-15 orthologs, revealed that these, despite their low sequence identity and belonging to the distant clades Zoopagomycota and Ascomycota, shared clear structural similarities. This structural resemblance was also observed across CCG-15 orthologs from different classes of fungi, a result that could not be retrieved from exclusively sequence analysis. Further results will be presented during the congress.

FORMULATION OF AN ORGANO-MINERAL FERTILIZER INOCULATED WITH PLANT GROWTH-PROMOTING BACTERIA

Melissa Nuñez-Valdés¹ and Antonino Báez Rogelio¹

¹Benemérita Universidad Autónoma de Puebla, Centro de Investigaciones en Ciencias Microbiológicas, Puebla, C.P. 72570
E-mail: melissa.nunezvaldes.icuap@viep.com.mx

95% of food comes from soils. Healthy soils provide the essential nutrients, water, oxygen, and support that plant roots need to grow and thrive. With an estimated global population of 9 billion by 2050 and the challenges of climate change, it is imperative to develop sustainable food production technologies¹. Intensive agriculture has led to soil degradation, threatening future food production. Only 30% of the nitrogen added to crops is absorbed by plants, causing environmental problems and economic losses². To mitigate soil degradation, more effective fertilizers need to be developed, such as organo-mineral fertilizers (OMF), which combine organic and mineral inputs and improve fertilizer absorption efficiency, reducing environmental impact³.

Currently, another alternative to improving agricultural productivity has been proposed, involving impregnating mineral fertilizers with plant growth-promoting bacteria (PGPB)⁴. PGPBs are free-living soil bacteria that have a positive effect on plant growth or development. Research on combining microbial and mineral resources aims to create efficient microbial formulations that are compatible with mineral inputs, benefiting crops and the environment. Microbial bioformulations are needed to improve plant yield and work complementarily and synergistically with mineral fertilization⁵.

This work consists of developing an organo-mineral fertilizer inoculated with a bacterial consortium including *Gluconacetobacter diazotrophicus* Pal5, *Paraburkholderia unamae* MTI-641, *Azospirillum brasilense* sp7, *Bradyrhizobium* sp. MS22, *Pseudomonas putida* KT2440, and *Sphingomonas* sp. OF178.

According to experiments conducted so far, an increase in the dry weight of lettuce has been observed when the organo-mineral fertilizer formulation consists of zeolite (mineral), compost (organic matter), DAP (chemical fertilizer), and microbial load inoculation of $\sim 10^4$ CFU/g of fertilizer.

References

1. FAO. Food and Agriculture Organization of the United Nations. (2015). Healthy soils are the basis for healthy food production. <https://www.fao.org/3/i4405e/i4405e.pdf>
2. Baez-Rogelio, A., Morales-García, Y. E., Quintero-Hernández, V., & Muñoz-Rojas, J. (2017). Next generation of microbial inoculants for agriculture and bioremediation. *Microbial Biotechnology*, 10(1), 19–21. <https://doi.org/10.1111/1751-7915.12448>
3. Anušauskas, J., Steponavičius, D., Romaneckas, K., Lekavičienė, K., Zaleckas, E., & Sendžikienė, E. (2023). The Influence of Bacteria-Inoculated Mineral Fertilizer on the Productivity and Profitability of Spring Barley Cultivation. *Plants*, 12(6). <https://doi.org/10.3390/plants12061227>
4. Ahmad, S., Imran, M., Hussain, S., Mahmood, S., Hussain, A., & Hasnain, M. (2017). Bacterial impregnation of mineral fertilizers improves yield and nutrient use efficiency of wheat. *Journal of the Science of Food and Agriculture*, 97(11), 3685–3690. <https://doi.org/10.1002/jsfa.8228>
5. Bargaz, A., Lyamlouli, K., Chtouki, M., Zeroual, Y., & Dhiba, D. (2018). Soil Microbial Resources for Improving Fertilizers Efficiency in an Integrated Plant Nutrient Management System. In *Frontiers in Microbiology* (Vol. 9). Frontiers Media S.A. <https://doi.org/10.3389/fmicb.2018.01606>

CHARACTERIZATION OF RECOMBINANT MODIFIED TRICHOCYSTATIN TC-2

Claudia Ivonne Flores Pucheta¹, Verónica Aranda-Chan¹, Octavio Montes-Flores¹,
Rossana Arroyo² and Jaime Ortega López¹

¹Centro de Investigación y de Estudios Avanzados del IPN, ¹Depto. de Biotecnología, ²Depto. de Infectómica y Patogénesis Molecular, Ciudad de México CP 07360
E-mail: jortega@cinvestav.mx

Cystatins are small proteins that act as reversible inhibitors of papain-like and legumain-like cysteine proteinases (CPs). Given their natural inhibitory effect on CPs, these proteins may be considered potential candidates for drug design. *Trichomonas vaginalis*, a sexually-transmitted protozoan parasite, encodes three cystatin-like genes dubbed trichocystatins 1, 2, and 3 (TC-1, TC-2, and TC-3). Two of them have been characterized. The TC-2 is an endogenous inhibitor of several trichomonad CPs that was found to be associated with TvCP39, one of the CPs involved in trichomonal cellular damage¹. This study aimed to obtain TC-2 without the first 11 amino acids of the N-terminal region to determine their participation in aggregation and CP inhibition. Therefore, the mutated protein was expressed in *Escherichia coli* BL21(DE3) and purified by Ni affinity chromatography. Its inhibitory function was evaluated against human cathepsin-L and compared with the native TC-2. A reduction in the propensity for dimer formation was observed, accompanied by a decline in its inhibitory activity. These data suggest that the missing N-terminus region may be involved in the inhibitory process and participate in dimer formation^{2,3}

References

1. Puente-Rivera, Jonathan; Ramón-Luing, Lucero; Figueroa-Angulo, Elisa; Ortega-Lopez, Jaime and Arroyo, Rossana. The International Journal of Biochemistry & Cell Biology, 54, 255-265.
2. Rzychon M, Chmiel D, Stec-Niemczyk. Acta Biochim Pol., 51(4), 861-73.
3. Klotz C, Ziegler T, Daniłowicz-Luebert E, Hartmann S. Adv Exp Med Biol.; 712:208-21.

SEPARATION AND CHARACTERIZATION OF THE CHARGE VARIANTS OF THE MONOCLONAL ANTIBODY BEVACIZUMAB

Maricela Olvera¹, Tiany Miranda¹, Luis Acosta¹, Irlanda Camacho¹, Arlene Calderón¹, Mayra Quiñonez²,
Ismael Jimenez², Juan de Dios², Laura Palomares¹, Tonatíuh Ramírez¹ and Mabel Rodríguez^{1*}

National Laboratory for Production and Analysis of Molecules and Drugs (LAMMB). National Autonomous University of México¹
Centro de Investigación Sophia²
E-mail: maricela.olvera@ibt.unam.mx
E-mail: operaciones@lammb.unam.mx*

Monoclonal antibodies, the best-sellers of biotechnological medicines, are immunoglobulins designed to act on defined therapeutic targets and are directed at a specific antigen to interfere with the action of a molecule or receptor in order to stop a specific pathogenic process or stimulate a cellular action or even divert a cellular mechanism towards another pathway.

The monoclonal antibody *PRO-169* targets a protein called vascular endothelial growth factor (VEGF), which is a protein that helps cancer cells develop a new blood supply. This monoclonal antibody binds to VEGF, blocking its binding to cells, stopping the development of new blood vessels and in consequence, reduces the oxygen and nutrients supply of cancer cells, so the tumor shrinks or stops growing.

Antibodies can exhibit changes in charge heterogeneity during production and purification caused by amino acid substitutions, glycosylation, and other chemical or post-translational modifications. These modifications can affect the charge on the surface of the monoclonal antibody causing the appearance of charge variants.

To demonstrate whether there are differences in the *in vitro* activity and pharmacokinetic profile between the charge variants: acidic, basic and neutral of *PRO-169*, the purification of the isoforms of a biosimilar product was carried out through ion exchange chromatography with a cation exchange column (cationic separation). In this way it was possible to separate the charge variants. In addition, analytical techniques were carried out to determine the concentration and potency of the monoclonal antibody variants by the inhibition of cell proliferation in the presence of VEGF.

Based on the results, it is concluded that only the neutral variant maintains the expected biological potency.

References

1. Gunjan, N., Rathore, A. & Kumar S. (2016). Should charge variants of monoclonal antibody therapeutics be considered critical quality attributes? *Electrophoresis*, 37, 2338-2346.

BIOINFORMATIC ANALYSIS OF THE RELATIONSHIP BETWEEN THE ENVIRONMENT AND MICROORGANISMS FOR MAIZE YIELD SIMULATION

Jimena Ortiz Laris¹, José Alfredo Mares Mora¹, Jorge Noé García-Chávez², Humberto Herrera-Ubaldo³, Cynthia Paola Rangel-Chávez¹, Ana Laura Alonso-Nieves⁴

¹Instituto Tecnológico Superior de Irapuato, Irapuato 36 821, Guanajuato, México. ²Instituto Potosino de Investigación Científica y Tecnológica A.C., Cam. a la Presa de San José 2055, Lomas 4ta Secc. CP 78216. San Luis Potosí, México. ³Department of Plant Sciences, University of Cambridge, Cambridge CB2 3EA, United Kingdom. ⁴Unidad de Genómica Avanzada, Centro de Investigación y de Estudios Avanzados, Irapuato 36 824, Guanajuato, México.
E-mail: ortizlarisjimena@gmail.com

Abstract. In Mexico, maize is the most important cereal, economically and nutritionally as well as in many communities' social and cultural realms. Mexico is the center of origin, domestication, and diversification of maize, which is reflected in the number of native varieties existing in our country. Native varieties have adapted to grow in environments with various temperatures, different altitudes, precipitation levels, soil nutritional conditions, etc. Some microorganisms associate with the plant's ecosystem, especially in the rhizosphere and endosphere, benefiting crop development. Drastic environmental changes can affect these associations. This project aims to determine the relationship between climatic variables and the changes in microbial diversity associated with maize plants to determine their influence on crop production.

To achieve this goal, first, we identified optimal zones for planting 158 families of maize varieties. We established two field experiments in two different years. We collected and analyzed environmental data available on the official website of the National Water Commission (CONAGUA) of Mexico for each year. These environmental data were our starting point for classifying initial growth conditions. In the second stage of the project, we obtained 700 samples of the rhizosphere and endosphere in total, corresponding to the years 2021 and 2022. Then, we isolate and quantify DNA from most of the samples following an established protocol, with specific modifications for our study.

Currently, we are working with the database to apply appropriate genomic prediction models that will allow us to predict yield based on the environmental variables we have collected. We will identify the microorganisms through sequencing, intending to add these metagenomic data to the yield simulation.

Keywords: Environmental variables, Maize, Microorganisms, Sequencing, Yield.

AGAVIN PROMOTES BENEFICIAL MICROBES IN THE SHRIMP MICROBIOTA

Adrián Ochoa-Leyva¹, Juan Pablo Ochoa-Romo¹, Fernanda Cornejo-Granados¹, Alonso Lopez-Zavala²,
María Teresa Viana³, Filiberto Sánchez¹, Luigui Gallardo-Becerra¹, Rogerio Sotelo-Mundo⁴,
Andrés Cota-Huizar⁵, Agustín López-Munguía⁶

¹Departamento de Microbiología Molecular, Instituto de Biotecnología, UNAM, México. ²Departamento de Ciencias Químico Biológicas, Universidad de Sonora, México. ³Instituto de Investigaciones Oceanológicas, Universidad Autónoma de Baja California, México. ⁴Laboratorio de Estructura Biomolecular, Centro de Investigación en Alimentación y Desarrollo, México. ⁵Camarones El Renacimiento SPR de RI, Sinaloa, México. ⁶Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, UNAM, México.

*adrian.ochoa@ibt.unam.mx

The use of prebiotics and probiotics have shown to prevent diseases in cultured shrimps. Most of these are fermentable oligosaccharides, disaccharides, monosaccharides, and polyols (FODMAPS), such as agavin, which promote microbiota biodiversity¹. We evaluated the growth performance and microbiota composition in the intestine and hepatopancreas of *Litopenaeus vannamei* using agavin as a diet supplement. Adult *L. vannamei* were fed a standard diet supplemented with agavin at 2% (AG2) or 10% (AG10) and compared to an agavin-free standard diet (BD). After 28 days, the feed conversion ratio, total feed ingested, and protein efficiency ratio were significantly improved only in shrimps fed with AG2. Surprisingly, profiling of the 16S rRNA gene showed higher microbial richness and diversity of the shrimps fed with the AG10 diet, while these parameters decreased in the AG2 diet. Finally, we gathered from the literature a list of 80 beneficial microbes for shrimp's health and observed 42 species in our data. Interestingly, *L. pentosus*, *P. putida* and *P. synxantha* were significantly increased in the hepatopancreas of AG10 while *R. palustris* and *S. thermophiles th1435* were significantly increased in the hepatopancreas of the AG2. If we analyzed the abundance of the 42 beneficial microbes as one community, their abundance increased as agavin concentration increased in the hepatopancreas. Additionally, we sequenced DNA extracted from agavin and found 9 of the 42 beneficial microbes. Our study proves that agavin supplementation is associated with increased beneficial microbes for the shrimp microbiota in farming conditions and that shrimp prebiotics can selectively modify the microbiota in an organ-dependent manner².

References

1. CM Chino de la Cruz, F Cornejo-Granados, L Gallardo-Becerra, ME Rodríguez-Alegría, et. al.. (2023) Complete genome sequence and characterization of novel Enterococcus faecium with probiotic potential isolated from the gut of *Litopenaeus vannamei*. Microb Genom 9,3.
2. JP Ochoa-Romo, F Cornejo-Granados, AA Lopez-Zavala, MT Viana, et. al.. (2022) Agavin induces beneficial microbes in the shrimp microbiota under farming conditions. Sci Rep 12, 6392.

SYNTHESIS OF XANTHAN GUM DERIVATIVES AND ITS BIOLOGICAL PROPERTIES

Karen Onofre Rentería¹, Luis Enrique Cobos Puc¹, Alejandro Zugasti Cruz¹, Francisco Javier Enríquez Medrano², Sonia Yesenia Silva Belmares¹, Ernesto Oyervides Muñoz^{1*}

¹Facultad de Ciencias Químicas, Universidad Autónoma de Coahuila, Saltillo, Coahuila, México. 25250.,

²Centro de Investigación en Química Aplicada, Saltillo, Coahuila, México. 25294.

E-mail: konofre@uadec.edu.mx

Xanthan gum (XG) is a biopolymer constituted of a D-glucose chain and ramifications of mannoses, with acetyl and pyruvic groups, and glucuronic acids¹. It is obtained by aerobic fermentation with *Xanthomonas* bacteria². It is mainly used in the food, pharmaceutical, and cosmetic industries thanks to its physicochemical properties. However, modifications are required to enhance or develop new properties for biomedical applications^{3,4}. It was synthesized two quaternary ammonium salts named 1-(2-aminoethyl) pyridinium bromide (PYB) and 1-(2-aminoethyl) trimethylammonium bromide (TAB), through quaternization reactions. These salts were chemically grafted onto xanthan gum to form Schiff bases. Then, the products were characterized by Fourier Transform Infrared Spectrometry (FTIR) and Nuclear Magnetic Resonance (NMR), in addition to carrying out Thermogravimetric Analysis (TGA) and evaluating the hemolytic, antibacterial, and anticancer properties. FTIR revealed some characteristic peaks corresponding to O-H and C=O attributed to xanthan gum and C=N attributed to ammonium salts. With NMR, it was determined the monosaccharides and methyl groups of XG and the carbons of trimethylammonium, pyridinium, and aminoethyl groups that correspond to the ammonium salts. TGA dictated the degradation point for XG-PYB (246 °C) and for XG-TAB (385 °C). The hemolytic test showed no toxicity in red blood cells. Breast cancer cells were not affected by XG, but XG-PYB showed cytotoxicity at 3 000 µg/mL and XG-TAB decreased cell viability from 100 µg/mL. *Escherichia coli* was found to be susceptible to XG-TAB and *Staphylococcus aureus* to both derivatives. To conclude, it was obtained two novel xanthan gum derivatives, checked by the presence of the molecules' characteristic groups and their corresponding carbons, in addition to demonstrating that XG-TAB is more thermostable, both compounds proved to be non-toxic in human blood, XG-TAB revealed to be toxic on breast cancer cells at low concentrations, besides both derivatives showed antibacterial properties.

References

1. E. Vázquez et. al., 2020. "Structural Analysis of Xanthan GUM-FE (III) Capsules," Academic Journal of Chemistry, no. 54, pp. 31-40.
2. Kure J.T., Ahmadu U., Gana M., Salubuyi S. B., and Ijah U. J. J., 2022. "Production and characterization of xanthan gum by bacterial isolates," Covenant Journal of Physical and Life Sciences, vol. 10, no. 1.
3. T. Riaz, M. W. Iqbal, B. Jiang, and J. Chen, 2021. "A review of the enzymatic, physical, and chemical modification techniques of xanthan gum," Int J Biol Macromol, vol. 186, pp. 472-489.
4. J. Patel, B. Maji, N. S. H. N. Moorthy, and S. Maiti, 2020. "Xanthan gum derivatives: review of synthesis, properties and diverse applications," RSC Adv, vol. 10, no. 45, pp. 27103-27136.

EFFECT OF SO₂ FROM SIMULATED FLUE GAS ON GROWTH AND GENE EXPRESSION OF S-COMPOUNDS IN THE MICROALGAE *DESMODESMUS ABUNDANS* RSM

Bryan Hernández¹, Laura Rodas-Zuluaga¹, Shirley Mora-Godínez², Rocío Díaz de la Garza², Adriana Pacheco¹

¹Tecnológico de Monterrey, Centro de Biotecnología, 64849 Monterrey, NL, México. ²Tecnológico de Monterrey, Institute for Obesity Research, 64849 Monterrey, NL, México
E-mail: adrianap@tec.mx

Desmodesmus abundans RSM is a microalgae adapted for 13 years to high CO₂ atmospheres. Previous studies have shown overexpression of genes associated with purines and folate biosynthesis under model flue gas (250 000 ppm CO₂, 700 ppm NO, and 100 ppm SO₂). Therefore, it is hypothesized that an excess of S in the growth medium generate biomass with desirable concentrations of S-containing compounds reflected in the overexpression of key genes involved in biosynthesis of purines and folate, and other associated pathways such as cysteine, methionine, and glutathione. In this study, *D. abundans* was grown with different S concentrations to analyze the differential gene expression of the above-mentioned pathways. *D. abundans* was cultured in Erlenmeyer flasks to 3/4 log phase (4 d) with a continuous flow of 25% v/v CO₂/air at 0.05 vvm (300 mL BG11 medium, 25 ± 2 °C, 100 rpm, 85 μmol PAR-photons m⁻² s⁻¹ of continuous light). Three experimental conditions, in triplicate and duplicated biologically, were tested: i) 100% S (28.6 mg L⁻¹ SO₄²⁻), original BG11 recipe; ii) 14% S (4 mg/L SO₄²⁻), limited condition; and iii) excess S, under a continuous supply of 100 ppm SO₂. Neither culture was inhibited by the growth condition (no lag phase) and exponential growth was similar among cultures; however, biomass production was the highest in the control (0.88 ± 0.11 g L⁻¹ d.w.), followed by excess S and limited S conditions (0.71 ± 0.06 and 0.51 ± 0.17 g L⁻¹ d.w. respectively). Therefore, S from simulated flue gas only reduced by 19% biomass production. A similar trend was observed for total protein content but with a greater reduction of 38% under excess S (56.2 ± 1.6%, 34.6 ± 0.41%, and 26.5 ± 3.7% d.w. for the control, excess S, and limited S, respectively). Amino acid quantification and transcriptome analysis will validate and reveal how the excess of S and, in general, S concentration affects protein content and other biochemical pathways of interest. Overall, it will be ideal to revalorize flue gas components into valuable biomass as a mitigation strategy using microalgae.

IN VITRO ASSISTED REFOLDING OF THE RECOMBINANT TRYPANOSOMA CRUZI ANTIGEN TSA-1

Esdras Enoc Pacindo Cabrales¹, Claudia Ivonne Flores Pucheta¹, Octavio Montes-Flores¹, Jaime Ortega López¹

¹Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Departamento de Biotecnología y Bioingeniería. Ciudad de México.
esdras.pacindo@cinvestav.mx

Chagas disease, caused by *Trypanosoma cruzi*, is a neglected disease for which the available drugs are not effective in the chronic phase¹. The search for new treatments leads to exploring alternatives, such as a therapeutic vaccine based on recombinant Tc24 and TSA-1 antigens expressed in *E. coli*. In preclinical tests, it has been found both proteins are good vaccine antigens. Nevertheless, the refolding of TSA-1, which is expressed as insoluble aggregates, is a complex process that requires further investigation².

Therefore, the aim of this work is to develop an *in vitro* refolding protocol assisted by the recombinant peptidyl-prolyl-isomerase TcCyP19 of *T. cruzi*, the disulfide oxidoreductase with chaperone activity DsbC and a minichaperone AD-GroEL from *E. coli* to improve the *in vitro* refolding of the TSA-1 antigen.

References:

1. World Health Organization. (2023). La enfermedad de Chagas (tripanosomiasis americana). Recuperado el 22 de mayo del 2023 de: [https://www.who.int/es/news-room/fact-sheets/detail/chagas-disease-\(american-trypanosomiasis\)](https://www.who.int/es/news-room/fact-sheets/detail/chagas-disease-(american-trypanosomiasis))
2. Villanueva-Lizama, L. E., Cruz-Chan, J. v., Aguilar-Cetina, A. del C., Herrera-Sanchez, L. F., Rodriguez-Perez, J. M., Rosado-Vallado, M. E., Ramirez-Sierra, M. J., Ortega-Lopez, J., Jones, K., Hotez, P., Bottazzi, M. E., & Dumonteil, E. (2018). Trypanosoma cruzi vaccine candidate antigens Tc24 and TSA-1 recall memory immune response associated with HLA-A and -B supertypes in Chagasic chronic patients from Mexico. PLoS Neglected Tropical Diseases, 12(1). <https://doi.org/10.1371/journal.pntd.0006240>

ANTIFUNGAL EFFECT OF CLOVE ETHANOL EXTRACT (*SYZYGIUM AROMATICUM*) ON THE IN VITRO AND BIOCHEMICAL INHIBITION OF *PHYTOPHTHORA SP.*

M.C. Adolfo Padilla Mendiola¹, Q.B.T. María José Reyes Chávez¹,
D.C. Verónica Dayali Gutiérrez Martínez¹, D.C. Maribel Cervantes-Flores¹

Facultad de Ciencias Químicas Durango, Universidad Juárez del Estado de Durango.
Antiguo camino a la ferrería # 89 int. 3, Granja Graciela, Durango, Dgo. cp. 34190.
Cel. 6181458028, adolfo.padilla@ujed.mx.

Fungi pose a significant threat to plants, each species targeting specific plant types. Their impact on agriculture is profound, causing economic and ecological damage while escalating production expenses due to the reliance on synthetic chemical fungicides. To address this challenge, we explored a natural solution: an ethanolic extract derived from cloves (*Syzygium aromaticum*). This extract, with a yield of 56.6%, demonstrated remarkable efficacy in inhibiting the growth of *Phytophthora sp.*, a notorious plant pathogen. Through meticulous evaluation, we determined that even at lower concentrations, the extract effectively combats the fungus without risking the development of resistance. Our findings suggest the potential of utilizing natural extracts as a sustainable and effective alternative to conventional fungicides in plant disease management. For which the concentrations were classified according to their parts per million and were named as; C1 =1000 ppm, C2 =1500 ppm, C3 =2000 ppm, C4 =2500 ppm and C5 =3000 ppm. The inhibitory effect of the ethanol extract of clove (*Syzygium aromaticum*) on the fungus *Phytophthora sp.* was evaluated. The ethanolic extract of cloves (*Syzygium aromaticum*) was obtained. The strain of the fungus *Phytophthora sp.* was obtained, isolated and purified. The efficiency of the ethanolic extract of cloves (*Syzygium aromaticum*) was evaluated, being this very efficient against the fungus *Phytophthora sp.* All concentrations were effective against the fungus, therefore, we recommend the lowest concentration to make its control more efficient and not cause future resistance to *Phytophthora sp.*

B. LICHENIFORMIS α -AMYLASE ENGINEERING GUIDED BY MACHINE LEARNING-COUPLED DIRECTED EVOLUTION TO INTRODUCE TRANSGLICOSIDIC ACTIVITY

José Alberto Parra Espinoza¹, Alejandro Garcíarrubio¹, Leticia Olvera¹ and Gloria Saab-Rincón¹

¹Biotechnology Institute, National Autonomous México University, Universidad Avenue 2001, Chamilpa C.P. 62210, Cuernavaca, Morelos, México.
E-mail: jose.parra@ibt.unam.mx

Introduction. Directed evolution gives us the opportunity to accelerate the evolutionary processes that guide the direction and function of proteins towards a desired task, either as improvements in the stability of the protein, improvement in its performance or modification of the original function: However, screening of large libraries in search of hits can be a daunting task. Therefore, the design, screening and selection processes must be improved for a more efficient engineering involving less experimental time and focusing efforts mainly on the experimental design of methodological strategy for the proposal of a limited library of variants to work, with the purpose to making such mutants efficient after expression, purification, and testing. Thus, in recent years and with the improvement of predictive tools applied to biologic fields, the use of machine learning tools and algorithms has been chosen for the design of enzymatic variants^{1,2,3}. Artificial intelligence paves the way for a significant contribution to this field in the coming years. α -amylases (E.C. 3.2.1.1) are extracellular hydrolases that act on D-glucose α -1,4 bonds within starch polymers (specifically in their amylose composition),⁴ contain 3 domains, one of them (A) composed of TIM barrel containing the catalytic triad. Some of them also have a double function, transglycosylation, which is the transfer of glycosidic group between a hydroxyl from one sugar to another acceptor different from water, and likewise some transferases contain a slight glycosylase activity⁵, in our laboratory we want to modify the reaction of a hydrolase (H) of *Bacillus licheniformis* (PDB ID: 1BLI) that itself hasn't a relevant transferase activity (T), and by analyzing enzyme sequences with both functions (H,T) we seek to train a machine learning method with a protein language model, to predict the important amino acids in the specificity of one or another reaction (H,T) and thus build variants with the ability to switch activity to transferase and test for the formation of alkylglucosides.

Method. We propose a phylogenetic analysis to determine homologies between GH13 family enzymes, sequence alignment focusing on the TIM barrel, identify preserved regions and perform an embedding using a protein language model to classify sequences of both functions (H,T) to project them into a two-dimensional space, perform in silico evolution to classify mutations in some region, train the model with different datasets to construct the variants, analyze their transglycosidic capacity, purify and analyze structurally the constructed variants.

References

1. Zachary Wu, Jennifer Kan, D Russell, Bruce J. Wittmann, Frances Arnold (2019). *PNAS*, 116 (18), 8852-8858.
2. Jonathan C. Greenhalgh, Sarah A. Fahlberg, Brian F. Pflieger, Philip A. Romero (2021). *Nature communications*, 12, 5825.
3. Sou Sugiki, Teppei Niide, Yoshiro Toya, Hiroshi Shimizu (2022). *ACS Synth. Biol.* 11, 3973-3985.
4. Machius M, Wiegand G, Huber R (1995). *Journal of molecular biology*, 246 (4), 545-559.
5. Sinnott Michael L (1990). *Chem. Rev.* 90, 1171-1202.

EVALUATION OF THE INTERACTION BETWEEN BACTERIA AND PHOTOBIONTS ISOLATED FROM LICHENS

Pérez-Barrón, B.I.¹, Rangel-Chávez, C.P.¹, Vázquez-Martínez, J.¹ and López-Ramírez, V.¹

¹Tecnológico Nacional de México/ ITS de Irapuato. Carr. Irapuato – Silao km 12.5 Col. El Copal. C.P. 36821
varinia.lr@irapuato.tecnm.mx

Lichens are symbiotic associations between an organism called a photobiont, which can be a cyanobacteria or green algae responsible for providing nutrients, and an organism called a mycobiont, which is a fungus that gives structure and characteristics to a lichen. Non-photosynthetic bacteria belonging to the families *Enterobacteriaceae*, *Pseudomonadaceae*, *Bacillaceae*, and *Rhizobiaceae* have been identified in this association. The role that non-photosynthetic bacteria play in the interaction within the lichen thallus is still unknown. The objective of our study was to observe the behavior of these non-photosynthetic bacteria in interaction with the photobiont, as well as to evaluate the phenotypic changes and understand the communication that exists between the two organisms. Through interaction assays, signaling molecules, vitamins, nutrients, and other metabolites can be identified. In the present work, two interaction strategies were carried out: the first based on photobiont-bacteria interaction, and the second on interaction in a culture by dispersion. The photobiont-bacteria interaction consisted of inoculating the algae strain in the form of a drop in four different sites of the culture medium and then inoculating the bacteria in the same way at different concentrations. The dispersal culture consisted of immersing the algae strain in agar and inoculating the bacterial strains similarly to the modified Kirby-Bauer method. The results of the photobiont-bacteria interaction demonstrated that only the strains of *Trebouxia sp.*, *Bracteacoccus sp.*, *Stichococcus sp.* and *Chlorella sp.* in interaction with the bacterial strains identified as *Bacillus mobilis*, *Enterobacter cloacae*, *Klebsiella oxytoca*, and *Stutzerimonas sp.* from all families, showed significant phenotypic changes. These changes included alterations in the colouring of the algae and the administration of nutrients in the culture medium, which allowed the algae to survive longer. Additionally, mutualism between the organisms predominated.

PURIFICATION AND CHARACTERIZATION OF CYTOTOXIC MOLECULES FROM A LIPID-RICH EXTRACT FROM MEXICAN AVOCADO SEED (*PERSEA AMERICANA* VAR. *DRYMIFOLIA*)

María Guadalupe Pérez-Esquivel¹, Marisol Báez-Magaña¹, Rafael Salgado-Garciglia²,
Alejandra Hernández-García², Ramón Cervantes-Rivera¹,
Alejandra Ochoa-Zarzosa¹ and Joel Edmundo López-Meza¹

¹Centro Multidisciplinario de Estudios en Biotecnología-FMVZ, Universidad Michoacana de San Nicolás de Hidalgo, Km 9.5 Carretera Morelia-Zinapécuaro, Posta Veterinaria. 58893, Morelia, Michoacán. ²Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Edificio B3, Ciudad Universitaria, Morelia, Michoacán.
E-mail: 2330749b@umich.mx

Cancer is one of the leading causes of death worldwide and is considered the third cause of death in Mexico. Although chemotherapy drugs are the primary cancer treatment, their side effects are still a significant problem. For this, searching for new bioactive compounds with enhanced cytotoxic activity and specificity on cancer cells is essential. Previously, our group reported a lipid-rich extract from Mexican avocado seed (LEAS) showed cytotoxic effects on different cancer cell lines: MCF-7 (IC₅₀: 33.09 µg/ml), Caco-2 (IC₅₀: 28 µg/ml), D-17 (IC₅₀: 15.5 µg/ml), and B16-F0 (IC₅₀: 40.85 µg/ml)¹. In addition, LEAS administration (0.2 mg/g body mass) inhibits tumor growth by up to 50% *in vivo* mouse melanoma tumors. LEAS contains long-chain fatty acids and acetogenins (avocatin, pahuatins, persins, and polyhydroxylated fatty alcohols)². However, the molecule(s) involved in the cytotoxic activity is (are) unknown. This work aims to identify the molecule(s) responsible for the cytotoxic effects of LEAS. LEAS was obtained from avocado seed by Soxhlet extraction with hexane. Further, LEAS was fractionated by silica column chromatography using a mobile phase with hexane, ethyl acetate, isopropanol, and acetic acid in a ratio of 40:20:5:1. Five fractions were recovered and analyzed by TLC. The fractions have different retention factors (RF) (0.76, 0.70, 0.53, 0.41, and 0.25). Then, the cytotoxicity of fractions was tested on murine melanoma cell line B16-F0 by MTT assays. The most significant effects were observed in fraction 2 (RF 0.53) at 30 µg/ml because the cell viability decreased by 40% at 24 h. According to the RF value and the standard, this result suggests that the cytotoxicity could be attributed to acetogenins C17, which could be responsible for the reduction in melanoma tumor growth previously reported. The chemical composition of fraction 2 is being determined by HPLC and GC-MS.

References

1. Ochoa-Zarzosa A., Báez-Magaña M., Guzmán-Rodríguez J., Flores-Alvarez L., Lara-Márquez M., Zavala-Guerrero B., Salgado-Garciglia R., López-Gómez R., and López-Meza J.E. (2021). *Plant Foods for Human Nutrition*, vol 76, pp. 133–142.
2. Lara-Márquez M., Báez-Magaña M., Raymundo-Ramos C., Spagnuolo P.A., Macías-Rodríguez L., Salgado-Garciglia R., Ochoa-Zarzosa A., and López-Meza, J.E. (2020). *Journal of Functional Foods*, vol 64, pp. 1-10.

STUDY OF AN ISOLATED *PENICILLIUM* SP THAT USES PLASTIC POLYMERS AS CARBON SOURCE

Martha Lizeth Pérez-Méndez, Lérica Liss Flores-Villavicencio,
Patricia Ponce-Noyola, Julio César Villagómez-Castro

Departamento de Biología, División de Ciencias Naturales y Exactas, campus Guanajuato, Universidad de Guanajuato, Noria Alta s/n Colonia Noria Alta, Guanajuato, Guanajuato, México. C.P. 36050, México. Tel. +52-4737320006-8167.
ml.perezmendez@ugto.mx; castroj@ugto.mx

The accumulation of waste from natural and synthetic polymers in the environment has increased in recent years, causing damage to the ecosystem and human health. Several strategies have been proposed to reduce their impact and, where possible, recycle them. However, chemical and physical recycling methods do not solve this problem, which is why efforts have been directed towards its use using microorganisms, fungi and bacteria mainly, or their secreted enzymes (esterase, lipase, cutinase, etc.). Particularly, in recent years, the isolation of bacteria and fungi capable of degrading polyethylene terephthalate (PET), a plastic highly resistant to biodegradation, has been described. In our laboratory, we have different fungal isolates from a consortium of microorganisms capable of growing in an aqueous suspension of pulverized PET, suggesting the presence of hydrolytic activities secreted at culture medium. Based on this background, this work aims to biochemically characterize the enzymatic activities of the isolated called C3, to analyze if they have potential for biotechnological use in the future. The isolate C3, was molecularly characterized using different DNA regions (β -tubulin, calmodulin, the 18S region, ITS, and, the second major subunit of RNA polymerase II) without being able to determine a specific species, but classifying it as *Penicillium* sp. This isolate can use plastic polymers as a carbon source when grown in a modified Mathur minimal medium. According to their preliminary electrophoretic and proteomic profiles performed from the secretome of *Penicillium* sp. (C3), the presence of at least one oxidoreductase and one esterase is suggested, when the isolate is grown with polyethylene (PE) as a carbon source; at least one esterase, in the presence of PET; and, one laccase, in the presence of polystyrene (PS). Additionally, this isolate showed the ability to degrade these plastic polymers (around 90%) after 5 weeks of incubation at 28 °C, under static conditions, using a modified Mathur minimal medium added with 3% w/v of the different polymers as a source carbon.

This work has been financed with funds from the person in charge and from CONAHCYT under the project CBF2023-2024-3070.

IMPACT OF PHOSPHITE ON GENE EXPRESSION IN THE FILAMENTOUS FUNGUS *TRICHODERMA ATROVIRIDE*

Camilo Pérez-Salazar¹ and Alfredo Herrera Estrella¹

¹UGA-LANGEBIO Cinvestav, km 9.6 Libramiento Norte carretera Irapuato-León, Irapuato, Gto.
E-mail: alfredo.herrera@cinvestav.mx

Abstract. Plant biostimulants are considered green solutions that improve crop production yields that are environmentally friendly. Phosphite (Phi: PO_3^{-3}) is a reduced form of phosphate (Pi: PO_4^{-3}) used in agriculture as a biostimulant and fungicide. Phi acts as a biostimulant in plants when well fertilized, inducing their defense response and improving their physicochemical characteristics¹. In addition, Phi is used as a growth inhibitor of oomycetes and phytopathogenic fungi and bacteria, such as *Fusarium oxysporum*, *Pseudomonas*, *Erwinia*, and *Xanthomonas*². Nevertheless, little is known about the effect of Phi on beneficial microorganisms found in the rhizosphere of plants or its impact at the gene expression level in these microorganisms. In this work, the objective was to understand and evaluate the effect of Phi on the filamentous fungus *Trichoderma atroviride*. Fungi of the *Trichoderma* genus are widely used in agriculture because they act as biostimulants in plants and are used as biocontrol agents against phytopathogenic fungi. The effect of Phi on the *in vitro* growth of *T. atroviride* was evaluated under different concentrations of Phi in the medium. Using RNA-Seq, it was possible to determine the gene expression pattern of *T. atroviride* when grown in the presence of Phi as the only source of phosphorus and in the presence of Pi + Phi in the medium. This work represents one of the first gene expression analyses in a beneficial fungus due to Phi.

References

1. McDonald, A. E., Grant, B. R. & Plaxton, W. C. (2001). Phosphite (phosphorous acid): its relevance in the environment and agriculture and influence on plant phosphate starvation response. *Journal of plant nutrition*. 24 (10), 1505-1519.
2. Huang, Y., Cai, S., Zhang, G. & Ruan, S. (2020). Transcriptome-Based Analysis of Phosphite-Induced Resistance against Pathogens in Rice. *Plants*. 9 (10), 1334.

HUMANIZATION OF CHIMERIC ANTI-CD20 ANTIBODY RITUXIMAB VIA CDR-GRAFTING

Daniel Pérez-Vega¹, Keyla Gómez-Castellano¹, Stefany Rodríguez-Luna², Ivette Mendoza-Salazar², Luis Elizarrarás-Rodríguez², Martha Pedraza-Escalona², Sonia Mayra Pérez-Tapia^{1,2,3} and Juan Carlos Almagro^{1,4}

¹Unidad de Desarrollo e Investigación en Bioterapéuticos (UDIBI), Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Mexico City, Mexico. ²Laboratorio Nacional para Servicios Especializados de Investigación, Desarrollo e Innovación (I+D+i) para Farmoquímicos y Biotecnológicos, LANSEIDI-FarBiotec-CONACyT. ³Departamento de Inmunología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional. ⁴Global Bio, Cambridge Massachusetts, United States. Zip code: 11350; Phone number: 729 182 5302; E-mail: 04daniel94@mail.com

Over the past 25 years, monoclonal antibodies (mAbs) have seen accelerated growth due to their significance in treating various diseases, including cancer and autoimmune disorders. Since the approval of the first mAb by the FDA in 1996, and up until December 2019, this regulatory entity had approved 79 therapeutic mAbs, a number that has since increased considerably¹. Rituximab is a chimeric anti-CD20 monoclonal antibody of the IgG1 subclass approved by the FDA in 1997 for treating B-cell malignancies and non-Hodgkin lymphoma (NHL). Nowadays it is routinely included in therapeutic regimens for conditions such as NHL, rheumatoid arthritis, pemphigus vulgaris, and systemic lupus erythematosus. However, immunogenicity in patients that receiving the treatment with rituximab has been reported, making it unsuitable for long time disorders such as autoimmune diseases². To mitigate this issue, in this study we humanized rituximab through complementarity-determining region (CDR) grafting onto the human acceptor germline frameworks, the most validated route to a successful humanised monoclonal antibody. Briefly, our methodology comprises four experimental phases. In the initial phase we performed the search in IMGT human database of germline sequences for the closest human germline VH and VL sequences, resulting in 4VH and 3VL-kappa chains. In addition, structural analysis of the humanized antibodies and the three-dimensional modelling included to assess the structural integrity. Subsequently, these genes as synthetic DNA fragments were cloned into mammalian expression vectors containing the human IgG1 heavy chain and kappa light chain constant domains. All possible combinations of the VH and VL chains were expressed in Expi293F cells. The yield, binding to CD20 were first assessed in the culture supernatant. The selected antibodies were large-scale expressed and purified by affinity chromatography using a Protein A column. Lastly, in the fourth phase, a physicochemical characterization of the obtained humanized antibodies was carried out, alongside biological characterization through CD20 binding assays, and immunological potential evaluation via *in silico* testing.

References

1. Lu, R., Hwang, Y., Liu, I., Lee, C. & Tsai, H. (2020). Development of therapeutic antibodies for the treatment of diseases. *Journal Of Biomedical Science*, 27(1).
2. Du, F. H., Mills, E., & Mao-Draayer, Y. (2017). Next-generation anti-CD20 monoclonal antibodies in autoimmune disease treatment. *Autoimmunity Highlights*, 8(1).

DESIGN AND GENERATION OF A GENETIC CONSTRUCTION FOR ECTOPIC EXPRESSION OF THE ARSR C₁₀₄S MUTANT REPRESSOR IN *BACILLUS SUBTILIS* FOR ORGANIC ARSENIC DETECTION

Miguel De Jesús Piña-Mena¹, Luz Idalia Valenzuela-García², Mario Pedraza-Reyes³
and Víctor Manuel Ayala-García¹

¹Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango, Unidad Dgo. Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur C.P. 34120., ²Centro de Investigación en Materiales Avanzados, Calle CIMAV n°110 Ejido Arroyo Seco Durango, Dgo. México CP.34147., ³Departamento de Biología. Universidad de Guanajuato. Noria Alta Guanajuato, Gto. México CP. 36050.
E-mail: 1103304@alumnos.ujed.mx

Arsenic is a toxic metalloid that has detrimental effects on all organisms. By evolutionary adaptations, most organisms on this planet have developed strategies to escape to the noxious impact of arsenic effects. The most notable are the *ars* operons, a set of ordered genes that give rise to proteins, which are used by bacteria and can work together to effectively eliminate arsenic in all its forms¹.

The *ars* operon, which is strongly conserved in bacteria, generally codifying a As-inducible repressor protein termed ArsR that regulates the operon transcription, and some proteins for metalloids detoxification. This repressor protein has specific As(III) binding motifs, which vary among the various bacteria studied.

Evidence has revealed that the *ars* operon of *B. subtilis* can be induced by arsenite, arsenate but not by DMA(V)². However, it has not been evaluated whether other forms of organic As such as phenylarsenic acid or trimethylarsine oxide are able to activate this operon response in *B. subtilis*, as occurs in *E. coli*. Furthermore, a previous study in the bacterium *Acidithiobacillus ferrooxidans* revealed that a single Cys₁₀₂-Ser amino acid change in ArsR through site-directed mutation, allowed a whole-cell biosensor designed in this bacterium to respond to aromatic arsenic and methylarsenic (III)³. Therefore, it is of interest to investigate whether point mutants of the putative As(III) binding domain in the ArsR repressor of *B. subtilis* would allow the binding of organic As species. In summary, this project aims to generate an ectopic copy of the ArsR repressor with point mutations in the As binding site.

References

1. Páez-Espino, D., Tamames, J., de Lorenzo, V., & Cánovas, D. (2009). *Biology, Biochemistry, and Medicine*, 22(1), 117-130.
2. Valenzuela-García, L. I., Alarcón-Herrera, M. T., Ayala-García, V. M., Barraza-Salas, M. (2023). *Microbiology Spectrum*, 11(4).
3. Chen, J., Nadar, V. S., & Rosen, B. P. (2017). 106(3)

MUSCLE GROWTH PROMOTION BY A CHIMERIC MYOSTATIN IMMUNOGEN DELIVERY BY AN ANTIGEN DISPLAY AND GENE DELIVERY BACULOVIRUS VECTOR

Vianey Ramírez-Andoney; Víctor D. Gonzalez-Fernandez; Amanda Gayosso-Vázquez; Alicia Ortega-Aguilar; Juan Pablo Pintor-Ríos; Julio Cesar Sánchez-Lemus, Ernesto Enríquez-Enríquez; Rogelio A. Alonso-Morales

Department of Genetic and Biostatistics, Faculty of Veterinary Medicine and Zootechnics, National Autonomous University of Mexico, Mexico City 04510, Mexico. Address: Av. Universidad 3000, Universidad Nacional Autónoma de México, C.U., Delegación Coyoacán, C.P. 04510, Ciudad de México. Phone: 55 39 34 96 88. E-mail: vianny102@hotmail.com

The current demographic and social improvement conditions in developing countries anticipate an increasing demand for animal products in the next few years. Projections for world meat demand, foresee a 44 percent increase by 2030. Meat production is an extraordinarily complex process where the knowledge in muscle physiology and animal biotechnology is crucial to designing novel strategies to growth promotion in livestock.

A promising approach to physiological intervention is the use of immune methods, i.e., inducing an immune response towards specific molecules, which will be blocked or activated. Some immunological targets may be negative regulatory factors, which naturally restrict biological processes, so that when these are blocked, a physiological response is promoted or increased.

A negative growth factor that has been evaluated as a good target to promote growth is myostatin, which is a negative regulator of muscle cell development and differentiation². Our working group has developed a strategy to target specific immune responses to endogenous proteins that consists of the fusion of immunostimulatory epitopes to specific proteins.

In this work, the inhibition of myostatin, in a murine model, was achieved by the immunization with a chimeric genetic construct encoding the bioactive region of the myostatin fused to the immunostimulatory peptides P2 and P30 from the tetanus toxin delivered by a novel recombinant baculovirus immunization vector. This genetic construct was evaluated in mice as recombinant antigen in DNA vectors, obtaining muscle growth between 20 and 40%.

Baculoviruses have been used as gene transfer vectors and immunization vectors with great success. Baculoviruses with dual gene expression (in insect and mammalian cells) are known as BacMam.

Baculoviruses as a vehicle for immunization may offer certain advantages, they are easier to produce, compared to the production of recombinant proteins or the production of plasmid DNA for genetic vaccines. In this work, we report the development and evaluation of a BacMam as a myostatin immunization vector with the potential to be used in therapeutic and animal production applications.

Keywords: Muscle growth, myostatin, baculovirus, immunization vector.

METABOLIC CHANGES IN HTB 177 LUNG CANCER CELLS IN RESPONSE TO THE ANTICANCER EXTRACT OF *COLEUS HADIENSIS*

Ana Laura Valdez Arellanes^{1,2}, Mónica Azucena Ramírez Cabrera², Emanuel Hernández Núñez³, Eder U. Arredondo Espinoza², Karla Ramírez Estrada^{1*}

¹Laboratorio de Metabolismo Celular. FCQ. UANL. Pedro de Alba S/N, Ciudad Universitaria, San Nicolás de los Garza, N.L. México., ²Lab. Farmacología Molecular y Modelos Biológicos, Posgrado, FCQ, Vicente Guerrero s/n, Treviño, Monterrey, N.L. México., ³Departamento de Recursos del Mar CINVESTAV Mérida. Carretera antigua a Progreso, km 6. CP 97310. Mérida Yucatán México
E-mail: karla.ramirezst@uanl.edu.mx

Plants are a source of bioactive compounds, some of them are of specific interest to the field of medicine. *Coleus hadiensis* is an aromatic semi-succulent plant. Organic extracts of this plant have shown anticancer activity against some types of cancer. This disease is characterized by the uncontrolled growth and spread of tumor cells in the body. Cancer cell metabolic reprogramming supports tumor growth, invasion, and immune escape. It has been reported that plant secondary metabolites can modify this abnormal metabolism and thus help in the treatment of cancer.

The aim of this project was to analyze the anticancer activity of the methanolic extract of *C. hadiensis* in HTB 177 lung cancer cells, and to study the metabolic changes in these cells in response to the extract.

We obtained the extract by maceration of the lyophilized aerial part of the plant with methanol. To study the anticancer activity and cytotoxicity of the extract, HTB 177 and Vero cell lines were grown independently in 96-well plates and exposed to different concentrations of the extract or the reference drug (vincristine) for 48 hours. Cell viability was measured by the MTT method and the inhibitory concentration 50 (IC₅₀) was calculated. The metabolic changes in the cells treated with the IC₅₀ were analysed by 1H-NMR. Three cell extracts were studied: extracellular, intracellular polar and lipophilic.

The *C. hadiensis* extract showed dose-dependent anticancer activity and low cytotoxicity in the healthy cell line. The IC₅₀ for the HTB 177 line was 192.85 µg/mL and for Vero cells it was 618 µg/mL. The main metabolites affected in cancer cells in response to the extract were those related to abnormal cancer metabolism. Our analysis revealed that the *C. hadiensis* extract treatment interfered with the cell energy metabolism, membrane phospholipid synthesis and amino acid metabolism. This suggests a metabolic reprogramming effect in the cell towards normal metabolism in response to *C. hadiensis* extract.

ONE WAY TO EVOLVE C FAMILY DNA POLYMERASES

Andrea Ramón-García¹, Sergio Romero-Romero^{3,4}, Rodrigo Reyes-Lamothe², Silvia Armenta^{1,2}

¹Instituto de Ciencias Agropecuarias, Universidad Km. 1 s/n Ex. Aquetzalpa, 43600 Tulancingo, Hidalgo. ²McGill University, Biology Department, Bellini Life Science Complex, Montreal, Canada. ³Instituto de Fisiología Celular, UNAM, Circuito exterior s/n, Ciudad de México. ⁴Universität Bayreuth, Bayreuth, Germany.
E-mail: ra439728@uaeh.edu.mx, silvia_armenta@uaeh.edu.mx

The DNA polymerases (DNAPs) are the key enzymes of DNA replication. The evolution of polymerases is a complex and ongoing process that involves genetic variations, natural selection, and adaptation to changing enzymatic features. One key aspect of polymerase evolution is the stepwise and cumulative nature of changes, where new features or functionalities are built upon pre-existing ones. The DNA polymerases (DNAPs) are the key enzymes of DNA replication and diverse DNA repair processes; these are distributed in all domains of life¹. To date, the DNAPs have been grouped into different families, using sequence alignments, mentioning PolA, PolB, PolC, PolD, PolX, PolY, and Reverse transcriptase. These different families of polymerases have evolved to perform specific tasks and have unique characteristics that enable them to function efficiently in different cellular contexts^{2,3}. Besides, it has been suggested that DNAPs of known structures could be divided into three groups based on the structural fold of their catalytic sites, which are klenow fold, Pol β -like polymerases, and two barrels fold. DNA polymerase from *E. coli* belongs to polC family and has a Pol β fold, which has some interesting features as they are the fastest DNA polymerases (700 nt/s), display high fidelity, and have very high processivity (50 kb)³. All these features are an exciting field to explore, trying to obtain polymerases useful for different biotechnological applications. Moreover, we are very interested in a better understanding of how DNA replication works; in this sense, having a PolC with different properties as the capacity of incorporation of modified NTPs could be a great option, as well as it could be interesting for many synthetic biology applications. Therefore, this work aims to know the catalytic characteristics and thermostability properties of some replicative polymerases, such as those from *Termus aquaticus*, *Aquifex aeolicus*, *Escherichia coli*, and some previously obtained mutants by establishing a protocol of catalytic activity of these, expressing them in a heterologous system. Previous efforts have been made to enhance the thermostability of these polymerases with limited success.

References

1. Akram, F., Shah, F. I., Ibrar, R., Fatima, T., Haq, I. U., Naseem, W., Gul, M. A., Tehreem, L., & Haider, G. (2023). *Analytical biochemistry*, 671, 115150.
2. Aschenbrenner, J., & Marx, A. (2017). *Current opinion in biotechnology*, 48, 187–195.
3. Johansson, E., & Dixon, N. (2013). *Cold Spring Harbor perspectives in biology*, 5(6), a012799.

ISOLATION AND CHARACTERIZATION OF NATIVE STRAINS FROM LITHIUM-CONTAINING MINING TAILINGS

Renteria-Ortiz, K. S.¹, Medrano-Roldan, H.¹, Zazueta-Alvarez, D. E.², Reyes-Jaquez, D.¹, Rojas-Contreras, J.A.¹

¹TecNM/Instituto Tecnológico de Durango. UPIDET. Blvd. Felipe Pescador 1830 ote. 34080 Durango, Dgo.México.

²Ingeniería en Tecnología Ambiental, Universidad Politécnica de Durango, Carretera Durango-México Km 9.5, Durango, Durango, 34300, México.

*23040244@itdurango.edu.mx

Introduction. Lithium, a silvery-white alkali metal in its pure form, is distinguished by its low density and rapid oxidation upon contact with water or air. It is the lightest metal and has a high melting and boiling point among the alkali metals. In recent years, lithium mining has increased considerably due to the demands of key industries such as aerospace, automotive, electronics, and medical sectors. **Objective:** The main objective of this study was the isolation and characterization of native strains present in lithium-containing mining tailings from Minera Noche Buena, Zacatecas, México. **Methodology:** Initially, an isolation process was carried out using three selective culture media commonly used in bioleaching: LB, API, and 9K. The tailings were incorporated into the medium at a concentration of 10% W/V by serial dilution. The surface spreading technique was used for isolation based on visual morphological differentiation. Each identified bacterium was re-cultured by cross-streaking, obtaining distinct strains. Subsequently, bacterial growth was characterized through kinetics and their macroscopic and microscopic properties were analyzed using the Gram staining technique. **Results:** In each of the selective media, disparate results were obtained. In the 9K medium, no bacterial growth was recorded. On the other hand, in the LB medium, two different strains were identified: PBLB and PRLB. PBLB was characterized by large diameter colonies with a hard texture, grayish-white color, elevation, and stellate shape, while microscopically they were identified as gram-negative bacilli. PRLB exhibited medium diameter colonies with a viscous texture, opaque pink color, no elevation, and wavy shape, also identified as gram-negative bacilli under microscopic examination. In the API medium, two additional strains were isolated: AAPI and BAPI. AAPI was characterized by small diameter colonies, viscous texture, yellow color, circular shape, and absence of elevation, being also identified as gram-negative bacilli in the microscopic analysis. Finally, BAPI presented medium-diameter colonies with a viscous texture, opaque white color, absence of elevation, and stellate shape, and were identified as gram-negative bacilli at the microscopic level. **Conclusions:** The results of this investigation highlight a significant diversity of bacterial strains present in the lithium-containing mine tailings from Minera Noche Buena. These strains, identified as PBLB, PRLB, AAPI, and BAPI, showed preferences for different selective culture media, underscoring the importance of proper media selection for isolation. Additionally, detailed descriptions of the macroscopic and microscopic characteristics of these strains provide valuable information on their morphology and behavior, which may be useful for further research as well as potential industrial applications in the field of bioleaching and lithium extraction.

MODIFYING THE PRODUCT SPECIFICITY BY PROTEIN ENGINEERING OF AN ARCHAEAL CYCLOMALTODEXTRIN GLUCANOTRANSFERASE FROM A DEEP-SEA HYDROTHERMAL VENT

Ana María Rincón-Murillo^{1,2}, Beatriz Velazquez-Cruz², Laura Espinosa-Barrera²,
Sara Centeno-Leija^{3*}, Hugo Serrano-Posada^{3*}

¹Instituto Tecnológico de Colima, Avenida Tecnológico 1, Colonia Liberación, 28976 Villa De Álvarez, Colima, México;

²Laboratorio de Biología Sintética, Estructural y Molecular, Universidad de Colima, Carretera Los Limones-Loma de Juárez, 28627 Colima, Colima, México; ³CONAHCyT, Laboratorio de Biología Sintética, Estructural y Molecular, Universidad de Colima, Carretera Los Limones-Loma de Juárez, 28627 Colima, Colima, México.

Corresponding authors: scenteno0@uacol.mx; hserrano0@uacol.mx

Abstract. Cyclomalto-dextrin glucanotransferases (CGTases) convert glucosyl-intermediates from the starch substrate to functional toroidal-shaped α -(1,4)-linked oligosaccharides, named cyclodextrins (CDs). Previously, our research group identified a novel thermoacidophilic CGTase (PlpA) from unknown archaea by (meta)genome mining of a deep-sea hydrothermal vent microbiome. As expected for extremophilic CGTases, the functional characterization of recombinant native PlpA showed a modest cyclization specificity and high hydrolytic/disproportionation activities from starch, limiting the CDs yield but making it an attractive target for rational protein engineering. Here, we designed a PlpA mutant through structural analysis and docking simulations to improve CDs production. The mutation includes a Gln residue at position 85, which is absent in native PlpA but highly conserved in mesophilic CGTases. The PlpA_{Q85} insertion mutant allowed the formation of a hydrogen bond between the side chain of Gln⁸⁵ and the main chain of Gly¹⁰³, which seems crucial for the function of His¹⁰² at the substrate binding subsite -2. Hence, the recombinant PlpA_{Q85} was obtained, and its functional characterization displayed differences in the product profile compared to native PlpA. This work is an example of rational protein engineering that can be used to play with the specificity of non-classic CGTases for biotechnological applications in the CDs industry, as well as suggests that the product diversification of extremophilic CGTases might be a convenient adaptation to allow archaeal survival in hot starch-poor environments.

SCL and HSP acknowledge financial support from CONAHCyT grants CB-2016-01-285001, INFR-2017-01-280608, CB-2018-A1-S-18011, CF-2019-33-549477, and INFR-2021-17-316456.

PRODUCTION OF A β -1,4-ENDOGLUCANASE ENZYME FROM *BACILLUS SUBTILIS* IN *SACCHAROMYCES CEREVISIAE*

Joel Ríos Alvarado¹, Juan Antonio Rojas Contreras¹, Perla Guadalupe Vázquez Ortega¹,
Ma. Adriana Martínez Prado¹, Javier López Miranda¹, Jesús Bernardo Páez Lerma¹, Norma Urtiz Estrada²

¹Tecnológico Nacional de México/Instituto Tecnológico de Durango, Blvd. Felipe Pescador 1830 Ote. Col. Nueva Vizcaya, 34080 Durango, Dgo., México. ²Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango, Av. Veterinaria S/N, Col. Valle del Sur, 34120 Durango, Dgo., México.
E-mail: jrojas@itdurango.edu.mx Cell phone: 618-109-7457

Bioethanol, which is produced from sugars fermentation obtained from lignocellulosic residues hydrolysis, is an alternative to replace petroleum-derived fuels. The hydrolysis of lignocellulosic residues requires the synergistic action of a group of three enzymes known as cellulases (endoglucanases, exoglucanases and glucosidases), produced by various microorganisms that are responsible for decomposing lignocellulosic biomass to use it as a source of energy. However, ethanologenic yeasts such as *Saccharomyces cerevisiae* do not produce these enzymes. The objective of this work was to generate a *Saccharomyces cerevisiae* strain capable of producing a β -1,4-endoglucanase from a *Bacillus subtilis* strain isolated from a paper effluent. First, codon compatibility of the native enzyme gene (*eglS*) with the translation mechanism of yeast *S. cerevisiae* was determined. An expression vector (named pITD06) containing the *eglS* gene into the *Bam*HI-*Not*I restriction sites of the yeast expression plasmid pYD1 was constructed and inserted by chemical transformation into the *Escherichia coli* BL21SI strain. Surprisingly, *E. coli* was able to produce the enzyme, a finding that has not been previously achieved with the pYD1 plasmid, that already has been reported by this working group (Ríos-Alvarado *et. al.*, 2024). The plasmid was extracted from *E. coli* and also transferred by chemical transformation to the *Saccharomyces cerevisiae* strain EBY100, this genetic background allows the selection of transformants by a tryptophan auxotrophy reversion assay induced when the plasmid is inserted. The ability of the modified strain to degrade carboxymethyl cellulose (CMC) was evaluated in Petri dishes stained with Congo Red dye. The *S. cerevisiae* strain EBY100-pITD06-*eglS* was found to have the expected enzymatic activity and a potency index of 0.9176, at 24 hours. This work concludes that a *S. cerevisiae* strain capable of producing β -1,4- endoglucanase and degrading CMC was obtained.

References

1. Ríos-Alvarado, J., Avitia-Rodríguez, O. N., Urtiz-Estrada, N., Zazueta-Álvarez, D. E., López-Miranda, J., Vázquez-Ortega, P. G., & Rojas-Contreras, J. A. (2024). Expression and characterization of a novel β -1, 4-endoglucanase from *Bacillus subtilis* strain isolated from a pulp and paper mill wastewater. *Protein Expression and Purification*, 106490.

NEWEST INSIGHTS INTO BIXIN BIOSYNTHESIS IN ACHIOTE, *BIXA ORELLANA* L.

Renata Rivera-Madrid¹, José A. Narváez-Zapata², Yair Cárdenas-Conejo³, Margarita Aguilar-Espinosa¹

¹Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, A.C. Calle 43 No. 130, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México. ²Plant Biology and Genome Center, University of California, Davis, CA, USA. ³CONACYT-Universidad de Colima, Colima, México.

*E-mail: renata@cicy.mx

Achiote (*Bixa orellana*) is a highly valued plant for its economic importance in bixin pigment production. For more than three decades, our research group has been focusing on the study of the biosynthesis of bixin. Thus, the first step of bixin biosynthesis involves a CCD oxidative cleavage of lycopene at the 5,6/5',6' double bonds, resulting in bixin aldehyde. A transcriptomic *B. orellana* analysis identified seven CCD gene types, CCD1 and CCD4, potentially involved in bixin synthesis. The production of bixin aldehyde was confirmed by LC-ESI-QTOF-MS/MS analysis of extracts of *E. coli* cells that accumulate lycopene and express the BoCCD1 and BoCCD4 proteins. In vitro assays also revealed the presence of bixin aldehyde, suggesting that cleavage activity at the 5,6/5',6' bonds of lycopene is present in all BoCCD1 and BoCCD4 proteins analyzed. Interestingly, in vivo, only four BoCCDs convert bixin aldehyde to norbixin, the second product of the bixin pathway. In order to conduct a deep analysis of the candidate bixin synthesis, we carried out the RNAseq analysis data from three achiote morphotypes using three developmental stages of achiote seeds. After enrichment analysis, our investigation revealed a crucial role of triterpenes, sesquiterpenes, and cuticular wax production pathways. We also found genes associated with these three pathways by WGCNA analysis, which strongly correlates with the amount of bixin. TPM and correlation analysis probed the significance of these particular genes in further detail. The study results emphasized the complexity of our findings by demonstrating the intricate role of genes associated with the triterpene and carotenoid pathways in the growth glands of cells and the accumulation of bixin in the achiote seeds. This suggests that isoprenoid formation is required for chemicals in growing seeds' reddish latex. In the last stage of seed development, there was a strong correlation between bixin and the BoCCD4 gene and certain BoALDH and BoMET gene members. This suggests that several genes may have a role in synthesizing apocarotenoids. The results of our study suggest that the synthesis of bixin involves the participation of several genes and metabolic pathways, making it a complex process. Our findings in this regard may contribute to a better understanding of this pigment's biosynthetic pathway and enable us to explore its potential applications in various industries.

ISOLATION AND FUNCTIONAL CHARACTERIZATION OF COLLAGEN FROM FISH SCALES

Crisalejandra Rivera-Pérez¹, Julie Martin

¹Centro de Investigaciones Biológicas del Noroeste, S.C., Instituto Politécnico Nacional 195, La Paz, BCS, C.P. 23096, México.
E-mail: crivera@cibnor.mx

Fish scales, often regarded as waste in the fishing industry, represent a significant source of discards. Annually, millions of tons of fish scales are discarded worldwide. However, this by-product holds great biotechnological potential due to its high collagen content, especially type I collagen. Type I collagen, abundant in fish scales, is a fibrillar protein composed of three polypeptide chains forming a triple helical structure. This type of collagen is like that found in mammals and plays a crucial role in the formation of connective tissues. Extracting and purifying type I collagen from fish scales is not only viable but also highly efficient and sustainable. Also, collagen and its hydrolysate have applications in the health and beauty industry and are a rich source of bioactive peptides with antioxidant and antimicrobial properties. These peptides are generated during the hydrolysis with exogenous enzymes of collagen and have shown considerable biotechnological potential. In this study, collagen was isolated from the scales of a commercial fish, *Seriola rivoliana*, using acetic acid and salting out precipitation. The isolated protein was confirmed as collagen by UV absorption spectrum at wavelengths of 220–240 nm, which was mainly attributed to peptide bond absorptions by $n \rightarrow \pi^*$ transitions of the groups of C=O, -COOH, and CONH₂ in the collagen polypeptides chains and, collagen was evaluated by SDS-PAGE. Collagen peptides were produced by hydrolysis with shrimp enzyme extract and wobenzym, respectively. The generated collagen peptides, using different units of activity, were assessed for antioxidant activity using DPPH for scavenging activity and antimicrobial activity was tested against *E. coli*, *Pseudomonas spp.*, *Salmonella spp.*, *V. diabolicus*, *V. parahemolyticus* and, *Photobacterium* using de radial diffusion method. Overall, collagen from the scales of *S. rivoliana* has promising functional activities.

METHODOLOGICAL STRATEGIES FOR BIOSYNTHESIS AND CHARACTERIZATION OF IRON NANOPARTICLES USING *TRICHODERMA HARZIANUM*

Angeles Rivera Román¹, Sergio Hernández-León¹, Oscar Arce-Cervantes¹, Silvia Armenta¹

¹Instituto de Ciencias Agropecuarias, Avenida Universidad Km. 1 s/n Exhacienda Aquetzalpa, 43600 Tulancingo, Hgo.

E-mail: ri379069@uaeh.edu.mx, silvia_armenta@uaeh.edu.mx

The use of microorganisms in nanobiotechnology is associated with the production of various secondary metabolites and bioactive compounds, which can be used as reducing, stabilizing, and capping agents during nanoparticle biosynthesis. Currently, there are publications^{1,2} that discuss the use of fungi to produce metal nanoparticles such as gold, silver, zinc, and iron. This approach involves using both extracellular and intracellular fractions to optimize the synthesis process and affect the characteristics, properties and potential applications of nanoparticles. It is considered a promising eco-friendly alternative to traditional synthesis methods³. *Trichoderma harzianum*, a fungus known for its rapid propagation and non-pathogenic nature, holds significant importance in agriculture by promoting plant growth and serving as a biocontrol agent against various plant pathogens. This makes it an intriguing candidate for nanoparticle biosynthesis. The development of methods for synthesizing nanoparticles using fungi necessitates understanding the fungal growth stage and identifying relevant reducing enzymes that play a role in metal ion reduction and nanoparticle stabilization. Therefore, the objective of this research is to establish optimized methodologies to evaluate the use of *T. harzianum* for the biosynthesis of iron nanoparticles (FeNPs) to subsequently evaluate its potential as an antimicrobial agent, as well as its characterization by UV-VIS spectroscopy, Scanning Electron Microscopy (SEM), Transmission Electron Microscopy (TEM) and Fourier Transform Infrared Spectroscopy (FTIR). The results of this study will provide valuable insights into the use of *T. harzianum* for the biosynthesis of iron nanoparticles and expand our understanding of the potential applications of these nanoparticles in various fields such as agriculture, biomedicine and environmental remediation.

References

1. Murillo-Rábago, E.I., Vilchis-Nestor, A.R., Juárez-Moreno, K., García-Marin, L.E., Quester, K. & Castro-Longoria, E. (2022). *Antibiotics*, 11, 1-19.
2. Molnár, Z., Bódi, V., Szakacs, G., Erdélyi, B., Fogarassy, Z., Sáfrán, G., Varga, T., Kónya, Z., Tóth-Szeles, E., Szucs, R. & Lagzi, I. (2018). *Scientific reports*, 8, 1-12.
3. Ramírez-Valdespino, C. & Orrantia-Borunda, E. (2021). *Frontiers in Fungal Biology*, 2, 1-16.

FACILE SYNTHESIS OF PEGYLATED Fe^0 AU HETERONANOPARTICLES, AS HIGH POTENTIAL BIOMEDICAL USE

¹Jose J Olivares-Trejo, ²Romero-Ibarra Josué, ³Fabiola V. Borbolla-Jiménez, ⁴Jonathan J. Magaña, ¹Rivera Soto Maria Ana

¹Universidad Autónoma de la Ciudad de México, ²Instituto de Investigaciones en Materiales, (UNAM) CDMX, ³Laboratorio de Medicina Genómica, Departamento de Genética (CENIAQ), Instituto Nacional de Rehabilitación-Luis Guillermo Ibarra Ibarra (INR-LGII) CDMX, ⁴Programa de Ciencias Biomédicas, Facultad de Medicina, (UNAM) CDMX.

Correspondence: jose.olivares@uacm.edu.mx, maria.ana.rivera.soto@uacm.edu.mx.

Synthesis of PEGylated Fe Au nanoparticles (Np's) is an increasingly topic given its great importance in the development of new smart drugs and their biomedical potential uses (magnetic, optical, radio imaging, and cell cancer radiosensitize) ¹⁻³. We synthesize an innocuous Np's PEGylated Fe^0 Au Np's by chemical NaBH_4 -reduction. UV-Vis, HRTEM, SEM, XPS and PPMS for Np's characterization. Functionalization assay and cell morphology (MTT and Confocal Microscopy) was performed in A549 cell line Np's exposed, in a time dose-response curve with concentrations 2-500 $\mu\text{g}/\text{mL}$ in 2-48h. Characterization of Np's showed Uv-Vis peaks at 320 nm and 370 nm and an absorbance band at 560 nm confirming Fe and Au Np's respectively. By XPS low B concentration (14 wt%) was determine.)(National Center for Biotechnology Information (2024) as the presence of $\text{Fe}^0/\text{Fe}_2\text{O}_3$ with 4.35 wt % and 27.67 wt % ⁸; Au^0 presents 4.2 wt %. SEM determined spherical agglomerates in ranges from 40-300 nm, EDS analysis observed Fe/Au ratio ~40 in wt. %; FFT in HRTEM identify $\text{Fe}^0/\text{Fe}_2\text{O}_3$, Au in a BCC and FCC crystal structure and a low contrast of the shell corresponds to PEG Np's was determined as heteronanocompound. PPMS showed 75 $\text{A m}^2\text{kg}^{-1}$ magnetization value in PEGylated iron gold heteronanocompounds ⁹⁻¹¹. Cellular viability, in cells expose at higher concentrations 200- 500 $\mu\text{g}/\text{mL}$ results in a significant decrease of viability (< 50%) after 24, 48h, and in lower exposition at 2- 5 $\mu\text{g}/\text{mL}$ presents higher viability despite time (~85v %.). Lower Np's concentration exposition (2-5 $\mu\text{g}/\text{mL}$) presents higher viability. Therefore only 5 $\mu\text{g}/\text{mL}$ concentration was chosen as the concentration for the cell morphology assay, exposition was performed in a time lapse curve (2- 48h), were after 24h and 48 of exposition, cells begin to display ruffles, which could be indicative of increased cellular motility and a round morphology cytoskeleton due to cellular stress, in the assay was no cell mortality indicators. Due the optical, magnetic properties, the high cell viability, and the small amount of damage or death indicators in cell morphology assay we propose as high potential for biomedical use (hyperthermia, photothermal therapy, radio sensitizing) Pegylated $\text{Fe}^0/\text{Fe}_2\text{O}_3$, Au heteronanocompunds in a 5 $\mu\text{g}/\text{mL}$ concentration.

References

1. Slama, Y. et. al. Evaluation of core-shell $\text{Fe}_3\text{O}_4@Au$ nanoparticles as radioenhancer in A549 cell lung cancer model. *Heliyon* 10, e29297 (2024).
2. Bansal, S. A., Kumar, V., Karimi, J., Singh, A. P. & Kumar, S. Role of gold nanoparticles in advanced biomedical applications. *Nanoscale Advances* vol. 2 3764–3787 Preprint at <https://doi.org/10.1039/d0na00472c> (2020).

3. Musielak, M. et. al.. Methodological and Cellular Factors Affecting the Magnitude of Breast Cancer and Normal Cell Radiosensitization Using Gold Nanoparticles. *Int J Nanomedicine* 18, 3825–3850 (2023).
4. Sun, Y. P., Li, X. Q., Zhang, W. X. & Wang, H. P. A method for the preparation of stable dispersion of zero-valent iron nanoparticles. *Colloids Surf A Physicochem Eng Asp* 308, 60–66 (2007).
5. Glavee, G. N., Klabunde, K. J., Sorensen, C. M. & Hadjipanayis, G. C. Chemistry of borohydride reduction of iron(II) and iron(III) ions in aqueous and nonaqueous media. Formation of nanoscale Fe, FeB, and Fe₂B powders. *Inorg Chem* 34, 28–35 (1995).
6. Al-kazazz, F. F. M., Al-imarrah, K. a F., Al-, I. A., Agelmashotjafar, L. & Abdul-majeed, B. A. A Simple Method for Synthesis, Purification and concentration Stabilized Goldnanoparticles. *Int J Eng Res Appl* 3, 21–30 (2013).
7. National Center for Biotechnology Information(2024). PubChem Compound Summary for CID 10219853, S. Borate. R. Sodium Borate. <https://pubchem.ncbi.nlm.nih.gov/compound/Sodium-Borate#section=Information-Sources> (2024).
8. Liu, A., Liu, J. & Zhang, W. xian. Transformation and composition evolution of nanoscale zero valent iron (nZVI) synthesized by borohydride reduction in static water. *Chemosphere* 119, 1068–1074 (2015).
9. Bedanta, S. & Kleemann, W. Supermagnetism. *J Phys D Appl Phys* 42, 013001 (2009).
10. S P Gubin, Yu A Koksharov, G B Khomutov, G. Y. Yurkov. Magnetic nanoparticles: preparation, structure and properties. *Russian Chemical Reviews* 489, (2005).
11. Issa, B., Obaidat, I. M., Albiss, B. A. & Haik, Y. Magnetic nanoparticles: Surface effects and properties related to biomedicine applications. *International Journal of Molecular Sciences* vol. 14 21266–21305 Preprint at <https://doi.org/10.3390/ijms141121266> (2013).
12. Rivas-García, L. et. al.. Ultra-small iron nanoparticles target mitochondria inducing autophagy, acting on mitochondrial dna and reducing respiration. *Pharmaceutics* 13, 1–18 (2021).

These results are consistent with those obtained by TEM and with previous experiments that revealed a faster solubilization of Venofer . These results are consistent with those obtained by TEM and with previous experiments that revealed a faster solubilization of Venofer® than the synthetic FeNPs. Such solubiliza-® than the synthetic FeNPs.

Such solubilization might increase the bioavailability of ionic Fe to get into the mitochondria once the NPs have been internalized as such, as can be observed in Figure 1. It is tion might increase the bioavailability of ionic Fe to get into the mitochondria once the NPs have been internalized as such, as can be observed in Figure1. It is known that iron is transported to the mitochondria, where it is utilized for synthesis of cofactors essentials for the function of enzymes involved in oxidation-reduction reactions, DNA synthesis and repair, and a variety of other cellular processes. Nowadays, the trafficking of iron to the mitochondria and normal mitochondrial iron metabolism, including heme synthesis and iron-sulfur cluster biogenesis, are being investigated [20].¹²

TREATMENT OF DAIRY WASTEWATER USING NATURAL COAGULANTS DERIVED FROM AGRO-INDUSTRIAL WASTES

Ana R. Rocha-Vallejo, Elizabeth Quintana-Rodríguez, David A. Camarena-Pozos

Centro de Innovación Aplicada en Tecnologías Competitivas (CIATEC)

Omega 201, Industrial Delta, 37545 León, Guanajuato, México.

E-mail: arocha.estudiantepicyt@ciatec.mx

Dairy industry in Guanajuato, Mexico has grown considerably in the last decades generating about 1,900,000 liters of wastewater per year¹. These effluents are characterized by their high content of chemical oxygen demand (COD), fats, oils and grease (FOG), total solids (TS) and variable pH². For the treatment of these effluents, the coagulation-flocculation process is used as primary treatment, using traditional chemical coagulants such as aluminum sulfate³. This coagulant causes negative impacts on the environment, since the treated water and the sludge generated contain traces of aluminum that can accumulate in groundwater and soil when used as irrigation or fertilizer^{4,5}. The objective of this project is to evaluate alternatives for wastewater treatment by coagulation-flocculation using natural coagulants derived from agro-industrial wastes.

For this study, *Opuntia ficus indica* and *Agave salmiana* residues were used, which were collected from the nopal and mezcal producing industries in the municipalities of Salamanca and San Luis de la Paz. These materials have a high content of carbohydrates and electrolytes such as Ca⁺², Mg⁺², K⁺¹ and Na⁺¹, which makes them candidates for use as natural coagulants^{4,6}. Preparation and extraction of the coagulants was carried out and characterized by quantification of total carbohydrates, identification of functional groups -OH, -COOH, NH₂ by Fourier transform infrared spectroscopy (FTIR) and measurement of the point of zero charge (PZC). For the evaluation of the coagulants, wastewater from a cheese manufacturing company was used. The wastewater concentration averaged 17,828 ppm COD, 3,877 ppm FOG, 15,666 ppm ST (n=3) and a variable pH of 5-11. Jar Tests were performed using as control a chemical coagulant of aluminum sulfate and a natural coagulant such as chitosan. Three concentrations were used for the *O. ficus indica* coagulant (0.5 g/L, 1 g/L and 1.5 g/L) and for the *A. salmiana* coagulant (1 g/L, 2 g/L, 3 g/L). According to the PZC, the jar tests were performed at pH=4 which showed a higher clarification of the wastewater and floc formation.

The results showed an average removal efficiency of COD, FOG and ST using the *O. ficus indica* coagulant of 48% at a dose of 1.5g/L, and for the *A. salmiana* extract of 40% at a dose of 3g/L, compared to 54% with the aluminum sulfate coagulant and 46% with the natural coagulant chitosan.

References

1. Servicio de Información Agroalimentaria y Pesquera (SIAP). (2022). Anuario Estadístico de la Producción Ganadera.
2. Ganta A, et. al.. (2022). *Energies* 15(23):9084.
3. Diver D, Nhapi I, Ruziwa WR. (2023). *Environmental Advances* 13:100421
4. Nimesha S, et. al.. (2022). *Global Journal of Environmental Science and Management* 8(1):101-116.
5. Kurniawan SB, et. al..(2020). *Int J Environ Res Public Health*. 17(24):1-33
6. Urias-Orona V, et. al.. (2010). *Int J Mol Sci*. 11(10):3686-3695

MODIFICATION OF NOPAL PECTIN WITH QUATERNARY AMMONIUM SALTS TO GIVE IT ANTIBACTERIAL, ANTICANCER AND BIOCOMPATIBILITY PROPERTIES

Rodríguez Amaya Tania Milena¹, Luis Enrique Cobos Puc¹, Silva Belmares Sonia Yesenia¹, Zugasti Cruz Alejandro¹, Enríquez Medrano Francisco Javier², Oyervides Muñoz Ernesto¹

¹Facultad de Ciencias Químicas de la Universidad Autónoma de Coahuila.

²Centro de Investigación en Química Aplicada

taniamilenarodriguezamaya@gmail.com / milena-rodriguez@uadec.edu.mx

Summary. Biopolymers have been of great importance, which is why their physical and chemical characteristics have been studied. Among biopolymers, pectin stands out because it has many applications in industry, but it is very prone to bacterial contamination, which is why modifications to its structure are required to improve its biological properties. In this work, two ammonium salts (1-(2-aminoethyl) trimethylammonium bromide and 1-(2-aminoethyl) pyridinium bromide) were synthesized using quaternization reactions and subsequently grafting these salts to pectin previously oxidized with periodate. of sodium. These new pectin derivatives were characterized by FTIR (Fourier Transform Infrared Spectroscopy) where the functional groups of the molecules could be appreciated and complemented with the NMR (Nuclear Magnetic Resonance) technique. The products obtained were also evaluated for their hemolytic, antibacterial and anticancer properties. For the antibacterial tests, the following strains were used: *S.aureus* ATCC 6538, *S.aureus* ATCC33592, *E.coli* ATCC 11229, *P. Aeruginosa* ATCC 15442, *Enterobacter* and *C.albicans*, *Fecalis*, obtaining an antimicrobial material. For anticancer tests, we worked with 3T3 and MCF7, demonstrating that these pectin derivatives inhibit cell proliferation of these cancer cell lines. Finally, the results of hemolytic tests demonstrated that these materials are biocompatible with human blood erythrocytes.

Keywords: pectin, quaternary salts, antibacterial, anticancer.

References

1. Alonso-Montemayor F., Zugasti-Cruz A. "Minimum inhibitory but maximum non-hemolytic concentration of plasma-treated catheters coated with *L. tridentata* and *O. vulgare* extracts", *Plasma Science and Technology*, Plasma Sci. Technol. 25 (2023) 125503 (14pp).
2. Rivera Álvarez A., Oyervides Muñoz E. "Modificación de pectina con 4-cloroanilina para otorgarle propiedades antibacteriales." *CienciAcierta*, No. 65 enero-marzo 2021.
3. Sharma, R., & Ahuja, M. (2011). "Thiolated pectin: Synthesis, characterization and evaluation as a mucoadhesive polymer". *Carbohydrate Polymers*, 85(3), 658-663.

EFFECT OF PROLACTIN AND 17 β -ESTRADIOL ON THE ADHESION AND INTRACELLULAR PERSISTENCE OF *STAPHYLOCOCCUS AUREUS* IN BOVINE MAMMARY EPITHELIAL CELLS

Gladys Romero-Corona¹, Marisol Báez-Magaña¹, Joel Edmundo López-Meza¹ and Alejandra Ochoa-Zarzosa¹

¹Centro Multidisciplinario de Estudios en Biotecnología, Facultad de Medicina Veterinaria y Zootecnia, Universidad Michoacana de San Nicolás de Hidalgo, Km 9.5 Carretera Morelia-Zinapécuaro, Posta Veterinaria. 58893, Morelia, Michoacán.

1237805c@umich.mx

Bovine mastitis (BM) is a mammary gland disease that causes an inflammatory response, causing significant economic losses in dairy farms worldwide. *Staphylococcus aureus* is the leading causal agent of subclinical MB, responsible for up to 70% of cases [1]. During the peripartum and lactation periods, hormonal fluctuations can compromise the animal's innate immune response (IIR), increasing the infection susceptibility. Prolactin (PRL) and 17 β -estradiol (E2) are the essential hormones associated with these changes. Bovine mammary epithelial cells (bMECs) are the target of these hormones, regulating different physiological processes, including milk production. Previously, we reported that physiological concentrations of bovine PRL (bPRL 5 ng/mL) and E2 (50 pg/mL) reduced *S. aureus* internalization into bMECs by ~40% [2]. In this work, the effects of both hormones on *S. aureus* persistence, adhesion, and virulence in bMECs were analyzed by two approaches: 1) bMECs treated 24 h with the hormones (bPRL, E2, and E2+bPRL) before *S. aureus* infection, and 2) *S. aureus* treated with the hormones (bPRL, E2, and E2+bPRL) for 2 h and then used to infected bMECs. The results showed that bMECs treated with the hormone combination (E2+bPRL) decreased *S. aureus* adhesion (~50%) and persistence (~80%). Also, *S. aureus* pretreated with the hormone combination (E2+PRL) increased internalization (~70%) and decreased adhesion (~64%) into bMECs at 2 h. Besides, to evaluate if biofilm production was regulated for hormones and its relationship with persistence and internalization, *S. aureus* was hormone-treated during 2–24 h. Results showed that *S. aureus* treated with the hormone combination decreased biofilm formation (46%) at 2 h. This effect coincides with the up-regulation of *srdC* expression (~9.5-fold), a gene related to adhesion and sepsis. Also, gene global regulators of adhesion and virulence were down-regulated, such as *RNAIII* (~0.9-fold) and *agr* (~0.8-fold) at 2 h. These results suggest that hormones affect the adhesion, internalization, and persistence of *S. aureus* in bMECs by regulating the expression of genes associated with virulence factors in *S. aureus*.

References

1. Kerro-Dego, O., Dijk, J. E. & Nederbragt, H. (2003). *The Veterinary Quarterly*, vol 24(4), 181-198.
2. Salgado-Lora, M. G., Medina-Estrada, I., López-Meza, J. E., & Ochoa-Zarzosa, A. (2020). *Pathogens (Basel, Switzerland)*, vol 9(7), 520.

STUDY OF INTERACTION MECHANISMS BETWEEN *PLEUROTUS OSTREATUS* AND HEAVY METALS (CHROMIUM AND LEAD) IN CULTIVATION MEDIA PRIMARILY COMPOSED OF LIGNOCELLULOSIC RESIDUES

Pamela Romo-Rodríguez*, Tania Marieel Ovalle-Ruiz¹, Oscar Abraham Flores-Amaro²,
Laura Yamamoto-Flores² and Francisco Javier Avelar-González²

¹Fungal Biotechnology Laboratory, Tecnológico Nacional de México Campus Pabellón de Arteaga, Carretera a la Estación de Rincón Km. 1, Pabellón de Arteaga 20670, Mexico. ²Environmental Sciences Laboratory, Department of Physiology and Pharmacology, Universidad Autónoma de Aguascalientes, Av. Universidad 940, Col. Cd. Universitaria, 20131, Aguascalientes, Mexico.

*E-mail: pamela.rr@pabellon.tecnm.mx

In the context of addressing the critical issue of heavy metal pollution, this research investigates the interaction mechanisms between *Pleurotus ostreatus* and heavy metals, specifically chromium and lead, within a liquid cultivation medium predominantly composed of pulverized lignocellulosic residues. The study underscores the importance of developing alternatives for the removal of heavy metals from the environment. Key findings reveal the remarkable capability of *P. ostreatus* to actively adsorb and remove heavy metals from the liquid medium. Particularly noteworthy is the chemical reduction observed in a significant portion of the chromium concentration, transforming it from its hexavalent form to the less toxic trivalent state. The interaction primarily operates through mechanisms of absorption and chemical reduction. These results contribute valuable insights into the potential application of *P. ostreatus* in the remediation of environments contaminated with heavy metals, emphasizing its dual role in absorption and chemical transformation. The study highlights the biotechnological promise of harnessing fungal interactions as an effective and sustainable solution for addressing the environmental challenges posed by heavy metal pollution.

IMPROVING XYLOSE METABOLISM IN SACCHAROMYCES CEREVISIAE

Jaime Rosas Díaz, Luis Caspeta Guadarrama

Instituto de Biotecnología. Universidad Nacional Autónoma de México.
Av. Universidad 2001, Chamilpa, CP 62210, Cuernavaca, Morelos.
Tel. 7773291601. jaime.rosas@ibt.unam.mx, luis.caspeta@ibt.unam.mx

The lignocellulosic biomass is a polymeric material composed of cellulose, hemicellulose, and lignin. After its hydrolysis, the most abundant monosaccharides are glucose and xylose, with roughly 50% and 30%, respectively. Therefore, this biomass is of great interest as it can serve as a sustainable raw material to produce biofuels and chemicals through microbial fermentation.

The yeast *Saccharomyces cerevisiae* is one of the most promising microbial strains for the conversion of lignocellulosic biomass. This yeast has been widely used to produce ethanol from glucose. However, it does not naturally catabolize the xylose. To overcome this issue, different heterologous pathways for its assimilation have been introduced, with the isomerase pathway being the most used. However, although the genetically modified *S. cerevisiae* strains are capable of xylose assimilation, they lack the ability to simultaneously ferment glucose and xylose. Recently, some independent studies have found that mutations in the Ras-cAMP-PKA signaling pathway contribute to enhance the xylose assimilation in the presence of glucose (Tran Nguyen Hoang *et. al.*, 2018; Sato *et. al.*, 2016; Wu *et. al.*, 2020).

In the present study, we focused on assembling a plasmid containing the xylose isomerase and xylulose kinase to transform the *S. cerevisiae* strain W3031A mutant in *GRE3* to generate the strain XYLEV, in which xylose consumption was improved by adaptive laboratory evolution. Strain XYLEV was then mutated in *ASC1*, which belong to the Ras-cAMP-PKA pathway, with the purpose of evaluating their effect on the co-fermentation of glucose and xylose. Analyzing how mutations in the Ras-cAMP-PKA signaling pathway influence xylose metabolism in the presence of glucose will provide new insights into how *S. cerevisiae* reprograms glucose signaling pathways for the co-consumption of different carbon sources.

References

1. Tran Nguyen Hoang, P., Ko, J. K., Gong, G., Um, Y., & Lee, S. M. (2018). Genomic and phenotypic characterization of a refactored xylose-utilizing *Saccharomyces cerevisiae* strain for lignocellulosic biofuel production. *Biotechnology for biofuels*, 11, 268
2. Sato, T. K., Tremaine, M., Parreiras, L. S., Hebert, A. S., Myers, K. S., Higbee, A. J., Sardi, M., McIlwain, S. J., Ong, I. M., Breuer, R. J., Avanesi Narasimhan, R., McGee, M. A., Dickinson, Q., La Reau, A., Xie, D., Tian, M., Reed, J. L., Zhang, Y., Coon, J. J., Hittinger, C. T., ... Landick, R. (2016). Directed Evolution Reveals Unexpected Epistatic Interactions That Alter Metabolic Regulation and Enable Anaerobic Xylose Use by *Saccharomyces cerevisiae*. *PLoS genetics*, 12(10), e1006372.
3. Wu, M., Li, H., Wei, S., Wu, H., Wu, X., Bao, X., Hou, J., Liu, W., & Shen, Y. (2020). Simulating Extracellular Glucose Signals Enhances Xylose Metabolism in Recombinant *Saccharomyces cerevisiae*. *Microorganisms*, 8(1), 100.

EVALUATION OF THE INTERACTION BETWEEN LICHENIC BACTERIA AND NON-LICHENIC FUNGI

Julieta Rosas-Vallejo¹, Cynthia Paola Rangel-Chávez¹, Divanery Rodríguez-Gómez¹ and Varinia López-Ramírez¹

¹Tecnológico Nacional de México/ ITS de Irapuato., Irapuato–Silao km 12.5 Col. El Copal. C.P. 36821.
varinia.lr@irapuato.tecnm.mx

Lichens are symbiotic consortia between a photobiont organism and a mycobiont. This stable and self-sufficient symbiosis allows them their survival in different environments. Various studies have reported the presence of bacteria associated with this symbiosis such as *Acinetobacter* and *Bacillus*, however, the role they play in the symbiosis has not been described yet. The objective of this study was to understand whether *Bacillus pumilus*, *Acinetobacter lwoffii*, *Caulobacter mirabilis*, *Paenibacillus cineri*, *Curtobacterium allii*, *Klebsiella pasteurii*, *Stutzerimonas sp.* Can interact with non-lichenizing fungi of the Ascomycota division such as *Aspergillus fumigatus*, *Penicillium catarractarum*, *Alternaria alternata*, *Penicillium subrubescens*, *Talaromyces pinophilus*, *Fusarium oxysporum* and *Trichoderma asperellum*, and the type of biological interaction that exists among them is determined. Confrontation assays make it possible to evaluate the nature of the biological interaction between various microorganisms, whether bacteria promote or inhibit fungal growth. In this work, diffusion tests were carried out in Petri plates with agar. The first test consisted of the inoculation in the center of a plate of the fungus and then placing a drop of four different bacterial inoculums around it. For the second confrontation test, those antagonistic interactions were selected by inoculating the fungus at one end and of the Petri dish and a bacterial inoculum at the other. The results of the first trial showed that lichen-associated bacteria have mostly antagonistic interactions with non-lichenizing fungi. It was found that 60% of cases bacteria inhibited fungal growth, which suggests that during the interaction both organisms compete for essential nutrients from the environment. The antagonism and phenotypic changes resulting from the assays will allow us to understand the mechanisms underlying these interactions. This information may be useful to develop new strategies to control fungal diseases.

IN VITRO ANTIFUNGAL ACTIVITY OF ETHANOLIC EXTRACTS FROM LEAF AND STEM RESIDUES OF MEXICAN OREGANO (*LIPPIA GRAVEOLENS* KUNTH) ON *FUSARIUM* SPP.

Estela Ruiz-Baca^{1*}, Martha Rosales-Castro², Leobardo Ibarra-Sánchez¹, Guillermo Moreno-Gándara¹, Sandra J. Velázquez-Huizar¹, Angélica López-Rodríguez¹, Iván Meneses-Morales¹, Alejandra García-Núñez¹, Juan P. Cabral-Miramontes¹, Ana Lilia Martínez-Rocha³

¹University Juárez of Durango State, Faculty of Chemistry Sciences. *Av. veterinaria s/n, Circuito Universitario, Col. Valles del Sur, C.P. 34120, Durango, Dgo. México. Tel: +52 6188271340. ²Interdisciplinary Research Center for Regional Integral Development CIIDIR-IPN Durango Unit ³ University of Guanajuato, Division of Natural and Exact Sciences, Department of Biology. *E-mail: eruiz@ujed.mx

The genus *Fusarium* contains several phytopathogenic species that cause substantial economic and yield losses in alimentary base crops [1]. Natural products based on plant extracts can be an option for sustainable management to control such pathogens. Therefore, in the search for new compounds that are easily accessible to the population, an alternative is the use of plants such as Mexican oregano (*Lippia graveolens* Kunth). Oregano leaves and stems no longer have commercial value once the essential oils are extracted. However, these remnant oregano leaves and stems contain flavonoids and phenolic acids with antifungal potential [2]. This work aimed to evaluate the *in vitro* antifungal effect of ethanolic extracts of leaf and stem residues of Mexican oregano on *Fusarium graminearum*, *Fusarium solani*, and *Fusarium oxysporum* f. sp. *lycopersici*. Ethanolic extracts of leaf and stem residues of Mexican oregano were obtained by maceration with aqueous ethanol 70:30 for 48 h, filtered, and taken to a rotary evaporator with a vacuum at 50°C and subsequently dried by freeze-drying. The antifungal activity of these extracts was carried out using 3 µL of a suspension of 1X10⁶ conidia/mL of the fungus inoculated on potato dextrose agar (PDA) medium and the oregano extracts (4, 8, and 16 mg/mL) and incubated at 28 °C. Tebuconazole was used as a growth control. Radial mycelial growth was quantified at 120 h. The inhibition of fungal growth was observed with leaf and stem residue extracts starting at a concentration of 4 mg/mL. At a concentration of 16 mg/mL, inhibition rates of 100% and 94% were observed for *F. graminearum*, respectively. While inhibition rates of 100% and 87% were observed for *F. oxysporum*, and 92% y 77%, were observed for *F. oxysporum*. Ethanolic extracts of leaf and stem residues of Mexican oregano demonstrated antifungal activity on *Fusarium* spp. Therefore, this could be an excellent alternative to control such pathogens.

References

1. Ríos-Saucedo, C.J., Ramírez-Valadez, G., Santana-Espinoza, M., Martínez-Fernández, S.M., Rosales-Serna, R. (2022). IntechOpen.
2. Frías-Zepeda, M.E., Rosales-Castro, M. (2021). Revista Chapingo Serie Ciencias Forestales y del Ambiente, 27(3), 367–381.

ANTIFUNGAL ACTIVITY OF *ERIGERON CANADENSIS* AND ITS ESTABLISHMENT OF CALLUS *IN VITRO* CULTURE

Andrea Ruiz-Betancourt¹, Ricardo Cocoltzi-Gutierrez¹, Karen Estefes-Carranza¹,
Virginia Mandujano González², Mariana Sánchez Ramos¹, Francisco Cruz Sosa¹

¹Universidad Autónoma Metropolitana Iztapalapa. Av. San Rafael Atlixco 186, Leyes de Reforma 1a secc. Iztapalapa, Ciudad de México, 09340. ²Universidad Tecnológica de Corregidora, Carretera estatal 413 Km 11.2, Santa Bárbara, Corregidora, Querétaro, 76924.
E-mail: cbs2223801912@izt.uam.mx

Erigeron canadensis is an invasive species and glyphosate resistant, well-known as arrocillo. It has been used in traditional medicine to treat stomach diseases and fever¹. Some essential oils and derivatives of cinnamic and benzoic acids were identified, also the activities related to these compounds are repellent (*Aedes aegypti*)² and antifungal (*Aspergillus flavus*)³. The present script aims of demonstrating antimicrobial activity of three organic extracts and establish the protocols to obtain callus cultures capable of producing bioactive compounds. The plant material was collected in Tetela del Volcán, Morelos, the leaves and stems were dried separately, and the extraction was performed by maceration with hexane, ethyl acetate and methanol (from lowest to highest polarity), they were revised by Thin Layer Chromatography (TLC) and tested through Kirby-Bauer method for antimicrobial activity. For the callus culture establishment, the fruits collected from the wild plant in Morelos were disinfected with ethanol 70% and sodium hypochlorite 1%, they were placed on MS media at 25°C with 16 hours of light; leaf, stem and root of plantlets were used as explants to induce callus with Picloram (PIC), 6-Benzylaminopurine (BAP) and Kinetin (KIN). The TLC of extracts shows different compounds in each extract with different polarity. The extracts with lower polarity from the leaf have antimicrobial activity against yeast *Candida albicans*, in concentrations of 100 and 50 mg/ml of hexane extract and 100 mg/ml of ethyl acetate extract, only the 100 mg/ml of leaf-hexane extract has an antimicrobial effect similar to fluconazole at 2 mg/ml. Regarding callus induction, treatments with 0.1 mg/L of PIC and 1 or 2 mg/L of BAP formed callus able to survive a constant change of media culture, in both cases the explant were internode, and the rest of the treatments and explants showed the formation of callus but it wasn't survived for one month of culture or died after replanting. Finally, TLC profiles of wild plants and callus showed some differences about the compounds produced *ex vitro* and *in vitro*. In conclusion, *Erigeron canadensis* could have antifungal compounds of slightly polar nature and could be found mainly in the wild plant but in the callus culture too, in less quantity. This species needs more investigation to discover the compounds in each extract and their antimicrobial activity, also increase the production of callus to obtain antifungal compounds in bigger quantities and constantly in a laboratory.

References

1. Argueta, A., & Gallardo, M. C. (1994). Atlas de las Plantas de la Medicina Tradicional Mexicana (A. Argueta & L. Cano, Eds.; 1a ed.). Instituto Nacional Indigenista.
2. Azeem, M., Zaman, T., Tahir, M., Haris, A., Iqbal, Z., Binyameen, M., Nazir, A., Shad, S. A., Majeed, S., & Mozūraitis, R. (2019). Industrial Crops and Products, 140, 111609.
3. Azeem, M., Zaman, T., Abbasi, A. M., Abid, M., Mozūraitis, R., Alwahibi, M. S., & Elshikh, M. S. (2022). Arabian Journal of Chemistry, 15, 103482.

SHINE3 AND MYB31 TRANSCRIPTION FACTORS REGULATE CUTICLE BIOSYNTHESIS AND CELL WALL REMODELING DURING SOURSOP (*ANNONA MURICATA*) FRUITS RIPENING

Héctor Ruiz-Ortega^{1*}, José Villalobos-Escobedo², María Islas-Osuna¹, Martín Tiznado-Hernández¹, Carlos Calderón-Vázquez³ y Miguel Hernández-Oñate^{4**}

¹Centro de Investigación en Alimentación y Desarrollo (CIAD). Gustavo Astiazarán Rosas #46, Ejido La Victoria, Hermosillo, Sonora, México CP 83304, ²Instituto de Investigación sobre Obesidad. Unidad de Biología Integrativa. Tecnológico de Monterrey. Monterrey, México, ³Instituto Politécnico Nacional, CIIDIR Unidad Sinaloa. Juan de Dios Bátiz-Paredes #250, San Joaquín, Guasave, Sinaloa, México CP 81000, ⁴CONAHCyT-Centro de Investigación en Alimentación y Desarrollo (CIAD). Gustavo Astiazarán Rosas #46, Ejido La Victoria, Hermosillo, Sonora, México CP 83304.
E-mail:*hruiz120@estudiantes.ciad.mx, **miguel.hernandez@ciad.mx

Soursop (*Annona muricata*) is a climacteric fruit of great interest due to its nutritional value, with México being one of the leading producers globally. However, our country has not been able to position itself as an important exporter of this crop internationally because of its short postharvest life. The structure and composition of the cuticle play important roles in the quality and postharvest life of fruits. The cuticle component biosynthesis is regulated at different levels, including the transcriptional level, where transcription factors are involved in gene regulatory networks. Studies on the elucidation of these gene regulatory networks have been carried out mainly in model fruits such as tomatoes. Nevertheless, due to the differences in the biochemical and physiological characteristics between fruits, the knowledge generated in model fruits cannot necessarily be generalized to other fruits. Therefore, it is necessary to perform studies on less studied fruits, such as soursop fruits. Therefore, the goal of the present study was to identify and characterize transcription factors (TFs) involved in cuticle biosynthesis in soursop fruits, to determine changes in the expression of these TFs during fruit ripening, and to elucidate the gene regulatory network in which the TFs participate. For this purpose, we searched the TFs in the transcriptome of the soursop fruit during ripening by homology search with BLAST using the iTAK database. Moreover, we identified the expression changes of TFs and genes involved in cuticle biosynthesis. We identified 1,887 TFs in the soursop transcriptome, of which 1,164 were differentially expressed during the postharvest ripening of the fruits, suggesting their regulatory role in the biochemical and physiological changes that the fruit undergoes during this process. Furthermore, SHINE3 and MYB31 were co-expressed with various genes, including genes related to cell wall remodeling, such as pectate lyase. This data suggests a possible relationship between the regulation of cuticle biosynthesis and cell wall remodeling in soursop fruits. Although more research is required to confirm this hypothesis, the present study contributes to the knowledge generation of soursop fruits that will be useful in designing strategies to extend the postharvest life of soursop fruits.

SOLUBLE EXPRESSION OF CHAGASIN CHIMERAS HARBOURING FOUR TSA-1 EPITOPES IN *ESCHERICHIA COLI*

Marco Ibrim Rodríguez Sánchez¹, Paola Eugenia Agustin Vélez¹, Claudia Ivonne Flores Pucheta¹, Octavio Montes-Flores¹, Rossana Arroyo² and Jaime Ortega López¹

¹Departamento de Biotecnología y Bioingeniería y ²Departamento de Infectómica y patogénesis Molecular Centro de Investigación y Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN), Ciudad de México, México, C.P. 07360. E-mail: marco.rodriguez@cinvestav.mx

Chagas disease is a zoonotic, parasitic, systemic, and chronic disease caused by *Trypanosoma cruzi*¹ with an annual incidence of 30,000 new cases and around 12,000 deaths, where the tropical zones of Latin America are the most vulnerable². Due to the low efficacy of available chemotherapy, therapeutic vaccines based on recombinant antigens have been proposed as a novel complementary therapy; One promising antigen is TSA-1, a surface antigen with five epitopes highly conserved in various lineages, which can induce a cellular immune response³. This protein has been expressed recombinantly in *E. coli* as inclusion bodies⁴, A soluble scaffold protein, such as chagasin, containing two TSA-1 epitopes is proposed as a novel approach to the soluble expression of TSA-1 epitopes⁵. Therefore, this project aims to design by bioinformatic tools chagasin chimeras containing up to four of the five conserved TSA-1 epitopes and recombinantly express them in *E. coli* as a soluble protein by the mutation of a hydrophobic exposed amino acid on chagasin 3D structure, as well as by using solubility fusion tags such as MBP (Maltose Binding Protein). Two chimeras with the best solubility scores were cloned and expressed in *E. coli* as soluble antigens.

References

1. Jansen, A., Xavier, S. and Roque, A. (2020). Landmarks of the Knowledge and *Trypanosoma cruzi* Biology in the Wild Environment. *Frontiers in Cellular and Infection Microbiology*, 10.
2. Organización Mundial de la Salud [OMS] (2015). Enfermedad de Chagas (American trypanosomiasis) <http://www.paho.org/en/topics/chagas-disease>
3. Wrightsman, R., Dawson, D., Fouts, D., and Manning, J. (1994). Identification of immunodominant epitopes in *Trypanosoma cruzi* trypomastigote surface antigen-1 protein that mask protective epitopes. *J Immunol.* 1;153(7):3148-54. PMID: 7522250
4. De la Cruz, J., Villanueva-Lizama, L., Dzul-Huchim, V., Ortega-Lopez, J., Flores-Pucheta, C. I., Gillespie, P., Zhan, B., Bottazzi, M. E., Hotez, P. J., and Dumonteil, E. (2019). Production of recombinant TSA-1 and evaluation of its potential for the immuno-therapeutic control of *Trypanosoma cruzi* infection in mice, *Hum Vaccin Immunother* 15, 210-219
5. Cárdenas, E., Montes, O., Nava, E., Flores, C. and Ortega, J. (2024). Chagasin from *Trypanosoma cruzi* as a molecular scaffold to express epitopes of TSA-1 as soluble recombinant chimeras. *Protein expression and purification*, 218:106458.

HUMANIZATION OF AN IGE ANTIBODY: RECOMBINANT EXPRESSION AND STRATEGY TO RECOVER AFFINITY TOWARDS ITS ALLERGEN

Rosas-Ramírez ED¹, García-Ramírez B², Muñoz-Herrera JC¹, Pelcastre-Gómez AY¹, Vázquez-Leyva KS¹, Rodríguez-Flores SN¹, Gómez-Velasco H², Pérez-Tapia SM¹, Rodríguez-Romero A², Pedraza-Escalona MM^{1,3}

martha.pedraza@udibi.com.mx; tel. +525557296000 ext. 62543

¹Unidad de Desarrollo e Investigación en Bioprocesos de la Escuela Nacional de Ciencias Biológicas del Instituto Politécnico Nacional, CDMX, C.P. 11340. ²Instituto de Química, Universidad Nacional Autónoma de México, Ciudad Universitaria, CDMX, C.P. 04510. ³CONAHCyT-UDIBI-ENCB-IPN, CDMX C.P. 11340.

Antibody humanization is a technic used to reduce the immunogenicity of those molecules with biotherapeutic potential. 2F5 is a murine monoclonal IgE antibody (2F5m), obtained by generation of hybridomas from spleen cells of immunized mice against Hev b 8, the profilin of rubber tree (*Hevea brasiliensis*). 2F5m recognizes its allergen with an affinity of 1.7 nM. This work reports the humanization of 2F5m antibody by CDR grafting. Each variable region was cloned on commercial vectors for heavy and light chains respectively. The humanized IgE 2F5 was expressed on sever eukaryotic systems; however, the highest yield was obtained by co-transfection on Expi 293F cells. Subsequently, the 2F5h affinity constant for Hev b 8 was determined by indirect ELISA and Surface Plasmon Resonance (SPR), finding that it had decreased by an order of magnitude (5.2×10^8 M). Residues that, when replaced, compromised the stability of 2F5h and its interaction with Hev b 8 were identified through molecular dynamics and structural analysis. To recover the affinity of 2F5h, some of them were selected to be reestablished by reverse mutation using PCR overlap. To find those variants with similar biological characteristics to the original murine antibody, six reverse mutations were performed in total: four on the light chain and two on the heavy chain. The mutations were inserted in pairs, so that the double mutants 2F5h-VLFQ (FQ) , 2F5h-VLQV (QV) and 2F5h-VHMS (MS) were generated. Even though it was not possible to stablish the affinity of the FQ mutant, it was possible for the QV and MS mutants (5.8×10^8 and 4.6×10^8 M respectively), finding that MS appear good candidate in the way to reestablish the affinity of 2F5h.

IMMOBILIZATION OF α -AMYLASE FOR ALKYL-GLUCOSIDES PRODUCTION

Gloria Saab-Rincón, Wendy Xolalpa-Villanueva, Fidel Ramírez Amador, Leticia Olvera-Rodríguez, Alfonso Miranda Molina y Agustín López-Munguía

Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, Universidad Nacional Autónoma de México.
Av. Universidad 2001, Col. Chamilpa, 62210 Cuernavaca, Mor., México.
E-mail: gloria.saab@ibt.unam.mx

The enzymatic production of alkyl glucosides is limited by the stability of the enzymes in the presence of alcohols. In the present study, we have investigated the alcoholysis reaction carried out by α -amylase from *Thermotoga maritima* (AmyA) immobilised on cross-linked fast flow Sepharose. The advantage of using an α -amylase for this reaction is the low cost of starch used by these enzymes instead of activated glucosides required by glycosyltransferases. The immobilised enzyme achieved the same concentration of butyl-glucoside as previously determined for the free enzyme, but the ability to reuse the enzyme for five cycles with more than 50% residual activity increases the efficiency of the process. In addition, the previously reported H222Q variant of AmyA was immobilised and retained at least 50% more transglycosidic activity than WT AmyA. Both variants showed an increase in residual activity after 24 h incubation at 85°C following immobilisation. Successful alcoholysis with longer chain alcohols has not been previously reported for α -amylases. We investigated alcoholysis with longer chain alcohols, such as hexanol and octanol, and optimised the reaction conditions. The addition of DMSO had a positive effect on the alcoholysis reactions, yielding 0.7 mg/mL of octylglucoside compared to 0.4 mg/mL in the absence of DMSO for the wild-type enzyme.

IDENTIFYING BACTERIA WITH CUTINASE ACTIVITY FOR THE DEGRADATION OF PLASTIC FROM THE TEPETILTIC LAGOON

Magdiel Salas-Montenegro¹, Yuma Ávila-Lopez¹, Leticia Casas-Godoy², Marcela Robles-Machuca*

¹Universidad Autónoma de Nayarit, Secretaría de Investigación y Posgrado. Ciudad de la cultura s/n centro, 63000, Tepic, Nayarit, Mexico. ²CONHACYT–Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A.C., Unidad de Biotecnología Industrial, Camino Arenero 1227, El Bajío del Arenal, 45019 Zapopan, Jalisco, Mexico. Contacto: marcela.robles@uan.edu.mx

Abstract. Plastic is a widely used material, with a mass production of 400 million metric tons by 2021. It has a prolonged degradation process, and its accumulation causes environmental and health problems by generating microplastics. The alternative for the degradation of plastics by bacterial enzymatic means is of interest after the discovery of *Ideonella sakaiensis* 201-F6 in PET. Cutinases have demonstrated plastic degradation activity due to their ability to cut ester bonds and their structure is related to PETase. This work aims to identify bacteria isolated from the Tepetiltic lagoon, Nayarit with cutinase activity for plastic degradation. Bacteria on the plastics from the lagoon were cultured and isolated in non-selective media at 30°C for 24 hours, and then identified by MALDI-TOF MS. A screening was carried out in solid minimal medium with 10g/L linseed oil as inducer at 30°C for 96 hours, determining the cutinase activity by the presence/absence of a halo around the bacteria growth. A second screening in a liquid minimal medium was realized at 30°C and 150 rpm using the same inducer, and quantified cutinase activity with p-NPB as substrate at 405nm for 20 minutes. 108 strains were isolated and identified, with the most predominant genera being *Bacillus* at 40.7%, *Pseudomonas* at 38%, and *Saccharomyces* at 3.7%. The first screening indicated 11 strains with cutinase activity, while in the second screening, only 4 strains showed activity up to 1.728×10^{-5} U/L. Both screenings allowed us to identify a higher proportion of bacteria capable of producing the cutinase enzyme. This will guide us to evaluate plastic biodegradability.

ANALYSIS OF CELL MIGRATION AND PROLIFERATION IN EXPOSURE TO MEFLOQUINE ON THE MDA-MB-231 CELL LINE

Andrey Salazar-Mendoza¹, Fernando Verduzco-Valdez¹, Maximiliano Díaz-Inzunza¹, Angélica López-Izquierdo¹

¹Laboratorio de Bioingeniería, Facultad de Ingeniería Mexicali, Universidad Autónoma de Baja California (UABC), Blvd. Benito Juárez S/N, 21280 Mexicali, Baja California. Tel. (653)1373768. E-mail: andrey.salazar@uabc.edu.mx

Breast cancer is one of the most diagnosed and deadliest cancer around the world. Among the many subtypes of breast cancer is the triple negative breast cancer (TNBC) subtype, characterized by the lack of expression of the three receptors that are commonly found in breast cancer. TNBC is highly aggressive, with greater metastatic potential and poorer clinical outcome than the other subtypes¹. Currently, there is no diagnosis or treatment standard due to the lack of a proper molecular target. It has been reported that voltage-gated ion channels are aberrantly expressed in breast cancer tissue and cells, modulating several key cell processes that promote cancer progression, such as cell proliferation and migration². The overexpression of these transport proteins may be useful for diagnosis and therapeutic purposes. However, the functional role of ion channels in cancer is still not completely known. Mefloquine is an antimalarial drug that has been described as an inhibitor for voltage-gated potassium channels, which are suggested to confer survival advantages to breast cancer cells. The main purpose of this project is to analyze the effect of mefloquine on the cell migration and proliferation processes of a TNBC cell line, MDA-MB-231. In this research, cell migration, proliferation and viability assays are used to examine and compare the cytotoxic impact of mefloquine on the TNBC cell line. Results obtained so far show that mefloquine decreases cell viability over time in a concentration-dependent manner and inhibits cell migration in the same way. This research will be used to propose possible adjuvant treatment methods and specific therapies that help fight triple negative breast cancer metastasis, as well as contribute in the search of potential biomarkers to support early detection of this highly aggressive subtype of cancer, to improve patients' quality of life.

Funding: Internal Call for Research Projects 2024-2025, UABC.

References

1. Lu, B., Natarajan, E., Balaji Raghavendran, H. R., & Markandan, U. D. (2023). *Technology in Cancer Research and Treatment*, vol. 22.
2. Lastraioli, E. (2020). *Frontiers in Pharmacology*, vol. 11.

IMMOBILIZATION OF *THERMOMYCES LANUGINOSUS* LIPASE (TLL) VIA IONIC-COVALENT INTERACTION ON HETEROFUNCTIONAL GLYOXYL-AGAROSE SUPPORTS FOR BIODIESEL PRODUCTION

Cristian Salcedo-Cuaran¹, Juan S. Pardo-Tamayo¹, César A. Godoy¹

Universidad del Valle, C.P. 76001, Cali, Colombia¹

E-mail: cristian.salcedo@correounivalle.edu.co

Increasing demand and dependence on depleted oil reserves have prompted the world to explore and develop new strategies for renewable energy development, including the use of biomass as a promising alternative to fossil fuels and contributing to the achievement of Sustainable Development Goals (SDGs).¹ Biocatalysis with *Thermomyces lanuginosus* lipase (TLL) emerges as a sustainable alternative to traditional processes for biodiesel production.² However, its industrial application is limited by low stability, high cost, and inability to be reused.³ These limitations can be overcome by immobilization on supports, such as Glyoxyl-agarose (Gx), which has increased stability up to 2 million times.⁴ by providing multipoint interactions between the -NH₂ groups of lysine residues and the aldehydes of the support. This requires a pH of 10.0, implying enzyme stability at this pH, and many lysine residues, conditions that are not common in the enzyme world.⁵⁻⁶ Therefore, heterofunctionalization of Gx with anionic modifier taurine (Tau) and cationic modifier Girard T Reagent (RGT) is proposed to obtain two types of supports (Gx-Tau) and (Gx-RGT) that allow for ionic and covalent enzyme-support interactions. This modification was achieved through a condensation reaction, with aldehyde modification of Gx reaching 66.08% with RGT and 65.42% with Tau. This methodology was optimized by reducing the reaction time from 72 hours to 25°C using pH 3.0 for 3 minutes, with microwave assistance at pH 10.0. Using the modified supports, TLL immobilizations were performed, achieving immobilization percentages of 58.04% and 4.25% for Gx-RGT and Gx-Tau, respectively, at 6 hours and pH 7.0, conditions where immobilization does not occur in Gx alone. This result improved to 88.18% for Gx-RGT with SDS as an additive in immobilization, approaching those obtained with the commercial support Q-sepharose®, which showed 96.70% immobilized activity even when Gx-RGT has 1.75 times fewer ionic groups and exhibits 1.31 times more expressed activity compared to Q-sepharose®. It is concluded that immobilization at neutral pH is possible on modified Gx supports. Finally, considering the previous results, the obtained derivatives will be used as biocatalysts in the production of ethyl esters, the main component of biodiesel.

References

1. An overview on microalgae as renewable resources for meeting sustainable development goals—ScienceDirect. <https://www.sciencedirect.com.bd.univalle.edu.co/science/article/pii/S0301479722014700?via%3Dihub> (accessed 2024-05-31).
2. Cavalcante, F. T. T.; Neto, F. S.; Rafael de Aguiar Falcão, I.; Erick da Silva Souza, J.; de Moura Junior, L. S.; da Silva Sousa, P.; Rocha, T. G.; de Sousa, I. G.; de Lima Gomes, P. H.; de Souza, M. C. M.; dos Santos, J. C. S. Opportunities for Improving Biodiesel Production via Lipase Catalysis. *Fuel* 2021, 288, 119577. <https://doi.org/10.1016/j.fuel.2020.119577>.
3. Sheldon, R. A. Biocatalysis and Biomass Conversion: Enabling a Circular Economy. *Philos. Trans. R. Soc. Math. Phys. Eng. Sci.* 2020, 378 (2176), 20190274. <https://doi.org/10.1098/rsta.2019.0274>.
4. Lyu, X.; Gonzalez, R.; Horton, A.; Li, T. Immobilization of Enzymes by Polymeric Materials. *Catalysts* 2021, 11 (10), 1211. <https://doi.org/10.3390/catal11101211>.
5. Zucca, P.; Fernandez-Lafuente, R.; Sanjust, E. Agarose and Its Derivatives as Supports for Enzyme Immobilization. *Molecules* 2016, 21 (11), 1577. <https://doi.org/10.3390/molecules21111577>.
6. Guisan, J. M.; Fernandez-Lorente, G.; Rocha-Martin, J.; Moreno-Gamero, D. Enzyme Immobilization Strategies for the Design of Robust and Efficient Biocatalysts. *Curr. Opin. Green Sustain. Chem.* 2022, 35, 100593. <https://doi.org/10.1016/j.cogsc.2022.100593>.

PHYTOTOXICITY AND GENOMIC INSTABILITY OF THE INTERACTION USING NANOPARTICLES WITH NITROGENATED FERTILIZERS IN THE IN VIVO *ALLIUM CEPA* AND *CAPSICUM* MODEL

Alejandro Sánchez Gonzalez¹, Rafael Bello Bedoy¹ and Evarista Arellano García¹

¹Facultad de Ciencias, Universidad Autónoma de Baja California, Ensenada Baja California, México. Carretera Transpeninsular Ensenada-Tijuana No. 3917 Colonia Playitas C.P. 22860 Tel: 646 152 8210
AlejandroSG@uabc.edu.mx

Abstract. The use of fertilizers with nanomaterials as nutritional agents in agriculture is a topic of interest in the field of polluting processes, because the use of nitrogenous chemical fertilizers (FQN), which became widespread since the second half of the last century, continues being one of the strategies that guarantee crop performance with the consequent environmental pollution derived from its accumulation and interaction with the natural environment. On the other hand, the availability of nanomaterials (NMs) to the environment increases as their use diversifies. This interaction has recently been a source of concern about so-called nano fertilizers, which offer nutritional advantages by increasing their absorption efficiency by the plant rhizome. The nutrients, applied alone or in combination, bind to nanometric adsorbents, which release the nutrients very slowly compared to conventional FQNs; This approach not only increases the efficiency of crop nutrient use, but also minimizes their leaching to groundwater. The purpose of this project is to evaluate the phytotoxicity and genomic instability of the interaction of silver nanoparticles (AgNPs) with FQN urea (46% N) and ammonium nitrate (NH_4NO_3) in the in vivo model of the onion *Allium cepa* and *Capsicum spp.* The test model (*Allium test*) is widely used to characterize the genotoxic and cytotoxic effects of a substance of interest. The present study will analyze whether nanoparticles associated with nutrients are phytotoxic and produce genomic instability in the applied vegetables, in a way that affects the quality of the products and leads to contamination for the consumer.

That is why the present study will analyze how nanoparticles affect plants by analyzing their phytotoxicity through the analysis of their genomic instability.

This project proposes to study silver nanoparticles associated with fertilizers and other organic growth promoters that allow local farmers to use them, reducing the use of chemical fertilizers. By incorporating non-phytotoxic or genotoxic silver nanoparticles in vegetable crops, the aim is to develop nanotechnology that can become a patent or utility model. Also, by investigating the effect of silver nanoparticles on germination, early growth and morphological parameters of wild and domesticated chili peppers and onions, we found a favorable effect of these nanoparticles on root and plant growth using concentrations of 50, 100 and 250 ppm.

NEUTRALIZATION OF THE NEUROTOXIN ACTIVITY OF ALPHA-LATROTOXIN FROM THE BLACK WIDOW SPIDER USING MONOCLONAL ANTIBODIES

Fátima Sánchez-Ramírez¹, Alejandro Olvera-Rodriguez¹ and Alejandro Alagón¹

¹Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Universidad 2001, Chamilpa, C.P. 62210 Cuernavaca, Mor., México.
Group leader E-mail: alejandro.alagon@ibt.unam.mx

In Mexico, the two spiders of medical importance belong to the genus *Loxosceles*, commonly known as the violin spider, and the genus *Latrodectus*, colloquially known as the black widow spider or capulina (in México). The venom of the black widow spider is composed of various toxins; among these are alpha-latrotoxin, latrocrustotoxin, latroinsectotoxin, and neurexins, which have an affinity for different receptors ^[1].

The alpha-latrotoxin is synthesized as a 157 kDa polypeptide that subsequently undergoes proteolysis at the ends, leaving a mature protein of 130 kDa that is made up of two important domains: the amino-terminal domain and the 22-type repeat sequence ankyrins domain. Alpha-latrotoxin is the venom component aimed at vertebrates. It is responsible for causing a neurological syndrome called latrodectism, characterized by local pain associated with diaphoresis and nonspecific systemic effects in bitten patients ^[2]. The commercial antivenom currently used to treat black widow poisoning in Mexico is produced from the venom of hundreds of spiders, obtained by electrical stimulation, and the venom is inoculated into horses in increasing concentrations. Subsequently, the horse's plasma is extracted, and the antibodies are purified and digested with pepsin to process them and obtain a biotherapeutic called Aracmyn®.

This project aimed to develop a murine monoclonal antibody using the complete venom of *L. mactans* as an immunogen and evaluate its neutralizing capacity in the complete venom of the black widow spider. In the present work, three monoclonal antibodies were obtained (mAb 3B7, mAb 2D11, mAb 4D12) capable of preventing the death of mice poisoned with 3DL₅₀ in preincubation experiments. In Western blot and ELISA assays, we determined mAb 4D12 and 2D11 recognize the ankyrin-like domain, and mAb 3B7 recognizes the amino-terminal domain of α -Latrotoxin. These findings are not just promising; they are a beacon of hope. We conclude that the monoclonal antibodies obtained show great potential for their possible expression in a recombinant manner and their possible use as antivenom, possibly offering a brighter future in the treatment of black widow spider venom poisoning.

References

1. Williams, M., Sehgal, N., & Nappe, T. M. (2023). Black Widow Spider Toxicity. In StatPearls. StatPearls Publishing
2. Ushkaryov, Y. A., Rohou, A., & Sugita, S. (2008). Alpha-Latrotoxin and its receptors. Handbook of experimental pharmacology, (184), 171–206. https://doi.org/10.1007/978-3-540-74805-2_7

CHARACTERIZATION OF PROTEASE ENZYMES FROM THE ZH2 STRAIN OF THE GENUS *GEOBACILLUS SPP.*

Eric Alejandro Sánchez Rosas¹, Juan Francisco Sánchez López, Silvia Díaz-Sandoval²

Unidad Profesional Interdisciplinaria de Ingeniería Campus Guanajuato. Instituto Politécnico Nacional (UPIIG IPN).
Avenida Mineral de Valenciana 200 Col. Fraccionamiento Industrial Puerto Interior, 36275 Silao de la Victoria, Gto. Mex.
C.P. 36275. Tel (472)7484638 Conmutador (55) 57296000 ext. 81324 y 81346.
esanchezr1910@alumno.ipn.mx¹ sidiaz@ipn.mx²

Protease enzymes from bacteria are used in detergents and in the skin de-wooning process in the tannery. However, its instability at high temperatures limits its use. Therefore, heat-resistant enzymes from bacteria of the genus *Geobacillus spp.* They are an implementation alternative. The objective of this research work is to obtain and characterize protease enzymes produced by a thermophilic strain of *Geobacillus spp.* designated with the name ZH2, isolated from a hot spring located in San Francisco, Silao, Guanajuato, Mexico, to identify the reaction time and the pH and temperature conditions in which its proteolytic activity is greater. To do this, the strain was inoculated in a liquid enzyme production medium added with 1% lactic acid casein and incubated for 48 and 72 hrs at 60°C to obtain the culture supernatant. Subsequently, a protein profile of the supernatant was performed using the SDS-PAGE technique to check the presence of enzymes with proteolytic activity. Afterwards, said enzymatic activity was evaluated using the Lowry technique in reaction times of 5 to 25 minutes, under pH conditions between values of 3 and 8, and under temperatures from 50 to 100 °C; in order to establish the optimal reaction conditions. Regarding the performance of the protein profile of the culture supernatant; This did not yield conclusive bands that could be associated with the presence of a protease enzyme, however, it did demonstrate the proteolytic activity present as the respective casein band shown in the control of the uninoculated supernatant was not observed. In relation to the enzymatic activity tests, it was found that, for a medium incubated for 72 hrs at 60°C, a protein concentration of 0.0868 mg/ml is obtained, being higher than that obtained after 48 hrs. Regarding the reaction times, the highest protease activity was observed at 5 minutes at 60°C with pH 5, being 0.077 ± 0.034 U/ml, where from that time on, its value drops. On the other hand, it was found that the best conditions for the reaction are at 60°C at a pH of 3, this being 0.083 ± 0.011 U/ml. In summary, the thermophilic strain ZH2 of the genus *Geobacillus sp.* manages to synthesize thermostable protease enzymes capable of reporting a proteolytic activity of 0.083 U/ml under the optimal conditions of 60°C at pH 3 in 5 minutes of enzyme-substrate reaction.

References

1. Ahmad W., Tayyab M., Nauman Aftad M. et. al. (2020). Optimization of Conditions for the Higher Level Production of Protease: Characterization of Protease from *Geobacillus SBS-4S*. Editorial Springer nature.
2. Naquiche Calero A. (2014). Aislamiento e identificación de bacterias termófilas productoras de proteasas provenientes de los geiseres de calientes, Candarave. Tacna, Perú.

DEVELOPMENT OF AN EDIBLE COATING AND FILM FROM CARBOXYMETHYLCELLULOSE, MESQUITE GUM, AND A DAIRY INDUSTRY WASTE PRODUCT

María Cristina Sánchez Aguirre*, Nathiely Ramírez Guzmán¹, Jorge Aguirre Joya², Cristian Torres León²

Facultad De Ciencias Biológicas, Universidad Autónoma De Coahuila. Carretera Torreón Matamoros Km. 7.5, Ejido el Águila, 27275, Torreón, Coahuila. Centro de Investigación y Jardín Etnobiológico, Universidad Autónoma de Coahuila. 24480, Viesca, Coahuila, México.

*msanchezaguirre@uadec.edu.mx

The application of edible coatings and films to fruits and vegetables represents an environmentally friendly approach and an innovative solution to extend the shelf life of export fruit products. Edible coatings and films can act as environmentally friendly and biodegradable packaging.[1] A biofilm is a thin layer of biodegradable compounds that protects harvested vegetables and increases shelf life by providing a barrier against gases and water vapor. [2] The objective of this research has been to develop a coating and edible film from an organic product endemic from the semi-desert region of the state of Coahuila, mesquite gum and whey, which is the residue of the dairy industry of the comarca lagunera. Mesquite gum was dissolved in 70% ethanol at 65°C, then carboxymethylcellulose and whey protein were mixed in a beaker at 1% (w/v); finally, the two solutions were mixed to obtain the coating. Avocado, a fruit of high economic relevance in Mexico, was used as a model fruit to evaluate the coating, and a control with uncoated avocado was used. The casting method was used in Petri dishes. The coated fruit was analyzed for physicochemical parameters such as pH, titratable acidity, color, and total soluble solids. The results of these parameters showed that the edible coating delayed the ripening of avocados compared to the control. The coated avocados had a minimum loss of lightness, L=35.22 on the first day and after seven days L=34.72, while the control samples had a significant loss from L=32.90 to L=28.86. Edible films have suitable characteristics for use as innovative biodegradable packaging in the food industry.

References

1. Sahraee, S.; et. al.. Protection of Foods against Oxidative Deterioration Using Edible Films and Coatings: A Review. *Food Biosci.* 2019, 32, doi:10.1016/j.fbio.2019.100451.
2. Torres-León, C.; Ramírez-Guzman, et. al.. Food Waste and Byproducts: An Opportunity to Minimize Malnutrition and Hunger in Developing Countries. *Front. Sustain. Food Syst.* 2018, 2, 1-17, doi:10.3389/fsufs.2018.00052.

CHARACTERIZATION OF DYP-TYPE PEROXIDASES AND THEIR USE AS BIOCATALYSTS IN TEXTILE DYES DECOLORIZATION

Jesús Alberto Segovia-Cruz¹, Andrés Zárate-Romero², Ayixon Sánchez Reyes¹

¹Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos,

²Department of Bionanotechnology, Centro de Nanociencias y Nanotecnología, Universidad Nacional Autónoma de México.
E-mail: alberto.segovia@ibt.unam.mx

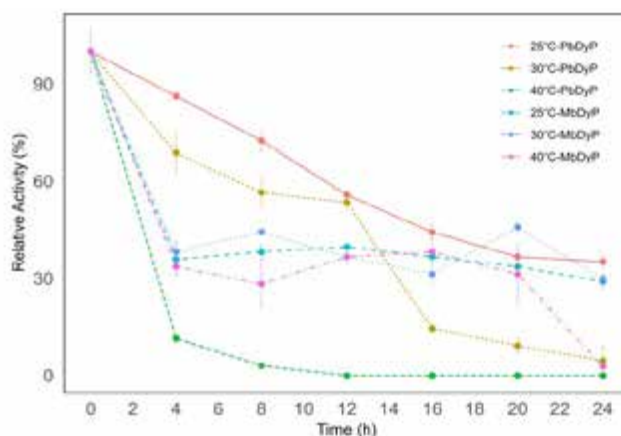
Keywords: *DyP peroxidases, characterization, textile dyes*

Introduction. Dye decolorizing peroxidases (DyP) have been proposed as potential biotransformers in treating wastewater containing dyes and other organic contaminants. Recently, we identified two DyP sequences from two bacterial genomes assembled from metagenomes (MAGs). The genes were synthesized de novo and subsequently inserted into individual constructs in expression vectors. These enzymes are referred to as MbDyP and PbDyP.

Methods. The two enzymes were purified using a HisTrap column. Subsequently, the thermal stability was determined by incubating each DyP with phosphate buffer at different temperatures (25, 30, 40, and 50 °C) for 24 hours. ABTS and H₂O₂ were used as substrates. Afterward, DyP's enzyme extracts were tested in the presence of H₂O₂ as an oxidizing agent against anthraquinone dyes like Vat red 10 and azo dyes like Disperse orange at a concentration of 100 µM at 25°C for 24 hours.

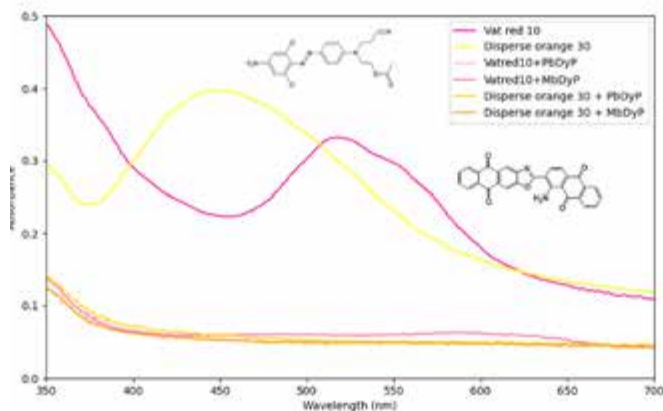
Results. Only selected results are presented in this abstract. Both enzymes were maintained at ~30% of their activity after 24 hours of incubation at 25 °C. MbDyP exhibited greater thermostability at 40 °C. This suggests these DyP enzymes are temperature-sensitive, demonstrating their viability for potential biotechnology applications (Fig. 1).

Figure 1. Effect of temperature on the activity of MbDyP and PbDyP.



PbDyP and MbDyP were assayed against textile dyes achieving biotransformations of 84.9% and 84.7%, respectively for Vat red 10, and 84.6% and 86.2% respectively for Disperse orange.

Figure 2. Dye biotransformation of anthraquinone and azo dyes by MbDyP and PbDyP



Conclusions. The enzymes PbDyP and MbDyP proved their effectiveness in biotransforming anthraquinone and azo dyes with efficiencies exceeding 84%. This underscores the potential of these enzymes as biocatalysts for treating wastewater contaminated with these pollutants.

GMO STUDY LANDSCAPE: CONTENT, CITATION, AND GEOGRAPHY

Cynthia V. Soberanes Gutiérrez¹, Erica L. Hagman Aguilar¹, Juan de Dios Ortega Alonso¹,
Eduardo Alexis Valencia Dorantes², Edgardo Galán Vásquez²

¹Secretaría Ejecutiva de la Comisión Intersecretarial de Bioseguridad de los Organismos Genéticamente Modificados, Av. Insurgentes Sur 1582, Col. Crédito Constructor, Benito Juárez, C.P. 3940, CDMX; ²Instituto de Investigaciones en Matemáticas Aplicadas y Sistemas, Circuito Escolar S/N, Ciudad Universitaria, Coyoacán, C.P. 04510, CDMX.
E-mail: soberanesvcv@gmail.com, cinthia.soberanes@conahcyt.mx

Transgenic organisms are those that have been genetically modified by introducing genetic material from another species, using genetic engineering techniques and are part of the so-called Genetically Modified Organisms (GMO).

Most genetically modified crops have been designed to be resistant to herbicides such as glyphosate or to be resistant to pests such as bollworm. While some studies suggest that GMO may be safe and beneficial, others indicate potential risks such as allergenicity, antibiotic resistance, and unintended harm to other organisms. Concerns about GMO include possible long-term health effects on humans and animals that consume these products. As well as environmental impacts that include the development of herbicide-resistant superweeds, loss of biodiversity and damage to non-target species such as insects and soil microorganisms.

All of this has resulted in a very great interest that has led to the publication of thousands of articles about GMO. Therefore, a comprehensive analysis of this information from the point of view of natural language processing can help us understand the general panorama of the institutions, researchers, organizations, collaboration networks and citations that most study topics related to GMO.

To do this, a search was carried out for publications related to GMO reported in Pubmed, which includes 66,000 articles from 1990 to 2024. To build a structured data set of the information published in PubMed, an approach was used data mining, which involves the recovery of information such as: PMID, title, name of the journal, authors, author affiliations, abstracts, DOI, references and in the case of open access publications, the recovery of the complete article. For this, Python 3.9 and regular expressions were used for data recovery and cleaning. Subsequently, natural language processing (NLP) techniques were used to tokenize the texts, perform a content analysis, identify keywords, recurring themes and carry out a sentiment analysis. NetworkX was used for the reconstruction and analysis of citation and collaboration networks. Finally, these networks were structurally characterized to better understand the landscape of GMO studies.

GENETIC DIVERSITY AND PHENOTYPIC VARIATIONS IN FOUR NATIVE STRAINS OF *BACILLUS* SPP. BIOCONTROL AGENTS THROUGH PAN-GENOME AND BGC ANALYSIS

Hilda Mabel Sosa Esquivel¹, Alejandro Rubén Martínez Espinoza¹, Yumiko De la Cruz Rodríguez¹
Janeth Lucero Hernández Gaytán¹, Alejandro Alvarado Gutiérrez¹, Saúl Fraire Velázquez¹

¹Lab Biología Integrativa de Plantas y Microorganismos, Unidad Académica de Ciencias Biológicas,
Universidad Autónoma de Zacatecas, Zac.
sfraire@uaz.edu.mx

Biocontrol of phytopathogens, an alternative to reduce pesticide usage and better care for the environment. The genus *Bacillus* is distinguished by providing valuable species and strains to counteract plant pathogens, with *Bacillus velezensis* standing out in this regard. In the present study, among 24 outstanding rhizobacteria, 4 were isolated showing aptitude and variability in inhibiting 4 root pathogens in chili pepper and one foliar pathogen in bean plants. Similarly, variations were observed in promoting growth in chili pepper and bean plants. Moreover, a strain with the capacity to induce the formation of pseudo-nodules in bean root was identified. With the genomic sequencing of these strains, the current aim is to identify the species, genetic diversity, and particularities among them and their possible relationship with the phenotype, with emphasis on the core genome, singletons, genes for antibiotic biosynthesis, and genes related to nodulation induction in roots. Genomic sequencing revealed that the genome size of these 4 strains ranged between 3.9 and 4.01 Mb. Surprisingly, comparative genomics showed that these four strains correspond to *B. velezensis* (strains 2A-2B, 2A-10A, 3A-6A, and 3A-25B); together, these 4 genomes yield a pan-genome of 4,224 genes, with a core genome of 3,398 genes, 440 unique genes (singletons), and 386 dispensable genes. An expanded analysis including 18 genomes of North American *B. velezensis* strains resulted in a pan-genome of 6,750 genes and a core genome of 2,870 genes. Analysis using antiSMASH¹ revealed that the four strains contain genes for the biosynthesis of 6 antibiotics: macrolactin H, bacillaene, fengycin, bacillibactin, bacilysin, and surfactin. The most outstanding strain, which is both a phytopathogen inhibitor and an inducer of pseudo-nodules, also possesses 3 gene clusters for antibiotic biosynthesis of the NRPS class, and no genes related to nodulation were identified through mining efforts. A genome alignment using Mauve² revealed an average of 212 collinear blocks, with 118 located on the antisense strand, and in strain 2A-2B, more than 6 regions of between 450 and 850 bp were identified with no homology in the other three genomes.

References

1. Blin K, Shaw S, Augustijn HE, Reitz ZL, Biermann F, Alanjary M, Fetter A, Terlouw BR, Metcalf WW, Helfrich EJN, van Wezel GP, Medema MH, Weber T. (2023). *Nucleic Acids Res.* 2023 Jul 5;51(W1):W46-W50.
2. Darling AC, Mau B, Blattner FR, Perna NT. (2004). *Genome Res.* 2004 Jul;14(7):1394-403.

PRODUCTION *IN SILICO* OF NEW CANCER IMMUNOTHERAPEUTIC CHIMERIC PROTEIN DESIGNED WITH PD-1 IGV-DOMAIN-FC

Eva Cristina Sosa-Sillas¹, Evangelina Beltrán-López¹, Kimberly Arisbeth Casal-Rodríguez¹,
Ulises Vega-Castillo¹ and Héctor Samuel López-Moreno^{1*}

¹Laboratory of Molecular Biomedicine, CAC-UAS-264; Graduate Program in Biomedical Sciences and Graduate Program in Biotechnology, Faculty of Chemical and Biological Sciences. Universidad Autónoma de Sinaloa, Cd. Universitaria, Culiacán, CP.80010, Sinaloa, México.

*hslmoreno@ms.uas.edu.mx; CP.80010; +526675747963

Cancer is a complex disease that employs different mechanisms to evade the immune system; one of these critical mechanisms is the PD-1/PD-L1 signaling pathway. Producing a monoclonal antibody is a very slow and expensive process, so chimeric Fc-proteins offer an alternative strategy to develop an immunotherapeutic molecule against cancer, that combine ligand specificity and humoral immunity effector activation like a MAb. In this study, an *in silico* chimeric protein was designed and produced using accessible bioinformatics programs to fuse IgG1 with the previously cloned PD-1 IgV-like sequence, which was recombinantly produced by our team. Initially, specific primers will be designed with the CLC Sequence Viewer and Primer 3 Plus programs to amplify the Fc sequence of human IgG for subsequent ligation into the pcDNA6 plasmid containing the IgV-like fragment of PD-1. The specific primers will then be used to amplify only the chimeric rFc-PD-1 gene for its subsequent prediction using the Swiss-Model bioinformatics program. This antibody can have a positive impact as a treatment for people with cancer by improving their quality of life, as it blocks the survival signaling mechanism of tumor cells.

Keywords: *Cancer, PD-1, Fc, chimeric protein, immunotherapeutic.*

ANTIOXIDANT AND ANTIMICROBIAL EVALUATION OF *IN VITRO* CULTURES OF *LUDWIGIA OCTOVALVIS* (JACQ.) RAVEN

Stephany Abigail Tadeo Cuenca¹, Valery Domínguez Villegas², Virginia Mandujano González³,
Francisco Cruz Sosa¹ y Mariana Sánchez Ramos¹

¹Departamento de Biotecnología, Universidad Autónoma Metropolitana Unidad Iztapalapa, Av. Ferrocarril de San Rafael Atlixco 186, Col. Leyes de Reforma 1ª. Sección, Alcaldía Iztapalapa, Ciudad de México 09310, México. ²Laboratorio de Investigación farmacéutica, Facultad de Química, Universidad Autónoma del Estado de Morelos, Av. Universidad 1001, Col. Chamilpa, Cuernavaca Morelos 62209, México. ³Laboratorio de Biología Molecular, Universidad Tecnológica Corregidora, Carretera Estatal 413 km, Santa Bárbara, Corregidora, Querétaro 76926, México. E-mail: cbs2223801823@izt.uam.mx

In Mexico, 85% of medicinal plants are extracted from the wild; therefore, a tentative alternative for the production of secondary metabolites is the use of *in vitro* culture techniques, which allow us to control abiotic and biotic factors, as well as production in less time.¹ *Ludwigia octovalvis* is an aquatic herb belonging to the Onagraceae family, used as a medicinal plant in Morelos, Veracruz and Oaxaca, for the treatment of urinary infections and oedemas. Previous studies have demonstrated its pharmacological effects as antimicrobial, antioxidant, cytotoxic and anticarcinogenic.² Therefore, the main objective of this work was to establish callus and seedling cultures of *L. octovalvis*, as well as its antioxidant and antimicrobial evaluation. From *L. octovalvis* seeds collected in Lomas de Zomplante, Morelos, Mexico, a disinfection protocol was established and germinated in Murashige and Skoog medium with 3% sucrose and 3 g/L phytigel. With the seedlings at one month of age, leaves and stems were used as explants, to carry out a matrix of treatments evaluating the response of morphogenesis and callogenesis, with auxins: 2, 4-D and ANA, cytokinins: BAP and KIN, at concentrations of 0, 0.1, 0.5 and 1 mg/L, were kept under observation for one month. From the treatments that showed a response, seedling culture lines obtained by indirect organogenesis and callus were established. Extracts were then obtained with hexane, ethyl acetate and methanol, and yields were calculated according to the weight obtained. Then, the antioxidant activity of the extracts was determined at 10, 100 and 1000 ppm by the DPPH method using quercetin as a positive control; finally, the antimicrobial activity was evaluated by the Kirby-Bauer diffusion disc method against *Staphylococcus aureus* (ATCC 6538), *Escherichia coli* (ATCC 35218) and *Candida albicans* (urine), evaluating the extracts at 50, 25, 12.5 and 5 mg/mL.

The highest yields were obtained for the methanol extracts. The ethyl acetate extract of seedling added with 0.1 mg/L BAP with 1 mg/L ANA showed the highest uptake of DPPH free radical with 91.4% at 100 ppm, while the methanol extract of callus obtained with 1 mg/L BAP with 0.1 mg/L of 2, 4-D, and 1 mg/L of 2, 4-D, and 1 mg/L of ANA showed the highest uptake of DPPH free radical at 100 ppm. 1 mg/L of 2, 4-D, showed 85.3% antioxidant activity at 1000 ppm, compared to the wild plant Pandey *et. al.* (2023) obtained an 83% uptake response at a lower concentration of 270 µg/mL. The three extracts of callus and seedling at different concentrations showed no activity against *E. coli* and *C. albicans*; however, the hexane extract evaluated at 50, 25, 12.5 and 5 mg/mL showed activity against *S. Aureus* at all concentrations;

however, the highest inhibition halos were obtained at 50 mg/mL with 16.07 ± 1.30 mm; on the other hand, methanol extract of seedling presented halos of 8.39 ± 1.47 mm; unlike Yakob *et. al.* (2012) who evaluated wild plant extract at 20 mg/mL obtained smaller sized halos (7.0 ± 0.6 mm). The results obtained suggest that the seedling and callus cultures biosynthesise different compounds to the wild plant, although, the phytochemical content of the extracts evaluated still needs to be determined in order and get fastness conclusions.

References

1. Tasiu Isah, Shahid Umar, Abdul Mujib, Maheshwar Prasad Sharma, P.E. Rajasekharan, Nadia Zafar, Arajmand Fruk (2017). *Plant Cell, Tissue and Organ Culture*, 132(2), 239-265.
2. Ana Carolina Oliveira Silva, Elidiane Fonseca Santana, Antonio Marcos Saraiva, Felipe Neves Coutinho, Ricardo Henrique Acre Castro, Maria Nelly Caetano Pisciotano, Elba Lúcia Cavalcanti Amorim, Ulysses Paulino Albuquerque (2013). *Evidence-based Complementary and Alternative Medicine*, 2013, 1-9.
3. Stuti Pandey, Pawan Kumar Pandey, Alakh Niranjana Sahu, Himanshu Verma, Manmath Kumar Nandi (2023). *Traditional Medicine Research*, 8(2), 9.
4. Haidar Kadum Yakob, Shaida F. Sulaiman, Abd M. Uyub (2012). *World Applied Sciences Journal*, 16(1), 22-29.

EVALUATION OF THE IMPLEMENTATION OF THE CRISPR-CAS9 SYSTEM FOR THE MODIFICATION OF THE ATP6 GENE OF *PARACOCCLUS DENITRIFICANS*

Sergio Aarón Tinajero-Vargas¹, Edgar Morales Ríos¹

¹Departamento de Bioquímica, CINVESTAV-ZACATENCO

E-mail: aaron.tinajero@cinvestav.mx and edgar.morales@cinvestav.mx

Paracoccus denitrificans is an α -Proteobacterium closely phylogenetically related to eukaryotic mitochondria. The main reason why it has been widely used as a model in its bioenergetic studies. In this sense, the general architecture and function of the final enzyme of oxidative phosphorylation, the ATP synthase, is comparable with its mitochondrial homolog¹. The ATP synthase has been established as the main producer of ATP used in several biochemical processes carried out by virtually all known organisms. The general architecture of the ATP synthase consists of two structurally and functionally coupled regions, a soluble (F₁) and a membrane region (F_o)². These two regions are composed of a different number of subunits, where the number of them depends on the complexity of the organism in question, being the prokaryotic enzyme the most simple. Currently, there is only one crystallographic structure reported in the PDB of a bacterial ATP synthase at high resolution, obtained from *P. denitrificans* at 3.9 Å by X-ray crystallography³.

Genetic studies focused on mitochondrial DNA (mtDNA) mutations, specifically those found in the α -subunit gene of human ATP synthase (ATP6 gene in *P. denitrificans*), have shown the association between these mutations and certain congenital diseases, such as NARP/MILS syndrome. It has been observed that these conditions arise from the mutations T8993G/C and T9176G, producing the substitution of two key amino acids within the α -subunit (L156R/P and L217R). However, although it has been reported a decrease in the activity of ATP synthesis and hydrolysis due to the presence of the described mutations, the molecular mechanisms have not been well established⁴.

In the present work, we propose to modify the ATP6 gene of the ATP synthase from *P. denitrificans* by inserting the two mutations associated with the NARP/MILS syndrome through the implementation of the CRISPR-Cas9 system, and by a homologous recombination technique to determine and evaluate the level of efficiency achieved by both techniques. So far, within the development of the project we have managed to successfully carry out the assembly and standardization of the CRISPR-Cas9 technique *in vitro*, which involved the design, synthesis, and purification of each of the components necessary for the DNA cleavage *in vitro* and *in vivo* (sgRNA, Cas9, and sDNA). From this, we have evaluated the level of endonuclease efficiency on the genes of interest (ATP6 and *atpD*) and reporters (sfGFP).

Furthermore, we have constructed a homologous recombination plasmid, which has served as a repair template (RT) to carry out the insertion of the specific mutations required within the *P. denitrificans* genome (R175P and L235R). The construction of this plasmid also involved the modification of the PAM regions adjacent to the DNA sequences to be modified to prevent cutting both, the TR and the already modified genome. The implementation of gene editing techniques will be performed by electroporation of the TR and the CRISPR-Cas9 system in its ribonucleoprotein form. Therefore, we need to standardize the optimal conditions for transfection in order to have the highest possible editing efficiency in ATP6.

Reference

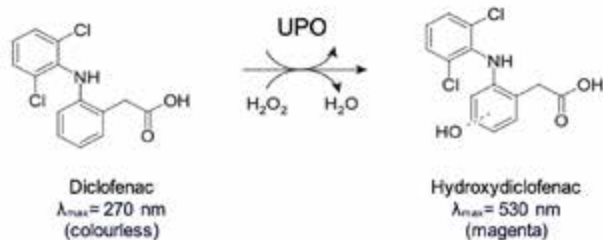
1. Robinson, G. et. al.. (2013). *Open Biology*. vol. 3. 120164
2. Deckers-Hebistreit, G. et. al.. (1991). *Annual Review of Microbiology*. vol. 50, 791-824.
3. Morales, E. et. al.. (2015). *PNAS*. vol. 112, 13231-13236.
4. Sgarbi, G. et. al.. (2006). *Biochemical Journal*. vol. 395. 493-500.

SCREENING METHOD FOR THE DIRECTED EVOLUTION OF A FUNGAL PEROXYGENASE ABLE TO CATALYZE THE OXIDATION OF EMERGING POLLUTANTS

Alina Elizabeth Torres Aguirre¹, Marcela Ayala Aceves¹

¹Institute of Biotechnology, Av. Universidad 2001, Chamilpa, 62210 Cuernavaca, Mor.
E-mail: alina.torres@ibt.unam.mx

Emerging pollutants (EPs) are defined as synthetic or naturally occurring chemicals that are not commonly monitored and regulated in the environment but have the potential to enter the environment and cause known or suspected adverse ecological and (or) human health effects¹. Pharmaceutical drugs (PDs) are a major category of EPs that have been detected in wastewater at low concentrations (ng/L or µg/L)². A green and biotechnological self-sufficient alternative is the use of enzymes to transform EPs, producing less active compounds that are less harmful³. Unspecific peroxygenases (UPOs) are versatile enzymes capable of catalyzing various oxidation reactions, including one- and two-electron oxidations of aromatic and heterocyclic compounds, inorganic halides, and organic heteroatoms, such as epoxidation, dealkylation and hydroxylation, using only hydrogen peroxide at low concentrations as an oxidizing agent⁴. Previous studies³, including those from our group (unpublished results), have demonstrated that a fungal UPO, derived from the basidiomycetes *Agrocybe aegerita*, is able to catalyze the oxidation of PDs. However, the reactions proceed with low catalytic efficiency. One powerful strategy to enhance specific enzyme characteristics is directed evolution⁵. We set up a novel colorimetric screening method for the selection of better variants, based on the oxidation of diclofenac which generates hydroxydiclofenac, a compound that can be monitored spectrophotometrically.



This work received funding from Project IN215423.

References

1. Geissen, V. et. al. *International Soil and Water Conservation Research* 3, 57–65 (2015).
2. Samal, K., Mahapatra, S. & Hibzur Ali, M. Treatment technologies, impact on environment and human health. *Energy Nexus* vol. 6 (2022).
3. Grunwald, Peter. *Chemoenzymatic Synthesis of Active Pharmaceutical Ingredients*. (2019).
4. Hofrichter, M., Kellner, H., Pecyna, M. J. & Ullrich, R. *Adv Exp Med Biol* 851, 341–368 (2015).
5. Arnold Frances H, V. A. A. *Curr Opin Chem Biol* (1999).

DESIGN OF A PCR-RFLP FOR MALARIA GENOMIC SURVEILLANCE IN MEXICO

^{1,2}Gabriel Trejo Sánchez, ¹Giovanna Barrera Aveleida, ¹Edgar José Rundquist Sánchez, ¹Rodrigo Ramos Monteagudo, ²Gabriela Meneses Ruíz, ¹Carlos Wong Baeza, ¹Isabel Baeza Ramírez, ¹Albany Reséndiz Mora and ²Juan Manuel Serna Velázquez

Laboratorio de Biomembranas, Departamento de Bioquímica, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Prolongación Carpio y Plan de Ayala s/n, Colonia Santo Tomás, Alcaldía Miguel Hidalgo, Ciudad de México, CP.11340. Tel +52 55 57296000 Ext 62326.

Departamento de Parasitología, Instituto de Diagnóstico y Referencia Epidemiológicos, Dirección General de Epidemiología, Secretaría de Salud, Francisco de P. Miranda 177, Colonia Lomas de Plateros, Alcaldía Álvaro Obregón, Ciudad de México, CP. 01480. E-mail: ramongabrieltrejosanchez@gmail.com

Malaria is a potentially fatal disease endemic to tropical or subtropical countries, with a higher incidence in African and sub-Saharan countries. It is caused by protozoa of the genus *Plasmodium*; *P. falciparum* and *P. vivax* are two of the most lethal species. This disease is transmitted through the bite of female mosquitoes of the *Anopheles* genus. Although it can be prevented and treated, it remains a significant worldwide public health problem ⁽¹⁾. Malaria is spread by the bite of female *Anopheles* mosquitoes. Although preventable and treatable, it remains a global public health concern.

The World Health Organization presented the Global Technical Strategy against Malaria, which aims to reduce mortality and morbidity rates by 90% and eliminate the disease in at least 35 countries where transmission was maintained in 2015 ⁽²⁾. In Mexico, the decline in indigenous cases in recent years makes it a candidate for disease elimination by 2025. However, due to the increasing migration phenomenon, the number of imported cases has risen ⁽²⁾. So, in this project, we will design a PCR-RFLP targeting the *Plasmodium pfs47* gene to determine the potential geographical source of imported malaria cases confirmed by the National Reference Laboratory between 2021 and 2023. This work will enable the establishment of genomic surveillance for malaria in Mexico, contributing to the fulfillment of the WHO Strategy objectives against this disease.

Referencias

1. Su, X.Z., Lane, K.D., Xia, L., Sá, J.M. & Wellems, T.E. (2019) *Plasmodium* Genomics and Genetics: New Insights into Malaria Pathogenesis, Drug Resistance, Epidemiology, and Evolution. *Clin Microbiol Rev*, 32.
2. World Health Organization. 2023. World Malaria Report 2023. WHO.71:114

GENERATION AND CHARACTERIZATION OF ALBUMIN-CYSTATIN MICROPARTICLES FOR THE CONTROL OF PLANT PATHOGENS

Silvia Valdés Rodríguez, Laura E. Zavala García, Lino Sánchez Segura,
Karina Manzo Valencia, Víctor Olalde Portugal

Centro de Investigación y de estudios avanzados-Unidad Irapuato. Km 9.6 Libramiento Norte.
Carretera Irapuato-León C.P. 36824, Irapuato, Gto., México.
silvia.valdes@cinvestav.mx

ABSTRACT. The development of nanotechnology and materials science has enabled the use of nanomaterials in agriculture. The use of nanoparticles and antimicrobial agents represents a novel biotechnological alternative for the control of fungi and nematodes in agriculture. Our research group isolated and characterized the gene encoding an amaranth cystatin. Recombinant amaranth cystatin produced in *Escherichia coli* was able to inhibit the growth and development of some phytopathogenic fungi and the *Meloidogyne incognita* nematode *in vitro* assays. Recently, direct application of amaranth cystatin to tomato plants was found to prevent and control infection by these pathogens (Cervantes-Juan *et. al.*, 2020, 2023). Based on the above, in the present work, it was proposed to immobilize amaranth cystatin in albumin microparticles to evaluate its potential in the control of phytopathogens. For this purpose, albumin-cystatin microparticles were prepared under sterile conditions by ethanol desolvation and cross-linked with different concentrations of cystatin by heating at 70 °C. The yield of microparticles decreased when high concentrations of encapsulated cystatin (500 to 840 $\mu\text{g}\cdot\text{mL}^{-1}$) were used. Transmission electron microscopy analysis showed that the size and shape of the microparticles changed with increasing cystatin concentrations. Microparticles without cystatin showed a predominantly circular shape, whereas, in treatments from 25 to 840 $\mu\text{g}\cdot\text{mL}^{-1}$, they showed a branched morphology with surface smoothing. The ζ -potential, the polydispersity index, and the hydrodynamic diameter of the aggregates, determined with a Zetasizer (Nano ZS90, Malvern Instruments), indicated that the electric charge of the surface of the microparticles (Z-potential) was negative. Nanoparticles with 50 $\mu\text{g}\cdot\text{mL}^{-1}$ cystatin showed the most negative charge, and their polydispersity index indicated a high homogeneity in size, ideal for subsequent antimicrobial evaluation.

References

1. Cervantes Juan M.M. *et. al.* (2020). Revista Mexicana de Ciencias Agrícolas 11, 567-580
2. Cervantes Juan M.M. *et. al.* (2023). Annals of Applied Biology, 183, 244-253
3. This research work is supported by CONAHCYT project CF-2023-I-1432.

EXPLORING FLUORESCENT PROTEIN ILOV AS VERSATILE INTRAGENIC REPORTER IN *CAPSICUM ANNUUM* L.

Lilian Gabriela Valencia-Turcotte¹, Felipe Leon-Garcilazo¹,
Manlio Alejandro González-Rivera¹ and Rogelio Rodríguez-Sotres^{*}

¹Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México,
Av. Universidad 3000, Col. Cd. Universitaria, Alc. Coyoacán, c.p. 04510. Cd. México

*E-mail: sotres@unam.mx

Plant phototropin is a blue light receptor kinase featuring LOV (light, oxygen, or voltage) domains. Their small size and quantum yield under anaerobic conditions make LOV fluorescent domains a feasible tag for plant protein. The green fluorescent protein (GFP) from a marine organism has been extensively used as tag, but here the base sequence from chili pepper LOV domain was tested as intragenic alternative tag, introducing only three-point mutations to enhance its quantum yield. Conceived as a viral infection reporter, iLOV has been expressed in bacteria, human cells, and *Arabidopsis*, but not in chili peppers. Suitable for subcellular protein localization studies in both plant and mammalian cells, the iLOV fluorescence did quickly recover from photobleaching and its intrinsic photochemistry rivals and may outperform GFP, in many applications¹.

Here, the coding sequences for one haloacid dehalogenase superfamily member (phosphate starvation 2; PS2), one soluble inorganic pyrophosphatase (PPa), or one membrane bound inorganic pyrophosphatase (PPv) were expressed in onion epidermis and callus of *Capsicum annuum* using a pCAMBIA 1300 vector with an intragenic cassette composed of the *C. annuum* promoter for fructose-1,6-bis-phosphate aldolase, one *C. annuum* ORF in frame with either *C. annuum* iLOV, or with GFP, and one of five putative terminators from the *C. annuum* genome.

The iLOV fluorescent signal in onion epidermis and chili pepper callus was of sufficient intensity in confocal microscopy. Transient expression of the construct in onion epidermis could be observed at both 24 and 48 hours, and constitutive expression was observed in calli after 60 days. The intensity and fluorescence distribution of iLOV signal was dependent on the fused protein used, in agreement with a vacuolar and cytoplasmic localization for chili pepper AVP and PS2, respectively. Based on these data, we propose iLOV as an intragenic non-toxic GFP replacement, for the generation of non-transgenic genetically modified plants.

References

1. Chapman, Faulkner, Kaiserli, et. al., (2008) PNAS 105, 20038-20043.

Funding. CONAHCyT-A1-S-8650, PAPIIT-DGAPA-IN219518, PAIP-FQ-5000-9122.

ESTIMATION OF ARSENIC CONCENTRATION IN DRINKING WATER WELLS USING THE AS BIOSENSOR *BSWCBGFPMUT3A*

Luz Idalia Valenzuela-García¹, María Teresa Alarcón-Herrera¹, Víctor Manuel Ayala-García²,
Luis Arturo Torres Castañón¹, José Rafael Irigoyen Campuzano¹,
Diego Armando Martínez Cruz¹, Mario Pedraza-Reyes³

¹Centro de Investigación en Materiales Avanzados, Calle CIMAV n°110 Ejido Arroyo Seco Durango, Dgo. México CP.34147,

²Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N.

Circuito Universitario. Col. Valle del Sur C.P. 34120., ³Departamento de Biología. Universidad de Guanajuato.

Noria Alta Guanajuato, Gto. México CP. 36050.

E-mail: luz.valenzuela@cimav.edu.mx

Arsenic (As) is a contaminant that causes serious health problems worldwide due to its presence in drinking water and its accumulation through the food chain; therefore, it is important to constantly monitor this element in the environment¹. A promising and low-cost strategy to quantify As in environmental samples is the use of whole-cell biosensors (WCBs). This approach takes advantage of the response to As in bacteria, in which As (III) inside the cell can unblock the repression of the *ars* operon to allow the expression of As defense genes².

In this work, we used a WCB for As detection, developed in *Bacillus subtilis* by fusing the promoter of the *ars* operon to the *gfpmut3a* gene in the pAD123 vector (*BsWCBgfpmut3a*)³. Using this biosensor, As concentrations in water samples, collected from 40 drinking water supply sites in the state of Durango, were estimated. For this purpose, a methodology was developed to quantify the As fluorescent response of *BsWCBgfpmut3a* after 2 hours of exposure to the water samples. The concentration of the pollutant in the water samples quantified with the biosensor were compared with those determined by ICP-MS, which allowed us to obtain parameters like precision and bias of the As estimation with the *BsWCBgfpmut3a*.

The results from both methods indicated that all the water samples analyzed exceeded the maximum permissible limits established by the WHO of 10 µg/L for drinking water, indicating a serious risk of exposure to As for the population of the region studied.

References

1. Genchi, C., Lauria, G., Catalano, A., M.S. Sinicropi (2022). Applied Sciences (Switzerland), vol.12. 6184.
2. Singh, M., Del Valle, M. (2015). Handbook of Arsenic Toxicology. 575-578.
3. Valenzuela-García, L. I., Alarcón-Herrera, M. T., Ayala-García, V. M. Barraza-Salas, J.M. (2023). Microbiology Spectrum.vol.11. N°4.

DESIGN AND GENERATION OF A GENETIC CONSTRUCTION FOR THE DISRUPTION OF THE *BACILLUS SUBTILIS* ARSR GENE

Evelin Aylin Valenzuela-Villalobos¹, Luz Idalia Valenzuela-García², Iván Meneses-Morales¹ and Víctor Manuel Ayala-García¹

¹Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur C.P. 34120., ²Centro de Investigación en Materiales Avanzados, Calle CIMAV n°110 Ejido Arroyo Seco Durango, Dgo. México CP.34147.
E-mail: valenzuelavi19@outlook.es

Abstract. *Bacillus subtilis* relies in two arsenic response operons: *ars* and *ase*. The *ars* operon, located in the skin element (intervening *sigma K*), is composed of the genes *arsR*, *yqcK*, *arsC*, and *arsB*. ArsR encodes the transcriptional repressor of the operon; *arsC* is an arsenate reductase, *arsB* product function as an arsenite efflux pump, and *yqcK* is an apparent C-As lyase; all these proteins conferring resistance to arsenate and arsenite¹. The transcription pattern of the *ars* operon involves intracellular arsenite binding to the repressor (ArsR), inducing a conformational change that dissociates the promoter, allowing continuous expression².

The interaction between arsenite and ArsR results in the dissociation of the repressive protein from the DNA, allowing the transcription of the operon. In the absence of arsenite, ArsR remains bound to the operator region, inhibiting the expression of the genes *arsR*, *yqcK*, *arsB*, and *arsC*. However, in the presence of arsenite, ArsR dissociates from the operator, enabling the expression of the three genes and the detoxification of the contaminant¹. Additionally, As (III) induces expression at lower concentrations than As(V), as As (V) must be reduced to As (III) before ArsR can bind to undergo a conformational change.

This study aims to create a genetic construct capable of disrupting the *arsR* repressor gene while allowing for a transcriptional fusion with the operon promoter and avoiding polar effects on downstream genes of the operon. The vector pMutin4 will be used for this purpose. Generating this mutant will enable the understanding of the effects of repressor absence on *ars* operon expression, modulation of other operon genes expression, and understanding its contribution to arsenic response.

References

1. Sato, T., & Kobayashi, Y. (1998). Journal of Bacteriology, vol 180.1655-1661.
2. Yu, X., Zheng, W., Bhat, S., Aquilina, J. A., & Zhang, R. (2015). PeerJ, 2015 vol 9. peerj.1230
3. Moore, C. M., Gaballa, A., Hui, M., Ye, R. W., & Helmann, J. D. (2005). Molecular Microbiology, vol 57.j.1365-2958.2005.04642

PHYSIOLOGICAL EVALUATION OF RECOMBINANT FORMS OF GRANULOCYTE COLONY-STIMULATING FACTOR

Miguel Ángel Valle Yañez¹, Leandro Alberto Núñez Muñoz¹, Berenice Calderón Pérez¹, América Padilla Viveros²
Beatriz Xoconostle Cázares^{1,2}, Roberto Ruiz Medrano^{1,2}

¹Depto. Biotecnología y Bioingeniería, ²Doctorado Transdisciplinario para la Sociedad. Centro de Investigación y de Estudios Avanzados del IPN. Av. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, 07360 Ciudad de México, CDMX
E-mail: rmedrano@cinvestav

Granulocyte colony-stimulating factor (G-CSF) is a hematopoietic cytokine that plays an important role in the stimulation, proliferation, mobilization, maturation, and activation of granulocytes, especially neutrophils¹. This pharmaceutical protein is mainly used as an adjuvant in the fight against neutropenia, which is the decrease in neutrophils in the blood. This condition is mainly caused by infectious diseases or is a secondary effect in patients under chemotherapy and radiotherapy². G-CSF presents four mRNA variants and, therefore, four isoforms (a, b, c y d), while isoform b is commercially produced.

In this study, synthetic genes of the G-CSF isoforms b and c were designed and expressed in *E. coli*, displaying a molecular weight of 17 kDa. The production of both isoforms in an aerobic bioreactor allowed to assess its biological activity employing a cell line culture. Subsequently, to increase the circulation time in mammals, the isoforms were PEGylated. The production of recombinant G-CSFs and their biological assays will be presented, including a strategy to scale up the synthesis for pharmaceutical use.

References

1. Anderlini, P., Przepiorka, D., Champlin, R., & Korbling, M. (1996). *Blood*. 88(8), 2819-2825.
2. Vanz, A. L., Renard, G., Palma, M. S., Chies, J. M., Dalmora, S. L., Basso, L. A., & Santos, D. S. (2008) *Microbial Cell Factories*. 7(1), 13.

IMPORTANCE OF UNDERSTANDING THE CORRELATION BETWEEN CHRONIC INFLAMMATION, THE IMMUNE RESPONSE AND MEMBRANE REMODELING, AS A RESTORATIVE TREATMENT STRATEGY

Alicia Reveles Espinoza¹, Lizeth Abad Alvarez², Iván Salgado¹, Isaac Chairez³, Andrés Rivera Chávez², Luis Figueroa-Yáñez⁴, Jesús Bernardino Velázquez-Fernández⁵, Karen Olivia Rojas², Saé Muñiz⁶, Flor Yohana Flores², Ernesto Prado Montes de Oca², Eduardo Padilla Camberos², Alba Adriana Vallejo Cardona^{2*}

¹Medical Robotics and Biosignals Laboratory, Centro de Innovación y Desarrollo Tecnológico en Cómputo, Instituto Politécnico Nacional (IPN), Mexico City. ²Medical and Pharmaceutical Biotechnology Unit, Center for Research and Assistance in Technology and Design of the State of Jalisco (CIATEJ) Jalisco, Mexico. ³Technologic of Monterrey, Institute of Advanced Materials for Sustainable Manufacturing, Jalisco, Mexico. ⁴Industrial Biotechnology Unit, Center for Research and Assistance in Technology and Design of the State of Jalisco (CIATEJ), Jalisco, Mexico. ⁵Consejo Nacional de Ciencia y Tecnología (CONACYT), Mexico City. ⁶ National Cancer Institute, Mexico City

During the generation of neoplasia and inflammation, some signaling processes are activated and correlate closely with both pathologies. Membrane remodeling occurs after activation of immune response, implying that membrane fluidity plays a role during protein function changes, which are derived from neoplasia and inflammation. Thus, the present work aimed to identify the possible association between different cell changes that occur during neoplasia by means of genetic and protein analysis using deep learning. Specifically, the analysis focused on pancreatic cancer cases found in the database of GDC Data Portal Homepage–National Cancer Institute. Besides, to get a better insight, experimental evidence was investigated. A proteomic study of three pancreatic cancer cell lines was carried out to analyze differential protein expression during proliferation, which may contribute to inflammation, immune response, and membrane remodeling.

Analysis by deep learning identified four genes EPHA2, H3C1, SRC and EGFR, which were also identified in the three cell lines, except for the SRC gene. Furthermore, with proteomic analysis of the three cell lines, a protein Tyrosine-protein kinase found only in the MIA PACA2 cell line was identified. These results highlight the genes and proteins associated with inflammation, the immune response, and the membrane. Such proteins represent potential therapeutic targets or biomarkers useful for diagnosis and therapy.

ELUCIDATION OF LACTOSE METABOLISM IN *ACTINOBACILLUS SUCCINOGENES* 130Z

Nadia Mariela Varela Pérez¹, Angélica Vallejo Giraldo¹, Juan Carlos Fragoso Jiménez¹, Luz María Martínez¹, Georgina Hernández Chavez¹, Claire Vielle², Alfredo Martínez Jiménez¹, Guillermo Gosset Lagarda¹

¹Instituto de Biotecnología, UNAM, Avenida Universidad 2001, Cuernavaca Morelos 62210.

²College of Natural Sciences, MSU, East Lansing, Michigan USA 48824
guillermo.gosset@ibt.unam.mx, Phone: 7773291601, 62210

Actinobacillus succinogenes is a Gram-negative bacterium with natural high-yield succinic acid production capacity. Analysis of the genome sequence of *A. succinogenes* showed genes with possible function in lactose transport and metabolism¹. The objective of this work was to study lactose consumption and the genes involved in this function. A mutant lacking gene *Asuc_1398*, which encodes a probably β -galactosidase was generated. It was determined that this gene is essential for lactose consumption and it encodes a protein with β -galactosidase activity. Transcriptomic analysis of *A. succinogenes* in culture with lactose as carbon source showed 24 overexpressed and 14 downregulated genes. Proteomic analysis showed 36 proteins with increased level and 58 with decreased level in the presence of lactose. Both analyses identified possible genes involved in lactose metabolism and the global response to this carbohydrate in *A. succinogenes*.

References

1. McKinlay, J. B., Laivenieks, M., Schindler, B. D., McKinlay, A. A., Siddaramappa, S., Challacombe, J. F., Lowry, S. R., Clum, A., Lapidus, A. L., Burkhart, K. B., Harkins, V., y Vielle, C. (2010). *BMC genomics*. Vol. 11, 680.

REVALUATION OF METZAL RESIDUE AS A TEXTURIZING AGENT IN THE VEGETATIVE DEVELOPMENT OF LETTUCE (*LACTUCA SATIVA* L.)

Carlos Getzrael Vazquez-Osorno¹, Josefa Espitia-López¹, Oscar Arce-Cervantes¹,
Alejandro Angel-Cuapio², Benito Flores-Chávez¹, Paul Misael Garza-López¹

¹Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo. Tulancingo, Hgo. 43600

²Tecnológico Nacional de México/TES de Ecatepec. Ecatepec, Edo. Mex. 55210

Número: 7714705040 E-mail: va277008@uaeh.edu.mx

The maguey pulquero (*Agave salmiana*) is the source of the residue known as metzal. This residue exhibits woody characteristics due to its lignocellulose content, which makes it an ideal substrate for growing vegetables, since it provides some physicochemical properties to the soil (1). Furthermore, some studies have demonstrated the effectiveness of using lignocellulosic waste as texturizing substrates in vegetable cultivation (2).

Lettuce seedlings (*Lactuca sativa* L. var. Iceberg) were transplanted in 7.85 L bags, five treatments were carried out with different concentrations of metzal (0%, 5%, 10%, 20% and 30%), each with five repetitions. At 70 days after transplanting, the following variables were measured: fresh weight (kg), root length (cm), pH, electrical conductivity (EC, mS/cm), and concentrations of Na⁺, Ca²⁺, NO³⁻ and K⁺ (ppm). A completely randomized design was used, and the data were analyzed using ANOVA, with significant differences determined by Tukey's multiple range test ($\alpha=0.95$). Additionally, a principal component analysis (PCA) was performed to identify behavioral trends between the different treatments and variables (XLSTAT by Lumivero® 2023.1).

It was observed that the treatment with 20% metzal was the most effective, with root growth measurements of 21.1 cm and fresh weight of 0.56 kg, these values being significantly higher ($P<0.05$) compared to the other treatments. On the other hand, the rest of the variables did not show significant differences between the treatments ($P>0.05$). In the PCA, the data presented a reliability of 88.69% with two main components, identifying two large clusters that relate the variables fresh weight, root length and K⁺ with the observations of the treatments at 10%, 20% and 30 %, while the observations of the 5% and 0% treatments showed a greater relationship with the rest of the variables.

The revaluation of metzal as a substrate is appropriate, since the observed trends indicate that the percentage of metzal directly influences lettuce development, with the treatment with 20% being the most effective.

References

1. Figueredo-Urbina, C. J., Medina-Pérez, G., Juárez-Muñoz, J., & Arce-Cervantes, O. (2023). *Mexican Journal of technology and Engineering*, 2, 10-20.
2. Reyes-Medina, A. J., Fraile-Robayo, D., & Álvarez-Herrera, J. G. (2019). *Revista Temas Agrarios*, 24, 34-41.

LIPID AND CAROTENOID PRODUCING YEAST RECOVERED FROM URBAN WASTEWATER: AN EVALUATION OF THEIR BIOTECHNOLOGICAL POTENTIAL

Dana Vázquez-de los Reyes, Viviana Escobar-Sánchez, Miguel Angel Orozco-González, Miguel Rosas-Paz, James González, Claudia Segal-Kischinevzky

Departamento de Biología Celular, Facultad de Ciencias, UNAM, Ciudad Universitaria, 04510, CDMX.
Tel. 5556224831; e-mail: dalorena@ciencias.unam.mx

Wastewater is a rich source of nutrients, such as nitrogen and phosphorus, which support the proliferation of organisms, including yeasts. Yeast offers substantial biotechnological, biomedical, and commercial value due to their ability to produce biomass, bioethanol, and lipids, which serve as precursors for biofuels, as well as pigments like carotenoids with applications in biomedical pharmacological and agricultural fields. This study aimed to identify the diversity of yeasts in a secondary effluent and to evaluate their biotechnological potential for lipid and pigment production. Yeasts were isolated using selective media and identified through PCR amplification of internal transcribed spacers (ITS), followed by sequencing and homology analysis with GenBank sequences. A total of 32 different yeast strains were isolated and identified from the “Atzintli” wastewater treatment plant; given the interest in discovering new oleaginous yeasts, an in-plate lipid production screening was carried out using Rhodamine B to stain intracellular oils under three different nutritional conditions and with two different carbon sources: glucose and glycerol. The yeasts were classified as non-oleaginous, poor, moderate, and good lipid producers. Predominantly, *Rhodotorula* strains, known for carotenoid production, were identified. These strains were further characterized through pigment production screenings conducted at different temperatures (10-37 °C) and with different carbon-nitrogen ratios: rich medium, diluted rich medium, and low-nitrogen minimal medium. Additional testing will expand our understanding of the biotechnological potential of these strains. The biological diversity observed in the wastewater samples revealed numerous strains with significant potential to produce lipids, biomass, and pigments. The strains can be exploited under different conditions of nutrient limitation, temperature, different carbon sources, and substrates, facilitating a circular economy and reinforcing the sustainability of diverse bioproducts.

IMMUNOPROTEOMIC IDENTIFICATION OF A TYPE 1 FIMBRIAE IN *KLEBSIELLA PNEUMONIAE*

Ulises Vega-Castillo¹, Carolina del Carmen Murúa López¹,
Salvador Velarde Félix² and Héctor Samuel López-Moreno^{1*}

¹Laboratory of Molecular Biomedicine, CAC-UAS-264; Graduate Program in Biomedical Sciences, School of Chemical and Biological Sciences., Universidad Autónoma de Sinaloa, Cd. Universitaria, Culiacán, CP.80010, Sinaloa, México.

²Genomic Medicine Unit; Hospital General de Culiacán

*hslmoreno@ms.uas.edu.mx; (52-6677137860)

Abstract: *Klebsiella pneumoniae* is a Gram-negative, anaerobic enterobacterium, known for causing various infections such as pneumonia, urinary tract infections, bacteriemia, and liver abscesses. The emergence of multiresistant hypervirulent strains has led to classifying this pathogen as a critical priority, suggest the imperative study of this bacteria. The immunoproteomic of *K. pneumoniae* allows us to identify antigenic proteins associated with resistance mechanism and other biological bacterial process, help us to clarify the host-bacteria relationship and identify antigens whit potential biomedical applications such specific diagnosis or vaccine design. First, we analyze the proteomic profile using three strains of *K. pneumoniae* (ATCC 13883, and two clinical isolates designed as Kp1 and Kp6) counting 158 individual spots-proteins distributed as follows: 36 for *K. pneumoniae* ATCC 13883, 71 and 51 for Kp1 and Kp6 respectively. Afterward, we detected 66 antigenic proteins in the immunoproteomic profile corresponding to 41.8% of the total proteomic profile. One antigenic spot of 18 kDa (p15) without previous report, was isolated and sequenced through mass spectrometry, obtaining two peptides AAVAFGLTAIDSAHPK and YYAIGEATPGAANADATFK which in silico homology analysis confirm it´s identify as type 1 fimbria of *K. pneumoniae*, a virulence factor involved in the host colonization, biofilm production and evasion of the host´s immune response.

EVALUATION OF THE EFFECT OF RNAI SILENCING ON THE PUTATIVE HYALURONIC ACID SYNTHASE (HAS) GENE OF *MUCOR LUSITANICUS*

Claudia Cecilia Vega-García, Ana Sofía Ramírez-Pelayo and Jorge Verdín*

Industrial Biotechnology Unit, Center for Scientific Research and Assistance in Technology and Design of the State of Jalisco (CIATEJ). Camino Arenero 1227, El Bajío, CP. 45019, Zapopan, Jal.

*jverdin@ciatej.mx

The cell wall is a dynamic structure that provides bacteria and fungi with several key properties that include rigidity, environmental stress protection, and, in some cases, also plays a role in pathogenicity. Certain pathogenic microorganisms build a pericellular capsule composed mainly of polysaccharides and proteins. This capsule plays a crucial role in adhesion and evasion of the host's defense systems. Particularly, *Cryptococcus neoformans* produces a pericellular capsule, which is rich in hyaluronic acid (HA) (Jong *et. al.*, 2007). Recent studies have identified proteins in filamentous fungi like *Coprinopsis cinerea* and *Mucor circinelloides* (*M. lusitanicus*, CBS 277.49) whose sequences are suggestive of having a hyaluronic acid synthase (HAS) activity (Franco-Herrera, Aranda-Barba, *et. al.*, in preparation). To characterize this putative activity, our research group employed both *in silico* and *in aquo* approaches that included heterologous expression in *Saccharomyces cerevisiae* and *in vitro* activity assays (Franco-Herrera *et. al.*; Aranda-Barba *et. al.*, unpublished). The present study aims to evaluate the biological role of this putative HAS in *M. lusitanicus*. To that end, RNA interference (RNAi) silencing of *M. lusitanicus* HAS encoding gene (HMPREF1544_11796) will be performed. Its impact on viability, pathogenesis, and cell wall composition in the silenced strain will be assessed. Progress will be shown during the congress. This work is being supported by CONAHCYT-Ciencia de Frontera 2019, grant 552259.

References:

1. Jong, A., *et. al.* (2007). "Identification and characterization of CPS1 as a hyaluronic acid synthase contributing to the pathogenesis of *Cryptococcus neoformans* infection." *Eukaryot Cell* 6(8): 1486-1496.
2. Trieu, T. A., *et. al.* (2017). "RNAi-Based Functional Genomics Identifies New Virulence Determinants in Mucormycosis." *PLoS Pathog* 13(1): e1006150.

EVALUATION OF THE MICROBIOLOGICAL AND ENZYMATIC SUSCEPTIBILITY OF ACRYLIC HYDROGELS CROSSLINKED WITH LIGNIN-MODIFIED

Gilberto Velázquez¹, Oscar-Kevin Reyes¹, Eulogio Orozco¹

¹Universidad de Guadalajara, Departamento de Química, Laboratorio de Bioquímica Avanzada, Marcelino García Barragán 1421, Colonia Olímpica, Guadalajara, Jalisco.
E-mail: gilberto.velazquez@academicos.udg.mx

Hydrogels are a family of crosslinked polymers that can capture large amounts of water within their structure, a phenomenon known as swelling. They are used in biomedical, personal care, bioseparations, metal extraction, and agriculture applications¹. Although it is convenient for hydrogels to be rigid and resistant to degradation in some circumstances, this property is detrimental on other occasions, especially those involving managing hydrogel environmental waste². Several strategies have been developed to make hydrogels biodegradable or biodegradation-susceptible. This work presents a suitable biotechnological option to promote the biodegradability of a hydrogel whose chemical crosslinker is lignin using the exposure to lignin-degrading microorganisms or the recombinant enzymes that allows accelerating the susceptibility to degradation and thus decreases the rigidity of the same. Two treatments were carried out, the first with *Streptomyces avermitilis* bacteria to degrade acrylic hydrogels crosslinked with modified lignin. After 90 days of exposition of the modified gel within a minimum nutrient culture of *S. avermitilis*, modifications in the hydrogel properties were observed: a reduction in the hydrogel swelling capacity and a decrement in its resistance to thermal degradation.

Intriguingly, enzymatic treatments using a recombinant laccase SilA from *Streptomyces ipomoeae* led to dramatic changes in the hydrogel's appearance. The characteristic yellow color of the lignin was lost, and the hydrogel's ability to shrink after heating was compromised. The original gel structure could not be restored, and the swelling properties and thermal susceptibility decreased by more than 50 %, indicating a substantial degradation of the crosslinker. This work provides a glimpse into the potential of biotechnological developments to facilitate the degradation of these materials.

Reference

1. Kasai, R. D., Radhika, D., Archana, S., Shanavaz, H., Koutavarapu, R., Lee, D. Y., & Shim, J. (2022). International Journal of Polymeric Materials and Polymeric Biomaterials, 72(13), 1059–1069.
2. Adjuik, T. A., Nokes, S. E., & Montross, M. D. (2023). Journal of Applied Polymer Science, 140(12), e53655.

DETERMINATION OF PARAMETERS FOR LOW-FREQUENCY ELECTROMAGNETIC STIMULATION OF MDA-MB-231 CELLS *IN VITRO*

Fernando Verduzco-Valdez¹, Jhonny Aguilar-Grajales¹, Andrey Salazar-Mendoza¹, Maximiliano Díaz-Inzunza¹, Angélica López-Izquierdo¹

¹Laboratorio de Bioingeniería, Facultad de Ingeniería Mexicali, Universidad Autónoma de Baja California, Blvd. Benito Juárez S/N, 21280, Mexicali, Baja California.
Tel: 686-243-67-18, e-mail: fernando.verduzco@uabc.edu.mx

Many biological systems, including humans, are exposed to extremely low-frequency electromagnetic fields (ELF-EMF) in the 1 to 300 Hz range¹. To date, the effects of ELF-EMF related to health remain unknown, as well as their possible therapeutic role for the treatment of major diseases such as breast cancer. Triple-negative breast cancer (TNBC) is one of the most aggressive cancer variants worldwide, accounting for 10-15% of all reported breast cancer cases². The lack of hormone receptors, its accelerated growth and metastatic capabilities, reduces treatment options and offers a worse prognosis for patients. The aim of this study was to determine the *in vitro* effects of low frequency electromagnetic radiation on the MDA-MB-231 (TNBC) cell line using different combinations of frequency and intensity in the generation of an ELF-EMF. Scratch wound assays and trypan blue exclusion tests were used to evaluate the migration and viability of MDA-MB-231 cells after exposure to low-frequency electromagnetic fields at different frequencies and intensities. A Helmholtz coil system, controlled by a graphic user interface, was used to stimulate the cells continuously for 48 hours prior to the analysis of the effects of EMR-BF on cellular properties. A decrease in cell viability accompanied by morphological changes was seen on the cells when stimulated with an electromagnetic field at a frequency of 60 ± 0.3 Hz and an intensity of $440 \mu\text{T}$. In addition, a slight reduction in the migration capabilities of the cells was observed after stimulation. These findings suggest that the parameters of 60 ± 0.3 Hz and $440 \mu\text{T}$ represent a combination of interest to further evaluate the effects of ELF-EMF as a new therapeutic basis for the development of non-invasive therapies targeting triple-negative breast cancer. This study is supported by the financial resources approved in the Convocatoria Interna de Proyectos de Investigación UABC 2024-2025.

References

1. Occupational Safety and Health Administration. (n.d). *Extremely Low Frequency (ELF) Radiation*. U.S DEPARTMENT OF LABOR. <https://www.osha.gov/elf-radiation>
2. American Cancer Society. (n.d). *Triple-negative breast cancer*. Information and Resources about for Cancer: Breast, Colon, Lung, Prostate, Skin | American Cancer Society. <https://www.cancer.org/cancer/types/breast-cancer/about/types-of-breast-cancer/triple-negative.html>

EFFECT OF VACCINE HB-ATV-8 ON CELL SIGNALLING IN A 3D *IN VITRO* MODEL OF ATHEROSCLEROSIS

Gloria Stephanie Villa-Jaimes¹, Sandra Calixto-Tlacomulco¹, Ismael Luna-Reyes¹, Blanca A. Delgado-Coello¹, Ma. Luisa Durán-Pastén², Danaí Montalván-Sorrosá³ and Jaime Mas-Oliva¹

¹Instituto de Fisiología Ceular, Universidad Nacional Autónoma de México, ²Laboratorio Nacional de Canalopatías, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, ³Instituto de Química, Universidad Nacional Autónoma de México. Apdo. Postal 70-245.
Phone: (+52) 55 5622 5584. E-mail: jmas@ifc.unam.mx

According to the World Health Organization, atherosclerotic cardiovascular diseases (ASCVD) are the leading worldwide cause of death, accounting for approximately 17.9 million deaths annually. The disease begins with the chronic accumulation of lipids in the arterial intima, due to the increase in plasma lipids where inflammation is a key component in the development of these diseases. ASCVD is related to other disorders such as metabolic-associated fatty liver disease (MAFLD).

Experiments performed in *in vivo* models of rabbit and pig by our group have shown promising results where the nasal vaccine HB-ATV-8, containing the peptide seq-1 derived from the C-terminus of the cholesteryl-ester transfer protein (CETP), prevents both atherosclerosis and fatty liver (1–3). In a 2D monoculture model of steatohepatitis, where HSC and HepG2 cells were treated with different fatty acid mixtures, we demonstrated that peptide Seq-1 decreases lipid internalization and downregulates profibrotic genes in HepG2 and hepatic stellate cells (HSC) exposed to Seq-1-treated steatotic HepG2 supernatants (4), suggesting that Seq-1 may indirectly regulate HSC activation.

In order to better understand the mechanisms by which Seq-1 helps prevent MAFLD and ASCVD, we developed a 3D *in vitro* model of atherosclerosis using microfluidic techniques that can mimic the structure of an artery. First, we encapsulated cells and Seq-1 in water-oil droplets to analyze the interaction between Seq-1 and endothelial cells. In a second microfluidic device used for cell growth we focused on the analysis of the communication among different populations of cells during the development of ASCVD. Following lipid overload of endothelial cells, the analysis under hydrodynamic, mechanical, and oxidative stress conditions was conducted using markers related to inflammation, lipid metabolism, and fibrogenesis.

References

1. García-González V, Delgado-Coello B, Pérez-Torres A, Mas-Oliva J. (2015). *Arch Med Res.* 427–37.
2. Gutiérrez-Vidal R, Delgado-Coello B, Méndez-Acevedo KM, Calixto-Tlacomulco S, Damián-Zamacona S, Mas-Oliva J. (2018). *Arch Med Res.* 49(7):456–70.
3. Carrillo-Tripp M, Reyes Y, Delgado-Coello B, Mas-Oliva J, Gutiérrez-Vidal R. (2023). *PPAR research.* <https://www.hindawi.com/journals/ppar/2023/8047378/>
4. Calixto-Tlacomulco S, Luna-Reyes I, Delgado-Coello B, Gutiérrez-Vidal R, Reyes-Grajeda JP, Mas-Oliva J. (2024). *Arch Med Res.* 55(2):102937.

PURIFICATION AND VALORIZATION OF URBAN EFFLUENTS USING PHOTOBIOREACTORS OPERATED WITH *ANABAENA INAEQUALIS*

Wendy Zárate-Hernández¹, Roger Emmanuel Sales-Pérez²,
Norma Alejandra Vallejo-Cantú³, Juan Manuel Méndez-Contreras⁴

Instituto Tecnológico de Orizaba¹, Ote. 9, Emiliano Zapata, 94320 Orizaba, Ver.
juan.mc@orizaba.tecnm.mx

In recent years, the global generation of wastewater has increased significantly. This rise is mainly attributed to population growth and the expansion of industrial processes. Such wastewater typically contains chemical and microbiological agents, with high levels of organic matter, nitrogen, and phosphorus being particularly notable. These components can cause environmental damage, the destruction of ecosystems, intestinal diseases in humans, and other serious issues¹. A biotechnologically viable alternative for wastewater treatment is the use of microalgae. This unconventional alternative is characterized by high efficiency, the absence of greenhouse gas emissions, low costs, and the production of biomass².

In this study, the microalga *Anabaena inaequalis*³ was used for the treatment of municipal wastewater. Before the experiments, the species was propagated in Soil-Water Medium⁴, a synthetic medium suitable for its growth. After 15 days of propagation, a concentration of 3.16×10^5 cells/mL was achieved. The wastewater treatment was evaluated over 21 days in batch photobioreactors, resulting in a reduction of organic matter concentration, analyzed through COD levels. Among the evaluated inoculum concentrations (5%, 15%, and 25%), the 25% inoculum achieved the highest COD removal (95%) and the greatest cell growth, reaching concentrations higher than those in the synthetic medium (1.82×10^6 cells/mL). Finally, the process successfully removed inorganic contaminants (nitrogen and phosphorus), which were utilized by the microalga for growth and the generation of microalgal biomass with potential clinical and energy applications, representing promising biotechnological alternatives for the future.

References

1. Ramírez, L. (2020). Agrobiología: Una visión general y sus aplicaciones.
2. Medina Mojica, I. Y. (2020). Las microalgas una alternativa para la remoción de materia orgánica de aguas residuales domésticas.
3. Cyanobacteria, and other aquatic objects. University of New Hampshire Center for
4. Freshwater Biology. <http://cfb.unh.edu/phycokey/phycokey.htm> 18 May 2022

PROTEOMIC AND METABOLITE PROFILE DURING THE GERMINATION OF AMARANTH SEEDS (*AMARANTHUS SPP.*)

Fernando Daniel Urbieta Villalobos, Antonio De León-Rodríguez, Ana Paulina Barba de la Rosa¹

¹IPICYT, Instituto Potosino de Investigación Científica y Tecnológica A.C., Camino a la Presa de San José 2055, Lomas 4ta Secc., 78216 San Luis Potosí, S.L.P., México.
E-mail: apbarba@ipicyt.edu.mx

The acceleration of climate change and the global population increase pose new challenges for humanity in agriculture and food security.¹ One of the recommended strategies to mitigate crop loss due to climate change is the introduction of stress-tolerant crops adapted to climate variability². Amaranth is a plant adapted to grow under various environmental conditions, including high temperatures, prolonged periods of drought, and poor soil conditions, making it an interesting crop for utilization². Seed germination occurs in three phases: the first involves rapid water uptake until full hydration, in the second a limited water uptake was observed, and the third phase begins with the rupture of the seed coat by the radicle, and characterized by another rapid increase in water uptake³. The second phase is crucial as it reactivates metabolism, including physiological and biochemical processes such as hydrolysis, macromolecule synthesis, respiration, subcellular structures, and cell elongation, leading to germination⁴. To date, there are no studies related to identify the molecular processes that occur during amaranth seeds germination. This study aimed to investigate the proteome profile and metabolites changes of cultivable amaranth seeds (*A. hypochondriacus* cv Nutrisol and *A. hypochondriacus* cv Cristalina) and wild seeds (*A. hybridus*) through the germination phases. Proteins were fractionated as polar and non-polar and differentially accumulated proteins were identified by 1-DE LC-MS/MS approach. Volatile and semi-volatile metabolites were analyzed by GC-MS. Differences in germination were observed, cultivable species showed faster germination and development compared to the wild species. Protein degradation was also observed at the third phase of germination. Similar metabolite profile, including fatty acids, alkanes, alcohols, phenols, and phytosterols were observed amongst different seed species; however, *A. hypochondriacus* cv Nutrisol showed a lower number of isoprenoids and different phytosterol composition compared to cv Cristalina and *A. hybridus*. A decrease in squalene and an increase in stigmasterol were observed as germination progressed. Changes in proteomic and metabolite profile will be discussed and compared amongst species and its possible relation with seed vigor and germination.

References

1. Kijak H, Ratajczak E. (2020). Int. J. Mol. Sci. 21(10), 3612. 2Alemayehu FR, et. al.. (2015). J. Agr. Crop Sci. 201(5), 321-329. 3Nonogaki, et. al.. (2010). Plant Sci. 179(6), 574-581. 4.Ali AS, Elozeiri AA. (2017). Adv. Seed Biol. 2017, 141-166.
2. Thanks to CONAHCYT, Fronteras de la ciencia 2023 (proyecto no. CF-2023-G-674)

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex of atoms, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

GENETICS, EPIGENETICS AND GENETIC REGULATION

CLINICAL VALUE OF GENE EXPRESSION PROFILING BY DIGITAL PCR FOR THE DETECTION OF LEUKEMIC CELLS IN PEDIATRIC POPULATION

Jesús García-Gómez^{1,2}, Aleyda G. Zamudio-Martínez², Josefina Dalia Ramírez-Ramírez³, Rosana Pelayo-Camacho^{3,4}, Luis Felipe Jave-Suárez² and Adriana del Carmen Aguilar Lemarroy²

¹Doctoral Program of Biomedical Sciences, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara, Guadalajara, Jalisco, ²Immunology Division, Centro de Investigación Biomédica de Occidente-IMSS, Guadalajara, Jalisco,

³Pediatric Oncoinmunology and Cancer Cytomics Laboratory, Centro de Investigación Biomédica de Oriente, IMSS, Metepec, Puebla, ⁴Research and Education Unit, IMSS, Ciudad de México.

E-mail: jesusgarmz@gmail.com

Acute lymphoblastic leukemia (ALL) is the most frequent pediatric malignancy with the gold standard for its diagnosis being immunophenotyping by flow cytometry. Our team recently identified a group of highly expressed genes in bone marrow samples of pediatric ALL patients through CGH arrays. In this work, we validated the elevated expression of some of those genes employing digital PCR (dPCR), an emerging technique that has gained relevance in the field for the enhanced sensibility and reproducibility it provides. In short, we analyzed 33 bone marrow samples, immunophenotype (EuroFlow) was determined and RNA was extracted to obtain cDNA. We conducted an initial assessment for the expression of 18 relevant genes using real-time PCR (qPCR), those genes with most notable overexpression in the leukemia group (n=23) compared to individuals without leukemia (n=5) and minimal residual disease (MRD) negative patients (n=3) were evaluated using dPCR. Thus, we measured mRNA expression of *JUP*, *CNP*, *NT5C3B*, *C-MYC* and *BIRC5* in the QIAcuity One instrument obtaining absolute quantification values (number of copies per reaction). Data was analyzed using RStudio and statistically significant differences were established using Mann-Whitney U test. Our results show that *JUP*, *CNP*, *NT5C3B*, *C-MYC* and *BIRC5* are significantly overexpressed in bone marrow samples of leukemia patients. This suggest that these genes possess potential value for detecting leukemia cells in pediatric population as a gene panel evaluated through dPCR. This method offers high sensitivity even in samples of limited volume, eliminates the need for calibrator samples, and provides absolute values for gene expression at the mRNA level.

EXPRESSION OF THE CCD4-3 GENE IN DIFFERENT TISSUES OF THE N4 AND P12 MORPHOTYPES OF *BIXA ORELLANA* L. BY QRT-PCR REAL TIME

Margarita Aguilar-Espinosa¹, Neimi Estrella-Casanova², Víctor Manuel Carballo-Uicab³,
Renata Rivera-Madrid¹, Ivan Estrada-Mota²

¹Unidad de Biología Integrativa. Centro de Investigación Científica de Yucatán, A.C. Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo, Mérida, Yucatán, México. CP 97205. ²Instituto Tecnológico Superior de Calkiní en el estado de Campeche. Campeche, Camp. México. ³Universidad de Guanajuato, División de Ciencias de la Vida, Posgrado en Biociencias. Ex Hacienda El Copal Km. 9, Carretera Irapuato-Silao, Irapuato, 36500 Guanajuato, Mexico.

Contact renata@cicy.mx

Summary. *Bixa orellana* L. is a plant of great commercial importance, due to the red-orange pigment accumulated in its seed coat, known as bixin. In the analysis of the transcriptome of *Bixa orellana* L., a group of genes belonging to the CCD (Carotenoid dioxygenases), ADH (Aldehyde dehydrogenases), and MET (methyltransferases) families were proposed, which could potentially be involved in the biosynthetic pathway of this pigment.¹ One of the most identified and analyzed families are the CCDs whose biological and chemical functions have been confirmed by in vitro assays in *E. coli* using achiote leaf and immature/ripe seed tissues as a study model.^{2,3} In this research, a comparative analysis of gene expression of the CCD4-3 transcript was carried out in 4 tissues, root, stem, leaf, and flower of achiote (*Bixa orellana* L.) in seedlings of morphotypes N4 and P12. The analysis was performed with the real-time polymerase chain reaction (PCR) quantification technique, taking as reference the expression of the 18S ribosomal gene and internal control immature seeds of stage S3 for the standardization of gene expressions in the tissues evaluated. The data obtained show a differential expression of the CCD4-3 gene in all tissues analyzed in this study, indicating that it tends to be expressed preferentially in non-photosynthetic tissues: root, flower, and seeds of the P12 accession whose expression levels are relatively higher than in N4, which produces more bixin content. The results reveal that possibly the CCD4-3 gene is the cause of the white flower phenotype due to the degradation of carotenoids caused mainly by the overexpression of this gene.

Bibliography

1. Cárdenas-Conejo, Y., Carballo-Uicab, V., Lieberman, M., Espinosa, M. A., Coma, L., & Madrid, R. R. (2015) doi:10.1186/s12864-015-2065-4
2. Carballo-Uicab, V. M., Cárdenas-Conejo, Y., Vallejo-Cardona, A. A., Aguilar-Espinosa, M., Rodríguez-Campos, J., Serrano-Posada, H., Rivera-Madrid, R. (2019). doi:<https://doi.org/10.7717/peerj.7064>, <https://doi.org/10.7717/peerj.7064>
3. Us-Camas, R., Aguilar-Espinosa, M., Rodríguez-Campos, J., Vallejo-Cardona, A. A., Manuel, C.-U. V., Serrano-Posada, H., & Rivera-Madrid, R. (11 de February de 2022). . Plant Sci. doi:<https://doi.org/10.3389/fpls2022.829089>

AGAVE AS A MODEL TO UNDERSTAND THE MOLECULAR GENETIC CIRCUITS CONTROLLING DEVELOPMENT AND CELL WALL METABOLISM OF HARD FIBERS

Fulgencio Alatorre-Cobos¹

¹Centro de Investigación Científica de Yucatán. Unidad de Biología Integrativa.
Calle 43 130 x 32 and 34, Chuburná de Hidalgo, 97205, Mérida, México
E-mail: fulgencio.alatorre@cicy.mx

Recently, the world economy is coming back to natural and renewable resources to decrease the negative impacts on the environment. Plant fibers are widely used in clothing, paper, building materials, textiles, mats, etc. Plant-based nanocelullose and lignin have potential in medicine and biopolymers. Plant hard fibers are composed of sclerenchyma cells, which have been little explored in science because of suitable model plants. Agave species have world relevance because of spirits like tequila and mescal; however, they are also a valued source of hard fibers (Morán-Velázquez et al., 2023). In America, Mexico and Brazil are the main producers of hard fibers obtained from *Agave fourcroydes* and *A. sisalana*, respectively. Our research group has contributed to gaining knowledge about fiber formation. Using cell biology, metabolomics, and RNAseq approaches, fiber development, and patterns have been revealed in *A. fourcroydes* (Morán-Velázquez et al., 2020), and by the first, our group reported all the expressed biosynthetic genes involved in cellulose and lignin formation in *A. tequilana* (Maceda-López et al., 2022). Tissue-specific transcriptome sequencing has revealed key players (biosynthetic genes and transcription factors) controlling fiber development in *A. fourcroydes*; in situ activity of peroxidases and laccases detected in leaves suggest autonomous and semi-autonomous mechanisms operating during Agave fiber lignification (Cruz-Balam et al., unpublished data).

References

1. Morán-Velázquez D.C., Maceda-López L. F., Lorena Rodríguez-López L., Vilaplana F., and Alatorre-Cobos F. 2023. Anatomía, composición química y genómica de las fibras de Agave spp. Los Agaves y sus derivados. R. M. Camacho Ruiz, A. Gutiérrez Mora, A. C. Gschaedler (Ed.). CIATEJ, Guadalajara, México. 93-109.
2. Morán-Velázquez D.C., Monribot-Villanueva J. L., Bourdon M., Thang J.Z., López-Rosas I., Maceda-López L. F., Villalpando-Aguilar J. L., Rodríguez-López L., Gauthier A., Trejo L., Azadi P., Vilaplana F., Guerrero Analco J. A. and Alatorre-Cobos F. 2020. Unravelling chemical composition of Agave spines: news from Agave fourcroydes Lem. *Plants* 2020, 9, 1642.
3. Maceda-López L. F., Góngora-Castillo E. G., Ibarra-Laclette E., Morán-Velázquez D. C., Girón Ramírez A., Bourdon M., Villalpando-Aguilar J. L., Toomer G., Tang J. Z., Azadi P., Santamaría J. M., López-Rosas I., López M. G., Simpson J., and Alatorre-Cobos F. 2022. Transcriptome mining provides insights into cell wall metabolism and fiber lignification in Agave tequilana Weber. *Plants*, 11 (11): 1496.

DETERMINATION OF *NR3C1* GENE METHYLATION AND EXPRESSION LEVELS IN PERIPHERAL BLOOD AND POST-MORTEM BRAIN TISSUE AND THEIR ASSOCIATION WITH SUICIDE.

Yazmín Mercedes Amador Segovia¹, Marcelo Barraza-Salas¹, Norma Urtiz Estrada¹, Víctor Manuel Ayala-García¹, Adolfo Padilla Mendiola¹, Juan Antonio Rojas Contreras², Jesús Bernardo Páez Lerma²

¹Facultad de Ciencias Químicas UJED, Av. Veterinaria S/N, Circuito Universitario, 34120 Durango, México. ²Instituto Tecnológico de Durango ITD, Blvd. Felipe Pescador 1830 Ote. Col. Nueva Vizcaya. Durango, Dgo. México C.P. 34080.

Address: Cecilia Valdez Luna 304, Col Amalia Solórzano, CP 34016.

Email: yazminamador22@gmail.com, cel. 6181578271

Suicide is a worldwide public health problem, due to the sustained increase in recent years, with around 800,000 people committing suicide annually, which represents one death every 40 seconds. In the last decade, the influence of environmental stimuli on the addition of marks in the genome, known as epigenetic marks, and how they limit gene expression, has been studied. One of these marks is the methylation of the CpG islands; the presence of methylation of these islands in the promoter region of the *NR3C1* gene, an important gene in the response to stress in suicidal subjects, can be considered a probable cause of the decrease in its expression, thus potentially leading to a decreased ability to cope with stressors, increasing the risk of suicide. Therefore, one of our objectives was to determine the methylation levels of the promoter zone of the *NR3C1* gene, as well as the expression of this gene in the prefrontal cortex and in peripheral blood in search of a possible biomarker, in suicidal individuals and control subjects. This analysis was performed using real-time PCR (qRT-PCR), and Taqman probes to determine gene expression, as well as using the OneStep qMethyl™ kit to determine the percentage of methylation in a specific part of the promoter region of the gene. It was determined if there is any difference in gene expression using the $\Delta\Delta C_t$ method, as well as to determine if there is any difference in the percentage of methylation between cases and controls that could influence the decision to commit suicide. The results showed significant differences in both groups in the relative methylation of the promoter region of the *NR3C1* gene in the prefrontal cortex ($p < 0.003$). The samples of our group of suicidal individuals showed a high methylation in the analyzed region. Also, according to the Mann Whitney U test, significant differences were found between cases and controls in the relative expression of the *NR3C1* gene in the prefrontal cortex ($p = 0.044$), with a higher expression of the gene in our group of individuals who died by suicide. However, no significant differences were found between cases and controls ($p < 0.057$) in the relative expression of the *NR3C1* gene in peripheral blood. The relative expression of *NR3C1* gene in the prefrontal cortex and peripheral blood did not correlate with each other, because the significance value obtained was > 0.05 . The relative expression of the *NR3C1* gene in the prefrontal cortex did not correlate with the percentage of methylation ($p > 0.05$). These results were evaluated with a 95% confidence interval.

ASSOCIATION OF THE SNP HLA-DRB1*07:01, HLA-B*55:01 AND IGE LEVELS WITH HYPERSENSITIVITY REACTIONS TO BETALACTAMIC ANTIBIOTICS IN COVID-19 PATIENTS IN THE MEXICAN POPULATION

Andrade Valdez Paola Nataly, Dra. Mónica Griselda Arellano Mendoza, Dr. Rodrigo Romero Nava

¹Laboratory of chronic degenerative diseases, Higher School of Medicine -IPN.
Plan de San Luis y Salvador Díaz Mirón S/N, Col. Santo Tomás, C.P:11340, Ciudad de México.
qfbpao78@outlook.com

Genetic factors play an important role in betalactam (BL) allergy, which are related to the synthesis of IgE and T lymphocytes. COVID-19 has also been associated with allergic sensitization, leading to increased susceptibility induced by this virus, triggering adverse reactions to certain medications such as BL. In this context, predisposing genetic factors have been found that influence severe adverse reactions caused by these medications. Therefore, the aim is to evaluate the association of SNPs HLA-DRB107:01, HLA-B55:01, and IgE levels with allergy in COVID-19 patients in the Mexican population. The association of polymorphisms found in the HLA-DRB1 and MICA genes with BL allergy in Mexican patients who had COVID-19 will be evaluated, as well as IgE levels in hypersensitivity reactions, using a non-probabilistic convenience sampling method. This association seeks to reflect the influence of IgE pathway genes and T lymphocytes in this pathology.

FREQUENCY OF *KDR* MUTATIONS IN THE VGSC OF *Aedes Aegypti* VECTOR

Annete Itzel Apodaca-Medina¹, Jesús Omar García-Jiménez², Praxedis Félix-Alcalá²,
Elisa Analí Camacho-Ureta¹, Sergio Alonso Durán-Pérez³

¹Unidad de Investigación en Biotecnología Biomédica, Universidad Autónoma de Occidente, Unidad Regional Culiacán, Boulevard Lola Beltrán y Rolando Arjona, colonia 4 de marzo, Culiacán, Sinaloa, México. ²Licenciatura en Ciencias Biomédicas, Universidad Autónoma de Occidente, Unidad Regional Culiacán, Boulevard Lola Beltrán y Rolando Arjona, Colonia 4 de marzo, Culiacán, Sinaloa, México. ³Posgrado en Biotecnología, Facultad de Ciencias Químico-Biológicas, Universidad Autónoma de Sinaloa, Boulevard Las Américas y avenida Universitarios, Ciudad Universitaria, Culiacán, Sinaloa, México.

E-mail: annete.apodaca@uadeo.mx

Phone: 6672064337

The *Aedes aegypti* (*Ae. aegypti*) mosquito is the primary vector of viral diseases including Zika, chikungunya and dengue¹. One of the strategies to reduce the incidence of these diseases is vector control using various insecticides such as pyrethroids. However, in recent years, vector resistance to these insecticides has been detected due to the presence of mutations in the voltage-gated sodium channel (VGSC) gene, known as *kdr* mutations, including V410L, F1534C and V1016G, which cause a change in the target site that prevents mosquito death upon exposure². For this reason, arboviral diseases transmitted by *Ae. aegypti* continue to be a serious public health problem worldwide; therefore, the search for the mutations that confer resistance to these vectors in Sinaloa is essential to propose new strategies to reduce the mosquito population and contribute to reducing the number of cases of diseases such as dengue. Therefore, in this study, AS-PCR and sequencing were used to search for *kdr* mutations in *Ae. aegypti* mosquitoes from the state of Sinaloa that are resistant to commonly used pyrethroid insecticides. As a result, high frequencies of V410L and V1016I mutations were found, showing complete dominance in the *Ae. aegypti* population from Culiacán Sinaloa, a fact that is related to resistance to pyrethroid insecticides such as permethrin and deltamethrin.

Referencias

1. Matthews JB (2019) *Aedes aegypti*. *Trends in Genetics* 35: 470-471.
2. Rochmijati WJ (2020) Frequency of *kdr* mutations in the voltage-sensitive sodium channel (VSSC) gene in *Aedes aegypti* from Yogyakarta and implications for Wolbachia-infected mosquito trials. *Parasites Vectors* 13: 429.

MEOX2 AS AN EPIGENETIC REGULATOR OF GENES INVOLVED IN LUNG CANCER CARCINOGENESIS

Leonel Armas, Mariana Garcia, Priscila Pineda, Federico Ávila

Universidad Nacional Autónoma de México (UNAM),
Facultad de Estudios Superiores (FES) Iztacala, Unidad de Biomedicina (UBIMED).
larmas@unam.mx, f.avila@unam.mx

Introduction. Homeobox-type transcription factors such as Mesenchymal Homeobox-2 (MEOX2), have been associated with the capacity for resistance to oncological drugs, progression and/or clinical prognosis in patients with lung cancer, however, the mechanisms involved are still remain to be elucidated. Objective: to analyze the positioning of MEOX2 and RNA Pol II on the epigenome of lung tumors. Methodology: Using ChIP assays, H3K4me3, H3K27ac, H3K9me3 and H3K27me3, as well as RNA Pol II on samples from patients with non-small cell lung carcinoma (NSCLC), and hybridization on DNA microarrays. Results: a profile of promoter sequences was identified by bioinformatic analysis in a significant way ($FDR \leq 0.1$) corresponding to 13 genes (ALDH1A2, MAN1A1, BTBD3, MMP24, DMD, RUFY3, FHL1, TANC1, GLI-1, TLX3, HRH1, WWP1, ZEB1). It was shown that the transcriptional factor MEOX2 is positioned in the GLI-1 promoter region at position -2,192 to -109 accompanied by RNA Pol II and activation histones H3K27ac and H3K4me3. Through siRNA assays, it was demonstrated that MEOX2 and GLI-1 are involved in cisplatin resistance, as well as involved in migration and colony formation. Finally, the analysis of survival of patients under pharmaco-oncological treatment allowed us to confirm that the expression of GLI-1 dependent on MEOX2 is associated with clinical progression and poor overall survival, this carried out in a cohort of patients with a diagnosis of NSCLC, EGFR- Wild Type, or EGFR-mutated under TKI-directed therapy.

Conclusion. Our results from the study of the epigenome of NSCLC patients have allowed us to identify the overexpression of the MEOX2-GLI-1 axis biologically and clinically, involved in drug resistance and the capacity to respond to targeted therapy based on TKIs of the EGFR receptor in patients. NSCLC.

DETERMINING THE LENGTH OF THE MAOA-UVNTR POLYMORPHISM IN A SAMPLE OF SUICIDAL SUBJECTS AND CONTROL SUBJECTS

Samuel Arroyo Carranza¹, Marcelo Barraza-Salas¹,
José Manuel Salas Pacheco² and Daniel Francisco Ramos-Rosales²

¹Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango, Unidad Dgo. Av. Veterinaria S/N, Circuito Universitario. Col Valle del Sur C.P 34120, ²Instituto de Investigación Científica, Universidad Juárez del Estado de Durango, Av. Universidad y Fanny Anitua S/N, Zona Centro, C.P 34000.
Email: sathdays@gmail.com

Suicide is a multifactorial phenomenon and a major public health issue, causing over 700,000 deaths worldwide in 2019¹. Understanding the biological mechanisms underlying suicide helps identify risk factors that predispose certain individuals to self-harm.

Evidence suggests an imbalance in brain monoamine levels in suicidal individuals, possibly mediated by monoamine oxidase type A (MAO-A) dysregulation². Specific alleles of the MAOA-uVNTR polymorphism have been linked to enzyme activity profiles, as well as to psychopathologies associated with aggression, impulsivity, and suicide. Recent findings in our population revealed increased MAOA gene expression profiles in the hypothalamus of suicide victims³. A subsequent analysis showed that the MAOA-uVNTR polymorphism may influence the expression of MAOA in these cohort.

This study aims to determine the length of the MAOA-uVNTR polymorphism in a sample of 5 suicidal subjects and control subjects with different repeat alleles. PCR tests amplified the MAOA-uVNTR polymorphism using samples with different numbers of repeated alleles. The fragments were inserted into the pGEMT-Easy plasmid and cloned into *E. coli* DH5α cells. Fragment sizes were determined using Sanger sequencing by capillary electrophoresis. From the 5 sequenced fragments, the length of at least two was obtained, which allowed confirming the presumed length of the MAOA-uVNTR polymorphism in the observed expression assays.

References

1. Esparza-Reig, J., & Julián, M. (2024). Association between suicidal ideation and burnout: A meta-analysis. *Death studies*, 1-12. Advance online publication.
2. Lung, F. W., Tzeng, D. S., Huang, M. F., & Lee, M. B. (2011). Association of the MAOA promoter uVNTR polymorphism with suicide attempts in patients with major depressive disorder. *BMC medical genetics*, 12, 74.
3. Du, L., Faludi, G., Palkovits, M., Sotonyi, P., Bakish, D., & Hrdina, P. D. (2002). High activity-related allele of MAO-A gene associated with depressed suicide in males. *Neuroreport*, 13(9), 1195-1198.

CHROMATIN STATE CHANGES BETWEEN PRIMARY CORTEX ASTROCYTES INDUCED TO SENESCENCE AND GLIOSIS WITH PALMITATE

Karla Estephania Ávila-Galicia^{1,2*}, Michel López-Teros^{1,2}, Raúl Librado-Osorio³, Armando Luna-López³,
Ernesto Soto-Reyes⁴, Adriana Alarcón Aguilar¹, Mina Königsberg¹

¹Laboratory of Bioenergetic and Cellular Aging, Department of Health Sciences, Metropolitan Autonomous University, Iztapalapa, CDMX. ²Postgraduate in Experimental Biology, Universidad Autónoma Metropolitana, Iztapalapa, CDMX.

³Department of Basic Research, National Institute of Geriatrics, CDMX. ⁴Department of Natural Sciences, Metropolitan Autonomous University, Cuajimalpa, CDMX.

*kfanyrc@gmail.com

Obesity and aging are risk factors for the development of inflammatory processes in the central nervous system, as well as the emergence of neurodegenerative diseases and a decline in cognitive functions. During these processes, the appearance and accumulation of senescent astrocytes and the development of astrogliosis have been observed. These cellular events, which occur simultaneously in the brain, have been linked to the exacerbation of functional decline observed during aging. However, the understanding of their development and establishment is still not fully comprehensive.

In the present study, we aim to identify the changes in the chromatin state of primary cortical astrocytes from neonatal rats induced to gliosis and senescence through treatment with differential doses of palmitate, by evaluating the epigenetic marks H3K9me3 and H3K9ac, which are associated with the presence of heterochromatin and euchromatin, respectively. Additionally, we assessed the content of the enzymes Suv39H1 and Sirt1, which are involved in the establishment and removal of these marks. Evaluations were carried out through immunofluorescence and western blot assays of these molecules.

The results show an increase in the marks in both gliosis and senescence, as well as in the enzymes involved in their establishment. However, senescent astrocytes exhibit a significantly greater increase compared to the increase observed during astrogliosis.

We thank Dr. María de los Ángeles Guerrero-Aguilera from UAM-I for providing the necessary animals for this project. This work was supported by FORDECYT- PRONACES/263957/2020. KSAG and MLT are CONAHCyT fellows.

ELLAGIC ACID MODULATES THE MRNA EXPRESSION OF TNF PATHWAY, NLRP3 INFLAMMASOME, NFKB1 AND FTO IN ADIPOSE TISSUE OF DIET-INDUCED OBESE WISTAR RAT

Antonio Ávila Guerrero¹, Ángel Miliar García¹, Alejandro Barrios Nolasco², Jorge Cornejo Garrido²,
María Eugenia Jaramillo Torres³, Aarón Domínguez López¹

¹Laboratorio de Biología Molecular, Escuela Superior de Medicina (ESM), IPN, CDMX, México. ²Laboratorio de Biología Celular y Productos Naturales, Escuela Nacional de Medicina y Homeopatía (ENMH), IPN, CDMX, México. ³Laboratorio de polímeros, Departamento de Ingeniería Bioquímica, Escuela Nacional de Ciencias Biológicas (ENCB), IPN, CDMX, México.
E-mail: aavilag2300@alumno.ipn.mx

Introduction. Ellagic acid (EA) is a polyphenol, it can be found as precursor by the name of Ellagitannin as secondary metabolite in fruits and vegetables, such berries, nuts and wine, which can be metabolized by microbiome gut to liberate EA, it sustains anti-oxidant, anti-inflammatory, antimutagenic and antiproliferative properties.

Since ellagic acid possess anti-inflammatory properties, we can explore the ability to regulate different effects in the inflammatory response, in a variety of markers such Fat mass obesity associated FTO, Inflammasome NLRP3 (NLRP3, P50, P65, IkkA, IL-1 β , IL-18 and Caspase1) and other pro-inflammatory genes like TNF pathway.

Clinical studies in animal models confirm the potential role of EA as a therapeutical approach in the inflammatory process. FTO have been shown to affect obesity, body mass index, type 2 diabetes, cardiovascular disease, energy homeostasis, and inflammation. FTO exhibits efficient oxidative demethylation activity of abundant N6-methyladenosine (m6A). FTO knockdown with siRNA leads to an increased m6A level in mRNA, whereas FTO overexpression resulted in decreased m6A level in human cells. FTO suppresses the transcription of a distinct set of Interferon-stimulated genes (ISGs), including many known pro-inflammatory genes, and that this regulation requires its catalytic activity but is not through the actions of FTO on m6A but through the depletion of FTO that produces the activation of the transcription factor STAT3. Here we propose that EA produce a sequential inhibition of FTO transcription and expression of pro-inflammatory genes like TNF pathway and Inflammasome NLRP3.

Objectives. Evaluate the mRNA expression of FTO, NLRP3 inflammasome, TNF pathway, NFkB in qPCR

Methodology. Animal model: Wistar rats was randomly distributed into three groups: standard diet (SD), high-fat diet (HD), and high-fat diet plus ellagic acid (EA), with n=6 for each group. The EA dose of 50 mg/kg per day via intragastrical was administrated.

Retroperitoneal adipose tissue was obtained, isolated total RNA with TriZol methodology, synthesized cDNA and evaluated the expression of the mRNA of FTO, Inflammasome NLRP3, IL-1 β , IL-18, TNF pathway, and NFkB in qPCR.

Results. Ellagic Acid reduces the expression of FTO, pro-inflammatory genes of inflammasome NLRP3: NLRP3, IL-1 β and IL-18, Caspase1, and others: TNF and NFkB which expressions is reduced in group EA vs HD.

Conclusions. Ellagic Acid possess anti-inflammatory effects in obese Wistar rat adipose tissue, modulating the expression of FTO, inflammasome NLRP3 genes, TNF pathway and NFkB.

GENERATION OF THE *LIBR/FLCA* MUTANT OF *AZOSPIRILLUM BRASILENSE* SP7

Ana Margarita Balderas Peña¹, María Luisa Xiqui Vázquez¹, Alberto Ramírez-Mata¹,
Beatriz E. Baca¹ and Sandra Reyes Carmona¹

¹Laboratorio de la Interacción Bacteria-Planta, Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla. Av. San Claudio S/N Edif. IC11, Ciudad Universitaria. Puebla, Pue. México.
E-mail: sandra.reyes@correo.buap.mx

The *Azospirillum* genus colonize the roots of a wide variety of plants and promote plant growth by producing plant hormones such as auxins¹. The indole-3-acetic acid (IAA) is the primary and most essential auxin in plants. Its far-reaching effects are not limited to regulating plant growth; IAA also plays a role in regulating bacterial physiology, adaptation to stress conditions, and interactions between microorganisms². The main IAA production pathway in *A. brasilense* is that of the indole-3-pyruvic acid intermediate, where indole-3-pyruvate decarboxylase, encoded by the *ipdC* gene, is involved. To study the transcriptional regulation of *ipdC* gene, we identified a protein named as LibR. The mutant in this protein showed a decrease in the IAA biosynthesis³. This protein belongs to LuxR transcriptional regulators wherein FlcA appears as the protein with the highest identity to LibR (63%). FlcA controls the production of capsular polysaccharides, flocculation process, and the colonization of the root surface of wheat⁴. In this work bioinformatic analyses have been carried out on the relationship between the characteristics of both proteins, generating knowledge about their structure and composition. In addition, a double mutant of the *libR* and *flcA* genes was developed to evaluate the effect on the loss of these LuxR proteins regulators.

References

1. Dubey, A. P., Pandey, P., Mishra, S., Gupta, P., & Tripathi, A. K. (2021). Role of a fasciclin domain protein in photooxidative stress and flocculation in *Azospirillum brasilense* Sp7. *Research in Microbiology*, **172**(6)
2. Duca, D., & Glick, B. R. (2020). Indole-3-acetic acid biosynthesis and its regulation in plant-associated bacteria. *Applied Microbiology and Biotechnology*, **104**(20), 8607–8619.
3. Jijón M. S. (2017). Estudio de la regulación transcripcional del gen *ipdC* que participa en la biosíntesis de ácido indol-3-acético en cepas de *Azospirillum brasilense*. Tesis doctoral, Benemérita Universidad Autónoma de Puebla (BUAP), Centro de Investigación en Ciencias Microbiológicas, Puebla.
4. Pereg G. L., Paquelin, A., Gounon, P., Kennedy, I. R., & Elmerich, C. (1998). A transcriptional regulator of the LUXR-UHPA family, FlcA, controls flocculation and wheat root surface colonization by *Azospirillum Brasilense* Sp7. *Molecular Plant-microbe Interactions*, **11**(3), 177-187.
5. Acknowledgments: We thank to Vicerrectoría de Investigación y Estudios de Posgrado (VIEP) for financial support

MEDIATOR 18 MEDIATOR COMPLEX SUBUNIT ASSOCIATION IN PHOSPHATE SCARCITY RESPONSE

María Fernanda Ballesteros-Barrera¹, Adrián Ávalos-Rangel¹,
León Francisco Ruiz-Herrera¹, José López-Bucio*

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo. Edificio B3, Ciudad Universitaria, 58030, Morelia Michoacán, México
Av. Francisco J, Mujica S/N Col. Felicitas del Rio, Morelia, Michoacán, C.P. 58030
jbcucio@umich.mx*

Through evolution, plants have acquired the ability of sensing and responding to changes in environment factors such as nutrient scarcity through a strict genetic expression control. Nevertheless, these adverse conditions could affect genetic material and lead to the activation of programmed cell death. This phenomenon is manifested mainly in the apical meristems, highly proliferative areas flanked by the stem cell niche.

Multiple mediator complex subunits have been reported as essential elements involved in Arabidopsis growth, development and immune response processes. MED18 subunit participates in different development aspects such as floriation, hormone response, immune response by pathogen assault and cell proliferation. In the present research, the effect of phosphate deficiency on cell death in the meristems of the *med18-1* mutant, deficient in the *MED18* gene, under contrasting phosphate availability was studied. A comparative analysis based on the cell integrity of the meristem, shows that phosphate scarcity suppresses cell death in the *med18-1* roots. Results obtained up to now suggest that *MED18* is involved in low phosphate levels perception, and therefore, in cell damage response processes such as cell differentiation. In this sense, it is suggested that MED18 may control downstream genes and transcriptional factors required to the scarcity response.

DETERMINATION OF *TPH1* GENE EXPRESSION LEVELS IN POST-MORTEM BRAIN TISSUE AND ITS ASSOCIATION WITH SUICIDE.

Norma Gabriela Barraza Mendez¹, Marcelo Barraza-Salas¹, Miguel Ángel Leyva-López¹,
Verónica Dayali Gutiérrez Martínez¹, Maribel Cervantes-Flores¹,
Francisco Xavier Castellanos-Juárez², José Manuel Salas Pacheco²

¹Facultad de Ciencias Químicas UJED, Av. Veterinaria S/N, Circuito Universitario, 34120 Durango, México. ²Instituto de Investigación Científica UJED, Av. Universidad esq. con Volantín. Zona centro, 34000. Durango, México.
Dress: Calle Rio Piaxtla No. 607, Col Valle del sur, 34120 Durango, México.
Telephone number: 618-111-14-94, E-mail: gabrielabarrazam@gmail.com

The World Health Organization defines completed suicide as the deliberate act of taking one's own life, causing injury to oneself, with a varying degree of intent to die. Suicide represents a health problem worldwide, since every year more than 800,000 people take their own lives. Suicide is one of the leading causes of death in young adults aged 15 to 29 years. In addition, suicide is one of the leading causes of death in young adults between the ages of 15 and 29. In Mexico, suicide rates have increased in recent decades, affecting mainly young men and older adults. In the year 2022, a suicide rate of 6.4 per 100,000 inhabitants was reported, while in Durango, 8.2 suicides per 100,000 inhabitants occurred in this sector of the population. There are several risk factors for suicide such as environmental, psychological, mental disorders and one of the least studied are the biological ones where it has been shown that certain genes involved in neurotransmission are involved in the risk of suicide. The neurotransmitter serotonin (5-HT) plays an important role in both the peripheral and central nervous system and several studies indicate that candidate genes for serotonergic pathways cause 5-HT dysfunction and are therefore implicated in suicidal behavior. One of the promising genes is *TPH1* (tryptophan hydroxylase 1), as it produces a major enzyme involved in the first and limiting step of 5-HT synthesis. Due to the above, the main objective of this research was to establish the expression levels of the *TPH1* gene in post-mortem brain tissue in suicidal and control subjects. In this study, RNA extraction was performed in samples of the prefrontal cortex (PFC) in postmortem brain tissue from suicidal subjects (n=25) and control subjects (n=25) from the state of Durango. To measure gene expression levels in the CPF, real-time PCR (qRT-PCR) and Taqman probes were performed for the gene of interest (*TPH1*) and a control gene (*B2M*) using the $\Delta\Delta C_t$ method to calculate the difference in expression, the results obtained were analyzed in SPSS version 25 statistical software. The statistical analyses obtained showed a $p < 0.034$ according to the Mann Whitney U test in the expression levels between both groups, this result was evaluated with a 95% confidence interval, so significant differences were found between cases and controls in the relative expression of the *TPH1* gene in prefrontal cortex.

Keywords: Suicide, Gene, *TPH1*, CPF, Expression.

EFFECT OF THE EXPRESSION OF THE LONG NON-CODING RNA XIST REGARDING THE ONCOGENE MYC, IN THE TREATMENT OF PATIENTS WITH ACUTE MYELOID LEUKEMIA

Susana Bernardo Hernández¹, Vanessa Iglesias¹, Gustavo Acosta Altamirano², Mónica Sierra Martínez¹

¹Genomic Medicine Laboratory of the Health Unit of the Regional High Specialty Hospital of Ixtapaluca dependent on the Health Services of the Mexican Institute of Social Security for Welfare, Mexico-Puebla Highway Km.34.5, Town of Zoquiapan, Ixtapaluca, State of Mexico.

²General Hospital of Mexico Dr. Eduardo Liceaga, Dr. Balmis 148, Doctors, Cuauhtémoc, 06720 Mexico City, CDMX.
E-mail: monica.sierra@hraei.com

Acute myeloid-type leukemia (AML) is a neoplasm that arises from the malignant transformation of myeloid-type hematopoietic precursor cells that proliferate uncontrollably in the bone marrow and sometimes invade the bloodstream. Among the main causes of this pathology are the epigenetic changes that intervene in leukemogenesis. This work aims to investigate the expression level of the lncRNA Xist and its effect on the MYC oncogene, as well as its implication in the treatment of patients with AML. **Objective:** To evaluate the expression of lncRNA Xist with respect to the MYC oncogene in bone marrow samples from patients with AML, using digital PCR to relate its involvement in the treatment of this oncohematological pathology. **Methodology:** Bone marrow samples with suspected leukemia were collected, confirming the diagnosis of AML by flow cytometry and searching for chromosomal rearrangements by qPCR with a commercial IVD kit. The groups with and without chromosomal alterations were stratified; and a control group of leukemia-negative patients was stratified. Finally, expression studies of the lncRNA gene Xist and MYC were performed using digital PCR. **Results:** Of 311 samples, 77 were compatible with AML, with expression of AML markers such as CD3, 33, 117. 23 patients were carriers of some chromosomal alteration, most frequently t(15;17), patients who were given ATRA as chemotherapy, some of them in combination with cytarabine, mercaptopurine or daunorubicin. Regarding the number of copies, the non-coding length XIST is significantly decreased with a p value less than 0.05, in the control group, around 400 (copies/ μ l) were obtained, while for the groups with the pathology, an average of 1.9 (copies/ μ l) were obtained. Regarding MYC, the control group obtained 0.3 to 3 (copies/ μ l) and the AML group mostly did not exceed 0.8 (copies/ μ l), the expression is decreased compared to the control, but to a lesser extent XIST. **Conclusion:** The underexpression of XIST is highly significant, the literature mentions that in those carriers of some chromosomal alteration in which the neoplasia is highly aggressive and despite having individual or combined chemotherapy treatments, the underexpression is forceful and considerable. Regarding MYC, it has been reported that when its activity is stopped it stimulates the activity of the retinoic acid receptor, thus allowing tumor cells to function normally, stopping the uncontrolled production of malignant cellularity, which is why AML patients with fused genes They are mostly treated with ATRA, which keeps MYC expression suppressed.

MIR124-3 POLYMORPHISMS AND NEURODEVELOPMENT IN CHILDREN FROM DURANGO, MÉXICO

Katzumy Lizbeth Blancas-Olvera¹, Muñoz-Agüero Jessica-Daniela¹, Francisco Xavier Castellanos-Juárez¹, La-Llave León Osmel¹, Méndez-Hernández Edna-Madaí¹, Elizabeth Irasema Antuna-Salcido¹, Oscar Eduardo Almeida-Ojeda², Salas-Pacheco Sergio-Manuel², Sandoval-Carrillo Ada¹, Salas-Pacheco-José Manuel¹

¹Instituto de Investigación Científica-UJED. ²Facultad de Odontología-UJED.
e-mail: katzumy.blancas@gmail.com

In recent years, there has been a growing interest in understanding the contribution of genetic factors to neurodevelopment during both embryonic and postnatal periods. One of the microRNAs that has been the focus of research regarding infant neurodevelopment is microRNA-124-3, encoded by the *MIR124-3* gene. Previous studies have demonstrated that this microRNA plays a crucial role in regulating cellular differentiation, neuronal morphology, as well as synapse formation and maturation, directly influencing the proper development of brain functions¹. Various polymorphisms have been reported in the *MIR124-3* gene, including rs34059726, rs67543816, rs35418153, rs1884338, and rs6011653.

The aim of this study was to genotype polymorphisms of the *MIR124-3* gene and establish their association with neurodevelopment in children from the city of Durango. Neurodevelopment was assessed using the BSID-III test, which evaluates cognitive (CD), language (LD), and motor (MD) development. Genotyping was performed by real-time PCR using DNA obtained from saliva samples of the children. Fifty children (24 females and 26 males) with a mean age of 9.9 months were included. When evaluating CD, 8% showed low evaluation, 50% normal, and 42% high. Regarding LD, 26% showed low evaluation, 60% normal, and 14% high. And in relation to MD, 30% showed low evaluation, 40% normal, and 30% high. Correlation analysis showed a positive correlation between the three areas of development. When comparing allelic and genotypic frequencies among CD, LD, and MD levels, statistically significant differences were only found in the allelic and genotypic frequencies of the rs67543816 polymorphism when evaluating CD ($p=0.009$ and $p=0.022$, respectively). Upon determining possible associations, we found that in a dominant inheritance model, the C/T genotype of the rs67543816 polymorphism is associated with higher DC ($OR=13.61$, $95\% CI_{95}=2.34-79.19$), and the G/A genotype of the rs35418153 polymorphism is associated with higher DM ($OR=0.07$, $95\% CI_{95}=0.01-0.72$). These results are similar to those reported by Park et al., who found that the rs67543816 and rs35418153 polymorphisms are significantly associated with a mental composite score calculated from the BSID-III in Bangladesh children². Future studies in our population with larger sample sizes are necessary to corroborate our findings.

References

1. Park HR, Sun R, Panganiban RA, Christiani DC, Lu Q. (2020). *Scientific reports*, **10**, 5934-5944.
2. Chafouri-Fard S, Shoorei H, Bahroudi Z, Abak A, Majidpoor J, Taheri M. (2021). *Biomedicine & Pharmacotherapy*. 135, 111198-111210.

LIGAND-DEPENDENT XRE BINDING PATTERNS OF ARYL HYDROCARBON RECEPTOR (AHR) ON *CYP1A1* GENE PROMOTER

Itzel Adriana Cabildo Delgado¹, Jorge Humberto Limón Pacheco¹ and Guillermo Elizondo^{1*}

¹Departamento de Biología Celular, Centro de Investigación y de Estudios Avanzados del I.P.N., Av. Instituto Politécnico Nacional 2508, Col. San Pedro Zacatenco, Del. Gustavo A. Madero, Mexico City, P.C. 07360. Tel: +52 (55) 5747 3800 Ext. 5555

*E-mail: gazuela@cinvestav.mx

The aryl hydrocarbon receptor (AHR) is a ligand-activated transcription factor belonging to the basic helix-loop-helix (bHLH) Per-Arnt-Sim (PAS) family. Upon ligand binding, AHR translocates to the nucleus and binds to specific DNA regions known as xenobiotic response elements (XREs) located in the promoter of its target genes, thereby inducing their transcription. By binding to xenobiotics, diet-derived compounds, tryptophan metabolites, and gut microbiota metabolites, the AHR plays multiple roles such as xenobiotic metabolism, immune responses, development, and neurogenesis. Although in general terms AHR activation by different agonists leads to the expression of same genes, it has been observed that gene induction mediated by AHR may be differential depending on the type of ligand. However, the causes that determine the AHR preferences towards the induction of one or other genes are still unknown. In the present study, we investigated whether AHR binds to different XREs located at *CYP1A1* gene promoter when is activated by two different ligands (TCDD and Kynurenine) in SHSY-5Y cells. Our results showed that both ligands promote the *CYP1A1* gene expression. However, chromatin immunoprecipitation (ChIP) analysis indicated that activation of AHR by TCDD favored its binding to a specific XREs, whereas Kynurenine favored binding to a different XREs. These data demonstrate that AHR has a higher affinity for one response element over another, depending on the nature of the ligand and likely influenced by the adjacent nucleotide sequence flanking the XRE.

INTERACTION BETWEEN GENETIC VARIANTS IN VITAMIN D METABOLISM GENES: IMPACT ON RHEUMATOID ARTHRITIS SUSCEPTIBILITY AND HYPOVITAMINOSIS D

Bertha Campos-López^{1,2}, Melissa Rivera-Escoto^{1,2}, Adolfo I. Ruiz-Ballesteros^{1,2}, Karen Pesqueda-Cendejas^{1,2}, Isela Parra-Rojas^{1,3}, Paulina E. Mora-García^{1,2}, Sergio Cerpa-Cruz⁴ and Ulises De la Cruz-Mosso^{1,2}

¹Red de Inmunonutrición y Genómica Nutricional en las Enfermedades Autoinmunes; Departamento de Neurociencias, Centro Universitario de Ciencias de la Salud; Universidad de Guadalajara; Guadalajara, Jalisco 44340, Mexico.

²Instituto de Neurociencias Traslacionales, Departamento de Neurociencias, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara, Guadalajara 44340, Jalisco, Mexico. ³Laboratorio de Investigación en Obesidad y Diabetes; Facultad de Ciencias Químico-Biológicas; Universidad Autónoma de Guerrero; Chilpancingo de los Bravo 39087, Guerrero, Mexico. ⁴Departamento de Reumatología; O.P.D. Hospital Civil de Guadalajara Fray Antonio Alcalde; Guadalajara 44280, Jalisco, Mexico.

E-mail: ulises.mosso@academicos.udg.mx.

Rheumatoid arthritis (RA) is an autoimmune disease of multifactorial etiology. One environmental-nutritional factor associated with RA pathophysiology is serum vitamin D deficiency (hypovitaminosis D). Different genetic association studies have reported that around 65% of hypovitaminosis D can be partially explained by the presence of single nucleotide variants (SNV) in key genes of its metabolism. This study aimed to determine the association of genetic variants in (rs10741657) *CYP2R1*, (rs10877012) *CYP27B1*, (rs4809959) *CYP24A1* and (rs731236 *TaqI*) *VDR* for the risk to RA and the hypovitaminosis D in the Mexican-Mestizo population. This study was conducted in 177 RA patients and 204 control subjects (CS), and allelic discrimination was performed with TaqMan probes. Vitamin D serum levels (calcidiol and calcitriol) were analyzed through ELISA commercial kits. SNVs were evaluated by multivariate dimensionality reduction (MDR) analysis. The rs731236 *TaqI* in *VDR* was presented in each of the models and this variant could have the most influence to RA. CT and CC *TaqI* genotypes were particularly associated with 1.8-fold more susceptibility to AR (OR=1.8; CI=1.2-2.7; p<0.01), as well as with 2.7-fold more susceptibility to activity of disease, according to DAS28-VSG (OR=2.7; CI=1.1-6.3; p=0.02). RA patients had higher calcitriol (47.83 vs. 36.85 pg/mL; p<0.001), and calcitriol/calcidiol ratio (2.07 vs. 1.48 pg/ng; p<0.001) compared to CS. Particularly, GG and TT genotypes on the rs10877012 *CYP27B1* were associated with 1.7-fold more susceptibility to lower serum levels of calcidiol (OR=1.7; CI=1.2-2.7; p<0.01) in both study groups. In conclusion: the CT and CC genotypes on rs731236 *TaqI* *VDR* confer genetic susceptibility to RA and activity of disease and the *CYP27B1* was associated with hypovitaminosis D in the Mexican mestizo population.

GENOTYPING OF THE RS737865 POLYMORPHISM OF THE COMT GENE IN SUICIDAL SUBJECTS AND CONTROL SUBJECTS USING REAL-TIME PCR

Jesús Eduardo Castañeda-Betancourt¹, Marcelo Barraza-Salas¹, Patricia Ponce-Peña¹, Iván Meneses-Morales¹ and Miguel Ángel Leyva-López¹

¹Facultad de Ciencias Químicas-Durango, Universidad Juárez del Estado de Durango. Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur C.P. 34120.
E-mail: rojesus_eduardo@hotmail.com

Suicide defined as the deliberate act of ending one's own life, poses a significant challenge in the realm of public health, with its impact making it one of the leading causes of death worldwide. Approximately 700,000 individuals are estimated to lose their lives each year due to suicide, ranking prominently in mortality statistics, surpassed only by incurable chronic diseases such as cancer¹.

In the Mexican context, approximately 8,000 annual deaths are attributed to this cause², underscoring the urgency of conducting thorough research to address the possible underlying causes of this behavior. By using genetic markers, based on the search for polymorphisms, it is possible to associate biological patterns that may have some possible relationship with suicidal behavior, stemming from alterations in physiological processes that can disrupt biochemical pathways at the neurological level.

Therefore, this study aims to associate suicidal behavior with the rs737865 polymorphism of the *COMT* gene, which encodes the catechol-O-methyltransferase protein³. This protein plays a crucial role in the metabolism of catecholamines such as dopamine, epinephrine, and norepinephrine, important neurotransmitters in the brain and peripheral nervous system, with the goal of finding an effective form of prevention or treatment.

References

1. Frequently Asked Questions About Suicide [Internet]. National Institute of Mental Health
2. (NIMH). [cited on December 6, 2023]. Available at: <https://www.nimh.nih.gov/health/publications/espanol/preguntas-frecuentes-sobre-el-suicidio>
3. National Institute of Statistics and Geography. INEGI. National Institute of Statistics and Geography (INEGI) [Internet]. Org. mx. [cited on December 6, 2023]. Available at: <https://www.inegi.org.mx/default.html>
4. Pelayo Terán j. M. Val158met Polymorphism of the Catechol-O-Methyltransferase Gene and Clinical Characteristics in First-Episode Psychosis [Internet]. [Santander, Spain]: University of Cantabria; 2010. Available at <https://repositorio.unican.es/xmlui/bitstream/handle/10902/1541/TesisJMPT.pdf>

INSULIN PATHWAY CHANGES ON THE GENE EXPRESSION IN ADIPOSE TISSUE MODULATED BY ELLAGIC ACID IN OBESE WISTAR RAT INDUCED WITH HIGH FAT DIET

Ramón Castillo Correa¹, Aarón Domínguez López¹, María Eugenia Jaramillo Flores², Antonio Ávila Guerrero¹, Ángel Miliar García¹

¹Laboratorio de Biología Molecular, Escuela Superior de Medicina, Salvador Díaz Mirón esq. Plan de San Luis S/N, Miguel Hidalgo, Casco de Santo Tomas, C.P. 11340, Miguel Hidalgo, CDMX #1, ²Laboratorio De Polímeros, Depto. de Ingeniería Bioquímica, Escuela Nacional de Ciencias Biológicas, Unidad Profesional Adolfo López Mateos, Zacatenco, Av. Wilfrido Massieu 399, Col. Nueva Industrial Vallejo, C.P. 07738, Gustavo A. Madero, CDMX #2, ³Laboratorio De Biología Celular y Productos Naturales, Escuela Nacional de Medicina y Homeopatía, Av. Luis Enrique Erro S/N, Unidad Profesional Adolfo López Mateos, Zacatenco, Alcaldía Gustavo A. Madero, C.P. 07738, IPN. 5627707488, E-mail: rcastilloc1101@alumno.ipn.mx

Introduction. Obesity is a multifactorial disease that is a global health problem, where Mexico is one of the countries with the highest rates of this condition, and which is associated with many diseases: insulin resistance, type 2 diabetes (T2D), arterial hypertension, metabolic dysfunction associated with fatty liver disease (MAFLD) and several types of cancer.

The high-fat diet has been associated with alterations in the insulin signaling pathway, related to glucose metabolism (Irs1, Irs2, Pi3k1r, Akt1, Tbc1d4 and Slc2a4).

Resistin (Retn) is a cytokine that alters glucose metabolism, and its name is related to the insulin resistance (IR), mainly in conditions of obesity and T2D.

Ellagic acid (EA) is a natural polyphenol found in various fruits and vegetables. Numerous studies have shown that ellagic acid has anti-inflammatory, antioxidant and anti-apoptotic effects, which may represent a novel treatment for IR.

Objective. Evaluate the mRNA expression of Retn, Insr, Glut4, Akt, Pi3k and Tbc1d4 and associated miRNAs in retroperitoneal adipose tissues in obese Wistar rat model treated with EA.

Materials and methods. 18 male Wistar rats randomly distributed in three groups n=6: standard diet (SD), high-fat diet (HF) and high-fat diet plus EA treatment dose of 50 mg/kg per day intragastrically (HF+AE). At the end sacrificed and retroperitoneal adipose tissue was obtained, total RNA was isolated and purified, cDNA synthesized and qPCR evaluation for Retn, Insr, Irs1, Irs2, Akt1, Pi3k, Tbc1d4 and Slc2a4. As an appreciation, a network analysis was performed in the miRNET 2.0 to identifying microRNAs and other interesting potential markers.

Results. Ellagic acid reduces the expression of Retn, and the increased the expression of Insr, Irs1, Irs2, Akt, Pi3k, Tbc1d4 and Slc2a4.

Network analysis shows Retn, Insr, Akt, Pi3k, Tbc1d4, Slc2a4 and associated microRNAs as potential markers of interest.

Conclusion. EA treatment reduces effect on inflammation and improve glucose metabolism in HF+AE rats modulating gene expression through upregulation of Irs1, Irs2, Akt1, Pi3kr1, Slc2a4, and Tbc1d4, and downregulation of Retn. Treatment with EA represents a possible alternative to insulin resistance associated with different clinical conditions.

SILENCING OF ABCG2 GENE EXPRESSION IN HCT15 COLON CELLS BY TRANSFECTION OF A SPECIFIC SGRNA USING THE CRISPR-CAS9 TECHNIQUE

Jesús Fabián Cervantes Meneses, Rosy Yunuen Velázquez Jiménez, Javier Fernández Torres, Yessica Eduviges Zamudio Cuevas, Karina Martínez Flores, Ambar López Macay

Synovial Fluid Laboratory, National Institute of Rehabilitation Luis Guillermo Ibarra Ibarra.

Phone Number: 55 5999 1000, Ext: 19501 and 19502

E-mail: lopez_macay@hotmail.com lopez_macay@yahoo.com.mx

Abstract. ABCG2 is a glycoprotein that contains 655 amino acids and has an approximate molecular weight of 70 kDa¹. The CRISPR-Cas9 system is composed of the Cas9 endonuclease, which uses a guide sequence within an RNA duplex; tracrRNA, also known as single guide RNA (sgRNA)².

General Objective. Develop and analyze a specific sgRNA sequence to silence the gene expression of ABCG2.

Methodology. The CRISPR Design Tool was used to obtain the sgRNA sequence of the ABCG2 gene. Subsequently, bioinformatic analysis was performed using BioEdit 7.7 with the ABCG2 sequence with the accession number NM_001257386.2 to identify the deletion site, then translate the new sequence, identifying a termination codon at amino acid 292. The SWISS-MODEL platform was used to model the resulting protein of 292 amino acids. Transfection was performed by seeding in 96-well plates until reaching 80% confluence, and then the culture medium was replaced with the transfection medium containing the CRISPR-Cas9 system and DharmaFECT as the transfection vehicle. After transfection, the efficiency was evaluated by assessing gene expression using qRT-PCR and ABCG2 protein expression by western blot and immunofluorescence.

Results. So far, a reduction in both gene expression and ABCG2 protein expression has been observed in some groups of transfected cells.

Conclusions. The transfection reduced the gene expression and protein expression of ABCG2.

References

1. Moinul, M., Amin, S. A., Jha, T. & Gayen, S. (2022) Updated chemical scaffolds of ABCG2 inhibitors and their structure-inhibition relationships for future development. *European Journal of Medicinal Chemistry* vol. 241 Preprint at <https://doi.org/10.1016/j.ejmech.2022.114628>.
2. Doudna, J. A. & Charpentier, E. (2014) *The New Frontier of Genome Engineering with CRISPR-Cas9*. *Science* 346,1258096. DOI:10.1126/science.1258096

ROLE OF HYPOXIA RESPONSE FACTORS FROM *PHASEOLUS VULGARIS* DURING A NODULE DEVELOPMENT

Ana I. Chávez-Martínez¹, Janet Palacios-Martínez, Luis Enrique Aguirre-Gil² and Luis Cárdenas¹

¹Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, México, ²Ingeniería en Biotecnología, Universidad Politécnica del Estado de Morelos, Jiutepec, Morelos, México. Av. Universidad 2001, Colonia Chamilpa. CP. 62210.
E-mail: luis.cardenas@ibt.unam.mx; Tel: 777-3291666

In plants, as in most organisms, oxygen is a necessary substrate for many of the reactions of energy metabolism. However, oxygen levels in plants vary according to the stage tissue and growth condition. In this sense, it is known that some tissues require a condition of low oxygen availability (hypoxia) for their correct development. Such as the meristematic zones, floral organs and seeds, with an oxygen availability of 1 to 50 μM . Particularly the symbiotic nodule is a clear example of a natural hypoxic niche, where low oxygen availability is crucial for nitrogenase, the enzyme responsible for catalysing the reduction of atmospheric nitrogen (N_2) to ammonium (NH_4). The response of plants to hypoxia conditions involves changes in the transcriptome that will lead to migration from aerobic metabolism to anaerobic fermentation in order to satisfy energy demand. The synergy that orchestrates the response to hypoxia is highly conserved in plants and is modulated by the group VII Ethylene Response Transcription Factors (ERFVIIIs): RAP2.2, RAP2.12, RAP2.3, Hypoxia Response ERF 1 (HRE1), and HRE2, also called Hypoxia Response Factors (HRF). However, under normoxia conditions, HRFs are targets for degradation through their Cys-Arg/N-terminal region in a ubiquitination-mediated proteolysis. In our working group we are interested in elucidating the mechanisms of perception and response to hypoxia conditions during nodule development in the interaction of *P. vulgaris*-*R. tropici*. In this regard, we have identified the orthologs in *P. vulgaris* of HRFs and their destabilizing elements. Furthermore, transcript accumulation analysis allowed us to generate a detailed profile of their transcriptional response during this process. Additionally, analysis of the activity of the HRF promoters as well as functional characterization by silencing and overexpression experiments revealed their involvement in different stages of nodule development.

EFFECT OF THE *FLIW*-GENE DELETION ON THE EXTRACELLULAR ELECTRON TRANSFER AND BIOFILM PRODUCTION IN *GEOBACTER SULFURREDUCTENS*

Jessica Cholula-Calixto, Katy Juárez and Alberto Hernández-Eligio

Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, UNAM.

Av. Universidad 2001, Chamilpa, 62210 Cuernavaca, Mor., México.

jessica.cholula@ibt.unam.mx, alberto.hernandez@ibt.unam.mx

Geobacter sulfurreducens is a bacterium that can reduce heavy metals and produce electricity through the extracellular electron transfer mechanism (EET). In *G. sulfurreducens*, biofilm formation, production of a conductive pili and *c*-type cytochromes are key elements in the production of bioelectricity. In our group, we found that in *G. sulfurreducens* the post-transcriptional regulator CsrA plays an important role in the regulation of TEE. The strain mutant in the *csrA* gene produces biofilms thicker up to twice and 45-50% more electron transfer than the wild type strain. RNA-seq transcriptome analysis revealed that CsrA regulates more than 280 genes, including those related to exopolysaccharide and *c*-type cytochromes. In Gamma-proteobacteria, CsrA is regulated by small regulatory RNAs, whereas in Firmicutes, the FliW protein interacts with CsrA to modulate its function. In *G. sulfurreducens*, the regulatory mechanism controlling CsrA function is unknown. To evaluate the involvement of the FliW protein in EET and in the regulation of CsrA function, the *DfliW* mutant strain was constructed and its phenotype was characterized in terms of EET and conductive biofilm formation. First, it was determined by RT-PCR that in *G. sulfurreducens*, *fliW* is transcribed in an operon with the *flgJKLMN-csrA-fliW* genes. A 2.4 kb chimeric fragment was generated by three-step PCR in which the *fliW* gene was deleted. This fragment was cloned into the pK18mobsacB plasmid to generate the plasmid pJCC. The pJCC plasmid was introduced into *G. sulfurreducens* by conjugation and kanamycin (Km)-resistant exconjugants were isolated. Km-resistant cells were counter-selected with 10% sucrose. To verify the correct genotype, chromosomal DNA was extracted from the sucrose-resistant and kanamycin-sensitive cells and the *DfliW* mutant strain was identified by endpoint PCR. The *DfliW* strain grows in the acetate-fumarate similarly to the wild type strain, but shows a delay in the reduction of soluble Fe(III). *c*-type cytochrome content and heme-staining assays show that the *DfliW* strain produces less cytochromes. Agglutination assays suggest that this strain produces less biofilm than the wild type strain. The data obtained so far suggest that FliW is involved in the regulation of biofilm formation, probably by regulating the function of CsrA. These data will be confirmed by RT-qPCR assays and scanning laser confocal microscopy to be presented at the congress.

MSN4 IS AN IMPORTANT REGULATOR OF THE ENVIRONMENTAL STRESS RESPONSE IN *CANDIDA GLABRATA*

Yazmín Contreras-Bravo¹, Alejandro De Las Peñas¹, Guadalupe Gutiérrez-Escobedo¹, Irene Castaño-Navarro¹

Instituto Potosino de Investigación Científica y Tecnológica
Camino a la Presa 2055, 78216 San Luis Potosí, San Luis Potosí
Tel. (444)-834-203 Email. cano@ipicyt.edu.mx

Microorganisms have development different intracellular strategies to survive under environmental stress conditions. These mechanisms are very important for pathogenic microorganisms such as *Candida glabrata*. During infection, this opportunistic pathogen is exposed within the host phagocytic cells to stressful conditions like low pH, nutrient starvation, and reactive nitrogen and oxygen species to kill or neutralize the pathogen. However, *C. glabrata* has an effective **E**nvironmental **S**tress **R**esponse (ESR) and can survive under extreme conditions. Msn2 and Msn4 are transcription factors that induce several genes in response to different types of stress. Msn2 and Msn4 recognize the *STRE* sequence in promoters of stress response genes. Msn2 and Msn4 have a conserved DNA binding domain, but the **N**uclear **L**ocalizations **S**ignal (NLS) and **N**uclear **E**xportation **S**ignal (NES) domains are not conserved, possibly indicating that Msn2 and Msn4 have a different regulation. In this work, we characterized *MSN2* and *MNS4* transcriptional regulation in basal conditions and during environmental stress. We found that in *C. glabrata* *MSN2* expression is low and does not respond to any stress. Conversely, *MSN4* has a high expression in normal conditions and this expression is induced by nutritional stress (Glucose 0.2%, Glycerol 3%, EtOH3%) and osmotic stress (NaCl 0.5 M). *MSN4* promoter does not respond to oxidative stress (Menadione or H₂O₂). Interestingly, we observed that Msn4 acts like a negative regulator of its own expression and this negative regulation probably is controlled by PKA and TOR pathways.

TRANSCRIPTION FACTORS AND DNA MOTIFS IN NASTEP PROMOTER REGULATE GENE EXPRESSION IN SELF-INCOMPATIBLE *NICOTIANA ALATA*

Yuridia Cruz-Zamora¹, Renata Salcedo-Sánchez¹, Humberto Herrera-Ubaldo², Vincent Cerbantez-Bueno², Sandra Ríos-Carrasco¹, Emilio García-Torres Caffarel¹, Stefan de Folter² and Felipe Cruz-García^{1*}

¹Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México. Cd. Mx, 04510, México.

² UGA-LANGEBIO, Cinvestav-IPN, Irapuato, Gto., 36824, México.

*fcg@unam.mx

Self-incompatibility (SI) is a genetic mechanism in angiosperms that has significantly contributed to maintaining genetic diversity through speciation in evolution time. SI allows discrimination between self-pollen and genetically unrelated pollen, allowing only the latter to fertilize successfully. SI is regulated by the multiallelic *S* locus, which harbors an *S*-RNase (female determinant) and SLFs (male determinant) genes required for the SI response. However, modifier genes (MG) are also essential for a proper pollen rejection response. One MG is *NaStEP* (*Nicotiana alata* Stigma Express Protein), a protease inhibitor with specific expression in the mature stigma of self-incompatible (SI) *Nicotiana* species. Self-compatible (SC) species, such as *N. plumbaginifolia*, encode this gene in their genome, but it is not expressed, probably because of a mutation on its promoter and/or because there are missing transcription factors (TFs). In this work, we evaluated *NaStEP* promoter (*pNaStEP*) in *Arabidopsis thaliana* and in *N. plumbaginifolia* by using *GUS* and *GFP* as reporter genes, respectively. Outcomes from T₃ transgenic *A. thaliana* lines indicated *pNaStEP* directs *GUS* expression in mature pistils. We also found that its tissue-specific expression is given by a silencer element that acts in pollen. Likewise, we demonstrated that *N. plumbaginifolia* has all the TFs needed for *StEP* expression and that the lack of expression is due to the deletion of 200 bp sequence, located near the start of transcription site. Finally, to identify TFs regulating *NaStEP* expression, we conducted a yeast one-hybrid assay using different versions of the *NaStEP* promoter as a bait vs. *A. thaliana* TFs library from anther and pistil. We identified 18 TFs that bind to *pNaStEP* and direct reporter gene expression.

PAPIIT IN204622; PAIP-FQ 5000-9128; CB-2017-2018-A1-S-10126

ASSOCIATION OF THE RS4986791 POLYMORPHISM OF THE *TLR4* GENE IN OUTPATIENTS WITH THE SEVERITY GRADE OF COVID-19

Ingrid Cuevas-Cárdenas¹, Marcelo Barraza-Salas¹, Jason Enrique Vazquez-Navarro¹, Daniel Francisco Ramos-Rosales² and José Manuel Salas Pacheco²

¹Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango, Unidad Dgo. Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur C.P. 34120, ²Instituto de Investigación Científica, Universidad Juárez del Estado de Durango, Av. Universidad y Fanny Anitua S/N, Zona Centro, C.P. 34000.
E-mail: 1101130@alumnos.ujed.mx

COVID-19 can present symptoms similar to those of a mild or moderate respiratory illness, which may escalate to severe symptoms in individuals with risk factors such as comorbidities or advanced age¹. Single Nucleotide Polymorphism (SNPs) in genes related to the innate immune system may affect its effectiveness². The SNP rs4986791 in the *TLR4* gene, which encodes the Toll-like receptor 4 essential for pathogen recognition and the activation of innate immunity, could influence susceptibility to severe symptoms of COVID-19 due to SARS-CoV-2 infection³.

This research aims to investigate the relationship between the genetic variant rs4986791 of the *TLR4* gene and the severity of COVID-19 symptoms among outpatients in the state of Durango. Genomic DNA was extracted from positive SARS-CoV-2 patients, classified into four groups based on symptom severity (asymptomatic, mild, moderate, and severe). Genotyping was carried out using a TaqMan probe on the QuantStudio 5 Real-Time PCR system. Statistical analyses were performed using SPSS v.22, with a *p* value < 0.05.

Differences in allelic and genotypic frequencies of the SNP were found, such as an association of the polymorphism with severity in women under a recessive inheritance model (OR = 1.19, I.C. = 0.37–2.00). In conclusion, the SNP rs4986791 of the *TLR4* gene is associated with symptomatic severity of COVID-19, acting as a protective factor.

References

1. Bakaros, E. et al., (2023). Innate Immune Gene Polymorphisms and COVID-19 Prognosis. *Viruses*, 15(9), 1784.
2. Aboudounya, M. M., & Heads, R. J. (2021). COVID-19 and Toll-Like Receptor 4 (TLR4): SARS-CoV-2 May Bind and Activate TLR4 to Increase ACE2 Expression, Facilitating Entry and Causing Hyperinflammation. *Mediators of inflammation*, 2021, 8874339.
3. Taha, S. I. et al., (2021). Toll-Like Receptor 4 Polymorphisms (896A/G and 1196C/T) as an Indicator of COVID-19 Severity in a Convenience Sample of Egyptian Patients. *Journal of inflammation research*, 14, 6293–6303.

GLOBAL DNA METHYLATION LEVELS ARE ASSOCIATED WITH CARDIOMETABOLIC RISK AND CLINICAL DISEASE ACTIVITY IN SYSTEMIC LUPUS ERYTHEMATOSUS PATIENTS

Ulises De la Cruz-Mosso^{1,2}, Karen Pesqueda-Cendejas^{1,2}, Isela Parra-Rojas^{1,3}, Bertha Campos-López^{1,2}, Paulina E. Mora-García^{1,2}, Adolfo I. Ruiz-Ballesteros^{1,2}, Melissa Rivera-Escoto^{1,2}, Sergio Cerpa-Cruz⁴

¹Red de Inmunonutrición y Genómica Nutricional en las Enfermedades Autoinmunes; Departamento de Neurociencias, Centro Universitario de Ciencias de la Salud; Universidad de Guadalajara; Guadalajara, Jalisco 44340, Mexico.

²Instituto de Neurociencias Traslacionales, Departamento de Neurociencias, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara, Guadalajara 44340, Jalisco, Mexico. ³Laboratorio de Investigación en Obesidad y Diabetes; Facultad de Ciencias Químico-Biológicas; Universidad Autónoma de Guerrero; Chilpancingo de los Bravo 39087, Guerrero, Mexico. ⁴Departamento de Reumatología; O.P.D. Hospital Civil de Guadalajara Fray Antonio Alcalde; Guadalajara 44280, Jalisco, Mexico.

E-mail: ulises.mosso@academicos.udg.mx

Epigenetic factors such as DNA methylation can affect the expression of immune related genes; consequently, could promote the development of autoimmune diseases such as systemic lupus erythematosus (SLE), and probably influence cardiometabolic risk. This study aimed to assess the association between global DNA methylation patterns with cardiometabolic risk and clinical disease activity in SLE patients. A comparative cross-sectional study was conducted in 204 female SLE patients and 201 control subjects (CS). SLE patients were classified by the 1997 SLE-ACR criteria, and the clinical disease activity by the Mexican-SLEDAI (Mex-SLEDAI). The global methylation level of DNA was determined by the 5-mC DNA ELISA Kit-Zymo. Active SLE patients had lower global DNA methylation than inactive SLE (Active SLE = 3.4% vs. Inactive SLE = 5.09%; $p=0.04$), the global DNA methylation also was lower in ANA-positive patients than in ANA-negative (ANAs positive = 4.09% vs. ANAs negative = 13.7%; $p=0.01$). A High global DNA methylation was associated with higher HDL levels (β coefficient = 6.8; IC=2.1-11.5; $R=0.05$; $p<0.01$) and lower LDL/HDL index (β coefficient = -0.37; IC = -0.66 - -0.08; $R=0.03$; $p=0.01$). In conclusion, global DNA methylation is associated with low cardiometabolic risk in SLE patients.

MICROEVOLUTION OF *CANDIDA GLABRATA* IN THE PRESENCE OF FLUCONAZOLE

Rosa Lilian Díaz-Chávez¹, Alejandro Juárez-Reyes¹, Alejandro De Las Peñas¹ and Irene Castaño-Navarro^{1*}

¹Institute for Scientific and Technological Research of San Luis Potosí, IPICYT, Camino a la Presa San José 2055, Lomas 4^a sección, ZIP. 78216, San Luis Potosí, SLP, México. Tel +52 (444) 8342000 Ext. 2038.

*E-mail: icastano@ipicyt.edu.mx

Opportunistic infections pose significant health risks for immunocompromised individuals, often leading to increased morbidity and mortality. *Candida glabrata*, a prominent opportunistic fungal pathogen, has demonstrated notable resistance to fluconazole (FLC), complicating clinical management. Understanding the underlying mechanisms of this resistance, including mitochondrial and epigenetic factors, is crucial to combat this type of infection. This study investigates the role of mitochondrial function and the impact of histone methylation, particularly H3K36, in FLC resistance (FLC^R) in *C. glabrata*. We conducted a microevolution experiment by chronically exposing three *C. glabrata* parental strains to FLC that generated evolved mutants. Using a growth spot assay, we assessed the stability of the FLC^R phenotype. We constructed a knockout plasmid targeting the *SET2* gene (pDC1) to evaluate the role of H3K36 methylation in response to FLC exposure. Mitochondrial function of the evolved mutants was analyzed through growth on non-fermentable carbon sources (Gly- phenotype), by mitochondrial staining with MitoTracker™ Green FM and MitoTracker™ Red CMXRos, and the presence of mitochondrial genes *COX2* and *COX3* was investigated. Additionally, we used a translational fusion of the mitochondrial protein Prx1 with GFP to observe mitochondrial morphology.

Our results showed variability in the stability of the FLC^R phenotype depending on the genetic background. Gly- mutants exhibited stable mitochondrial dysfunction that could not revert to Gly+, with potential loss or mutations in *COX2* and *COX3* genes. Additionally, Gly- mutants showed diffuse fluorescence signals with mitochondrial stains, suggesting mitochondrial structural and functional alterations compared to Gly+ strains. The Prx1-GFP fusion did not give a fluorescent signal in the Gly- strains, suggesting defects in mitochondrial integrity. Spot growth assays revealed a complex interplay between genetic background, phenotype, and FLC resistance stability. In addition, we generated a *set2Δ* mutant in different genetic backgrounds. Data on the FLC resistance phenotype of *set2Δ* mutants will be presented. Our results highlight the intricate relationship between mitochondrial function and FLC^R in *C. glabrata* and the need for further investigation into the underlying epigenetic mechanisms.

Keywords: *Candida glabrata*, fluconazole resistance, mitochondrial function, Gly- phenotype, epigenetic regulation.

LOSS OF THE TUMOR SUPPRESSOR MIR-122 PROMOTES CELL MIGRATION AND UP-REGULATION OF BORIS/CTCF IN TRIPLE-NEGATIVE BREAST CANCER

Mauricio Flores-Fortis^{1,2}, Isidro X. Perez-Añorve², Carlos C. Patiño-Morales³, Arturo Aguilar-Rojas⁴, Nicolas Villegas⁵, Elena Aréchaga Ocampo^{2,6*}

¹Posgrado en Ciencias Naturales e Ingeniería, Unidad Cuajimalpa, Universidad Autónoma Metropolitana. 05348, Mexico City, Mexico. ²Departamento de Ciencias Naturales, Unidad Cuajimalpa, Universidad Autónoma Metropolitana. 05348, Mexico City, Mexico. ³Laboratorio de Biología del Desarrollo y Teratogenesis Experimental. Hospital Infantil de Mexico Federico Gomez. 06720, Mexico City, Mexico. ⁴Unidad de Investigación Médica en Medicina Reproductiva, IMSS, Unidad Médica de Alta Especialidad No. 4, Mexico City, Mexico. ⁵Departamento de Biomedicina Molecular, Centro de Investigación y de Estudios Avanzados (CINVESTAV). 07360, Mexico City, Mexico. ⁶Red de investigación en Salud, Universidad Autónoma Metropolitana. Mexico City, Mexico

*E-mail address: earechaga@cua.uam.mx

Abstract. miRNAs are short non-coding RNAs that negatively regulate the expression of target genes post-transcriptionally. In human cancer, miRNAs are key regulators of pathways to promote or suppress carcinogenesis. miR-122 is aberrantly expressed in breast cancer and plays both a tumor suppressor and oncomiR role. In triple-negative breast cancer (TNBC) the function of miR-122 is still unclear. This study was conducted to understand the molecular mechanism of miR-122 in TNBC patients submitted to neoadjuvant chemotherapy. The expression and clinical association of miR-122 in TNBC patients, and the differentially expressed genes were evaluated throughout using the TNBC-related RNA-seq from TCGA and KM-plotter datasets. Functional analysis of miR-122 in TNBC cells was performed by knocking down or overexpressing miR-122 through cell transfections, RT-qPCR, immunoblotting, cell viability, and cell migration assays. The results showed that chemotherapy can deregulate the miR-122 expression levels and the downregulation of miR-122 was associated with tumor recurrence after NAC in TNBC patients. Lower levels of miR-122 in rapid-relapse TNBC patients allows the activation of a gene co-expression network enriched in genes associated to migration, invasion, and cell differentiation. Among these genes, DNA-binding protein BORIS was identified as target gene of miR-122. Functionally, exogenous overexpression of miR-122 impaired cell migration and BORIS expression. In addition, BORIS promoted a gene expression profile related to cell differentiation and cytoskeletal components in TNBC patients with loss of miR-122 expression. In conclusion, miR-122 negatively regulates cell migration at least in part by regulates BORIS expression in relapse TNBC patients.

Keywords: tumor suppressor, miR-122, BORIS, cell migration, TNBC

ASSOCIATION OF THE DNMT1 PROTEIN WITH LONG NON-CODING RNAs: MALAT1, UCA1 AND HOTAIR IN CERVICAL CANCER

Janis Aislinn García Flores, Carlos Manuel Contreras Romero, Carlos Pérez-Plasencia, Eloy-Andrés Pérez Yépez

Instituto Nacional de Cancerología
Laboratorio de Genómica, Instituto Nacional de Cancerología, Tlalpan, México.
carlos.pplas@gmail.com

Abstract. Cancer is defined as the sum of a series of alterations in cellular processes that lead to the acquisition of distinctive characteristics by healthy cells for the transition to neoplastic growth stages and their ability to form a malignant tumor. In 2022, cervical cancer ranked fourth in terms of incidence and mortality worldwide; in Mexico, cervical cancer is the second most diagnosed cancer and the second leading cause of death in women. Although the role of HPV is important for the development of CC, 75% of cases of infection by this virus are transient, so it can be deduced that this etiological factor is necessary for the development of the disease, but not decisive, since additional genetic and epigenetic changes are required for its progression. An example of this is non-mutational epigenetic reprogramming, within this reprogramming we can find DNA methylation, this process can be regulated by long non-coding RNAs. It has been demonstrated that some LncRNAs can interact with the DNMT1 protein, and that this interaction helps to maintain the aberrant methylation pattern characteristic of different types of cancer, thus promoting tumorigenesis and progression processes.

Aim. To analyze if there is an association between LncRNA: MalaT1, UCA1 and HOTAIR with DNMT1 protein in biopsies of patients with locally advanced stage and cervical cancer cells.

Results. RIP assays were performed using HaCaT non-tumor cells and HeLa tumor cells to analyze the level of association of DNMT1 with LncRNAs. The result showed that the DNMT1-LncRNA interaction is not significant in non-tumor cells while in tumor cells a rate of at least 2-fold change was achieved. Simultaneously to this assay, expression analysis of LncRNAs HOTAIR, MALAT1 and UCA1 was performed in patients, HeLa and HaCaT cell lines and in the TCGA database. In patients, only MALAT1 LncRNA showed a significant difference, from the data analyzed in the TCGA database no significant differences were found between cervical cancer tissue and dysplasia-free tissue, while in the cell lines MALAT1 and HOTAIR LncRNAs were found to be over-expressed, while UCA1 was found to be under-expressed with respect to the non-tumor control. From the RIP assay performed with patient biopsies, it was found that for MALAT1, in 8 of the 10 samples there was a strong association of the protein with the LncRNA, while for LncRNA UCA1 and HOTAIR in all samples this association was observed. To identify whether these LncRNAs were associated with methylated DNA, the CHIRP technique was performed, where MALAT1 and HOTAIR were found to be associated with methylated DNA, unlike UCA1 for which no evidence of this interaction was found. Finally, immunofluorescence was performed to analyze whether these molecules colocalize together. The resulting images show that we have been able to find areas where we can focus these two molecules together.

Conclusion. There is a relationship between LncRNAs: HOTAIR, MALAT1 and UCA1 with DNMT1 protein in biopsies of patients in locally advanced stage and in cells with cervical cancer, which indicates that this interaction could intervene in cellular processes such as DNA methylation promoting a distinctive aberrant pattern of cancer.

EXPLORING THE TRANSCRIPTOME OF MCF-7 BREAST CANCER CELLS TREATED WITH *CAPSICUM ANNUUM* L. VAR. FASCINATO

Roberto Jorge Garcia-Mendoza¹, Verónica Morales-Tlalpan^{1,2}, Joel Hurtado Patiño¹,
Adriana González Gallardo³, Angélica Feregrino-Pérez⁴ & Carlos Saldaña^{1,2}

¹Laboratorio de Biofísica de Membranas y Nanotecnología, ²Laboratorio Nacional de Visualización Científica Avanzada UNAM-UAQ. ³Unidad de Proteogenómica del Instituto de Neurobiología UNAM; ⁴Laboratorio de Metabolitos y Nanocompositos, Facultad de Ingeniería UAQ. Facultad de Ciencias Naturales, Universidad Autónoma de Querétaro. Juriquilla, Qro., México C.P. 76230. Phone +52 442 192 1200, ext. 65332, carlos.saldana@uaq.mx

Calcium (Ca²⁺) ion acts as a second messenger to regulate cellular homeostasis. Among the mechanisms to maintain Ca²⁺ homeostasis, a variety of Ca²⁺-regulated transporter proteins, Ca²⁺-binding proteins, and enzymes exist. Dysregulated Ca²⁺ homeostasis triggers a series of pathophysiological processes as: neurological diseases, cancer and others. Our laboratory studies the spatial organization of Ca²⁺-regulating proteins located in the endoplasmic reticulum (ER) in MCF-7 breast cancer cells. It's observed that the arrangement of the IP₃ receptor and RyR was homogeneous in the ER, however, SERCA (a protein that can canonically transport Ca²⁺ into the ER), was found to be localized towards one pole of the cell. To understand how Ca²⁺ dynamics could be modulated by bioactive molecules, we are using extracts from three varieties of *Capsicum annum* L.: Baselga, Fascinato, and Orangela; we determine IC₅₀ and Ca²⁺ levels (extracts could mobilize intracellular calcium dose-response and their kinetics are different). Next, we decide to characterize Fascinato extract, due to promoting the expression of SERCA 3, analysed by qPCR. The continuation of this project involved obtaining the transcriptome of MCF-7 cells treated with Fascinato. The results were analysed using bioinformatic software. The transcriptome analysis confirmed that Fascinato promotes an increase in SERCA 3 transcripts. Similarly, it was observed that Fascinato modulates the transcription of genes related to intracellular calcium dynamics, either up or down. Additionally, the transcriptome data will enable us to investigate a wide range of genes and how they were regulated.

Thanks to the Institutions that financed: FONDEC-UAQ, FONFIVE-UAQ and CONAHCyT A1-S-26966 to CS. CONAHCyT 1032842.

RS 2278163 VARIANT OF THE DLX3 GENE AND ITS RELATIONSHIP TO THE SEVERITY OF DENTAL FLUOROSIS IN WOMEN IN DURANGO CITY

Oscar Martín García Rosales¹, Luis Antonio Ortiz López¹, José Manuel Salas Pacheco², Leslie Karina Gamboa Guerrero¹, Nelly Molina Frechero³, Sergio Manuel Salas Pacheco¹, Omar Tremillo Maldonado¹

¹Facultad de Odontología, Universidad Juárez del Estado de Durango. ²Instituto de Investigación Científica, Universidad Juárez del Estado de Durango. ³Universidad Autónoma Metropolitana
Email: racsogarcia@hotmail.com

Keywords: dental fluorosis, single nucleotide variation, DLX3

Introduction. Dental fluorosis (DF) presents as an alteration in the development of dental enamel, due to high and prolonged concentrations of fluoride (F) during the development of dental organs. Clinically, dental fluorosis is characterized by opaque white areas in the enamel, up to a striated, mottled appearance and loss of enamel continuity. The DLX3 gene belongs to the homeobox family, which plays an important role in the regulation of embryonic development including the formation of dental organs. **Objectives.** To establish the relationship between the rs 2278163 variant of the DLX3 gene and its relationship with the severity of dental fluorosis in women from the city of Durango. **Materials and Methods.** A cross-sectional, observational, case-control, prospective and analytical study was carried out in which 72 women from the city of Durango, Durango, were included. The degree of severity of dental fluorosis was classified according to the modified Dean index with TF. The quantification of the concentration of fluoride present in drinking water, home water and urine was determined by potentiometry. Genotyping was performed with real-time PCR. The estimation of association between the rs2278163 polymorphism of DLX3 and the severity of fluorosis was carried out by means of the χ^2 test. **Results.** The average fluoride concentration in home water and consumption was 2.80 ± 1.28 ppm and 1.21 ± 1.41 ppm respectively. The average concentration of F⁻ in the urine samples was 2.66 ± 2.29 ppm. The frequency of distribution of dental fluorosis among the women participating in the study it was observed that the highest percentage (40.3%) presented a TF 4, 16.7%, for TF 3 and 6, 1.4% was obtained, in TF 8. There is a low and statistically significant correlation between fluoride levels in drinking water and fluoride levels in urine. Genotype G/G is associated as a protective factor against the most severe degrees of dental fluorosis. **Conclusion.** The results obtained reflect that the rs2278163 polymorphism of DLX3 is associated as a protective factor with the severity of dental fluorosis in women in the city of Durango.

References

1. FeleJ JA, Morasso MI, Jang SI, Sargent TD. Transcriptional activation by the homeodomain protein Distal-less 3 [Internet]. Vol. 27, Nucleic Acids Research. 1999. Available from: <http://nar.oxfordjournals.org>

“LA” PROTEIN IN *LEISHMANIA MAJOR*: ANALYSIS OF ITS INTERACTIONS WITH OTHER PROTEINS AND RNA MOLECULES

Sergio García De la Cruz¹, Luis E. Florencio Martínez¹, Rebeca Manning Cela²,
Tomás Nepomuceno Mejía¹ and Santiago Martínez Calvillo¹

¹Facultad de Estudios Superiores Iztacala, UNAM. Av. de los Barrios 1, Col. Los Reyes Iztacala, Tlalnepantla, Edo. de México, CP 54090, México; ²Departamento de Biomedicina Molecular, CINVESTAV-IPN, México
E-mail: sergdlc18@gmail.com

Leishmaniasis represents a public health problem around the world, affecting more than a million people in tropical and subtropical regions. The protozoan parasites of the genus *Leishmania* are the etiological agents of leishmaniasis and are transmitted to vertebrate hosts, including humans, through the bite of infected sandflies. In addition to their medical importance, *Leishmania*, and other trypanosomatid parasites, have atypical gene expression mechanisms, like trans-splicing and polycistronic transcription. While transcriptional peculiarities of RNA polymerases (RNAP) I and II have been described in these parasites, information about RNAP III transcription is still scarce. In *Leishmania*, RNAP III transcribes all snRNAs, in addition to 5S rRNA and tRNAs. All RNAP III transcripts contain an exposed UUU-OH-3' trailer that is protected from degradation by the La protein. The La protein contains multiple RNA binding domains and is involved not only in RNAP III-dependent transcript protection but also in mRNA processing. Since the La protein has not been studied in *L. major*, we generated a cell line that expresses the recombinant La protein attached to the PTP tag. Tandem affinity purifications revealed that La protein interacts with several ribosomal proteins, including the *kinetoplastid-specific ribosomal protein* (KSRP), and proteins involved in RNA processing, like tRNA methyltransferases and pseudouridine synthases, as well as other mRNA binding proteins. We also found, through RT-qPCR, that the purified La protein binds to RNAP III-dependent transcripts like the 5S rRNA, U4 snRNA and tRNA^{Ala}. We are currently exploring the association of the La protein with mRNAs. This work was supported by grants IN208224 (PAPIIT, UNAM) and CF-2023-I-820 (CONAHCYT).

INTERPLAY BETWEEN CHAPERONES AND THE SORTING PLATFORM FOR HIERARCHICAL SECRETION OF PROTEINS THROUGH THE INJECTISOME

Ricardo Gaspar Lino, Norma Espinosa and Bertha González Pedrajo

Departamento de Genética Molecular, Instituto de Fisiología Celular, UNAM. Circuito Exterior s/n, Ciudad Universitaria, Coyoacán, Ciudad de México, 04510
bpedrajo@ifc.unam.mx; Tel.: +52-55-5622-5965; Fax: +52-55-5622-5611

Abstract. Enteropathogenic *Escherichia coli* (EPEC) causes persistent diarrhea in children aged 0 to 11 months and is a major cause of infant mortality in developing countries. EPEC colonizes the intestinal epithelium using a type III secretion system (T3SS) or injectisome, which is a molecular machinery composed of more than 20 proteins, which translocates virulence effectors into the cytoplasm of eukaryotic cells, causing a histopathological damage known as attaching and effacing lesion. The injectisome is encoded within a 35.62 kb pathogenicity island called the locus of enterocyte effacement (LEE). One of the structural components of the LEE-encoded T3SS is the sorting platform located at the base of the injectisome, which consists of the proteins SctK, SctQ, and SctL. Chaperone proteins are also encoded within the LEE and are cytoplasmic proteins that bind, protect, direct and control ordered substrate secretion through the T3SS. The sorting platform is a dynamic structure and can be modulated in the presence of effectors and chaperones at the onset of secretion. Therefore, the proposed function of the sorting platform is to recruit and increase the local concentration of the type III substrates at the base of the injectisome, where the sorting platform proteins bind effectors in the cytosol and deliver the cargo to the export gate in the membrane-bound injectisome. It is believed that the presence of chaperone proteins for early, intermediate, and late substrates may influence the secretion hierarchy. Therefore, our interest lies in determining the role of chaperone proteins in differential substrate recognition by components of the sorting platform. In this work, we will present our results of overproduced substrate secretion in the absence of the sorting platform and a chaperone, as well as protein interaction assays between chaperones alone and in complex with their substrate, and the sorting platform.

Acknowledgments. This work was supported by DGAPA/PAPIIT (IN229023) and CONAHCYT (284081) grants.

IDENTIFICATION OF GENETIC VARIANTS ASSOCIATED WITH TREATMENT TOXICITY IN PEDIATRIC PATIENTS WITH ACUTE LYMPHOBLASTIC LEUKEMIA

Francisco Javier Gaytán Cervantes¹, Itzel Peralta Salguero¹, Carolina González Torres¹, Andrea Martínez Marroquín¹, Haydeé Rosas Vargas², Elva Jiménez Hernández³, Juan Carlos Núñez Enríquez⁴, Omar Alejandro Sepúlveda Robles², Silvia Jiménez Morales⁵, Minerva Mata Rocha², José Refugio Torres Nava⁶, Jorge Alfonso Martín Trejo⁷, Luz Victoria Flores Villegas⁸, María De Lourdes Gutiérrez River⁷, Laura Elizabeth Merino Pasaye⁸, Karina Anastacia Solís Labastida⁷, María Raquel Miranda Madrazo⁸, Janet Flores Lujano⁴, María Luisa Pérez Saldivar⁴, Juan Manuel Mejía Arangure⁵

¹Laboratorio de Secuenciación, Coordinación de Investigación en Salud, CMNSXXI, IMSS, CDMX. ²Unidad de Investigación Médica en Genética Humana, Hospital de Pediatría "Dr. Silvestre Frenk Freund", CMNSXXI, IMSS, CDMX. ³Servicio de Hematología Pediátrica, Hospital General, Centro Médico Nacional La Raza, IMSS, CDMX. ⁴Unidad de Investigación Médica en Epidemiología Clínica, Hospital de Pediatría "Dr. Silvestre Frenk Freund", CMNSXXI, IMSS, CDMX. ⁵Laboratorio de Genómica del Cáncer, Instituto Nacional de Medicina Genómica, Secretaría de Salud CDMX. ⁶Servicio de Oncología, Hospital Pediátrico "Moctezuma", Secretaría de Salud, CDMX. ⁷Servicio de Hematología Pediátrica, Hospital de Pediatría "Dr. Silvestre Frenk Freund", CMNSXXI, IMSS, CDMX. ⁸Unidad de Hematología Pediátrica, Centro Médico Nacional "20 de Noviembre", ISSSTE, CDMX. Email: Laboratorio de Secuenciación, CMNSXII, IMSS, CDMX 06720, alternativesplicing@gmail.com

Acute lymphoblastic leukemia (ALL) is one of the most common types of childhood cancer worldwide. In Mexico, it is the leading cause of death in patients under 15 years old, with a survival rate ranging from 50 to 80%. In recent years, there has been growing interest in the genomic study of ALL. This methodology has elucidated genetic variations that are correlated with treatment-induced toxicity, predisposition to disease development, and early relapse risk in pediatric patients. However, it is noteworthy that the majority of these investigations have primarily centered on non-Latin populations, thus resulting in a gap in our understanding of how genetic variants affect pediatric patients within our pediatric patients. Hence, the identification of genetic variants within our population may assume a pivotal role in diagnosis and therapeutic decision-making, aimed at mitigating adverse effects such as treatment toxicity. The objective of this study is to identify and characterize genetic variants within genes implicated in drug metabolism and assess their correlation with early treatment susceptibility in pediatric patients diagnosed with B-ALL. A custom sequencing panel targeting genes associated with treatment toxicity, B-ALL pathogenesis, and maintenance was devised. 95 samples from patients with B-ALL were meticulously chosen and subjected to sequencing. Subsequent classification based on toxicity grade was conducted utilizing the CTCAE classification. Variant annotation, ancestry analysis, and association studies were conducted employing tools such as GATK v4.0, PLINK 1.9, Admixture 1.3.0, and Eigensoft. A total of 1986 variants were identified, categorized into 1541 known variants and 445 De Novo variants. Among these, 60 variants were detected within genes such as *ABCC4*, *SLC22A1*, and *MTHFD1*, which are recognized for their involvement in drug transport and metabolism pathways. These genes harboring variants are linked to crucial biological processes like drug transport and metabolism. Hence, the results of this study have the potential to uncover biomarkers that can improve both diagnosis and treatment for patients, consequently reducing adverse effects associated with treatment.

EPIGENETIC ANALYSIS IN ADIPOCYTES DIFFERENTIATED FROM MESENCHYMAL STEM CELLS OF THE HUMAN UMBILICAL CORD OF NEWBORNS FROM HEALTHY, OBESE, AND DIABETIC MOTHERS UNDER HIGH GLUCOSE AND CHOLESTEROL CONDITIONS

Emily Jocelyn Gómez Flores¹, Fatima Yexalen Bermudez Urquiza¹, Mauricio Domínguez Castro¹, Jessica Hernández Pineda¹, Diana Soriano Becerril¹, Ismael Mancilla Herrera¹, Ursula Torres¹, Arturo Flores-Pliego¹, Araceli Montoya Estrada¹, Enrique Reyes Muñoz¹, José Romo Yáñez^{1*}

National Institute of Perinatology¹. Montes Urales 800, Lomas Virreyes CP 11000 Mexico City. Telephone 5555209900 X 245.

*Email jryz@yahoo.com

In Mexico, overweight and obesity in women of childbearing age ranges from 26 to 38%, with a greater risk of type 2 diabetes mellitus (DMT2). Children of obese or diabetic mothers are usually more likely to develop metabolic diseases and obesity, conditions that could be caused by epigenetic mechanisms. Because of this, this work analyzes the possible epigenetic changes in differentiated adipocytes from mesenchymal stem cells, from human umbilical cord Wharton gelatin (hUC-WJMSC) of newborns of obese mothers (RNMO), with DMT2 (RNMD) and healthy mothers (RNMS). The hUC-WJMSC were grown and differentiated into adipocytes under control conditions (5mM D-glucose), hyperglycemia (40mM D-glucose), hypercholesterolemia (10µg/mL cholesterol) and together (40mM D-Glucose and 10µg/mL cholesterol). Tests were carried out for global methylation and hydroxymethylation of DNA, global trimethylation of histones H3K4me3 and H3K9me3, in addition to the expression of the mRNAs of DNMT3A, EHMT2, EP300 and KAT2B.

Preliminary results in DNA methylation show that in the control condition, the hUC-WJMSC of RNMS expose the lowest values and are gradually increasing in the NMRN and RNMD groups, respectively. On the other hand, in the hydroxymethylation of DNA the hUC-WJMSC that have the lowest values in all treatments are those of NMR, followed by those of NMRN and presenting the highest values those of RNMD showing more evident in treatments with 40mM of D-Glucose and in that of 40mM of D-Glucose + 10µg/mL of cholesterol. In the trimethylation trial of the histones H3K4me3 and H3K9me3, a very similar pattern was presented for most treatments, in which the hUC-WJMSC of RNMS presented the highest values, followed by the group of NMR, to reach the group with the lowest values corresponding to the RNMD group, the most noticeable differences were in the treatment of 40mM of D-Glucose+10µg/mL of cholesterol which had an opposite pattern. As for the relative expression of mRNAs, different changes in genetic expression could be noted in the hUC-WJMSC from the three study groups (RNMS, RNMO, RNMD) and among the different treatments.

Our results indicate that there are epigenetic changes in the hUC-WJMSC of RNMO and RNMD cultured in vitro, indicating that the conditions of obesity and DMT2 during pregnancy are factors that can change the global profile of the methylation and hydroxymethylation of DNA, the trimethylation of the histones H3K4me3 and H3K9me3 and the genetic expression in the cells of newborns, changes related to An increased risk of developing short- and long-term metabolic diseases.

GENE REGULATION OF *CTA1* IN *C. GLABRATA* IS CONTROLLED BY DIFFERENT *CIS* AND *TRANS*-ACTING ELEMENTS AFTER BEING EXPOSED TO DIFFERENT STIMULI

Carlos Ricardo González Ruiz¹, Gabriel Guillermo Luna Arvizu², Israel Cañas-Villamar¹,
Guadalupe Gutiérrez-Escobedo¹, Irene Castaño-Navarro¹ and Alejandro De Las Peñas¹

¹IPICYT, División de Biología Molecular, Instituto Potosino de Investigación Científica y Tecnológica, Camino a la Presa San José, #2055, Col. Lomas 4^a Sección, San Luis Potosí CP 78216, San Luis Potosí, México. ²Department of Biology, Institute of Molecular Biology, University of Oregon, Eugene, OR, USA.

E-mail: carlos.gonzalez@ipicyt.edu.mx, Tel: (444)-834-2040

Candida glabrata is an important opportunistic fungal pathogen that causes infections in immunocompromised patients. *C. glabrata* has evolved different virulence factors, such as a robust oxidative stress response (OSR) that depends on enzymatic and non-enzymatic mechanisms to counteract oxidative stress. Pathogens use these highly conserved antioxidant mechanisms to survive inside phagocytes; as a result, the expression of these enzymes is associated with the level of virulence. *C. glabrata* possesses a single catalase enzyme (Cta1) that provides protection against high levels of H₂O₂ *in vitro*. *CTA1* expression is induced by different stress conditions such as oxidative stress and starvation, and the signaling molecule alkene 1-dodecene (C12). In addition, expression of *CTA1* is controlled by the transcription factors Yap1, Skn7, Msn2 and Msn4. Based on a 5' to 3' deletion mapping of the intergenic region between *OYE2* and *CTA1*, we previously identified three positive *cis*-acting regulatory elements in the *CTA1* promoter when exposed to H₂O₂. From the ATG of *CTA1*, these elements are located between -4.56 kb and -4 kb, -4 kb and -3.3 kb, and -1.34 kb and -1 kb, and the basal promoter is located between at -1.0 kb and -0.75 kb. In addition, we determine that both Yap1 or Skn7 are required for the induction of the *CTA1* promoter in the presence of H₂O₂. To define these *cis*-acting regulatory elements, we generated 3' to 5' deletions of this intergenic region. We identify nine different *cis*-acting regulatory elements, the basal promoter, six positive elements and two negative elements that regulate the expression of *CTA1* after exposition to H₂O₂. The positive elements are located between -4.07 kb and -4.0 kb, between -4.0 and -3.3 kb, between -3.06 kb and -2.41 kb, between -1.59 kb and -1.39 kb, between -1.22 kb and -1.1 kb, and between -1.1 kb and -1.0 kb, while the negative elements are located between -1.80 kb and -1.59 kb, and between -1.39 kb and -1.22 kb. In addition, we determined that Yap1 is required for the C12 mediated induction of the *CTA1* promoter in the absence of oxidative stress. These results show that the expression of *CTA1* in *C. glabrata* is highly regulated by *trans*-acting factors with their cognate *cis* acting elements in response to different stress stimuli.

ANALYSIS OF SMALL NON-CODING RNAs EXPRESSION PROFILES AND THEIR IMPACT ON MALIGNANT PLEURAL MESOTHELIOME DEVELOPMENT

Carolina González Torres¹, Andrea Martínez Marroquin¹, Francisco Javier Gaytán Cervantes¹, Saé Muñiz Hernández², Oscar Gerardo Arrieta Rodríguez², Itzel Peralta Salguero¹

¹Sequencing Laboratory, Research Development Division, National Medical Center Siglo XXI, Mexico City 06720, Mexico,

²Experimental Oncology Laboratory, Basic Research Subdirection, National Cancer Institute, Mexico City 14080, Mexico.

E-mail: gonzaleztorrescaro@gmail.com

Malignant Pleural Mesothelioma (MPM) is an aggressive type of cancer affecting the mesothelium of the lung's pleura, characterized by nonspecific symptoms and an unfavorable prognosis due to the complexity of early diagnosis. In the pursuit for new biomarkers to support diagnostic accuracy or serve as molecular targets, the role of small non-coding RNAs (sncRNAs) becomes significant. The aim of this study is to establish the differential expression profile of sncRNAs in samples from MPM patients, to determine their potential molecular targets, as well as the mechanisms involved in the development and maintenance of the tumor phenotype. We obtained the differential expression profile of sncRNAs in samples from MPM patients by sequencing. miR-28, miR-193a, miR-345, SNORD8, and piR-hsa-158814 were selected for validation of their expression by qPCR, all of which were found to be underexpressed. In the *in silico* model, their potential miRNA target genes were identified. Of the selected sncRNAs in this study, the miRNAs had not been validated in MPM patients, while snoRNAs and piRNAs have not yet been reported in MPM. Through *in silico* analysis, the potential role of miRNAs in cellular invasion, migration, and proliferation was determined through their interaction with target genes in samples from MPM patients.

TARGETING CTCFL ONCOGENE USING CRISPR-CAS13 TECHNOLOGY

Paula González-Hernández, Aylin del Moral Morales and Ernesto Soto-Reyes

Departamento de Ciencias Naturales e Ingeniería, Universidad Autónoma Metropolitana Unidad Cuajimalpa
E-mail: esotoreyes@cua.uam.mx; Vasco de Quiroga 4871, Contadero, Cuajimalpa de Morelos, 05348 Ciudad de México, CDMX

Cancer remains a prevalent disease, with emerging molecular targets critical for developing innovative treatments. BORIS, a protein encoded by the *CTCF* transcript, suppresses its paralog *CTCF* -a key regulator of chromatin's three-dimensional organization- promoting tumorigenesis and metastasis. However, targeted therapies to inhibit BORIS without compromising cellular and organismal viability are yet to be developed. Here we show the high efficiency of CRISPR-Cas13 system achieving significant knockdown of both *CTCF* mRNA and BORIS protein. Post-treatment analysis confirmed the substantial decrease in the expression levels, suggesting an effective gene silencing outcome. We employed the CRISPR-Cas13 system *in vitro* alongside specifically designed guide RNAs (gRNAs) that target *CTCF*, achieving a reduction in its expression within HeLa cell lines model. The expression levels of both the transcript and the protein were evaluated using different techniques like Western Blot, qPCR, RT-PCR and PCR. This approach validated the potential application of CRISPR-Cas13 to selectively downregulate *CTCF* highlights its potential as a gene therapy tool in cancer treatment. This study supports further investigation into CRISPR-based therapies targeting key cancer biomarkers, offering new avenues for the management of cancer.

CHROMATIN REMODELING PROTEIN ABF1 REGULATES EXPRESSION OF ADHESIN ENCODING GENES IN *CANDIDA GLABRATA*

Grecia Hernández-Hernández¹, Laura A. Vera-Salazar¹, Guadalupe Gutiérrez-Escobedo¹, Nicolás Gómez-Hernández¹, Osney Leiva- Peláez¹, Alejandro De Las Peñas¹ and Irene Castaño-Navarro¹

¹División de Biología Molecular, Instituto Potosino de Investigación Científica y Tecnológica, Camino a la Presa San José, #2055, Col. Lomas 4ª sección. San Luis Potosí, San Luis Potosí 78216, México.
E-mail: grecia.hernandez@ipicyt.edu.mx

The DNA spatial arrangement inside the nucleus depends critically on the stimuli outside the cell and plays a fundamental role in gene expression. In the fungal opportunistic pathogen *Candida glabrata* (*Nakaseomyces glabrata*) gene regulation plays an essential role on its ability to colonize and disseminate within the host, in part through the regulation of the expression of adhesins that are involved in its adherence to epithelial cells. For the most part, this characteristic depends on the gene *EPA1* which is subject to subtelomeric silencing and encodes the cell wall protein Epa1. Subtelomeric silencing in *C. glabrata* depends on the SIR complex, yKu70/yKu80, CgRap1, and CgAbf1. Moreover, we previously found that the telomere E-R that harbors the genes *EPA1*, *EPA2*, and *EPA3*, can be remodeled into loop-like secondary structures through the interaction of *cis*-acting elements like the protosilencer Sil2126, the Negative Element (NE), and other intergenic regions. In this work we found that Rap1 and Abf1 proteins bind at different positions throughout this telomere using chromatin immunoprecipitation assays (ChIP-qPCR). Moreover, we found that CgAbf1 binds near the promoter region of *EPA1* at two different positions, and that the expression of *EPA1* is negatively regulated by Abf1. Consistently, the truncated mutant *Cgabf1-43* (that lacks the last 43 amino acids at its C-terminal) was more adherent to epithelial cells *in vitro* in comparison to the parental strain. We also determined the binding sequences for Abf1 and Rap1 in *C. glabrata*, using the data from the ChIP assays and bioinformatic tools. In addition, we found that CgAbf1 co-immunoprecipitates with other silencing proteins like CgRap1, Sir3, Sir4, yKu70/80, which suggests their interaction.

On the other hand, we observed that Sir4 and yKu70/80 colocalized with the nuclear periphery which suggests that the telomeres might be localized at this region thereby reinforcing the silencing of the genes adjacent to the telomere.

Keywords: Adhesins, *EPA1*, CgAbf1, CgRap1, chromatin, telomere, silencing.

EPIGENETIC REGULATION OF *ATP2A3* AND *CACNA1H* GENES IN HEPG2 CELLS

Guadalupe Hernández-Martínez^{1,2}, Ángel Zarain Herzberg³,
Andrés Hernández-Oliveras⁴, Juan Santiago-García²

¹PhD Program in Health Sciences, Veracruzana University, ²Institute of Biological Research, Veracruzana University, Xalapa, Veracruz, México, ³Department of Biochemistry, School of Medicine, National Autonomous University of Mexico, Mexico City, ⁴Laboratory of Immunoncology, Science & Life Foundation, Santiago Chile.
E-mail: jusantiago@uv.mx Tel: (228) 841-8900 ext 13410

ATP2A3 gene is downregulated in several cancer types and cancer cell lines, including breast, lung, colon, gastric, choroid plexus, and hepatocellular carcinoma (HCC).¹⁻⁷ *CACNA1H* is also downregulated in brain, breast, kidney, and lung cancer⁸, and its altered expression was associated with progression from non-alcoholic hepatic steatosis to HCC in mice.⁹ Histone deacetylase inhibitors (HDACi) increase *ATP2A3* expression in gastric, colon, and lung cancer cells. Sodium butyrate and trichostatin A, two HDACi, induce *ATP2A3* expression by increasing K9 and K27 acetylation in rat HCC cells.⁶ In this study, we investigated whether LBH589, a HDACi, and 5-Azacytidine, a DNA hypomethylating molecule, regulate *CACNA1H* and *ATP2A3* expression in human HCC HepG2 cells. Our results show that LBH589 induced *ATP2A3* and *CACNA1H* expression through acetylation of lysine 9 and 27 of histone 3, which suggest that histone acetylation and deacetylation regulate the expression of these genes. Treatment with 5-Azacytidine induced *ATP2A3* and *CACNA1H* expression, suggesting that DNA methylation may regulate the expression of these genes. To evaluate the methylation status of their promoters, five CpG sites were explored by MS-PCR; the results showed that *ATP2A3* promoter remained methylated even after 5-Aza treatment, whereas the *CACNA1H* promoter was unmethylated. Furthermore, the combined treatment with LBH589 and 5-Aza induced, but did not potentiate the expression of both genes. In conclusion, LBH589 and 5-Aza induce the expression of *ATP2A3* and *CACNA1H* genes.

References

1. Papp B, Brouland J.P (2011). *Breast Cancer (Auckl)*, **5**, 163-174.
2. Gélébart P, Kovacs T, Brouland J.P (2002). *J Biol Chem*, **277**, 26310-26320.
3. Brouland J.P, Gélébart P, Kovacs, et al (2005). *American J Pathol*, **167**, 233-242.
4. Arbabian A, Brouland J.P, Apáti Á, et al (2013). *FEBS J*, **280**, 5408-5418.
5. Ait-Ghezali L, Arbabian A, Jeibmann A, et al (2014). *Neuropathol App Neurobiol*, **40**, 726-735.
6. Hernández-Oliveras A, Izquierdo-Torres E, Meneses-Morales I, et al (2019). *Int J Biochem Cell Biol*, **113**, 8-16.
7. Contreras-Leal E, Hernández-Oliveras A, Flores-Peredo L, et al (2016). *Mol Carcinog*, **55**, 1477-1485.
8. Phan NN, Wang C, Chen et al (2017). *Oncology Lett*, **12**, 2059-2074.
9. Liang JQ, Teoh N, Xu L, et al (2018). *Nature communications*, **9**, 4490.

FUNCTIONAL STUDY OF MAF1 IN THE PROTOZOAN PARASITE *LEISHMANIA MAJOR*

Aldo R. Hernández Zamarripa¹, Luis A. Rivera Rivas¹, Luis E. Florencio Martínez¹, Rebeca Manning Cela², Tomás Nepomuceno Mejía¹ and Santiago Martínez Calvillo¹

¹Facultad de Estudios Superiores Iztacala, UNAM. Av. de los Barrios 1, Col. Los Reyes Iztacala, Tlalnepantla, Edo. de México, CP 54090, México; ²Departamento de Biomedicina Molecular, CINVESTAV-IPN, México
E-mail: aldo_dodo1@hotmail.com; scalv@unam.mx Tel.: +52-55-5623-1333 (ext. 39781)

RNA polymerase III (Pol III) is responsible for synthesizing small essential RNA molecules, such as tRNAs and 5S rRNA. Maf1 was originally studied in *S. cerevisiae* as a transcriptional repressor of Pol III in response to multiple stress conditions. It was later found that Maf1 also regulates Pol I and Pol II transcription and that it is involved in several other functions in yeast and other organisms. Its activity and subcellular localization are regulated by phosphorylation. Little is known about transcriptional regulation of Pol III in *Leishmania major*, a flagellated protozoan parasite that presents atypical genetic expression mechanisms, such as polycistronic transcription and the maturation of nuclear pre-mRNAs by trans-splicing. By mass spectrometry analysis, five phosphorylated residues that had not been described in other eukaryotes were experimentally identified, including one located in the domain known as box B. Through tandem affinity purification experiments with a cell clone of *L. major* that expresses the LmMaf1 protein fused to the PTP flag, possible interactors of Maf1 were identified under optimal and unfavorable growth conditions. Purified proteins were identified by mass spectrometry and bioinformatic analysis. Interestingly, in both growth conditions, proteins with a great diversity of functions were identified, such as subunits of RNA Polymerases, transcription regulators, RNA-binding proteins, lipid metabolism, and ubiquitination and interaction with the proteasome. Additionally, ChIP assays showed that LmMaf1-PTP associates with Pol III promoter regions and suggested the binding of the protein to some other genomic regions. Therefore, these results indicate that Maf1 plays multiple cellular roles in *L. major*. Immunofluorescence assays are currently being carried out to determine the subcellular localization of Maf1 in this parasite. This work was supported by grants IN208224 (PAPIIT, UNAM) and CF-2023-I-820 (CONAHCYT).

METABOLIC PROCESSES ASSOCIATED WITH CHEMOTAXIS RESPONSES IN *KOSAKONIA COWANII*

M. Inés Hernández-Castellanos¹, Verónica Morales-Tlalpan^{1,2}, Karla Fernandez-González¹, Regina Ruiz-Barrios¹, J. Campos Guillen³ & Carlos Saldaña^{1,2*}

¹Laboratorio de Biofísica de Membranas y Nanotecnología, ²Laboratorio Nacional de Visualización Científica Avanzada FCN-UAQ. Facultad de Ciencias Naturales, ³Facultad de Química. Universidad Autónoma de Querétaro. Juriquilla, Qro., México C.P. 76230. Phone +52 442 192 1200, ext. 65332. *carlos.saldana@uaq.mx

Kosakonia cowanii is a Gram-negative, facultative anaerobic, motile bacterium belonging to the Enterobacteriaceae family and characterized by the presence of flagella. In recent years, it has gained significance in fields such as agriculture, biotechnology, and biomedicine. Genome sequencing of *K. cowanii* has provided valuable insights into its metabolic capacity and genes associated with antibiotic resistance. Recent research has revealed that *K. cowanii* exhibits chemotactic response towards capsaicin concentrations, a substance naturally present in chili seeds and tissues. (Gonzalez Espinosa et al., 2023) The primary objective of this study is to identify, through microbiological, physiological, and molecular biology techniques, the metabolic process of *K. cowanii*. To achieve this, we will design a specific operon and conduct metabolomics to determine if secondary metabolites produced by capsaicin are directly linked to the bacterial receptor response triggering the chemotaxis pathway. Consequently, we anticipate that the results obtained will provide sufficient information on heterologous gene expression, aiding us in constructing knockout in specific bacterial genes to identify related swimming patterns that correlate with the structure-function induced by capsaicin. These patterns can be visualized using functional fluorescence microscopy. Subsequently, we will perform the *ex vivo* expression of the same experiment in mammalian cells to potentially identify the response mechanisms to their calcium dynamics.

Acknowledgments. FONDEC-UAQ, FONFIVE 2024 and CONAHCyT A1-S-26966 for their generous support and financial backing provided to Carlos Saldaña.

Referencias:

1. González Espinosa J, et al. (2023). *Kosakonia cowanii* Ch1 Isolated from Mexican Chili Powder Reveals Growth Inhibition of Phytopathogenic Fungi 11(7):1758.

DETECTION OF SINGLE-NUCLEOTIDE POLYMORPHISMS IN *BLK*, *STAT4* AND *IRF5* GENES ASSOCIATED WITH PEDIATRIC ANTIPHOSPHOLIPID SYNDROME BY REAL-TIME PCR GENOTYPING

Tanya Paola Hidalgo Romero¹, Charmina Aguirre Alvarado^{1,2}, Vilma Carolina Bekker Méndez¹, Alfonso Ragnar Torres Jiménez³ y Francisco Xavier Guerra Castillo¹

¹Unidad de Investigación Biomédica en Inmunología e Infectología, Hospital de Infectología "Dr Daniel Méndez Hernández", CMN "La Raza", Instituto Mexicano del Seguro Social (IMSS), México City, México. ²Laboratorio de Bioquímica Farmacológica. Escuela Nacional de Ciencias Biológicas. Instituto Politécnico Nacional, México City, México. ³Unidad Médica de Alta Especialidad Hospital General "Gaudencio González Garza". Servicio de Reumatología Pediátrica. México City, México. E-mail: tanyahidalgo.11@gmail.com

Antiphospholipid syndrome (APS) is an autoimmune disease characterized by the positivity of antibodies against phospholipid-binding proteins in the presence of recurrent arterial/venous thrombotic events and/or pregnancy morbidity.¹ Although the etiology of APS is unknown, it has been suggested that it may arise in a genetically predisposed subject following antigenic stimuli from various sources as in other autoimmune diseases. Association studies with various alleles of genes near the coding region of the major histocompatibility complex, in particular the *BLK*, *STAT4* and *IRF5* have been studied as the genetic predisposition of APS.^{1,2} The aim of this study is to analyze the association of single-nucleotide polymorphisms (SNPs) in *BLK* (rs2736340), *IRF5* (rs2070197) and *STAT4* (rs7574865) with APS in a cohort of 53 Mexican pediatric patients with primary APS compared with a control group of 132 healthy subjects from the same geographic area. Genotyping of these SNPs was performed by real-time polymerase chain reaction (qPCR) using TaqMan predesigned SNP genotyping assays, then genotypic frequencies and odds ratios (OR) were calculated. The results were analyzed using the codominant, dominant, recessive, over dominant and additive genetic models; according to the additive model, significant associations were found for the SNP rs7574865 in *STAT4* (OR=2.05, P = 0.002), for the SNP rs2736340 in *BLK* (OR=2.1, P = 0.007) and for the SNP rs2070197 in *IRF5* (OR=3.17, P < 0.001). Also, the frequency of the risk allele (T) of the SNP rs7574865 in patients with APS (60.6%) was higher compared to healthy controls (41.7%). For SNP rs2736340, the risk allele (T) was also found more frequently in patients with APS (80%) compared to controls (64.5%). The same way, in the SNP rs2070197, the risk allele (C) was more frequently present in patients with APS (53.3%) in contrast to the group of controls where it was present in 25.2%. The results obtained indicate that the SNPs evaluated in the *BLK*, *STAT4* and *IRF5* genes are likely to be a crucial component in the pathogenesis of pediatric APS, the discovery of these associations could improve the understanding of the molecular pathways involved in the disease and provide new therapeutic targets.

References

1. Yin H, et al. (2009). *Arthritis Rheum*, **60**, 2468-2471.
2. Fredi M, et al. (2010). *Arthritis Rheum*, **62**, 1201-1202.

MCF-7CELLS MODIFY CAPACITIVE CALCIUMENTRY (CCE) BY EXPOSURE TO METHANOLIC EXTRACTS OF CAPSICUM ANNUU ML. VAR . FASCINATUM (FAS)

Joel Hurtado Patiño¹, Roberto Jorge Garcia-Mendoza¹, Verónica Morales-Tlalpan^{1,2}, Adriana González Gallardo³, Angélica Feregrino-Pérez⁴ & Carlos Saldaña^{1,2*}

¹Laboratorio de Biofísica de Membranas y Nanotecnología, ²Laboratorio Nacional de Visualización Científica Avanzada UNAM-UAQ. ³Unidad de Proteogenómica del Instituto de Neurobiología UNAM; ⁴Laboratorio de Metabolitos y Nanocompuestos, Facultad de Ingeniería UAQ. Facultad de Ciencias Naturales, Universidad Autónoma de Querétaro. Juriquilla, Qro., México, C.P. 76230.

*carlos.saldana@uaq.mx

Cancer is a multiple disease, which causes cells to have the ability to divide efficiently without completing the cell cycle, affecting the proper distribution of organelles, DNA and membranes. Cancer develops successfully because various mechanisms function abnormally: nutrient absorption, cellular senescence, inflammatory processes, immune system, cellular molecular activity, and epigenetic marks, cell migration and colonization, promoting the maintenance of the cancer phenotype. The evolution and the risk of developing it is influenced by age,

sex, genes, environment and, of course, nutrition (calories, type of nutrients, biomolecules, alcohol, among others). Many studies suggest that including plant compounds in the human diet can modify the risk of developing cancer. Therefore, we are studying the effects of a methanolic extract of *Capsicum annuum* L. var. *Fascinatum* (FAS) which is rich in antioxidants: phenolic compounds (epicatechin, chlorogenic acid, quercetin and others), carotenoids (beta-carotene, lycopene, beta cryptoxanthin, lutein and tocopherol) and flavonoids (anthocyanins and tannins). These antioxidants regulate the cell cycle, proliferation and apoptosis (mechanisms mediated by the Ca²⁺ ion). Our experiments in breast cancer cells (MCF-7): 1) demonstrated that this cell line exhibits an altered CCE (a mechanism that quantifies Ca²⁺ in the lumen of the endoplasmic reticulum, essential in maintaining the phenotype), 2) CCE decreases with incubation (24 h) with FAS extract and, 3) transcriptomic indicates that many molecular compounds of CCE are altered when cells are incubated with FAS. These data suggest that FAS extract can modify CCE and gene expression in this cell type.

Acknowledgements. We extend our sincere gratitude to the funding institutions: FONDEC-UAQ, FONFIVE 2024 and CONAHCyT A1-S-26966 for their generous support and financial backing provided to Carlos Saldaña.

EVALUATION OF THE EXPRESSION OF MIRNAS CONTAINED IN EXTRACELLULAR VESICLES OF PATIENTS DIAGNOSED WITH COVID-19

Nancy Vanessa Iglesias Vázquez¹, Susana Bernardo Hernández¹, Gustavo Acosta Altamirano², Norma Estela Herrera González³, J. Carlos Fernández¹, Mónica Sierra Martínez²

¹Unidad de Investigación en Salud Hospital Regional de Alta Especialidad de Ixtapaluca IMSS-Bienestar, Carr. Federal México-Puebla Km. 34.5, 56530 Ixtapaluca, Méx. ²Dirección de Investigación, Hospital General de México, Dr. Balmis 148, Doctores, Cuauhtémoc, 06720 Ciudad de México, CDMX. ³Escuela Superior de Medicina, Instituto Politécnico Nacional, Salvador Díaz Mirón esq. Plan de San Luis S/N, Miguel Hidalgo, Casco de Santo Tomas, 11340 Ciudad de México, CDMX. monica.sierram@hraei.gob.mx

Extracellular vesicles (EVs) consist of lipid bilayers that encapsulate a complex cargo. They are secreted by various types of cells, allowing intercommunication by delivering biomolecules, lipids, proteins, and microRNAs (miRNAs) to recipient cells. Researchers have focused on the study of miRNAs because they are molecules responsible for regulating genetic expression. Yanbo Wang et al. conducted research on serum miRNAs that included 3 miRNAs (miR-24-3p, miR-7-5p, miR-145-5p), concluding that they decrease their expression in elderly patients, in addition to directly inhibiting the S protein and the replication of SARS-CoV-2 in vitro. Therefore, our work group aimed to evaluate the level of expression of the aforementioned miRNAs contained in extracellular vesicles of COVID-19 positive patients. Methodology: Peripheral blood was collected from SARS-CoV-2 positive patients with mild, moderate and severe symptoms (n=10) and a group of healthy individuals. EVs were isolated using the ultracentrifugation technique. EVs were characterized by flow cytometry as well as by electron microscopy. miRNA extraction was performed using the miRVana Kit (Invitrogen), then the detection of miRNAs of interest was performed through digital PCR (QIAcuity One). Results: The copy number of each of the miRNAs miR-24-3p (13.25 copies / μ l), miR-7-5p (0.42 copies / μ l), miR145-5p (2.18 copies / μ l) was obtained in the healthy group, as well as in the groups stratified as mild, moderate and severe; observing that the 3 miRNAs were under-expressed in the presence of SARS-CoV-2; noting a greater expression difference in the group of severe patients, for miRNA-24-3p the expression was 4.5 times lower, miRNA-7-5p decreased 2.3 times less and miRNA-145-5p decreased 2.83 times less compared to the control group, being statistically significant $p=0.001$. Conclusion: We show that miRNAs from extracellular vesicles that were released from the blood plasma of patients with mild, moderate and severe COVID-19 disease were under-expressed in the 3 miRNAs of interest, especially in the group of severe patients. The preliminary results obtained from this work coincide with what has been published in the literature. It is known that the miRNAs under study are targets of the S protein of the SARS-CoV-2 virus, so the poor prognosis in severely ill patients is probably due to the low levels of these miRNAs since they are associated with various processes.

GENETIC VARIANTS RELATED WITH NEOADJUVANT CHEMOTHERAPY RESPONSE IN BREAST CANCER PATIENTS

Carlos Alfredo Barrón-Gallardo^{1,2}, Adriana del Carmen Aguilar Lemarroy^{3,4}, Luis Felipe Jave Suárez^{3,4}

¹Departamento de Ciencias de la Salud, Universidad Autónoma de Guadalajara. ²Unidad de Investigación, GeneScience.

³División de Inmunología, Centro de Investigación Biomédica de Occidente – IMSS. ⁴Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara.

Cancer is a leading cause of death worldwide, accounting for 10 million deaths in 2022. Among all types of cancers, breast cancer ranks first in mortality and incidence worldwide. The standard treatment for locally advanced breast cancer is neoadjuvant chemotherapy, notwithstanding some patients do not respond to this treatment. A factor involved in chemoresistance is the patient genetic background. DNA variants (DVs) are supposed to be important in the chemotherapy response. Gene expression patterns of responder and non-responder patients to neoadjuvant chemotherapy have been previously reported. However, DVs related to the responder's or non-responders' phenotype remain to be addressed. In this study we aimed to identify DVs related to the neoadjuvant chemotherapy response.

Methods. RNAseq Breast cancer fasta files from GEO DataSets GSE162187, were analyzed for variant calling among responder and non-responder to chemotherapy. GRCh38.p13 was used as the reference genome for fasta alignment with the STAR tool. For data processing and variant calling, the GATK and the HaplotypeCaller algorithm were used. VCF files were analyzed with R studio to find common mutations among responders and non-responders.

Results. A total of 116 DVs were found in 90% of the non-responder patients but not in the responder; of those variants, 21 were present in all non-responder patients. On the other hand, 44 mutations were present in 90% of responder patients but not in the non-responders; of those, 7 mutations were present in 100% of responder patients. Mutations exclusive and present in all non-responder patients were most prevalent in chromosome 15, followed by chromosomes 16 and 19. In addition, 9 DVs present in 90% of non-responders were missense with impact at protein level. Moreover, 42% of the DVs were exclusive of responder patients and were in chromosome 11; interestingly, 1 of the latter induced a missense codon.

Conclusions. We found DVs exclusive of responder and non-responder phenotypes, which could be used as predictive chemotherapy response biomarkers with high specificity.

ESTRADIOL, MEDROXYPROGESTERONE, AND cAMP DIFFERENTIALLY REGULATE UNFOLDED PROTEIN RESPONSE GENES DURING DECIDUALIZATION IN HUMAN IMMORTALIZED ENDOMETRIAL STROMAL CELLS

Miguel Ángel Jiménez-Beltrán¹, Jesús Iván Jiménez-Rivera¹, Alejandra Monserrat Retis-Resendiz¹,
Moisés Leon-Juárez² and Edgar Ricardo Vázquez-Martínez^{1*}

¹Unidad de Investigación en Reproducción Humana, Instituto Nacional de Perinatología-Facultad de Química, UNAM. ²Departamento de Inmunobioquímica, Instituto Nacional de Perinatología.

*Corresponding author: vamer@comunidad.unam.mx

The implantation of the blastocyst in the uterus requires the differentiation of the endometrium, a process called decidualization. Several molecules, such as progesterone, estradiol (E2) and cAMP collaborate to regulate gene expression during this process, which has been reproduced in vitro in endometrial stromal cell lines. On the other hand, it has been suggested that the unfolded protein response (UPR) plays a role in decidualization. Furthermore, the expression of UPR-related genes is regulated during in vitro decidualization. However, the individual roles of ovarian steroid hormones and the second messenger cAMP in the regulation of UPR genes have not been explored. Thus, the aim of this work was to determine the participation of each component of the decidualization cocktail in the expression regulation of UPR genes. For this, immortalized Human Endometrial Stromal Cells (t-HESC, ATCC CRL-4003) were treated with a combination of E2, MPA, and cAMP (EMC) for 48 h. Cells were also treated with each molecule alone or in pairwise combinations. RT-qPCR was performed to quantify the expression levels of decidualization biomarkers and UPR genes. Our results showed that EMC treatment induced the expression of *XBPI* and *XBPIs* isoforms, *GRP78*, and *PERK*, while decreasing *CHOP* expression. cAMP induced *XBPIs* and *GRP78* expression while downregulating *CHOP*. E2 and cAMP concurrently regulated *PERK* expression. From this, we conclude that this study provides novel insights into the regulation of UPR-relevant genes during in vitro decidualization of HESCs, which results from a complex interaction of hormones (E2, MPA) and the second messenger cAMP.

Acknowledgments. We acknowledge UNAM-PAPIIT (Grant Numbers IA203822 and IA203822) and Instituto Nacional de Perinatología 'Isidro Espinosa de los Reyes' (INPer, grant number 2017-2-93).

ROLE OF FLOTILLIN FROM *PHASEOLUS VULGARIS* DURING A MUTUALISTIC INTERACTION WITH *RHIZOBIUM TROPICI*

Pamela Jiménez-Chávez¹, Ana I. Chávez-Martínez¹, Janet Palacios-Martínez¹, Fernando Lara-Rojas², Ana Yasmín Martínez-Ocampo³ and Luis Cárdenas¹

¹Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, México, ²Departamento de Interacciones Planta-Insecto, Centro de Desarrollo de Productos Bióticos Instituto Politécnico Nacional, Yauhtepec, Morelos, México, ³Facultad de Ciencias, Universidad Autónoma de Morelos, Cuernavaca, Morelos, México. Av. Universidad 2001, Colonia Chamilpa. C.P. 62210. Tel. 777-3291666
E-mail: luis.cardenas@ibt.unam.mx

The symbiotic relationship between bacteria of genus *Rhizobium* and the plant of common bean (*Phaseolus vulgaris*), is one of the most common successful plant-microbe interactions. The bacteria are internalized in the root through specialized cells such as the root hair; to do so, they induce the invagination of the plasma membrane and cell wall of root hairs resulting in the formation of a structure called infection thread and subsequently penetrate the root cortex to develop a new organ known as nodule, where the bacteria will be release and internalized, so they can differentiate into bacteroides and be able to carry out the nitrogen fixation. It was shown that membrane microdomains are important for membrane shaping, trafficking and signal transduction. Flotillins are lipid microdomains components in both vegetal and animal cells and in plants were shown to play important role in plant-microbe interaction. In this study, thanks of bioinformatic analysis, we identified one flotillin gene in *P. vulgaris* genome, also we identified flotillin genes in different legumes. Later, we conducted transcriptional profiling in common bean roots to determine the specific expression patterns of flotillin during nodulation. Also, we determinate the subcellular localization of flotillin during the interaction with rhizobia. In addition to, we analyzed the promotor activity of flotillin during different nodulation stages. Our results reveal that flotillin is expressed in the early stages of nodulation and throughout the development of the nodule. Subcellular localization indicates that flotillin have a role in vesicular trafficking and root hair polar growth.

IDENTIFICATION OF SNORNAS THAT INTERACT WITH LIPIDS USING THE LIPID-RNA SEQ TECHNIQUE IN *SACCHAROMYCES CEREVISIAE*

Irma A. Jiménez-Ramírez¹*, Andrea Bayona-Hernández¹, Alma Laura Rodríguez-Piña¹,
Wilma A. González Kantun¹, Enrique Castaño¹*

¹Centro de Investigación Científica de Yucatán, Unidad de Biología Integrativa. Calle 43 No. 130 x 32 y 34,
Col. Chuburná de Hidalgo. C.P 97205, Tel. (999)-981-39-43, Ext: 242
E-mail: anggye.apple@gmail.com*; enriqueC@cicy.mx*

Small nucleolar non-coding RNAs (snoRNAs) are mainly located in the nucleolus of eukaryotic cells. These snoRNAs are 60 to 300 nucleotides long and are encoded within intronic regions of protein-coding and non-coding genes by RNA polymerase II. Its function includes is to guide the chemical modification of other RNA molecules, such as ribosomal RNAs (rRNA). The architecture of snoRNAs is characterized by having conserved elements, such as the H/ACA and C/D boxes. H/ACA boxes guide rRNA pseudouridylation, while C/D boxes form a ribonucleoprotein complex that guides rRNA methylation. snoRNAs play a fundamental role in modifying and processing rRNA´s within the nucleus, and is crucial for proper function in protein synthesis.

The lipids are an essential part of the cell due to their important role in maintaining the cellular membrane. Phosphatidylinositol 4,5-bisphosphate (PIP₂) is an amphipathic lipid that regulates various cellular processes such as phagocytosis, endocytosis, vesicle trafficking, polarization, and cell migration. In addition, to being found in the plasma membrane, PI(4,5)P₂ has been localized within nuclear compartments such as the nucleolus, and nuclear speckles. Studies have revealed that non-coding RNAs can interact with different molecules, including phosphoinositides, through specific binding sequence, an example is the long non-coding RNA LINK-A, which interacts with PI(3,4,5)P₃, was shown to facilitate the recruitment of AKT. Our research group designed a new method that allows us to identify ncRNAs that interact with lipids. In this research, we identified 921 types of *S. cerevisiae* RNAs that interact with PI(4,5)P₂, including snoRNA191, an H/ACA box snoRNAs that is involved in the pseudouridylation process. We also identified the possible RNAs interaction site with PI(4,5)P₂ by predicting RNA secondary structures. This work could potentially help understand the role of *S. cerevisiae* snoRNAs and their interacting partners as they form liquid-liquid phase separation process those allowing specific aggregation and increase concentration to carry out specific functions, thereby presenting a fresh perspective in the field of molecular biology and genetics.

Reference:

1. Bayona-Hernandez, A., Guerra, S., Jiménez-Ramirez, I. *et al*. LIPRNAseq: a method to discover lipid interacting RNAs by sequencing. *Mol Biol Rep* **50**, 6619–6626 (2023). <https://doi.org/10.1007/s11033-023-08548-5>
2. Lin, A., *et al* (2017). The LINK-A lncRNA interacts with PtdIns(3,4,5)P₃ to hyperactivate AKT and confer resistance to AKT inhibitors. *Nature cell biology*, **19**(3), 238–251. <https://doi.org/10.1038/ncb3473>

UNVEILING THE LANDSCAPE OF LINC00052 MOLECULAR MECHANISMS IN BREAST CANCER CELLS BY BIOINFORMATIC AND EXPERIMENTAL ANALYSES

Miguel Ángel Juárez Mancera, José Manuel Sánchez López, Gisela Ceballos Cancino, Magali Espinosa Castilla, Benjamín Bustamante Gómez, Araceli Ruiz Silvestre, Gretel Mendoza Almanza, Jorge Meléndez Zajgla, Vilma Maldonado Lagunas and Floria Lizárraga Sánchez

Laboratorio de Epigenética y Laboratorio de Genómica Funcional del Cáncer. Instituto Nacional de Medicina Genómica (INMEGEN)–Posgrado en Ciencias Biológicas, UNAM. Periférico Sur 4809 Col. Arenal Tepepan, Tlalpan, CDMX, CP 14610.

Background. Recent studies have shown that long-non-coding RNA (transcripts with lengths>200nt) play key roles in tumor progression. Previous data reported that LINC00052 expression is diminished in triple-negative human breast cancer (TNBC) samples, while it is expressed in luminal subtypes¹. Furthermore, our research group found that LINC00052 expression was repressed during the formation of breast cancer (BC) multicellular spheroids and that and its downregulation inhibits BC cell migration^{2,3,4}. However, as for most lncRNA, cellular and molecular mechanisms through which LINC00052 modulates BC cells functions remain unknown.

Methods/Results. Bioinformatic and experimental strategies were followed to assess this aim. First, LINC00052 expression was examined in BC patients` RNA samples (TCGA) and inferred its cellular and molecular mechanisms. Interestingly, higher LINC00052 levels are related to better BC patient`s overall survival in samples of all molecular subtypes. A similar LINC00052 expression pattern was observed in an in-house patient cohort. Surprisingly, LINC00052 expression increased when MCF- 7 and ZR-75-1 cells were treated with estradiol. In agreement with bioinformatic analyses, we observed that MCF-7 cells with low LINC00052 levels showed better cellular protection against DNA damage and diminished growth capacity. Furthermore, in cisplatin-resistant MCF-7 cells⁵, LINC00052 expression was downregulated. To get further insight about LINC00052 molecular mechanisms, a prediction of the possible molecular partners was made (RNAInter V4.0). Additionally, we performed ChIRP assays from MCF-7 cell extracts, a test based on the affinity capture of a target lncRNA using specific biotinylated oligonucleotides. Overall, this study highlights the need for further research to unravel LINC00052 molecular mechanisms and potential clinical applications in BC.

References

1. Mingming L., et al. (2016). lncRNAs as new biomarkers to differentiate triple negative breast cancer from non-triple negative breast cancer. *Oncotarget*. 7(11): 13047 – 13059.
2. Muñoz-Galindo, L., et al. (2019). Changes in the transcriptome profile of breast cancer cells grown as spheroids. *Biochemical and Biophysical Research Communications*. 516(4), 1258 – 1264.
3. Pacheco-Marín, R., et al. (2016). Transcriptome profile of the early stages of breast cancer tumoral spheroids. *Sci Rep*. 6(1), 23373.
4. Sanchez-Lopez, J. M., et al. (2021). Integrative analysis of transcriptional profile reveals LINC00052 as a suppressor of breast cancer cell migration. *Cancer Biomarkers*. 30(4), 365–379.
5. Ruiz-Silvestre A, et al. (2024) Transcriptomic Changes in Cisplatin-Resistant MCF-7 Cells. *Int J Mol Sci*. 25(7):3820.

THE INCREASE IN METHYLATION OF THE ABCG2 GENE IS RELATED TO THE PRESENCE OF ITS Q141K ALLELE AND LOWER EXPRESSION OF THE GENE IN PERIPHERAL BLOOD OF PATIENTS WITH GOUT AND CONTROLS

Ambar López Macay¹, Yunuen Velázquez Jiménez¹, Javier Fernández Torres¹,
Yessica Eduviges Zamudio Cuevas¹, Karina Martínez Flores¹, Denisse Clavijo Cornejo²

¹Synovial Fluid Laboratory, National Institute of Rehabilitation Luis Guillermo Ibarra Ibarra,

²Rheumatology laboratory National Institute of Rehabilitation Luis Guillermo Ibarra.

Phone Number: 55 5999 1000, Extensión: 19501 E-mail: lopez_macay@hotmail.com,

Gout is a disease characterized by joint pain due to the response activated by the precipitation of monosodium urate crystals in the joints, due to a persistent elevation of serum uric acid. ABCG2 is a urate transporter that belongs to the ATP-binding ABC cassette protein family. ABCG2 is important for the protection of cells against drugs and organic molecules. Polymorphisms (SNPs) of ABCG2 have been associated with hyperuricemia and gout, Q141K and Q126X are the most frequent SNPs in various populations studied. OBJECTIVE: To analyze the presence of the SNP rs2231142 (Q141K) of ABCG2 in a cohort of patients with gout and controls and its possible relationship with the methylation profile of the CpG island of the ABCG2 gene and its gene expression. METHODS: This case-control study included 560 subjects of Mexican origin, 269 of whom were patients from a well-characterized and drug-controlled cohort of the INRLGII outpatient clinic. The ABCG2 rs2231142 SNP was analyzed from peripheral blood by extraction of genomic DNA. The expression and methylation analysis of ABCG2 as well as the variant analysis was performed by RT-PCR. RESULTS: After adjustment for age and sex, there were significant differences in the distribution of genotypes of the rs2231142 variant between cases and controls. In the codominant model, the homozygous TT genotype was more common in patients than in controls (35.4% vs. 4.6%) and was associated with a higher risk of gout (OR=24.23, 95%, CI=12.01 – 49.71, $P \leq 0.0001$). The methylation percentage of ABCG2 was higher in the GT and TT genotype than in the GG in patients and controls and we found a negative correlation between the methylation percentage and gene expression in patients ($p \leq 0.05$). CONCLUSION: Our patients have a high frequency of the rs2231142 SNP compared to controls. The presence of the Q141K allele was associated with greater methylation of ABCG2 and lower expression of the gene.

COMPARATIVE GENOMIC ANALYSIS OF *CANDIDA GLABRATA* CLINICAL ISOLATES TO UNDERSTAND ANTIFUNGAL RESISTANCE

Ana Lizbeth López-Marmolejo¹; Alejandro Juárez-Reyes¹; Guadalupe Gutiérrez-Escobedo¹; Alejandro De Las Peñas¹; Enrique Merino-Pérez²; Irene Castaño-Navarro¹

¹División de Biología Molecular, Instituto Potosino de Investigación Científica y Tecnológica. Camino a la Presa San José 2055, 78216 San Luis Potosí, México; Tel: (444) 834-2038;

²Instituto de Biotecnología, UNAM Av. Universidad 2001, Chamilpa, 62210 Cuernavaca, Morelos, México; Tel: (777) 329 1600. E- mail: icastano@ipicyt.edu.mx

Candida glabrata is an emergent and opportunistic fungal pathogen that colonizes and survives in different niches within its host. The objective of this work is to determine the phenotypic and genotypic changes that occur in a group of sequential clinical isolates of *C. glabrata* throughout the course of an infection. These isolates were obtained from the same patient (P7), from blood (four isolates) and urinary tract (one isolate) cultures. We performed the sequencing and assembly of the complete genome of three of these isolates that are from the same patient but from different anatomical isolation sites, P7-1, P7-5 (blood) and P7-3 (urinary tract). We used Oxford Nanopore technology, with a subsequent error correction with Illumina sequencing, to determine the structure and variation of the genomes of these isolates during the infection in different anatomical sites. We compared the nucleotide sequence of the complete genomes by Average Nucleotide Identity (ANI) analysis, and we observed that clinical isolates P7-1, P7-3 and P7-5 have greater than >99% percentage of identity and are grouped in the same clade and therefore have a clonal origin. We identified the subtelomeric regions of the chromosomes of each clinical isolate (regions within ~20 Kb from the telomeres) and found rearrangements and variations, which occur in the regions closest to the telomere. In addition, the ORFs found in the subtelomeric regions encode adhesin-like proteins (ALP). Innate or acquired drug resistance is an important public health problem, particularly in *C. glabrata*, and we recently found that P7-3 (from urine) is the only isolate from this patient that is highly resistant to the echinocandin caspofungin, while the other four isolates from P7 are sensitive. However, this same isolate P7-3, is the only one that displays susceptibility to fluconazole (FLC), while the rest of the isolates are resistant to this antifungal. We looked for SNVs that are unique to P7-3 and are related to drug resistance that could explain the particular phenotype of P7-3 compared to the other isolates from P7.

Taken together, our results show phenotypic and genotypic variability between clonal isolates from different niches within a single host, suggesting microevolution of *C. glabrata* during an infection.

ANALYSIS OF THE INTERACTION NETWORK BETWEEN NON-CODING RNAs AND THE TRANSCRIPTION FACTORS C-JUN AND C-FOS AND ITS EFFECT ON THE EXPRESSION OF GENES THAT MEDIATE RENAL HYPERTROPHY

Isaac Virgilio Martínez Cárdenas, Yussel F. Pérez Navarro, Elisa I. Azuara Liceaga,
Máximo B. Martínez Benítez, Lilia Lopez-Canovas

Posgrado en Ciencias Genómicas, Universidad Autónoma de la Ciudad de México, San Lorenzo 290,
Col. del Valle, Benito Juárez 03100, Ciudad de México, México.
E-mail: lilia.lopez.canovas@uacm.edu.mx

Diabetic nephropathy (DN) is one of the main microvascular complications of diabetes mellitus, a condition that affects people worldwide. The interaction between advanced glycation end products (AGEs) and their receptors (RAGE) initiates signaling cascades that lead to renal hypertrophy, fibrosis, and inflammation, which in turn mediate the etiopathogenic process of DN. These signaling pathways could be modulated by the interaction of the molecules that make up them with non-coding RNAs. The transcription factors c-Jun and c-Fos are upregulated during hyperglycemia and are molecular effectors of the Janus kinase 2 pathway, which is involved in the hypertrophy process associated with DN. Despite reports of a deregulated expression of miRNAs and circRNAs linked to the molecular process in diabetes and DN, it is unclear how these molecules interact with c-Jun and c-Fos and how they contribute to the pathophysiology of DN. In this work, we predicted the potential interactions between the circRNAs expressed in renal cells and miRNAs targeting c-Jun and c-Fos. We also discovered that circRNAs, c-Jun, and c-Fos directly interact. We built a potential interaction network between circRNAs expressed in renal cells at high glucose (HG) concentrations (25 mM) with miRNAs and the mRNA of *c-fos* and *c-jun*. We discovered the presence of RNA-potential binding domains in c-Jun and c-Fos. At least one RNA recognition motif was identified in those regions, which can interact with 17 circRNAs expressed in renal cells at HG. Then, we analyzed whether the circRNAs' direct interactions with c-Jun and c-Fos could alter the expression of genes coding for extracellular matrix proteins in renal cells exposed to HG and 50 µg/ml AGEs of lysine.

The findings of this study indicated that c-Jun and c-Fos might be able to bind both DNA and RNA, suggesting that they have a dual role. They will also increase our understanding of the role that circRNAs and miRNAs play in the pathophysiology of DN as well as their prospective applications as biomarkers or therapeutic targets.

POST-TRANSLATIONAL REGULATION OF TISSUE-SPECIFIC BASIC HELIX-LOOP-HELIX TRANSCRIPTION FACTORS THROUGH HETERODIMERIZATION

Kenya Lizbeth Mena-Garcés^{1,2}, Carina Becerril Berrocal¹ and Ana Lilia Torres-Machorro¹

¹Laboratorio de Biología Celular. Departamento de Investigación en Fibrosis Pulmonar. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas. Calz. de Tlalpan 4502, Belisario Domínguez Secc 16, Tlalpan 1480, CDMX.

²Posgrado en Ciencias Bioquímicas. Circuito de Posgrado, Ciudad Universitaria, Alcaldía Coyoacán, C.P. 04510, CDMX, México
E-mail: kenyamengarcs@gmail.com

The basic Helix-Loop-Helix (bHLH) Transcription Factor (TF) superfamily is critical for vertebrate tissue development and maintenance¹. The family is subclassified into multiple classes, some being ubiquitously expressed, whereas others are only expressed in certain tissues. The function of the tissue-specific bHLH TFs is regulated in part through heterodimerization with other bHLH TFs². Our research focuses on the Scleraxis (SCX), Paraxis (TCF15), and TCF21 bHLH TFs, which are essential class II TFs involved in mesoderm development that are usually dysregulated in fibrosing diseases such as pulmonary and cardiac fibrosis³. These factors function by heterodimerizing with E-proteins, like E47.

In our lab, we study the post-translational regulation of these Class II bHLH TFs with the aid of adenoviral transductions. We found that E47 differentially stabilizes the protein levels of Class II bHLH TFs through heterodimerization. We utilized kinetic tests to measure the half-life of SCX, TCF15, and TCF21 and contrasted the data with the stability of the same TFs when E47 was present. The data obtained is important to understand regulation mediated by bHLH TFs.

References

1. Lee DG, Kim YK, Baek KH (2022). *Int J Mol Sci*, 23, 1-18
2. Jolma A, Yin Y, Nitta KR, Dave K, Popov A, Taipale M, et al (2015), *Nature*, 527, 384-388
3. Burgess R, Cserjosi P, Ligon KL, Olson EN (1995). *Developmental Biology*, 168, 296-306

CHARACTERIZATION OF ABC-TYPE TRANSPORTERS IN *CANDIDA GLABRATA*

Mariana Montserrat Morales Avila, Ma. Guadalupe Gutiérrez Escobedo and Alejandro De Las Peñas

Instituto Potosino de Investigación Científica y Tecnológica, División de Biología Molecular. Camino a la Presa San José 2055, Lomas 4ª sección, CP. 78216, San Luis Potosí, SLP., México. Tel. +52 (444) 834 2040.
mariana.morales@ipicyt.edu.mx

Candida glabrata is responsible for more than 25% of invasive clinical candidiasis cases worldwide. It is an opportunistic human pathogen that particularly affects immunosuppressed patients. One of its main virulence factors is its innate and acquired resistance to a wide variety of xenobiotics, including heavy metals, like cadmium (Cd), and antifungals such as azoles. Resistance to azoles, including fluconazole (FLC), is mediated by the transcription factor Pdr1, which in turn induces the expression of ATP-binding cassette (ABC) transporter encoding genes. The purpose of this study is to characterize the ABC-type transporters in *C. glabrata*: Pdr12, Yor1, Ycf1 and Snq2. We determined that the efflux pumps Pdr12, Yor1 and Ycf1 have a minor role in azole resistance in comparison to the main ABC transporter Cdr1. Whereas Snq2 provides fluconazole resistance in the BG14 background. Finally, we confirm that tolerance to Cd is specifically conferred by the vacuolar transporter Ycf1.

STUDY OF PROMOTER ACTIVITY, SUBCELLULAR LOCALIZATION AND OVEREXPRESSION OF ERULUS DURING MUTUALISTIC INTERACTIONS

Rogelio Morales-Sotelo¹, Janet Palacios-Martínez¹, Ana I. Chávez-Martínez¹ and Luis Cárdenas¹

¹Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México. Cuernavaca, Morelos, México.
E-mail: luis.cardenas@ibt.unam.mx

The establishment of mutualistic interactions such as nodulation occurs in several steps, in which there must be an exchange of chemical signal between nitrogen-fixing bacteria and legumes, this exchange of chemical signals activates different signalling pathways that allow the establishment of these interactions. The perception of these chemical signals is carried out in the plant through kinase-like receptors, such as LysM, LRR and malektin like domain. Within the malektin like domains, there is a subfamily called CrRLK, which in *Arabidopsis thaliana* has been reported to be involved in perception and response during plant-microorganism interactions, modulating the maintenance of cell wall integrity, in addition to their participation as key regulators during the cellular expansion of various tissues. CrRLK ERULUS/CAP1 (ERU/CAP1) is a receptor required for the formation of *A. thaliana* root hairs, as well as being shown to be a key regulator of cell wall composition by modulating pectin composition. However, in *Phaseolus vulgaris* the involvement of this receptor during the formation of root hair, as well as in the process of nodulation establishment, has been poorly studied. Because of this, in this paper we focus on the study of ERU/CAP1 in *Phaseolus vulgaris* (PvERU/CAP1) to determine its role during nodulation. Based on our data from the transcript accumulation analysis, as well as the promoter activity during nodulation kinetics and in addition to functional genomics approach, in this work we demonstrate that the member of CrRLK PvERU/CAP1 subfamily participates in the development of the infection threads, as well as in the development of the nodule.

IN SILICO AND CELLULAR DIFFERENCES ASSOCIATED TO THE CELL DIVISION PROCESS BETWEEN THE A AND B RACES OF THE COLONIAL MICROALGA *BOTRYOCOCCUS BRAUNII*

Xochitl Morales de la Cruz¹, Alejandra Mandujano Chávez², Daniel R. Browne^{3,4}, Timothy P. Devarenne³, Lino Sánchez Segura¹, Mercedes G. López⁵ and Edmundo Lozoya Gloria¹

¹Genetic Engineering Department, CINVESTAV-IPN Irapuato Unit, Irapuato 36824, Mexico. ²Instituto Tecnológico y de Estudios Superiores de Monterrey, Campus Irapuato, Irapuato 36670, México. ³Department of Biochemistry and Biophysics, Texas A&M University, College Station, TX 77843, USA. ⁴ Pacific Biosciences, Chicago, IL 60606, USA.

⁵ Biochemistry and Biotechnology Department, CINVESTAV-IPN Irapuato Unit, Irapuato 36824, Mexico.

E-mail: xochitl.morales@inecol.mx; xochitl.morales@cinvestav.mx

Botryococcus braunii produce liquid hydrocarbons able to be processed into combustion engine fuels. Depending on the growing conditions, the cell doubling time can be up to 6 days or more, which is a slow growth rate in comparison with other microalgae. Few studies have analyzed the cell cycle of *B. braunii*. In this work, we compared the sequences of the cell cycle regulators RBR and CDKs from *O. tauri*, a unicellular microalga that reproduces through binary fission; *C. reinhardtii*, an unicellular microalga that reproduces through multiple fission; *C. pectorale*, a colonial microalga that also reproduces through multiple fission; and *V. cartieri nagariensis* (male and female), a multicellular microalga that reproduces through multiple fission, along with the currently available data of the colonial *Botryococcus braunii* race A (Yamanaka strain) and *B. braunii* race B (Showa or Berkeley strain). Previous information about the cell cycle of *B. braunii* assumed a binary fission type of cell division. Differences in the number of cyclin-dependent kinases and potential retinoblastoma phosphorylation sites between the A and B races were found. Some cyclin-dependent kinases from both races seemed to be phylogenetically more similar to *A. thaliana* than to other microalgae. Microscopic observations were done using several staining procedures. Race A colonies, but not race B, showed some multinucleated cells without chlorophyll. An active mitochondrial net was detected in those multinucleated cells, as well as being defined in polyphosphate bodies. There are two mechanisms of multiple fission in microalgae: the grouped one that does not present multinucleated forms, such as *C. reinhardtii*, and the consecutive one in which there are multinucleated cells, such as *Scenedesmus* sp. and *Chromochloris* sp. Our results suggest that *B. braunii* race A colonies may have cells that undergo multiple fission in multinucleated cells.

GENOMIC SEQUENCING OF FOUR PATHOTYPES OF COLLETOTRICHUM LINDEMUTHIANUM AND ANALYSIS OF VIRULENCE GENES

Ma. Irene Morelos-Martínez¹, Horacio Cano-Camacho¹, Karla Morelia Díaz-Tapia¹,
Everardo López-Romero², María Guadalupe Zavala-Páramo¹

¹Centro Multidisciplinario de Estudios en Biotecnología, FMVZ, Universidad Michoacana de San Nicolás de Hidalgo, Km 9.5 Carretera Morelia-Zinapécuaro, Posta Veterinaria, Tarímbaro, Mich., México. ²Departamento de Biología, División de Ciencias Naturales y Exactas, UG, Noria Alta SN, Guanajuato, Gto. México.
E-mail: irene.morelos@umich.mx

Colletotrichum lindemuthianum is a pathogenic fungus of *Phaseolus vulgaris*, that presents 298 pathotypes with different levels of virulence, worldwide. During host infection, it secretes virulence proteins, such as plant cell wall degrading enzymes (PCWDEs), effector proteins, peptidases, etc., which can be affected by genomic rearrangements attributed to transposable elements (TEs). In this work, we analyzed the genes that encode virulence proteins and TEs in genomes of four *C. lindemuthianum* pathotypes. The genomes were sequenced with the Illumina NovaSeq600, assembled and annotated using SPAdes v.315.4 and funannotate respectively, and compared in the MEROPS, fungal TFs, CAZy, and PHI databases. Effector proteins were predicted using EffectorP 3.0 and transposable elements were analyzed with Earl Grey v4.03. Genome sizes ranged from 98.1 to 101.7 Mpb and the number of genes and proteins annotated were from 11,859 to 12,225 and 11,434 to 11,797 respectively. Around 400 peptidase-coding genes were found, with higher serine peptidases content in the four pathotypes. The genes encoding TFs range from 326 to 352, with significant differences in the Zn₂Cys₆ family, that include XlnR, AraR, and ACE2, among others, involved in the transcriptional regulation of CAZymes. About 630 genes were identified for CAZymes, with significant differences in the AA7 and CE10 families that include many PCWDEs. The number of virulence factors and effector genes was from 649 to 711 and 432 to 475, respectively. The content of TEs was highly variable from 70,899 to 78,182 among the four pathotypes. The two pathotypes with the highest level of virulence showed a greater number of genes annotated in the different databases.

References

1. Nunes, M. P. B. A., Gonçalves-Vidigal, M. C., Martins, V. S., Xavier, L. F... & Vidigal Filho, P. S. (2021). *Ciencia de los Cultivos*, 61(6), 3877-3893
2. Baroncelli, R., Amby, D. B., Zapparata, A., Sarrocco, S...& Thon, M. R. (2016). *BMC genómica*, 17, 1-17

AN INTERPLAY BETWEEN HISTONE EXCHANGE AND TRNA EXPRESSION IMPACTS CELLULAR PROTEOSTASIS AND AGING

Ericka Moreno-Méndez^{1,2}, Jimena Meneses-Plascencia¹, Ivón Salazar-Martínez¹, Judith Ulloa-Calzonin¹, Laura N. Balarezo-Cisneros³, Nuria Sanchez-Puig⁴, Daniela Delneri³, Soledad Funes² and Alexander De Luna¹

¹Unidad de Genómica Avanzada, Centro de Investigación y de Estudios Avanzados, 36824 Irapuato, Mexico,

²Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, 04510 Cd.Mx., Mexico, ³Manchester Institute of Biotechnology, The University of Manchester, Manchester M1 7DN, UK, ⁴Instituto Química,

Universidad Nacional Autónoma de México, 04510 Cd.Mx., Mexico
emoreno@ifc.unam.mx; sfunes@ifc.unam.mx; alexander.deluna@cinvestav.mx

Chromatin remodeling stands at the center of the intricate web of factors influencing cellular aging across all eukaryote living organisms. Evidence shows that activity of the highly conserved SWR1C histone-exchange complex underlies aging in *Saccharomyces cerevisiae*, yet the mechanisms of $\Delta swr1$ longevity remain largely elusive. Here, we show that impairment of most SWR1C subunits and the H2A.Z histone variant robustly extends the chronological lifespan of budding yeast. Using high-resolution competitive-aging epistasis screening of over 570 aging factors and non-coding RNAs, we show that $\Delta swr1$ longevity is functionally associated with the cytosol translation machinery and with several transfer RNAs. Moreover, deletions of *SWR1* suppressed the lifespan phenotypes of several tRNA deletions, which was also consistent with increased expression of specific tRNAs. Importantly, the sensitivity to unfolded-protein stress of such tRNA deletions was strongly modified in the $\Delta swr1$ background, suggesting that increased proteostasis and lifespan extension result from altered tRNA pools. Our systems-genetics approach reveals a novel functional link between chromatin remodeling, RNA biology, and cellular proteostasis, with impacts on aging and longevity.

DETECTION OF HUMAN PAPILLOMAVIRUS USING PCR *IN SITU* IN PROSTATIC CANCER

Guadalupe Morquecho Ramos¹, Rodolfo Benjamín Ocadiz-Delgado², Mario García Solís³,
María del Consuelo Gómez García¹, María Olivia Medel Flores¹ and David Guillermo Pérez Ishiwara¹

¹Escuela Nacional de Medicina y Homeopatía (ENMYH) IPN, Av. Guillermo Massieu Helguera 239, La Purísima Ticomán, Gustavo A. Madero, 07320 Ciudad de México, CDMX. ²Departamento de Genética y Biología Molecular; Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (IPN), Av. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, Gustavo A. Madero, 07360 Ciudad de México, CDMX. CP: 55050. ³Hospital General Dr. Darío Fernández Fierro ISSSTE, Av. Revolución 1182, San José Insurgentes, Benito Juárez, Ciudad de México, CDMX, México. CP: 03900.
Email: ishiwaramx@yahoo.com.mx, Tel: 5538993877

Prostate cancer (PCa) continues to be a serious health problem in Mexico with 9.8 deaths per 100 thousand men and is characterized by abnormal cell growth in the prostate gland¹. Risk factors include advanced age, family history and inflammation. Recent studies suggest that persistent infection with High-Risk Human Papillomavirus (HR-HPV) is an important risk factor in the development of PCa². The aim of this study was to determine *in situ* the presence High- and Low-Risk HPV in prostate samples and its correlation with histopathological diagnosis. For this study, 17 paraffin-embedded prostate tissue samples diagnosed with PCa were included. Internal areas of the same samples, showing no histopathological alterations neither HPV presence, were used as internal controls. Samples were subjected to *in situ* Polymerase Chain Reaction (*in situ*-PCR) for HPV detection using specific primers for HPV-LR or HPV-HR². The samples were observed under bright-field microscopy to identify HPV-infected cells and digitally analyzed³. It was found that 88.23% of the samples diagnosed with PCa were HPV positive. Of this infected percentage, 33% were positive for HR-HPV, 27% for LR-HPV. Interestingly, 40% of samples showed coinfection. These results suggest that HPV infection may be related to prostate cancer development. Additional studies, including molecules related to different cellular events such as microRNAs, will be necessary to elucidate the molecular mechanisms related to the malignant progression of HPV-associated prostate cancer.

References

1. García-Perdomo, H. A., Zapata-Copete, J.A., & Sánchez, A. (2018). A comprehensive and updated overview of prostate cancer. *Journal of the Faculty of Medicine*
2. Medel-Flores O, Valenzuela-Rodríguez VA, Ocadiz-Delgado R, Castro-Muñoz LJ, Hernández-Leyva S, Lara-Hernández G, Silva-Escobedo JG, Vidal PG, Sánchez-Monroy V. Association between HPV infection and prostate cancer in a Mexican population. *Genet Mol Biol.* 2018 Oct-Dec;41(4):781-789.
3. Ocadiz-Delgado R, et al. Expression of miR-34a and miR-15b during the progression of cervical cancer in a murine model expressing the HPV16 E7 oncoprotein. *J Physiol Biochem.* 2021 Nov;77(4):547-555.

REGULATION OF THE ANTIFUNGIC ALKYLRESORCINOL LIPID PRODUCTION BY PHOSPHATE IN *AZOTOBACTER VINELANDII*

Andrea Moyao¹, Daniel Segura González², Josefina Guzmán Aparicio³, Soledad Moreno⁴, Guadalupe Espín⁵

Instituto de Biotecnología, UNAM, Avenida Universidad 2001, Chamilpa, 62210 Cuernavaca, Mor.

E-mail: andrea.moyao@ibt.unam.mx

Azotobacter vinelandii is a gram negative bacterium that has two main states during its life cycle; vegetative and cyst. Vegetative cells are metabolically active, ovoid-bacillary and motile while cysts are latent cells where lipid metabolism is very active. During this stage, alkylresorcinols (ARs) and alkylpyrones (APs) replace phospholipids, and represent the 95% of total membrane lipids¹. In *Azotobacter vinelandii*, the operon *arsABCD* encode ARs and APs biosynthetic proteins, and its expression is regulated by ArpR, an activator of *arsABCD* transcription during encystment, and in turn is regulated by the alternative sigma factor RpoS².

It is known that in some bacteria, phosphate limitation induces changes in membrane composition, leading to phospholipid replacement with phosphorus-free lipids. This happens in order to optimize the distribution of the available phosphorus, using it for the synthesis of other cellular components that cannot be replaced, such as nucleotides and nucleic acids³. Here we report that phosphate starvation induces alkylresorcinols production under vegetative growth in *A.vinelandii*. ARs synthesis was proportionally increased while the phosphate concentration was limited. On the other hand, it is well known that the two-component system PhoR/PhoB regulates genes relevant for the response to phosphate starvation. PhoB is the response regulator that controls the expression of its target genes when it recognizes a specific sequence known as Pho box and depending on its phosphorylation state⁴. We found a possible Pho box sequence at the regulatory region of *arsA*, the first gene of the biosynthetic operon, which suggests that PhoB could be involved in the control of ARs production during phosphate limitation through the regulation of *arsABCD* expression.

In this work, through the use of transcriptional fusions in mutants inactivated in *phoB*, *rpoS* and *arpR* we confirmed that the positive effect on ARs production during phosphate limitation requires the participation of PhoB, the transcriptional activator ArpR and the sigma factor RpoS.

References

1. Noar (2018). *Microbiology*, **164**, 421-436.
2. Romero, Moreno, Guzmán, Espín, Segura (2013). *Journal of bacteriology*, **195**, 1834-1844.
3. Sohlenkamp, Geiger (2016). *FEMS Microbiology Reviews*, **40**, 133-159.
4. Lamarche (2008). *FEMS Microbiology*, **32**, 461-473.

MOLECULAR STUDY OF TAU131, SUBUNIT OF TRANSCRIPTION FACTOR TFIIC, IN THE HUMAN PATHOGEN *TRYPANOSOMA BRUCEI*

Leonardo Munguia Aguirre¹, Luis E. Florencio Martínez¹, Rebeca Manning Cela²,
Tomás Nepomuceno Mejía¹ and Santiago Martínez Calvillo¹

¹Facultad de Estudios Superiores Iztacala, UNAM. Av. de los Barrios 1, Col. Los Reyes Iztacala, Tlalnepantla, Edo. de México, CP 54090, México; ²Departamento de Biomedicina Molecular, CINVESTAV-IPN, México.
Email: leomunguia99@gmail.com; scalv@unam.mx

RNA polymerase III (RNAP III) transcription is of fundamental importance in all eukaryotes because its products (that include tRNAs, 5S RNA, and U6 snRNA) are essential for protein synthesis, RNA processing, protein transport, and other cellular processes. To start RNA synthesis, RNAP III requires three transcription factors known as TFIIIA, TFIIIB and TFIIC. In tRNA genes, the internal promoter elements, the A and B boxes, are recognized by TFIIC. In yeast and vertebrates, TFIIC is composed of six subunits that are distributed into subcomplexes TauA and TauB. Subunits Tau131, Tau95 and Tau55 are part of the TauA subcomplex, which associates directly with the A box in tRNA genes. Little is known about RNAP III transcription in the early-diverging eukaryote *Trypanosoma brucei*, the etiological agent of human sleeping sickness. In this work, we initiated the study of the Tau131 subunit in *T. brucei* (TbTau131). *In silico* analyses showed that, despite low sequence conservation, TbTau131 possesses the characteristic tetratricopeptide repeats (TPRs) throughout the protein. Also, the predicted three-dimensional structure of TbTau131 is similar to that of the human orthologue. To assess whether TbTau131 is necessary for the viability of procyclic forms of *T. brucei*, we generated a cell line in which the knockdown of the protein by RNAi could be induced with doxycycline. We are currently analyzing the growth of this cell line. This work was supported by grants IN208224 (PAPIIT, UNAM) and CF-2023-I-820 (CONAHCYT).

PEROXIDASE 35 GENE IS INVOLVED IN LATERAL ROOT PRIMORDIUM MORPHOGENESIS AND IS REGULATED AT THE EPIGENETIC LEVEL

Selene Napsucialy-Mendivil, Janett Ríos-González, Svetlana Shishkova and Joseph G. Dubrovsky*

Depto. de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Av. Universidad 2001, Col. Chamilpa, Cuernavaca, Morelos, Mexico. C.P.62210. Tel. 3291625.
Email: selene.napsucialy@ibt.unam.mx, joseph.dubrovsky@ibt.unam.mx

The Reactive Oxygen Species (ROS) signaling is central to many processes in plant development, and peroxidases, among others, maintain the cellular redox state. The *PEROXIDASE 35* (*PER35*) gene belongs to a classical plant (class III) peroxidase subfamily. It is known that peroxidase activity is essential to promote cell wall remodeling during Lateral Root (LR) primordium (LRP) development¹. We found that this gene was downregulated in a transcriptomic analysis of the root in the *atx1setm* mutant deficient in methyltransferase activity of a histone methyltransferase ARABIDOPSIS HOMOLOG OF TRITHORAX1 (*ATX1*). *ATX1* catalyzes the H3K4 trimethylation to maintain a transcriptionally active chromatin state and controls root growth by regulating cell cycle duration, maintaining stem cell niche, and patterning during primary and LR development². Here, we explored the role of the *PER35* gene in root development. We found that by 8 days after germination (dag), the primary root of the loss-of-function *per35-1* mutant was longer than the Wt (Col-0) root; this data correlated with a longer RAM in the *per35-1* mutant. Although no LRP density changes were found in the *per35-1* mutant, the LRP morphogenesis was abnormal in 8 dag *per35-1* seedlings compared to the Wt. These abnormalities were similar to those found in the *atx1setm* mutant: in both mutants, the LRPs formed had asymmetric and flat-shaped domes. The quantification of the abnormal LRPs after treatment of *per35-1* plants with 2mM H₂O₂ showed an increment of these abnormalities (54% vs 29% in untreated control). Interestingly, a low identity (35-42%) at the amino acid level was found between *PER35* and five other peroxidase proteins previously reported as involved in LR development in *A. thaliana*. This suggests no or low functional redundancy among them and further supports the idea that *PER35* activity is important for normal LRP morphogenesis. Research was supported by DGAPA-PAPIIT-UNAM (IN203024, IN208824) and CONACyT (A1-S-9236).

References

1. Orman-Ligeza B, Parizot B, De Rycke R, Fernandez A, Himschoot E, Van Breusegem F, Bennett MJ, Périlleux C, Beeckman T, Draye X. 2016. *Development*, 136:465.
2. Napsucialy-Mendivil S, Alvarez-Venegas R, Shishkova S, Dubrovsky JG. 2014. *Journal of Experimental Botany*, 65: 6373-6384.

THE FORGOTTEN SIDE OF THE GPN-LOOP GTPASE NPA3: THE CARBOXY-TERMINAL DOMAIN IS A CRITICAL REGULATOR OF THE GTPASE CORE FUNCTION

Manuel de Jesús Ochoa-Valdez^{1,2}, Martín Mora-García¹, Edgar D. Páez-Pérez³, Beatriz E. González-Contreras¹, Yolanda Reboloso-Gómez¹, Lina R. Riego-Ruiz², Mónica R. Calera¹, Roberto Sánchez-Olea¹

¹Instituto de Física, Universidad Autónoma de San Luis Potosí, ²División de Biología Molecular, Instituto Potosino de Investigación Científica, ³Instituto de Metalurgia, Universidad Autónoma de San Luis Potosí, Av. Parque Chapultepec #1570, Privadas del Pedregal, C.P. 78295, San Luis Potosí, San Luis Potosí, México.

Tel: 444-5036-499. Emails: rsanchez@ifisica.uaslp.mx (R. Sánchez-Olea) y mcalera@ifisica.uaslp.mx (M.R. Calera).

In eukaryotes and archaea, a family of GTPases with a highly conserved Glycine-Proline-Asparagine (GPN) motif has been described. The GPN-loop GTPases are critical for the cytosolic assembly and subsequent nuclear import of RNA polymerase II (RNAPII), the enzyme responsible for the synthesis of all messenger RNAs and some non-coding RNAs. Npa3 is the ortholog in yeast of human Gpn1 but only scarce information is available on the cellular function of these proteins. In addition to its GTPase core, Npa3 has a carboxy terminal domain (CTD) which is present in all the eukaryotic orthologs, but absent in the single GPN ortholog in archaea. In our research group, a *npa3ΔC Saccharomyces cerevisiae* yeast strain has been generated. The elimination of Npa3's CTD, the last 106 amino acids, did not affect the stability of the protein and only caused a subtle reduction in proliferation rate in *npa3ΔC* cells. Interestingly, RNAPII nuclear translocation and transcription levels were not affected in *npa3ΔC* cells. However, defects in cellular size, cell cycle progression and microtubule assembly were detected. Recently, it was suggested that Npa3 could be involved in the protein synthesis process, as it is co-expressed with protein synthesis and ribosomal biogenesis genes. Additionally, *TIF11* (eIF1A) rescued the growth of the *npa3ΔC* strain on plates containing hygromycin B. In this study, we evaluated the effect of several mutations in the Npa3 CTD on the sensibility of the corresponding strains growth to translation inhibitors, such as hygromycin B. Our results support the proposal that the CTD is a critical regulator of Npa3 cellular function. This work was supported by Consejo Nacional de Humanidades, Ciencias y Tecnologías (Conahcyt) grant number A1-S-21070 to RSO.

UNVEILING MOLECULAR DIFFERENCES IN 3D CELL CULTURE OF TRIPLE NEGATIVE BREAST CANCER CELLS AND THEIR RESEMBLANCE TO TUMOR TISSUE

Mercedes Olvera-Valencia¹, Rosalío Ramos-Payán², Eloy-Andrés Pérez Yépez³, Alejandro López-Saavedra⁴, Samuel Trujano Camacho^{3,5}, Laurence A. Marchat¹, César López-Camarillo⁶, Carlos Pérez-Plasencia^{3,7}

*Corresponding author: Av. San Fernando 22, Belisario Domínguez Secc 16, Tlalpan, 14080, CDMX, 55 3357 2955, carlos.pplas@gmail.com.

¹Doctorado en Ciencias en Biotecnología. Escuela Nacional de Medicina y Homeopatía, Instituto Politécnico Nacional, CDMX, México. ²Facultad de Ciencias Químico Biológicas, Universidad Autónoma de Sinaloa. ³Laboratorio de Genómica. Subdirección de Investigación Básica. Instituto Nacional de Cancerología, CDMX, México. ⁴Advanced Microscopy Applications Unit (ADMIRA). Instituto Nacional de Cancerología. CDMX, Mexico. ⁵Experimental Biology PhD Program. DCBS, Universidad Autónoma Metropolitana, CDMX, Mexico. ⁶Posgrado en Ciencias Genómicas, Universidad Autónoma de la Ciudad de México, CDMX, Mexico. ⁷Laboratorio de Genómica Funcional, Unidad de Biomedicina, FES-Iztacala, UNAM. Tlalnepanitla, Estado de México. Mexico.

Triple-negative breast cancer (TNBC) is the type of breast cancer with the highest mortality in women worldwide (REF). Its clinical classification arises from the lack of hormone and HER-2 receptors; however, this type of cancer has heterogeneous molecular characteristics that require further study. It is known that the development and progression of TNBC involved epigenetic and genetic alterations, some of which are modulated by lncRNAs. Several cell culture applications allowed the development of cancer research knowledge especially the three-dimensional culture methods. However, these methods still have technical challenges that preclude their wide use as a cancer study model. Here we established a novel organotypic 3D culture using TNBC cell lines. This model is based on the hanging-drop method with substantial modifications. The validation of morphological characteristics of organotypic structures was performed using fluorescent microscopy. Different transcriptomic pattern in cell lines cultured in 2D vs 3D conditions was observed. Specifically, the lncRNA expression profile in 3D structures was shared with triple-negative breast cancer tissues. Further, transcriptomic analysis revealed that both coding and non-coding RNAs in the 3D cultures were more associated with proliferation and invasion. Central signaling pathways implicated in proliferation such as PI3K and WNT/ β -catenin pathways were analyzed. Whereas the PI3K signaling pathway is activated, the β -catenin signaling pathway remains inactive due to b-catenin being located at cell junctions. This contrasts with the constitutive activation for both signaling pathways reported in the same cells cultured in 2D conditions. These results support the idea of organotypic 3D structures as a powerful tool that emulates morphological and genetic characteristics of tumors. This could allow the development of several studies of potential therapeutic targets that remain important clinical challenges in breast cancer therapy.

Acknowledgments: This research was funded by Consejo Nacional de Humanidades Ciencia y Tecnología CONAHCyT (Grant F003 #51207/2020) and the Advanced Microscopy Applications Unit (ADMIRA)

DIFFERENTIAL DISPLAY OF EXPRESSED GENES OF *TRICHODERMA ASPERELLUM* DURING GROWTH ON PET (POLYETHYLENE TEREPHTHALATE)

Adriana Guadalupe Orozco García¹; Andrea Román Delgado¹; Josué Raymundo Solís Pacheco¹; Ayixon Sánchez Reyes²; Alfredo Herrera Estrella³; Edgar Balcázar López¹

¹Universidad de Guadalajara-Centro Universitario de Ciencias Exactas e Ingenierías. Blvd. Gral. Marcelino García Barragán 1421, Olímpica, 44430 Guadalajara, Jal., 33 1378 5900 ext.26 27649; ²Instituto de Biotecnología, UNAM, Av. Universidad 2001, Chamilpa, 62210 Cuernavaca, Morelos; ³Centro de Investigación y de Estudios Avanzados del IPN, Unidad Irapuato (Cinvestav) Libramiento Norte León Km 9.6, 36821 Irapuato, Gto. edkaizerbal@gmail.com

Polyethylene terephthalate (PET) is a versatile and durable plastic used to manufacture water bottles, food packaging, and textiles, becoming an essential material in modern society (2). However, only 10% of plastic products are recycled, 12% are incinerated, and over 70% are discarded into the environment, where some particles can be transported by air and water, contributing to marine pollution (1). Improper disposal of PET has led to a global crisis in environmental impact (2). Nevertheless, biodegradation, especially through the application of enzymes secreted by microorganisms such as hydrolases (cutinases, lipases, esterases, and polyethylene terephthalate hydrolases (PETases and MHETases)), emerges as an effective and sustainable method to control PET generated pollution (4). In our laboratory, we reported that *Trichoderma asperellum* has capability to grow on PET as a sole carbon source, besides *T. asperellum* has a gene that codes for a Tannase enzyme, which has a similar structure to *Ideonella sakaiensis* MHETase (6), crucial enzyme for degrading MHET (the intermediate product) into TPA (Terephthalic acid) and ethylene glycol. However, the underlying molecular mechanisms of how *Trichoderma asperellum* degrades PET are not fully understood. The transcriptomic analysis allowed us to identify gene expression changes associated with PET degradation; the obtained differential genes will help elucidate the mechanism by which *T. asperellum* can utilize PET as a carbon source. Functional analysis of *T. asperellum* transcriptomes under PET conditions will provide valuable information to develop and design strategies to mitigate plastic pollution on the planet, harnessing the application of fungi in plastic bioremediation through the linkage of metabolic networks from genomics and transcriptomic interconnection (3).

References

1. An, R., Liu, C., Wang, J., y Jia, P. (2023). Recent Advances in Degradation of Polymer Plastics by Insects Inhabiting Microorganisms. *Polymers* (20734360), 15(5), 1307. <https://doi.org/10.3390/polym15051307>;
2. Joseph, T., Azat, S., Ahmadi, Z., Jazani, O. M., Esmaeili, A., Kianfar, E., Haponiuk, J. T., & Thomas, S. (2024). Polyethylene terephthalate (PET) recycling: A review. *Case Studies In Chemical And Environmental Engineering*, 100673. <https://doi.org/10.1016/j.cscee.2024.100673>
3. Kumari, A., Bano, N., Bag, S. K., Chaudhary, D. R., y Jha, B. (2021). Transcriptome-Guided insights into plastic degradation by the marine bacterium. *Frontiers in Microbiology*, 12. <https://doi.org/10.3389/fmicb.2021.751571>;
4. Li, X., Zhou, Y., Lu, Z., Shan, R., Sun, D., Li, J., y Li, P. (2023). Switchable enzyme mimics based on self-assembled peptides for polyethylene terephthalate degradation. *Journal of Colloid and Interface Science*, 646, 198-208. <https://doi.org/10.1016/j.jcis.2023.05.017>;
5. Palm Gottfried, Reisky Lukas, Böttcher Dominique, Müller Henrik, Michels Emile, Walczak Miriam, Weber Gert. Structure of the plastic-degrading *Ideonella sakaiensis* MHETase bound to a substrate. *Nature Communications*, 10: 1-10, 2019. DOI: <http://dx.doi.org/10.1038/s41467-019-09326-3>;
6. Pelayo, V. A. (2022). Análisis bioinformático de genes en *Trichoderma spp.* relacionados con la degradación de PET (Tesis profesional). Universidad de Guadalajara, Centro Universitario de Ciencias Exactas e Ingenierías.

STUDY OF SYCP3 AND ITS PERINUCLEAR ACCUMULATIONS IN PRIMARY SPERMATOCYTES DURING THE RAT FIRST SPERMATOGENIC WAVE

Pacheco-Gutierrez Sebastian¹, Echeverría-Martínez Olga M.¹, Torres-Ramírez Nayeli¹, Espinoza-Simón Emilio², Ortiz-Hernández Rosario¹

Cellular division processes are essential for life as we know it. For matters of sexual reproduction, meiosis is a key event that increases genetic diversity through a complex process called recombination. During recombination, homologous chromosomes exchange genetic information. This exchange, in a large number of species, is facilitated by a multiproteic structure called Synaptonemal Complex (SC). The main function of this structure is to bring each pair of homologous chromosomes close enough to allow exchange.

Synaptonemal Complex Protein 3 (SYCP3), is one of the principal components of SC. Studies have shown that alterations in SYCP3 result in alterations of sexual reproduction, such as infertility, pregnancy loss, fetal death or pathologies derived from aneuploidy. Moreover, our team work has demonstrated that, in prepuberal rats (*Rattus norvegicus*) more than in adult rats, SYCP3 generates aggregates, without affecting meiosis. We have named these aggregates as Perinuclear SYCP3 Accumulations.

Surprisingly, even when we can find these accumulations in the four stages of meiotic prophase I, accumulations are more present in pachytene-stage spermatocytes. Till the date, there is no certain explanation for the almost prepuberal exclusive existence of these accumulations, nor a potential function proposed for them.

To increase the understanding of this protein during the four stages of meiotic prophase I in prepuberal rats, we removed the testis from rats in 4 ages. These 4 ages (13, 16, 20 y 27 days) match with the major presence of cell in the 4 stages of meiotic prophase I. Testis were processed for Western blot and Real-Time PCR. We also obtained blood from the rats and tested it with ELISA kits to assess the level of sexual hormones (FSH, LH and testosterone) during meiotic prophase I. Finally, with bioinformatic tools, we attempted to resolve the tertiary and quaternary structure of rat SYCP3 in order to explore its ability to self-assemble.

In our results, we found particular behavior for SYCP3, this protein shows a bigger presence at the age of 27 days. On the other hand, expression of *sycp3* gene shows an homogenous behavior at the ages of 16, 20 and 27 days. The hormonal levels of LH were homogenous, not like testosterone and FSH, that showed a particular behavior. Our *in silico* analysis shows similarity between rat SYCP3 and human SYCP3.

Together, our findings suggest that the formation of Perinuclear SYCP3 Accumulations has no relation with an overproduction of SYCP3 nor with an overexpression of *sycp3*. The level of the sexual hormones seems to not be involved with the formation of accumulations either. Finally, the similarities between our predicted model of rat SYCP3 and the human SYCP3 reported with crystallographic techniques, suggest that they share a common mechanism of self-assembly.

GST, NQO1, AND CC16 POLYMORPHISMS AS POTENTIAL DISEASE SEVERITY BIOMARKERS IN INFLUENZA-RELATED PNEUMONIA

Margarita Isabel Palacios-Arreola¹, Brayan Alexis Jacinto-Arellano¹, Manuel de Jesús Castillejos-López²
and Martha Patricia Sierra-Vargas¹

¹Departamento de Investigación en Toxicología y Medicina Ambiental. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas, Calzada de Tlalpan 4502, Sección XVI, Tlalpan, Ciudad de México, C.P. 14080. ²Oficina de Investigación en Epidemiología Clínica e Infectología. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas, Calzada de Tlalpan 4502, Sección XVI, Tlalpan, Ciudad de México, C.P. 14080
E-mail: margarita.palacios@iner.gob.mx

Introduction. Influenza is a recurrent and highly contagious viral respiratory disease with epidemic and pandemic potential. Disease severity depends on several factors and some people develop a life-threatening pneumonia. There are currently scarce markers able to predict which patients are at risk of developing severe pneumonia.

Hypothesis. During influenza-related pneumonia both oxidative stress and inflammation are produced, so it is possible that genetic polymorphisms in genes coding for antioxidant defense proteins (GST and NQO1) and anti-inflammatory protection (CC16) influence disease progression and severity, thus serving as prognostic markers.

Methods. A descriptive, observational, transversal, retrospective study with data from patients that were admitted to the Instituto Nacional de Enfermedades Respiratorias, diagnosed with influenza-related pneumonia during the 2018-2019 influenza season. Present/null genotypes for GSTM1 and GSTT1 were determined by end-point PCR, while single nucleotide polymorphisms NQO1 C609T and CC16 A38G were determined by PCR-RFLP. Chi square and Cramer's V analysis were performed to assess the association between genotype and disease severity variables (Intensive Care Unit, intubation) and survival.

Results. The A allele of CC16 A38G was correlated with a more severe pneumonia in the studied patients. This might be explained because this allele is associated to a reduced expression of the CC16 protein, which possesses protective anti-inflammatory properties in the lung epithelium [1].

Conclusion. CC16 A38G polymorphism appears as a potential biomarker of disease severity in influenza-related pneumonia. Further studies are needed to validate its prognostic potential by itself or together with other polymorphisms.

References

1. Cheng, D., Di, H., Xue, Z., & Zhen, G. (2015). *Internal Medicine*, 54(2), 155-162.

TRIPARTITE MOTIF CONTAINING 25 IS UPREGULATED IN GLIOBLASTOMA

Eva Guadalupe Palacios-Serrato, Luis Fernando Jacinto-Alemán, Jesús Zepeda-Cervantes,
Marina Macías-Silva, Marcela Sosa-Garrocho and Angeles C. Tecalco-Cruz*

Postgraduate in Genomic Sciences, Autonomous University of Mexico City, Mexico City, 03100, Mexico.

*Correspondence: Angeles C. Tecalco-Cruz, Postgraduate in Genomic Sciences, Autonomous University of Mexico City,
Mexico City, 03100, Mexico.
angeles.tecalco@uacm.edu.mx

Abstract. Tripartite motif containing 25 (TRIM25) is a protein that acts as an E3-ligase for ubiquitin and ISG15 to post-translationally modify proteins through ubiquitination and ISGylation, respectively. In addition, TRIM25 seems to be a transcriptional and post-transcriptional modulator, since it can interact with RNA and DNA. Deregulation of TRIM25 expression has been reported in some cancer types. Here, we analyzed the subcellular distribution, expression, and abundance of TRIM25 in the glioblastoma context. **Methods.** We analyzed the expression and abundance of TRIM25 in glioblastoma cell lines and cancer samples from patients, and the relationship between TRIM25 expression and survival was examined using several databases. We explored the subcellular distribution of TRIM25 through subcellular enrichment and by prediction analysis. We used a tissue microarray containing glioblastoma tissue and normal brain tissue to immunodetect the TRIM25 protein using immunohistochemistry. **Results.** High levels of TRIM25 were detected in glioblastoma tissue compared to normal tissue, and this upregulation of TRIM25 was associated with lower survival rates. Our results also suggest that while the TRIM25 protein has a mainly nuclear distribution, it can also be found in extranuclear compartments. **Conclusions:** A high expression and abundance of TRIM25 may be considered a useful potential biomarker of glioblastoma.

Keywords: TRIM25, E3-ligase for ISGylation, E3-ligase for ubiquitination, glioblastoma.

This work was supported by UACM-CCyT (CCyT-2022-08). EGP-S receives a fellowship from CONAHCYT and is a student at Posgrado en Ciencias Genómicas-UACM

A COMPARATIVE STUDY IN THE REGULOME OF GALLIBACTERIUM ANATIS STRAIN ESV200 AGAINST PASTEURELLACEAE FAMILY AND E. COLI BACTERIA

Nain Gabriela Pedroza Viveros¹, María Patricia Sánchez Alonso¹, María Elena Cobos Justo¹, Erasmo Negrete Abascal², Candelario Vázquez Cruz¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla, C.U., Edificio IC11, 24 Sur y Av. San Claudio, CP. 72570, Puebla Pue. ²Carrera de Biología, FES Iztacala UNAM, Av. de los Barrios no. 1. Iztacala, 54090 Edo. de México.
e-mail nain.pedroza@outlook.com, ecobacilos@yahoo.com

Gallibacterium anatis is an opportunistic pathogen bacterium of poultry. This microorganism affects the respiratory tract and reproductive organs of birds, causing a reduction in egg-laying and increasing mortality. *G. anatis* has several virulence and resistance factors, so vaccination is the best treatment option for gallibacteriosis. However, the antigenic diversity of *G. anatis* leads to constant bacterin actualization because the molecular basis of effective antigens is not entirely known. In previous research, based in *E. coli* heterologous expression of *G. anatis* antigens under the control of native or inducible promoters, the expression of interest proteins has been inconsistent, probably because of differences in transcriptional and post-transcriptional control regulation, as we reported with an *in silico* comparative analysis in the regulome of *Pasteurellaceae* family bird's pathogens. For this work, we perform an *in silico* analysis to determine regulome-associated proteins conserved in *Pasteurellaceae* family bacteria, including *G. anatis*. To make the comparison, we select three protein groups: central metabolic (48), regulatory (150), and conserved with identity >70% (353) associated proteins, taken through alignments with the annotated proteins in the genomes of *E. coli* O157:H7 Sakai, *A. paragallinarum* ESV135 y *G. anatis* ESV200. We got 551 total proteins; 93.29% (514) were present in 3 genomes, and 6.71% (37) only in ESV200. We compared ESV200 proteins (551) with 35 selected *G. anatis* proteomes. The strains shared an identity rate of 94.01%, and in each protein group analysis, we observed less variation in metabolic (5.95%) and conserved (3.63%) groups but significant variation in the regulatory proteins group (16.88%). Also, we analyzed ESV200 variation against genomes of 9 *Pasteurellaceae* family genera, detecting considerable variation in the regulatory group (55.69%), and with *E. coli* genomes, the variation rate rose to 58.60%. This molecular diversity could explain the failures related to *G. anatis* recombinant proteins in *E. coli* heterologous expression.

EXPLORING THE GENETIC BASES OF RETROTRANSPOSON EXPRESSION AND ITS IMPACTS ON CELLULAR AGING

Cesia Daniela Pérez Aguilar^{1,2} and Alexander De Luna¹

¹Unidad de Genómica Avanzada and ²Posgrado en Biología Integrativa,
Centro de Investigación y de Estudios Avanzados, 36824 Irapuato, Mexico
cesia.perez@cinvestav.mx; alexander.deluna@cinvestav.mx

Transposable elements constitute a substantial fraction of eukaryotic genomes, contributing to its diversity and complexity. Activity of these mobile genetic elements are known to impact genome stability and cellular senescence, suggesting that they may also influence aging and longevity. In the budding yeast *Saccharomyces cerevisiae*, Ty1 are long terminal repeat retrotransposons which replicate via an RNA intermediate; Ty1 transposition leads to genetic diversity and potentially affects gene expression. Here, we aim to understand the effects on Ty1 expression on the chronological lifespan of yeast. We will control Ty1 expression by modulating the Tec1 transcription factor, which is known to be the main regulator of Ty1 expression in yeast. In addition, we will describe the way in which Ty1 expression affects the expression of neighbouring genes, specifically genes encoding transfer RNAs. Based on previous studies in our group, we hypothesize that altered tRNA pools resulting from changes in Ty1 expression drive a proteostasis response, leading to enhanced or decreased chronological longevity. Our study will provide novel mechanistic on the possible broad-scale effects of retrotransposon expression on aging yeast cells, shedding light on the genetic logic of aging in other organisms.

THE ROLE OF *ARABIDOPSIS* sRNA2 AND sRNA3 AND THEIR POTENTIAL TARGET *AT_INCRNA1* IN ESTABLISHING A MUTUALISTIC RELATIONSHIP WITH *TRICHODERMA*

Kumari Rashmi¹, Maria Montserrat Rosendo-Vargas¹ and Sergio Casas-Flores¹

Instituto Potosino de Investigación Científica y Tecnológica (IPICYT)
Camino a la Presa San José #2055 Col. Lomas 4ta. SLP 78216
Email: scasas@ipicyt.edu.mx

Small RNAs (sRNAs) are regulatory non-coding molecules that regulate gene expression by mRNA degradation, or even interfering with translation. In plants, sRNAs regulate disease resistance against phytopathogenic microorganisms and play key roles in symbiotic association with beneficial microorganisms. Fungi belonging to the *Trichoderma* genus induce beneficial effects in their host plants, including growth promotion and induction of plant defense resistance. A previous study by our group revealed that *Trichoderma atroviride* induces the accumulation of sRNAs in *Arabidopsis thaliana*. However, if some of these *Trichoderma*-induced sRNAs regulate gene expression in the host, remains to be investigated. In this work, two sRNAs of 24 nucleotides in length were differentially accumulated in *Arabidopsis* in response to *T. atroviride* and were named *At_sRNA2* and *At_sRNA3* and selected for further analysis. Stem-loop RT-qPCR experiments confirmed that both sRNAs are induced by *T. atroviride* at 24, 48, 72, and 96 h of co-culture. *At_sRNA2* and *At_sRNA3* were predicted to target a subset of *Arabidopsis* genes involved in several biological processes. Among these targets, both sRNAs potentially regulate the *Arabidopsis long noncoding RNA1* encoding gene (*At_IncRNA1*). RT-qPCR experiments indicated that *At_IncRNA1* is downregulated in *Arabidopsis* during several time points of interaction with *T. atroviride*. Phenotypic screening of a T-DNA insertion mutant of *IncRNA1* showed that *At_IncRNA1* is required in *Arabidopsis* for growth stimulation by beneficial microorganisms, including *T. atroviride*, *Pseudomonas stutzeri*, and *Azospirillum brasilense*. In addition, the *At_IncRNA1* mutant exhibited enhanced susceptibility to *B. cinerea*, which suggests that the *At_IncRNA1* positively regulates plant immunity against necrotrophic pathogens. Taken together, our results provide evidence that *Trichoderma* regulates plant growth and defense against phytopathogens by inducing the accumulation of sRNAs in their host involved in the regulation of plant growth- and defense-related genes.

Keywords: Small RNAs; symbiotic association; *Trichoderma*; *Arabidopsis*; long non coding RNA, systemic resistance, priming

EVOLVING THE THEORY OF HYPERMOBILE EHLERS-DANLOS SYNDROME FROM CONNECTIVE TISSUE DISEASE TO A MECHANOSENSING PATHOLOGY

Ishar Arad Retana-Arellano¹, Lourdes Vadillo-Pérez^{1,2}, Patricia Grether-González³, Roberto Richeimer-Wohlmuth⁴, Ian Ilizaliturri Flores⁵, Alma Lilia Hernández-Olvera¹, Elahe Mirzaeichesmeh⁶, Felipe Vadillo-Ortega^{1*}

¹Unidad de Vinculación de la Facultad de Medicina UNAM, Instituto Nacional de Medicina Genómica, ²Programa de Doctorado en Ciencias Biomédicas UNAM, ³Laboratorio Diagen S.C., ⁴Grupo Pediátrico de Santa Fe, S.C. Centro Médico ABC, ⁵Unidad Profesional Interdisciplinaria de Ingeniería Campus Hidalgo, IPN ⁶Instituto Nacional de Medicina Genómica. Periférico Sur 4809, Arenal Tepepan, 14610 Mexico City, CDMX.

*felipe.vadillo@gmail.com

Hypermobile Ehlers-Danlos Syndrome (hEDS) appears as an array of clinical phenotypes, with no correlation between gene mutations and the corresponding clinical manifestations. We studied a patient with hEDS carrying a variant of alpha-1 type XII collagen gene (COL12A1) that demonstrated structural and functional repercussions when analyzed by *in silico* molecular dynamics (MSD). Specifically a single mutation, c.6718G>A p.(Ala2240Thr), in the COL12A1 gene, inherited from the patient's father, disrupts the final FN3 region located at the carboxyl-terminal of the NC3 globular domain of the protein. This results in conformational and mechanical modifications in the NC3 domain's structure, as predicted by MSD. Consequently the presence of this protein variant is associated with major alterations in the histological structure of the skin extracellular matrix (ECM). Additionally, two staining techniques with high specificity and sensitivity for collagen revealed different spatial arrangements of collagen-containing fibers, appearing as irregular and loosely dispersed fibers in the dermis of hEDS patient. A differential transcriptional response of genes related with mechanosensing response in skin-derived fibroblasts, including integrins and ECM proteins, was found. This response correlated with a differential pattern of methylation in most of these genes. Overall, an area of interest in the understanding of the physiopathology of EDS is how single mutations in a scarce extracellular matrix protein, such as COL12A1, may result in pleiotropic clinical phenotypes involving severe multisystemic manifestations beyond musculoskeletal components, as those present in the patient we studied. Collectively, our findings suggest that the modification in the structure of the variant protein may explain changes in assembling of the ECM, resulting in differential mechanosensing by local cells and further altered epigenetically-mediated expression of other ECM proteins, which may add further changes in ECM composition/structure as a part the mechanisms of the disease.

IDENTIFICATION OF DIFFERENTIAL EXPRESSED MICRORNAS IN GASTRIC CARCINOGENESIS

Diego Reyna Colín^{1,2}, Mauricio Flores-Fortis¹, Isidro X. Perez-Añorve¹, Carlos C. Patiño-Morales³, Gabriela Delgado⁴, Rosario Morales Espinosa⁴, Elena Aréchaga Ocampo¹

¹Departamento de Ciencias Naturales, ²Licenciatura en Biología Molecular; Universidad Autónoma Metropolitana, Unidad Cuajimalpa, C.P. 05348, CDMX. ³Hospital Infantil de México Federico Gómez, C.P. 06720, CDMX.

⁴ Facultad de Medicina, UNAM, C.P. 04360, CDMX.

Correo: earechaga@cua.uam.mx

Gastric cancer is currently one of the most common tumour types worldwide with a poor prognosis and a low survival rate in advanced states. Gastric adenocarcinoma results from a series of preneoplastic lesions ranging from chronic gastritis, atrophic gastritis, intestinal metaplasia, and dysplasia. Several studies have reported the deregulated expression of microRNAs (miRNAs) in gastric carcinogenesis; therefore, they are proposed as a tool for the identification of patients in early stages of gastric cancer. The aim of this work was to identify progressively deregulated miRNAs throughout the preneoplastic lesions until tumour tissues. miRNAs expression profile in gastric cancer tissues relative to normal gastric tissues was identified from TCGA data bases. Differentially expressed miRNAs (DEMs) were evaluated in preneoplastic lesions. Ten up-regulated miRNAs and five down-regulated miRNAs were identified in the progression to premalignant lesion to adenocarcinoma. To determinates the clinical predictive potential, a ROC curves were performed for up- and down-regulated DEMs, obtaining a mean diagnostic value for both groups. Likewise, ten miRNAs (miR-187, miR-490, miR-9-3, miR-188, miR-194-1, miR-194-2, miR-196, miR-200, miR-429, miR-96) were associated with prognostic to overall survival of gastric cancer patients. To examine the value of *Helicobacter pylori* infection in the deregulation of miRNAs, the expression of the 10 miRNAs associates to gastric carcinogenesis were evaluated in preneoplastic lesions and tumor tissues positive or negative to bacterial infection. miR-196 was differentially expressed after *Helicobacter pylori* infection in the different premalignant lesions and tumors. Therefore, given that *Helicobacter pylori* infection modulates miR-196 expression, it is inferred that this miRNA could be used as a distinctive marker of *Helicobacter pylori* infection, while the set of 10 DEMs could play a key role in predicting the development of gastric cancer from preneoplastic lesions and the prognostic of the cancer gastric patients.

This work was supported by Molecular Biology Career, and the Department of Natural Science of Autonomous Metropolitan University Cuajimalpa (UAM-C).

LONG NON-CODING RNA LINC00115 IN RESPONSE TO 5-FU PHARMACOLOGICAL TREATMENT IN COLORECTAL CANCER

Rodríguez-Aboy Lil, García Castillo Verónica, López-Urrutia Eduardo y Pérez-Plasencia Carlos

Facultad de Estudios Superiores Iztacala, UNAM, Av. De los barrios 1, Los Reyes Iztacala, Hab Los Reyes Iztacala Barrio de los Árboles/ Barrio de los Héroes, 54090, Tlalnepantla de Baz, México
lilrodriguezaboy@gmail.com and carlos.pplas@unam.mx

Colorectal cancer (CRC) is the third most common cancer in the world. The standard treatment of CRC consists of surgical resection and adjuvant therapies such as radiotherapy and chemotherapy. 5-fluorouracil (5-FU), a frequent chemotherapeutic agent, is an anti-metabolite that inhibits thymidylate synthase (TS). Despite advances in therapeutic methods for CRC treatment, the prognosis and overall survival rate are unfavorable due to chemoresistance. Thus, it is currently necessary to understand the molecular elements and processes related to chemoresistance. Non-coding RNAs (ncRNAs) play a predominant role among these regulatory elements. Within them, our research group is particularly interested in long non-coding RNAs (lncRNA) associated with cancer development and maintenance. LINC00115 lncRNA is significantly overexpressed in CRC, it promotes tumor progression by targeting miR-489-3p via the PI3/AKT/mTOR pathway.

This work aimed to evaluate the pharmacological response to 5-FU in CRC-derived cell lines expressing LINC00115 differentially. First, the 5-FU inhibitory concentration 50 (IC₅₀) was determined in the HCT116 and RKO cell lines. Next, LINC00115 was underexpressed through two different methods: Short hairpin RNAs (shRNAs) and Antisense Oligonucleotides (ASOs). The results confirmed that LINC00115 is overexpressed in HCT116 and RKO lines relative to non-tumoral cells; however, it was not possible to achieve silencing by any of the methods used. Unexpectedly, housekeeping genes such as β -actin and GAPDH, commonly used for data normalization in qRT-PCR experiments, presented altered expression patterns in our experiments. This suggests that modifying the expression levels of LINC00115 possibly affected the expression patterns of β -actin and GAPDH, which calls for an alternative measurement approach to study the role of LINC00115 in CRC.

TFIIIC SUBUNIT TAU131 IN *TRYPANOSOMA BRUCEI*: IDENTIFICATION OF ITS PROTEIN INTERACTORS

Mariani Guadalupe Rodríguez López¹, Luis E. Florencio Martínez¹, Rebeca Manning Cela², Tomás Nepomuceno Mejía¹ and Santiago Martínez Calvillo¹

¹Facultad de Estudios Superiores Iztacala, UNAM. Av. de los Barrios 1, Col. Los Reyes Iztacala, Tlalnepantla, Edo. de México, CP 54090, México; ²Departamento de Biomedicina Molecular, CINVESTAV-IPN, México.
E-mail: marianigr1412@gmail.com, scalv@unam.mx

Trypanosoma brucei is the protozoan parasite that causes sleeping sickness in Sub-Saharan Africa. In addition to its medical relevance, *T. brucei* is important in the molecular biology field because it exhibits atypical mechanisms of gene expression, including polycistronic transcription and trans-splicing. The knowledge about RNA polymerase III (Pol III) transcription in this pathogen is scarce. Pol III transcribes 5S rRNA, tRNAs, U6 snRNA and other small non-coding RNAs that play critical roles in all organisms. To start RNA synthesis, Pol III needs transcription factors TFIIIA, TFIIIB and TFIIIC. In yeast and vertebrates, TFIIIC is composed of six subunits. One of these subunits is Tau131, which is required for the recruitment of TFIIIB and Pol III to the promoter regions to initiate transcription. In our group, we have recently found that TFIIIC is composed of only four subunits in *T. brucei*, including Tau131 (TbTau131). To characterize this protein, we have produced a *T. brucei* transgenic line where TbTau131 was tagged with a carboxy-terminal PTP tag. The growth of this cell line was very similar to that of the wild-type culture. With the transgenic line, we have performed tandem affinity purification experiments to isolate the proteins that interact with TbTau131. These proteins were sent to mass spectrometry analysis to determine their identity. The subcellular location of TbTau131 is also currently being studied in the transgenic cell line. This work was supported by grants IN208224 (PAPIIT, UNAM) and CF-2023-I-820 (CONAHCYT).

EVOLUTION OF THE EARLY RECOMBINOSOME IN PLANTS

Nahid Rafiei¹, Janet Palacios-Martínez¹, Patricia Rueda¹ and Arnaud Ronceret¹

¹Department of Plant Molecular Biology, Instituto de Biotecnología (IBT), Universidad Nacional Autónoma de México (UNAM), Cuernavaca, Morelos, México.

E-mail: arnaud.ronceret@ibt.unam.mx

During meiosis, replicated chromosomes are reshuffled at various crossing-over (CO) sites. The creation of CO starts with the formation of DNA double strand breaks (DSBs) during early prophase I^{1,2}. DNA DSBs are created by the enzymatic activity of a topoisomerase-like complex containing SPO11 known as the early recombinosome³. In plants, two SPO11 proteins SPO11-1 and SPO11-2 form a heterodimer also associate with additional factors called MTOPVIB, PUTATIVE RECOMBINATION DEFECT 1 (PRD1), PRD2, PRD3, DSB FORMING (DFO)⁴. All these factors are individually required for the formation of DSB formation in Arabidopsis and rice. Some of them are demonstrated to be also required in maize, barley and wheat. The SPO11 complex was described to form three sub-complexes. The core complex contains the SPO11-1/SPO11-2/MTOPVIB that conserve the conformation of a topoisomerase type VI⁴. The PRD1 protein allow to link the different subcomplexes together. PHS1, PRD2, DFO, PRD3 can contain unstructured domains. We analyze the evolution of these proteins in plants. The core complex is more conserved than the additional subcomplexes. Several of these proteins contain unstructured regions. We explore the modeling of these early recombinosome proteins through Alfold2. We propose a 'multi-key lock' model for each subunit of the early recombinosome complex to secure the formation of risky but necessary meiotic DSBs. We will also discuss the hypothetical implications that the established topoisomerase-like nature of the SPO11 complex can have in creating DSB in only one of the two replicated chromatids of the early prophase I meiotic chromosomes. This hypothetical 'one per pair of chromatids' DSB formation model could optimize the faithful repair of the self-inflicted DSBs. Each DSB could use three potential intact homologous DNA sequences as repair template: one from the sister chromatid and the two others from the homologous chromosome.

References

1. Rafiei N and Ronceret A (2023). *Frontiers in Cell and Developmental Biology* **11**, 1-13.
2. Gutiérrez PY, González JK, Rueda P, Ronceret A (2021) *Frontiers in Plant Science* **12**, 717423.
3. Ku JC, Ronceret A, et al. (2020) *PLoS Genetics* **16** (4), e1007881.
4. Vrielynck N, et al. (2021). *Nucleic Acid Research*, **49**,9821-9835.

EPIGENETIC CHANGES IN THE GAPDH GENE DURING HYPOXIA AND REOXYGENATION STRESS IN *PENAEUS VANNAMEI*

Jesús Adelmo Rosas Anaya¹, Silvia Gómez Jiménez¹, Alma Beatriz Peregrino Uriarte¹,
Gloria Martina Yepiz Plascencia¹

¹Centro de Investigación en Alimentación y Desarrollo, A.C.
E-mail: adelmorosas190995@gmail.com

Epigenetics regulates genome activity by altering gene expression, without changing the underlying nucleotide sequence, allowing organisms to respond to environmental changes. Currently, one of the most serious environmental changes is global warming, which increases ocean temperatures and decreases dissolved oxygen, causing hypoxia, a stressor for marine organisms and aquaculture. One of the most important species in aquaculture is the shrimp *Penaeus vannamei*, that uses physiological, cellular and biochemical strategies during hypoxia. In this crustacean, the expression of glyceraldehyde 3-phosphate dehydrogenase (GAPDH), an enzyme of glycolytic metabolism, is altered by oxygenation levels. Therefore, it is possible that an epigenetic change of DNA methylation in the genome and the promoter of this gene could contribute to regulate its expression. This work aimed to obtain and characterize the GAPDH promoter, quantify its gene expression and evaluate the global methylation of gill genomic DNA under hypoxic and reoxygenation conditions. First, three ~1000 bp fragments of the GAPDH promoter were analysed for putative binding sites for transcription factors HIF-1 α , p53, and CpG islands, obtained by PCR, and sequenced. Shrimp were exposed to hypoxia and reoxygenation, global DNA methylation was evaluated by ELISA and RT-qPCR was used to quantify gene expression. Using bioinformatics consensus sequences for the binding of HIF-1 α , p53, TBP and five potentially methylated CpG islands in the GAPDH promoter were found. Hypoxia and reoxygenation increased ($p < 0.05$) the expression of GAPDH at 6, 12 and 24 h compared to the normoxia 0 h control, but reoxygenation decreased ($p < 0.05$) the expression at 6 and 12 h compared to the 0 h normoxia control. Global methylation increased in response to 12-h hypoxia and during 24-h reoxygenation, compared to 0-h normoxia ($p < 0.05$). The putative sites for HIF-1 α , p53, TBPs identified and the CpG islands in the promoter suggest a complex regulation of the GAPDH gene responding to changes in oxygenation levels by induction under hypoxic and reoxygenating conditions. The changes in global methylation suggest that this gene and possibly others, are epigenetically regulated since hypoxia and reoxygenation increase methylation indicating transcriptional repression under hypoxic conditions.

RS2070935, RS2289671 AND RS17027 VARIANTS OF GFAP AND THEIR ASSOCIATION WITH NEUROLOGICAL SEQUELAE OF COVID-19

Alma Cristina Salas-Leal¹, Laura Guadalupe Stopani Quiñones¹, Francisco Xavier Castellanos-Juárez¹, Ada Sandoval-Carrillo¹, Lilia Martina Velez-Velez¹, Osmel La Llave León¹ and José Manuel Salas Pacheco¹

¹Institute of Scientific Research, Universidad Juárez del Estado de Durango. Av. Universidad esq. con Volantín. Col. Centro CP 34000. Tel: (618) 111-58-72
E-mail: alma_sleal@hotmail.com

Introduction. Since December 2019, COVID-19 disease has rapidly spread worldwide. Alongside respiratory and cardiovascular symptoms, the disease has been observed to impact the central and peripheral nervous system. Evidence suggests that COVID-19 can lead to neurological sequelae including cognitive impairment, anxiety, and depression¹. Glial fibrillary acidic protein (GFAP) encoded by the *GFAP* gene, is a glial cytoskeletal protein. Studies have shown that elevated levels of GFAP are associated with neurological damage in patients with COVID-19². However, gene variants in *GFAP*, such as rs2070935, rs2289671 and rs17027 have not been thoroughly investigated in relation to susceptibility to neurological sequelae of COVID-19.

Objective. To determine the association of the rs2070935, rs2289671 and rs17027 variants of the *GFAP* gene with neurological sequelae of COVID-19. **Materials and Methods.** A cross-sectional, analytical, observational study was performed involving 382 individuals who had recovered from COVID-19. Cognitive status, anxiety, and depression were assessed in participants. Whole blood was used for DNA extraction and genotyping of variants was performed by real-time PCR using TaqMan assays. **Results.** We observed a prevalence of 68.3% of cognitive impairment, 40.8% of anxiety, and 53.1% of depression. Patients with cognitive impairment were found to be older compared to those without cognitive impairment ($p=0.001$). Regarding the association of *GFAP* gene variants with neurological sequelae, TT genotype of rs2070935 was associated with a lower risk of cognitive impairment (OR=0.49, 95% CI=0.26-0.94, $p=0.033$). No significant associations were found with the rs2289671 and rs17027 polymorphisms. **Conclusions.** The prevalence of post-COVID-19 cognitive impairment is high, followed by anxiety and depression. The rs2289671 and rs17027 polymorphisms showed no significant association with the risk of neurological sequelae. The TT genotype of the rs2070935 polymorphism was associated with a lower risk of cognitive impairment, suggesting a possible protective effect.

References

1. Taquet, M., Geddes, J. R., Husain, M., Luciano, S., & Harrison, P. J. (2021). 6-month neurological and psychiatric outcomes in 236379 survivors of COVID-19: a retrospective cohort study using electronic health records. *The Lancet. Psychiatry*, 8(5), 416–427
2. Sahin, B. E., Celikbilek, A., Kocak, Y., Saltoglu, G. T., Konar, N. M., & Hizmali, L. (2022). Plasma biomarkers of brain injury in COVID-19 patients with neurological symptoms. *Journal of the neurological sciences*, 439, 120324.

MODULATION OF THE KILLER EFFECT BY REPLACING K⁺ WITH NA⁺ IN THE CULTURE MEDIUM

Salinas-Celestino J.¹, Morales-Tlalpan V.^{1,2}, Saldaña C.^{1,2*}

¹Laboratorio de Biofísica de Membranas y Nanotecnología. ²Laboratorio Nacional de Visualización Científica avanzada (FCN-UAQ). Facultad de Ciencias Naturales, Universidad autónoma de Querétaro. Juriquilla, Qro., México. CP 76230. Phone + 52 4421921200 x65332. *carlos.saldana@uaq.mx

Saccharomyces cerevisiae, is a model of scientific study, it is also used: food industry (bread, beer, wine and dairy products), pharmaceutical industry (antibiotics, cosmetics and drug design, food supplements and bioethanol production). In the wine-related industry it has been used in the fertilization and protection of crops and wine production; strains with specific characteristics are selected and used, among which the Killer phenotype stands out. It has been observed that the effect of the strain is capable of secreting toxic proteins into the environment and producing death in sensitive cells. Different types of strains with similar characteristic have been described, although in this work we will study the K1 (Killer) toxin. K1 is produced by the concatenated expression of two extrachromosomal elements: viruses (M1 and HA), both double-stranded RNA; and recently genomic sequences have been identified which can modulate their expression. According to the literature, the efficiency of the toxin depends on several parameters: temperature, pH, the biomolecules present and the nature and quantity of the sensitive microorganism. Now, we determine how the environment modifies the expression of the Killer phenotype. To achieve this, we use microbiological and molecular biology techniques. We carried out experiments to corroborate the effect of temperature and pH, as expected, we found that at 28°C strain 42300 (producer: MAT α Ade2/+Thr1/+sKi2-1/+ [KILK1]) and strain 5X47 (sensitive; 5x47 MAT α /MAT α his1/+ trp1/+ +/ura3 [KIL-o]), in YDPagar pH 4.7 they generate zones of growth inhibition; these phenomena are negatively altered when the temperature increases to 35°C. On the other hand, when we change the conditions of the culture medium (replacing K⁺ with Na⁺), the enhanced Killer effect is observed. Strongly suggesting that salt concentration also affects overexpression. We are performing growth curves, isolation of mRNA and genomic DNA to determine how the salt change is modulating the K1 phenomenon.

Acknowledgments. FONDEC-UAQ, FONFIVE 2024 and CONAHCyT AI-S-26966

ROLE OF THE ONCOGENE BORIS (*CTCF*L) IN TRANSCRIPTIONAL REGULATION OF THE *VEGFA* GENE PROMOTER

Ana Sofía Salmerón Oms, Aylin del Moral Morales and Ernesto Soto-Reyes*

Universidad Autónoma Metropolitana, Unidad Cuajimalpa, Departamento de Ciencias Naturales e Ingeniería, CDMX, C.P. 05348, 55 5558146500 ext. 3879.

*esotoreyes@cua.uam.mx

Cancer is one of the leading causes of death worldwide. This disease is defined as a group of diseases characterized by defects in cell proliferation and cell death. The molecular signatures of cancer, also known as the hallmarks of cancer, refer to a set of fundamental characteristics that are common in most types of cancer. Among these characteristics is angiogenesis, which is the stimulation of new blood vessel formation, providing the tumor with the nutrients and oxygen necessary for its growth and cellular survival, a process primarily mediated by vascular endothelial growth factor A (*VEGFA*). The transcriptional regulation of this gene is mainly mediated by hypoxia-inducible factor 1 alpha (*HIF-1 α*), where under hypoxic conditions, stabilization of *HIF-1 α* leads to its activation. Recently, our research group has proposed that the oncogene Brother of the Regulator of Imprinted Sites (*BORIS*), encoded by the *CTCF*L (*CTCF like*) gene, participates in the transcriptional regulation of *VEGFA*. *BORIS* has garnered attention because this gene has been reported to be overexpressed in multiple cancers and associated with poor prognosis and lower survival rates. Here we demonstrate that in a cellular model with *BORIS* overexpression, there is also increased expression of *VEGFA*, suggesting that *BORIS* may be involved in the transcriptional regulation of this gene. To demonstrate the participation of *BORIS*, a chromatin immunoprecipitation assay (ChIP) was performed against *BORIS*. We observed that *BORIS* binds to the *VEGFA* gene promoter, suggesting its potential involvement in transcriptional regulation. However, the precise mechanism by which *BORIS* could participate in the transcriptional regulation of *VEGFA* has not been fully addressed. Therefore, elucidating the molecular mechanism underlying the activation of this gene via *BORIS* could contribute to the understanding of *VEGFA* transcriptional regulation in cancer. These findings could provide new insights into cancer molecular mechanisms and reveal novel therapeutic strategies for cancer treatment.

TRANSCRIPTIONAL EVALUATION OF KEY GENES INVOLVED IN THE TISSUE REGENERATION PROCESS OF *AMBYSTOMA MEXICANUM*

Cynthia Sámano Salazar^{1*}, Jossephlyn Hernández Alcántara², Aylin del Moral Morales¹, José Antonio Ocampo Cervantes³, Ernesto Soto-Reyes^{1*}

¹Depto. de Ciencias Naturales e Ingeniería, Universidad Autónoma Metropolitana, Unidad Cuajimalpa; ²Posgrado en Ciencias Naturales e Ingeniería, Universidad Autónoma Metropolitana, Unidad Cuajimalpa; ³Centro de Investigaciones Biológicas y Acuícolas de Cuemanco (CIBAC), Universidad Autónoma Metropolitana, Unidad Xochimilco
*csamano@cua.uam.mx; * esotoreyes@cua.uam.mx

The Mexican axolotl (*Ambystoma mexicanum*) is a vertebrate amphibian endemic to the lake region of Mexico City. *A. mexicanum* has served as an important model organism in scientific research due to its remarkable ability to regenerate damaged tissues, limbs, and nearly any part of its body. Limb regeneration occurs through the initial formation of a mass of undifferentiated cells capable of growth and differentiation into specific tissues or organs, known as the blastema. Transcriptomic profiling of the blastema pinpointed key genes involved in the limb regeneration process. Recently, our research group has identified a group of genes (*ADAMTS17*, *FSTL1*, *GPX7*, and *CTHRC1*) with higher expression in regenerating tissue of 8-month-old juveniles compared to aged axolotls of 8-year-old individuals (that showed no signs of regeneration 10 days after amputation). *ADAMTS17* and *FSTL1* contribute to skeletal growth by encoding a matrix metalloproteinase and a glycoprotein, respectively, both of which are associated with extracellular matrix elements. *GPX7*, and *CTHRC1* are involved in epidermal wound closure. However, the underlying molecular mechanisms of transcriptional regulation are not yet fully understood. In this project we focused on the expression patterns of *ADAMTS17*, *FSTL1*, *GPX7*, and *CTHRC1* during limb regeneration in *A. mexicanum*. Here, we found in juvenile limb but not in aged organisms the genes *ADAMTS17*, *FSTL1*, *GPX7*, and *CTHRC1* are highly expressed. However, following limb amputation, *Adamts17* exhibited high expression in the blastema at 10 days post-amputation, with expression shifting at 20 days post-amputation. In contrast, *FSTL1*, *GPX7*, and *CTHRC1* showed lower expression levels in the blastema at 10 days post-amputation, with no significant change at 20 days post-amputation. These findings suggest that *ADAMTS17*, *FSTL1*, *GPX7*, and *CTHRC1* are key elements during the limb regeneration process of *A. mexicanum*. This approach provides a significant contribution to the research on tissue regeneration in *A. mexicanum*, addressing the limited understanding of the involvement of transcriptional mechanisms in this process.

The project was approved by the Comité de Ética en Investigación (CEI), UAM Xochimilco (CEI.2023.007), by the Consejo Nacional de Ciencias Humanidades y Tecnología (CONAHCyT, CB-SEP-CONACyT 284748 and CF-2023-G-1558), approved by the Consejo Divisional de Ciencias Naturales e Ingeniería (DCNI-10-240-22) and by the Rectoría de la Unidad Cuajimalpa (RC.153.2022).

TELOMERE AND SUBTELOMERE ORGANIZATION IN *USTILAGO MAYDIS* CHROMOSOMES

Adriana Inclán Popoca, Cintya Atonal Águila, Juan Antonio Sanpedro Luna, Helena Cobos Justo,
Candelario Vázquez Cruz, María Patricia Sánchez Alonso

Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla
E-mail: maria.sanchez@correo.buap.mx

Abstract. Telomere DNA is composed of a tandem of repeated DNA motifs with high G+C content, with the TTAGGG hexanucleotide as the most representative repeated unit among several distant taxonomic groups. These DNA, along with specific telomere-binding proteins, build telomeres as a seal that protects the chromosome termini from the DNA damage response machinery, avoiding their recognition as DSB DNA and hindering resection and fusion events. Average telomere length is species-specific and chromosome-end-specific in yeast and humans, maintaining its differences in length and distribution as telomeres shorten. Adjacent to them, telomere-associated sequences (TAS) are arranged in a mosaic of interspersed, species-specific middle-repeated sequences composed of polymorphic segmental duplications. Telomere elongation is kept by telomerase and, to a lesser extent, by recombinational events; TAS seems to be maintained by conventional replication and recombinational events and by not yet fully elucidated mechanisms.

Here, we present the results of the chromosome-end analysis from the genome sequences of three *U. maydis* strains sequenced by the PacBio platform. Large sequence reads generated a better depiction of the organization of the telomere and subtelomeric domains in *U. maydis* chromosomes.

Results. The previously reported *UTASa* and *UTASb* (*Ustilago's* TAS type a or b) have a conserved orientation of 5' to 3' OH toward the chromosome end, either as a whole genetic element or as pieces of truncated versions of them. *UTASa* harbored the telomere-linked helicase (TLH) *USHER*, which has a RecQ-like domain, this element can be absent or present in one or up to two copies on each chromosome end. No clear conserved promoter was found for transcription of the *USHER* helicase or TERRA-like lncRNAs. *UTASb* is less conserved and often is composed of divergent piecework DNA harboring less conserved ORF fragments that contain a 5' OrsD domain with two ZNFs. *UTASb* is mainly located at the utmost *CEN* proximal side of the TAS. Although the GIY-YIG-type homing endonuclease previously reported in the 521 strain is absent in the WT PGA2.1 strain, which harbors few copies of *UTASa*, we agree on the hypothesis that *UTASb* is the 5' side of a putative larger and broken gene, disconnected from *UTASa* and independently evolved. Dynamics of *UTASa* amplification in telomerase negative strains were explored in the PGA2.1trt1-4 derived strain. Preliminary analysis suggests recombinational events as translocations and conserved telomere lengths in chromosomes that do not experience translocations.

IDENTIFICATION OF POST-TRANSCRIPTIONAL REGULATORY SEQUENCES ERE-TYPE IN CANCER-RELATED GENES

Sánchez Bocanegra César Augusto¹, Rodríguez Torres Angelina¹, Acevedo Whitehouse Karina Alethya¹, Fausto Arellano Carvajal¹, María Elizabeth Álvarez Sánchez², Bertha Isabel Carvajal Gámez¹

¹Universidad Autónoma de Querétaro Campus Aeropuerto, Carretera a Chichimequillas s/n, Ejido Bolaños, Querétaro, Qro.,

²Universidad Autónoma de la Ciudad de México, San Lorenzo No. 290, Col. Del Valle, Delegación Benito Juárez.

E-mail: bicarvajal@uaq.mx

Cancer is characterized by the abnormal growth of cells and is considered one of the most significant non-communicable diseases in recent years. Polyamine-mediated regulation plays a crucial role in the cancer process, and its dysregulation suggests that they are of great importance in tumor formation and progression. The transcription initiation factor eIF-5a, dependent on polyamines, is essential in cell proliferation and the translation of mRNA necessary within the cell cycle. This regulation, based on post-translational modification of hypusination, allows eIF-5A to bind to mRNA at its 3'UTR end in eIF-5A response elements (ERE). This project focuses on searching for new strategies to understand and combat cancer, aiming to identify ERE-like sequences at the 3'UTR end of mRNA from cancer-related genes, as well as determining the proteins that bind to these sequences for their regulation. The methodology was based in an in-silico analysis of candidate genes. Through the GenBank platform, the sequence of the selected genes mRNA was obtained, which were analyzed at the 3'UTR end, locating the presence of previously reported ERE-type regulatory sequences. Using RNAFold software, it was determined if the found sequences possessed the stem-loop regulatory structure. BRCA1, gene involved in breast cancer was selected. All the present ERE-type sequences, the most conserved one was selected, and based on it, probes were designed for mRNA synthesis, which was obtained through in vitro transcription. The BRCA1 transcript was incubated with total protein extracts from the MCF7 breast cancer cell line. An electrophoretic mobility shift assay (EMSA) was conducted to visualize the protein-transcript interaction. The result showed that the BRCA1 ERE-type probe interacts with proteins from MCF7 cells. We can suggest some proteins that may be interacting with the regulatory sequence, one of them being the eIF-5A transcription factor, which, if confirmed, could serve as a new pharmaceutical target for developing new cancer treatments.

ROLE OF CREBA AND CREM τ IN THE REGULATION OF THE MURINE CATSPER3 PROMOTER

Diego Eduardo Sánchez Jasso^{1,2}, Rosa María Bermúdez Cruz¹, Norma Angélica Oviedo de Anda²

¹Centro de Investigación y Estudios Avanzados del Instituto Politécnico Nacional, Departamento de Genética y Biología Molecular. ²Unidad de Investigación Médica en Inmunología e Infectología, CMN La Raza. ¹Av. Instituto Politécnico Nacional 2508, Col. San Pedro Zacatenco, Alcaldía Gustavo A. Madero, Ciudad de México, Código Postal 07360. Apartado Postal: 14-740, 07000. Tel 55 5747 3800 ext. 5334, roberm@cinvestav.mx; ²Av. Jacarandas s/num esq con Seris, Col. La Raza, Alcaldía Azcapotzalco, Ciudad de México, Código postal 02990 Tel 5557821088 ext. 24315. naoviedoa@yahoo.com.mx

The *Catsper3* gene encodes one of the four α -subunits that form the CatSper calcium channel, which is expressed and localized in the principal piece of the sperm flagellum. *Catsper3* has been shown to be essential for the proper functioning of CatSper, sperm hyperactivation and egg fertilization. Although its functionality and physiological relevance to male fertility have been characterized, nothing is known about its regulation at the transcriptional level. cAMP response elements (CRE) are crucial for gene regulation through spermatogenesis and are bound by CREM-CREB-ATF transcription factors (TF). Loss of *Cremt* results in an infertile phenotype in mice and dysfunction of CREB significantly reduces spermatid count and fertility in rats. In addition, CREM τ and CREBA can directly regulate the gene expression of *Catsper1-2* and *Catsper1*, respectively. Here, we first identified the murine *Catsper3* promoter and evaluated the role of CREBA and CREM τ TF on the regulation of *Catsper3* gene promoter. Bioinformatics tools predicted TF binding sites and a putative promoter region for *Catsper3*, which was cloned, and strategic regions were eliminated by PCR. Also, the predicted TATA box, DPE and two CRE sites were mutated by site-directed mutagenesis. These constructs alone or with plasmids expressing CREBA and CREM τ were transiently co-transfected into GC-1spg and MSC1 cells. Its transcriptional activity was then determined by luciferase activity assays. Finally, the binding of CREBA and CREM τ to the *Catsper3* promoter was assessed in vivo by ChIP-qPCR in mouse testis and liver tissues. Deletion analysis indicates that the *Catsper3* core promoter is located at -157..+152 relative to the transcription start site (TSS). Notably, CREBA and CREM τ can increase the *Catsper3* promoter activity in the presence of the +268..+439 region, where two CRE sites were predicted. Mutagenesis of the CRE1 and CRE2 sites prevents transactivation by CREBA and CREM τ , respectively, suggesting that CREM τ and CREBA may bind to these sites and promote *Catsper3* transcription. Mutation of a TATA box and DPE did not alter luciferase activity, indicating that this promoter is independent of these elements. Finally, the ChIP-qPCR assay demonstrated an enrichment of CREBA and CREM τ binding at the *Catsper3* promoter in the testis, but not in the liver, indicating their tissue-specific binding. Altogether, these results strongly suggest that *Catsper3* gene has a TATA-less, DPE independent promoter encompassed by -157 to +152 region around TSS and that *Catsper3* gene expression is directly regulated through two CRE sites by the transcription factors CREBA and CREM τ .

MIR-193B AND ITS POSSIBLE INFLUENCE OVER DDR GENES THROUGH ANRIL REGULATION IN TRIPLE-NEGATIVE BREAST CANCER-DERIVED CELL LINES

Sánchez-Pérez Julio Alejandro, Brito-Elías Luis, García Castillo Verónica,
Pérez-Plasencia Carlos y López-Urrutia Eduardo

Facultad de Estudios Superiores Iztacala, UNAM, Av. De los barrios 1, Los Reyes Iztacala, Hab Los Reyes Iztacala Barrio de los Árboles/ Barrio de los Héroes, 54090, Tlalnepantla de Baz, México.
julio.98.bio@gmail.com and e_urrutia@unam.mx

Breast cancer is the malignant neoplasm with the highest incidence worldwide and a leading cause of death. This heterogeneous disease encompasses several types based on its histologic and molecular characteristics; among them, triple-negative breast cancer (TNBC) has the highest recurrence, metastasis risk, and mortality rate. TNBC is characterized by the absence of estrogen, progesterone, and HER2 receptors, which limits the available therapeutic options, rendering its treatment usually inefficient. Another critical characteristic of TNBC is the prevalence of mutations in breast cancer susceptibility genes like BRCA and other genes associated with the DNA damage response (DDR), which have been used to propose therapies that disrupt the DNA repair processes. Homologous recombination (HR) is one of the main mechanisms of DNA double-strand break (DSB) repair, and its regulation involves the intervention of non-coding RNAs (ncRNAs). Recent research has revealed two ncRNAs involved in the DNA repair process mediated by HR: ANRIL, a lncRNA transcript antisense by the locus CDKN2A/B report as a positive regulator of DSB repair, and miR-193b, a miRNA transcript of 16p13.12 chromosomal region, identified as a tumor suppressor and negative regulator of proteins involved in HR. In this study, we analyzed the expression of miR-193b and its effect on its targets involved in DDR through ANRIL regulation. We found that miR-193b and ANRIL expression were negatively correlated in TNBC cell lines. Additionally, ANRIL knockdown increased miR-193b expression and substantially decreased that of its targets BRCA1, RAD51 and MDM2. These data suggest the ANRIL/miR-193b axis regulating DDR in TNBC.

CXCR4 SPLICING ISOFORM EXPRESSION IN RESPONSE TO TYPICAL HYPERSENSITIVITY PNEUMONITIS SIGNALING

Daniela Fernanda Sánchez-Sánchez¹, Carina Becerril Berrocal¹, Everardo Hernández-Plata², Mariel Maldonado³, Erika Rubí Luis García¹, Iliana Herrera³, José Cisneros³ and Ana Lilia Torres-Machorro¹

¹Laboratorio de Biología Celular and ³Laboratorio de Biopatología Pulmonar INER-Ciencias-UNAM. Departamento de Investigación en Fibrosis Pulmonar. Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas. Calz. de Tlalpan 4502, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México, CDMX. ²Investigador por México, Consejo Nacional de Ciencia y Tecnología; Instituto Nacional de Medicina Genómica, Ciudad de México, México CONACYT, INMEGEN, Ciudad de México 14610, México.
E-mail: fer.sanchez2410@gmail.com

Hypersensitivity Pneumonitis (HP) is an inflammatory lung disease caused by the inhalation of a wide variety of antigens that induce an exaggerated immunological response in genetically susceptible individuals. About 40% of the HP patients evolve to fibrosis, an incurable condition with a high mortality rate.

Besides the known global gene expression changes in HP, the pathogenesis of the diseases can also be affected by the altered expression of protein isoforms generated by alternative mRNA splicing. CXCR4 is a G-protein coupled chemokine receptor essential for cell homeostasis. In our lab, we found that lung fibroblasts derived from HP patients expressed three CXCR4 splicing isoforms that were not expressed in control fibroblasts. These splicing isoforms were variants one, three, and four, which had different N-terminal ends located on the cell surface. Previous work found that the N-terminal region of CXCR4 is critical for the interaction with its ligands. To dissect the relevance of these isoforms in HP, we characterized CXCR4 isoform expression in lung fibroblasts in response to characteristic cytokine signals, or to the artificial overexpression of HP-specific variants. The splicing isoform expression changes presented here are potentially relevant for the diagnosis, prognosis, or therapeutics of HP.

LNCRNAS LANDSCAPE DURING THE HYPOXIA-INDUCED VASCULOGENIC MIMICRY IN BREAST CANCER CELLS

Gricelda Sanchez Sanchez¹, Mireya Cisneros Villanueva², Alfredo Hidalgo-Miranda², Mercedes Olvera-Sanchez³, César López-Camarillo^{1*}

¹Posgrado en Ciencias Genómicas, Universidad Autónoma de la Ciudad de México, México. ²Instituto Nacional de Medicina Genómica, CDMX, México. ³Instituto Nacional de Cancerología, CDMX, México.

*Corresponding author: César López-Camarillo. San Lorenzo 290, Colonia del Valle, CDMX, 03100, Mexico. Tel: 5511070280, Email address: cesar.lopez@uacm.edu.mx

Vasculogenic mimicry is a microcirculatory system generated by tumor cells in hypoxic condition that proliferates and aligns to grow in three-dimensional (3D) architectures, forming structures modeled in the form of channels. This type of microvasculature can provide the tumor with the necessary nutrients, oxygen and blood. for its growth. Several microRNAs can modulate vasculogenic mimicry through post-transcriptional regulation of key genes, but whether long non-coding RNAs (lncRNAs) are involved in the regulation of this cellular process is largely understood. These findings change our view that angiogenesis is the only mechanism by which tumors acquire the nutrients necessary for their growth, suggesting an explanation for resistance to anti-angiogenic therapies. Positive vasculogenic mimicry tumors are associated with poor prognosis, low survival, resistance to therapy and metastasis, which is why this mechanism is considered a therapeutic target. In the present investigation, we studied the lncRNA landscape of breast cancer cells during the early stage of hypoxia-induced vasculogenic mimicry. Our genome-wide analysis identified that 350 lncRNA genes were differentially expressed ($p < 0.05$, $FC > 2.0$) under hypoxic conditions in MDA-MB-231 breast cancer cells. Of these, 215 genes were upregulated and 135 genes were downregulated. Functional bioinformatics analyzes of differentially expressed genes suggested that selection of the top 10 such as: RP1-261G23.7, LINC01291, Y-RNA, AP001429.1 and HIF1A-AS2, mainly showed an activation of survival and communication function cell-cell in a state of hypoxia, creating a positive environment for cellular transformation, cell migration, metastasis, resistance to therapy and the ability to modify lncRNA genes with increased risk of therapy failure and prognosis. Also, downregulated genes such as: CTD-2033D15.1, LYRM4-AS1, RP11-119K6.6, RCC2, their participation in proliferative and cell survival mechanisms was demonstrated. Subsequently, in the correlation analyzes of the top 30 between mRNA and lncRNAs, the intrinsic deregulation in hypoxic environments of MDA-MB-231 cells and the tumor transforming processes were determined. In conclusion, lncRNAs are directly related to cellular transformation including metastasis and vasculogenic mimicry in mda-mb231 triple negative breast cancer cells. The data obtained from the lncRNA microarrays and the classification with mRNA were also analyzed, building an interaction network involved in vasculogenic mimicry, as well as the functions they play in biological and molecular processes, localization on chromosomes and signaling pathways in what a participant. Genes with high and low expression were located, the relationship they have in hypoxic conditions and therefore their participation in MV AND METASTASIS. The top 10 overexpressed and underexpressed lncRNAs were validated by determining how lncRNAs play a circumstantial role in hypoxia and MV in triple-negative breast cancer tumors; together they could be an innovative and effective targeted therapeutic target for patients with diseases advanced cancer.

Acknowledgments: This work was funded by Consejo Nacional de Humanidades Ciencia y Tecnologia CONAHCYT. Grant A3-S-33674.

EFFECT OF PANOBINOSTAT (LBH589) ON GENE ACETYLATION REGULATION OF CA²⁺ SIGNALING DURING THE EPITHELIAL-MESENCHYMAL TRANSITION IN THE BREAST MCF10A CELL LINE

Ana Cecilia Sánchez Trujillo, Pablo Gómez González, Ángel Salmerón Hernández,
Gabriela Rodríguez Rodríguez, Ángel Zarain Herzberg

Departamento de Bioquímica, Facultad de Medicina, UNAM
Av. Universidad 3000, Mexico city, Mexico.
E-mail: zarain@unam.mx

Cancer metastasis is regulated, in part, by the epithelial-mesenchymal transition (EMT), where epigenetic mechanisms such as histone acetylation play an important role. Another regulator of EMT is Ca²⁺ signaling, which comprises the expression of 1670 genes. *In silico* analysis showed an aberrant expression of these genes during EMT¹. The compound Panobinostat (LBH589) is an FDA-approved drug against the treatment of multiple myeloma and is classified as a pan histone deacetylase inhibitor (HDACi)². However, little is known about the effect of LBH589 on regulating Ca²⁺ gene expression during EMT in breast cancer. This project aims to analyze the differential expression of Ca²⁺ signaling genes in the normal breast MCF10A cell line with an EMT induction model, a treatment with the iHDAC LBH589, and a combination of both treatments.

We established an EMT induction model with the addition of TGF-β (5 ng/mL) and EGF (50 ng/mL) growth factors in non-tumor breast cells (MCF10A). Subsequently, we performed treatments with LBH589 at 40 nM alone and in combination with TEM inducers. RNA-Seq data from such conditions were analyzed *in silico* to evaluate changes in Ca²⁺ signaling gene expression patterns.

Our results show that the TEM induction model yielded 249 overexpressed genes and 145 underexpressed Ca²⁺ signaling genes. Treatment with LBH589 overexpressed CAMK2N, whose silencing has been reported in different types of cancer and associated with tumor progression. On the contrary, GPER1 is underexpressed, and its silencing is associated with a better prognosis in breast cancer. The EMT process induced in non-tumor epithelial cells activates the expression of genes involved in tumor development, but treatment with LBH589 induces the expression of Ca²⁺ signaling genes associated with tumor suppressor activities.

References:

1. Eleutherakis-Papaïakovou, E., Kanellias, N., Kastritis, E., Gavriatopoulou, M., Terpos, E., & Dimopoulos, M. A. (2020). Efficacy of Panobinostat for the Treatment of Multiple Myeloma. *Journal of Oncology*, 2020. <https://doi.org/10.1155/2020/7131802>
2. Hernández-Oliveras, A., Izquierdo-Torres, E., Hernández-Martínez, G., Zarain-Herzberg, Á., & Santiago-García, J. (2021). Transcriptional and epigenetic landscape of Ca²⁺-signaling genes in hepatocellular carcinoma. *Journal of Cell Communication and Signaling*, 15(3), 433–445. <https://doi.org/10.1007/s12079-020-00597-w>

STUDY OF MITOCHONDRIAL DNA VARIANTS ASSOCIATED TO BREAST CANCER DEVELOPMENT

Jessica Jazmin Saucedo-Rivas^{1,2}, Carlos Jhovani Pérez-Amado^{2,3}, Amellalli Bazan-Cordoba^{2,3}, Laura Gómez-Romero², Julián Ramírez-Bello⁴, Verónica Bautista-Piña², Alberto Tenorio-Torres⁵, Eva Ruvalcaba-Limón⁵, Felipe Villegas-Carlos⁵, Alfredo Hidalgo-Miranda², Silvia Jiménez Morales^{2*}

¹Facultad de Estudios Superiores Iztacala, UNAM, Edo. De México 54090, México, ²Instituto Nacional de Medicina Genómica, CDMX 14610, México, ³ Programa de Maestría y Doctorado en Ciencias Bioquímicas, UNAM, CDMX 04510, México, ⁴Instituto Nacional de Cardiología Ignacio Chávez, CDMX 14080, México, ⁵Instituto de Enfermedades de la Mama, FUCAM, CDMX 04980, México.

*Tel. (+52)55 53501900, Exts. 1155 y 1225, sjimenez@inmegen.gob.mx

Background: Breast cancer is the most common and deadliest cancer in Mexican women, posing a major public health issue. Mitochondrial genome (mtDNA) variants, particularly the A10398G variant in the *MT-ND3* gene, have been implicated in breast cancer risk, though results are controversial^{1,2}. **Objective:** Determine if the A10398G mtDNA variant is associated with breast cancer in the Mexican population. **Methods:** We included DNA blood samples from women with breast cancer and healthy controls. The A10398G variant was genotyped using *DdeI* restriction enzyme after amplifying the *MT-ND3* gene by TD-PCR. The frequency of the variant was determined in cases and controls, and breast cancer risk association was estimated by odds ratio (OR) at 95% confidence intervals. **Results and Discussion:** A total of 104 breast cancer cases and 100 controls were genotyped. The mean age of patients was 54 years old (interval: 29-85). Predominant tumor characteristics included ductal invasive histological subtype, in grade II, clinical stage II, and luminal A molecular subtype. The 10398G allele was identified in 27.9% (n=29) of patients and 26% (n=26) of controls. No significant association between the A10398G variant and breast cancer risk was observed (OR=1.1; 95% CI: 0.05-2.04, p=0.76). The analysis of mtDNA variants is challenging due to the high mutation rate and heteroplasmy of the molecule, complicating genotyping accuracy³. Factors such as ethnicity, environmental exposure, and clinical variables might also influence results. **Conclusion:** Our data suggest that the A10398G mtDNA variant is not associated with breast cancer. Further studies in wider and well characterized cohorts are needed to fully understand the role of this variant in Mexican population with breast cancer.

References

1. Setiawan, V., Chu, L., John, E., Ding, Y., Ingles, S., Bernstein, L., Press, M., Ursin, G., Haiman, C. & Neuhasen, S. (2008). *Cancer Genet Cytogen*, 181, 16-19.
2. Jahani, M., Azimi, M., Karimi, T., Banoei, M., & Houshmand, M. (2020). *Mitochondrial DNA Part A*, 31, 11-16.
3. Pérez-Amado, C., Tovar, Gómez-Romero, L., Beltrán-Anaya, F., Bautista-Piña, V., Dominguez-Reyes, C., Villegas-Carlos, F., Tenorio-Torres, A., Alfaro-Ruiz, L. A., Hidalgo-Miranda, A., & Jiménez-Morales, S. (2020). *Front oncol*, 10, 572954.

IS THERE A CONSERVED MACHINERY OF EXECUTIVE FUNCTION EVOLUTION AND DEVELOPMENT ACROSS MAMMALIAN SPECIES? A COMPARATIVE TRANSCRIPTOMICS PERSPECTIVE

María Fernanda Shehin^{*1}, Humberto Gutiérrez^{*}

^{*}Departamento de Genómica Computacional, Instituto Nacional de Medicina Genómica (INMEGEN)
Periférico Sur 4809, Tlalpan, Ciudad de México, CP:14610, México.

¹Posgrado de Ciencias Biológicas, Universidad Nacional Autónoma de México.

Development of neurological functions is the result of the collaborative action of hundreds or thousands of genes working together in tight functional coordination. The executive function is a set of higher cognitive abilities, mediated by the prefrontal cortex, enabling decision-making, adaptation to environmental changes, inhibition of automatic responses, resistance to distraction, social interactions, and planning. Self-control is a key component of executive function, defined as the ability to inhibit impulsive or automatic behavior during decision-making processing. While conserved aspects of executive function have been identified in various mammalian species, including self-control, here we asked whether an underlying conserved molecular machinery behind the evolution and development of this complex actually exists. In order to detect a signal of conserved transcriptional determinants of executive function development across species, we conducted a comparative study using available gene expression data for 11 thousand orthologous genes in the prefrontal cortex of 8 mammalian species exhibiting variations in self-control tasks scores. We find a statistically significant bias in the number of genes displaying a strong association between level of expression and two independent metrics of self control. In particular, there is a larger than expected number of genes with a negative association between expression and self control. Using phylogenetic generalized regression models we confirmed the existence of a significantly large number of genes displaying both a strong association between expression and self control scores, and statistical independence from phylogenetic relationships. Taken together, our results suggest the existence of a conserved genetic machinery across species, whose collective expression level is significantly associated with metrics of self-control, opening the prospect of identifying specific transcriptional determinants of executive function development and evolution in the human brain.

SOLANUM LYCOPERSICUM CV. MICRO-TOM AS A TOOL TO CHARACTERIZE CAROTENOID CLEAVAGE DIOXYGENASES FROM *BIXA ORELLANA* L.

Diana Laura Sierra Ulín¹, Cinthia Berenice Romano Martín¹, Luisa López Ochoa¹, Renata Rivera-Madrid¹

¹Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, A.C.
Calle 43 No. 130, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México.
E-mail: renata@cicy.mx

Bixa orellana L. is a neotropical domesticated species commonly known as achiote, has a significant industrial importance since large amounts of carotenoid pigments accumulate in the aril of its seeds, of which approximately 80% corresponds to the apocarotenoid bixin. Due to the efficacy and demand for bixin as a natural dye worldwide, it is necessary to implement approaches aimed at manipulating its biosynthesis to improve its production yield. However, this approach has been limited since the biosynthetic pathway of bixin has yet to be fully elucidated. Data analysis of achiote transcriptomes allowed the identification of three gene families responsible for the synthesis of bixin, the family of carotenoid cleavage dioxygenases (CCDs) accountable for carrying out the first step of the proposed pathway. The large number of products derived from carotenoid cleavage is due to the great promiscuity of carotenoid substrates that characterize this family of enzymes, making their study and characterization difficult. Functional analysis in a heterologous system has been proposed to characterize BoCCD candidate genes in *S. lycopersicum* cv. Micro-Tom since it produces large amounts of lycopene, a precursor of bixin, in its ripe fruits. This tomato cultivar, for its advantageous characteristics, has been selected as a heterologous plant system to use for the expression of BoCCDs genes under the control of a tomato fruit-specific promoter (E8), which will allow evaluation of the alterations of carotenoid and apocarotenoid products caused by the heterologous expression of achiote CCDs, and thus, determine their implication in bixin biosynthetic pathway.

EPIGENETIC CHARACTERIZATION OF ADAMTS17 GENE PROMOTER DURING TISSUE REGENERATION IN *AMBYSTOMAMEXICANUM*

Violeta Guadalupe Silva Díaz¹, Aylin del Moral Morales¹, José Antonio Ocampo Cervantes²,
Cynthia Sámano Salazar^{1*}, Ernesto Soto-Reyes^{1*}

¹Universidad Autónoma Metropolitana, Unidad Cuajimalpa, Departamento de Ciencias Naturales e Ingeniería, CDMX, C.P.05348,55 5558146500 ext. 3879, ²Centro de Investigaciones Biológicas y Acuícolas de Cuernavaca (CIBAC), Universidad Autónoma Metropolitana, Unidad Xochimilco *csamano@cua.uam.mx; *esotoreyes@cua.uam.mx

The Mexican axolotl (*Ambystomamexicanum*), is an endemic species of the Valley of Mexico, that possesses an extraordinary capacity to completely regenerate a variety of organs and tissues throughout its life cycle. Recently, our research group identified a group of genes related to the regeneration process, including *ADAMTS17* gene, which encodes for a metalloproteinase and play a crucial role in skeletal growth. This gene shows positive regulation during the tissue regeneration process but negative regulation in aged axolotls. However, the underlying molecular mechanisms of transcriptional regulation are not yet fully understood. In our project, we focused on characterizing the *ADAMTS17* gene promoter in *A. mexicanum* from an epigenetic perspective, using DNA from two stages during the blastema formation (a structure formed prior to the regeneration process), as well as in samples of aged axolotl tissue. Our goal was to propose a possible mechanism that explains how this gene is regulated during tissue regeneration. As a first approach, we evaluated the DNA methylation status and post-translational modifications of histones such as H3K4me3 related to gene activation and H3K27me3 involved in gene repression. Through methylation-sensitive PCR (MSP-PCR) assays and chromatin immunoprecipitation (ChIP) against H3K4me3 and H3K27me3 histone marks here we demonstrate that DNA methylation does not participate in the transcriptional regulation of this gene. However, significant changes in the enrichment of H3K4me3 and H3K27me3 histone marks were found in the promoter region. These suggest that *ADAMTS17* gene promoter may be regulated by histone marks having an impact at the transcriptional level in *ADAMTS17* during the tissue regeneration process in *A. mexicanum*. This approach represents a valuable contribution to research on tissue regeneration in *A. mexicanum*, as there has been limited knowledge regarding the involvement of epigenetic mechanisms in this process.

The project was approved by the Comité de Ética en Investigación (CEI) from UAM Xochimilco (CEI.2023.007) and by the National Council of Humanities Science and Technology (CONAHCyT, CB-SEP-CONACyT 284748 and CF-2023-G-1558, and approved by the Divisional Council of Natural Sciences and Engineering (agreement DCNI-10-240-22) and by the Rectorate of Unit Cuajimalpa (agreement RC.153.2022).

EFFECT OF PHARMACOLOGICAL INHIBITION OF CRM1 ON CARDIAC DYSFUNCTION IN A PROGERIA MOUSE MODEL

Angélica Soberano Nieto, Marlon De Ita Ley y Bulmaro Cisneros

Department of Genetics and Molecular Biology, CINVESTAV, Zacatenco Unit. Mexico City
9934257130. angelica.soberano@cinvestav.mx

Cardiovascular diseases are a leading cause of morbidity and mortality in older adults. The study of these conditions requires the use of experimental models of aging, such as Hutchinson-Gilford progeria syndrome (HGPS). HGPS is a rare disease that causes premature and accelerated aging due to the expression of progerin, a mutant variant of lamin A, which causes various cellular alterations that eventually lead to cellular senescence. In our laboratory, we recently described that progerin induces the overexpression of CRM1, an exportin that regulates the nuclear export of proteins, consequently altering proteostasis. Interestingly, treatment with selinexor, a selective inhibitor of CRM1, reduces progerin levels and reverses cellular alterations associated with senescence in primary fibroblast cultures from HGPS patients.

Considering these findings, the objective of the present study was to identify the effect of Selinexor on cardiac dysfunction in $Lmna^{G609G/G609G}$ mice, a mouse model of HGPS. To achieve this objective, $Lmna^{G609G/G609G}$ and WT ($Lmna^{+/+}$) mice were treated with selinexor (2.5mg/kg) or vehicle only for eight weeks, and then their hearts were dissected for histopathological analysis through hematoxylin-eosin staining and immunohistochemistry and western blot assays.

Results: The cardiomyocytes of $Lmna^{G609G/G609G}$ mice presented nuclei with aberrant morphology and a smaller longitudinal area compared to WT. Interestingly, these alterations improved after treatment with selinexor. Likewise, pharmacological treatment decreased the number of cardiomyocytes positive for progerin staining. Furthermore, the loss of the nucleus/cytoplasm ratio present in the hearts of mutant mice recovered in response to selinexor. Finally, various markers of cardiac damage, including BNP, Mef2d, and Mef2a, were evaluated. We found that the elevated level of BNP present in the myocardium of $Lmna^{G609G/G609G}$ mice decreased after treatment with selinexor. Regarding the transcription factors Mef2d and Mef2a that control cardiac function, we identified a decrease in their levels in progeroid mice and a trend towards recovery in mutant mice treated with selinexor.

Conclusions: The hearts of $Lmna^{G609G/G609G}$ mice present alterations suggesting cardiac dysfunction, including morphological and biochemical aberrations, and notably, these alterations are reversed by treatment with selinexor.

References

1. García-Aguirre I, Alamillo-Iniesta A, Rodríguez-Pérez R, Velez-Aguilera G, Amaro-Encarnación E, Jiménez-Gutiérrez E, et al. Enhanced nuclear protein export in premature aging and rescue of the progeria phenotype by modulation of CRM1 activity. *Aging Cell*. 2019;18(5):e13002.
2. Benedicto I, Dorado B, Andrés V. Molecular and Cellular Mechanisms Driving Cardiovascular Disease in Hutchinson-Gilford Progeria Syndrome: Lessons Learned from Animal Models. *Cells*. 2021;10(5)

STUDY OF THE SIGNALING PATHWAYS THAT MAINTAIN THE DNA INTEGRITY IN MITOCHONDRIA AND CHLOROPLAST IN PLANTS

Diana Solano-Arguello¹, Daniela Camacho-Manriquez² and José Antonio Pedroza-García*

¹Departamento de Bioquímica, Facultad de Química, UNAM, ²Langebio-Cinvestav Irapuato
e-mail: 315274877@quimica.unam.mx; *joped@quimica.unam.mx

Due to their sessile lifestyle, plants are inevitably exposed to adverse conditions present in their environment, which can induce DNA damage, altering the stability of their genome, which affects their survival and development¹. For all organisms, it is essential to develop efficient mechanisms in the genome integrity maintaining, that consist of highly coordinated cellular networks collectively denominated as DNA Damage Response (DDR)¹. In eukaryotes, the mechanisms are conserved and involving two protein kinases with a key role in the initiation of signaling pathways: ATM (Ataxia Telangiectasia Mutated) which recognizes double strand breaks (DSBs) and ATR (ATM-Rad3-related) that is involved in the detection of replicative stress and the collapse of replication forks². While ATM and ATR kinases are conserved in plants, they lack of orthologous genes for important regulators such as the transcription factor p53, which acts downstream of both kinases in yeast and mammals; instead, they have a plant specific transcriptional factor, SOG1 (Suppressor of Gamma Response 1) who has homologous functions to p53 as it activates DNA repair mechanisms, cell cycle arrest and programmed cell death when the damage is severe³. Recent advances have improved our understanding the DDR in plants, specifically in the nuclear signaling cascade. In animals, it has been reported that ATM and p53 contribute to maintaining the mitochondrial genome integrity^{4,5}. Nevertheless, until now the role of the ATR and ATM kinases, and the transcription factor SOG1 in the retrograde signaling that occurs from the organelles to the nucleus to respond to lesions in the chloroplast and mitochondrial DNA remains unknown. Therefore, our work was focused on to elucidate the role of these regulators of DDR in mitochondria and chloroplast, first the *Arabidopsis atm*, *atr* and *sog1* mutants were exposed to specific genotoxic stress in organelles caused by CIP and NOV, showing hypersensitivity to treatment in root, and area part in comparison with wild-type plants, apparently the signalling ATR-SOG1 axe depend contributes majority in response to organelle DNA damage. Finally, we carried out genetic crosses with organelle replisome machinery mutants, obtaining smaller plants than the simple mutants, with alterations in their proper development and fertility.

References

1. Pedroza-García, J. A., Xiang, Y., De Veylder, L. (2022) *The Plant Journal*, 109, 490-507
2. Nisa, M. U., Huang, Y., Benhamed, M. and Raynaud, C. (2019) *Frontiers in Plant Science*, 10, 653.
3. Yoshiyama, K., Conklin, P.A., Huefner, N.D. and Britt, A.B. (2009) *Proc Natl Acad Sci USA*, 106, 12843-12848
4. Shimura, T., Sasatani, M., Kawai, H., Kamiya, K., Kobayashi, J., Komatsu, K. and Kunugita, N. (2017) *Cell Cycle*, 16, 2345-2354.
5. Wong, T.S., Rajagopalan, S., Townsley, F.M., Freund, S.M., Petrovich, M., Loakes, D. and Fersht, A.R. (2009). *Nucleic Acids Research*. 37, 568-581.

ANALYSIS OF DNMT2 FUNCTION DURING DENGUE VIRUS SEROTYPE 2 INFECTION IN *Aedes aegypti* MOSQUITOES

Tapia-Rodríguez, A.^{1*}, Fabiola Claudio Piedras¹, Recio-Tótoro, B^{1,2}, Pacheco-Guillen, S², Lanz-Mendoza, H^{1*}

¹National Institute of Public Health, Center for Research on Infectious Diseases, Cuernavaca, Mexico.

²Biotechnology Institute, National Autonomous University of Mexico, Cuernavaca, Mexico.

Dengue fever is a viral disease caused by dengue virus (DENV) transmitted to humans by the bite of infected mosquitoes, mainly *Aedes aegypti*. It is currently a public health problem in tropical regions of developing countries. Between 2000 and 2019, the WHO reported that the number of reported cases worldwide increased tenfold, from 500 000 to 5.2 million. In Mexico during the 30th epidemiological week of 2023, 7,295 confirmed cases were reported compared to the same week in 2022 (2,518); an increase of 183%. There is currently no specific treatment to tackle this infection and the development of strategies based on the understanding of virus-vector molecular interactions is urgently needed. Methylation of cytosines in nucleic acids is a modification catalysed by cytosine-C5-methyltransferase enzymes. DNMT2 is the most conserved methyltransferase among eukaryotic organisms, and is present as a single copy in *Aedes aegypti*. Recent work from our research group showed that dengue virus-infected mosquitoes: (1) overexpress *dnmt2* at the transcriptional level, (2) double the 5-methylcytosine content in total RNA, (3) overexpress antiviral system genes if RNA methylation is inhibited with azacitidine, and (4) reduce the prevalence of viral infection from 83% to 30% in azacitidine-treated mosquitoes compared to the control group. Although the use of methylation inhibitors is a classic strategy to analyse the function of methylation in nucleic acids, an approach with greater specificity that limits the indirect effects associated with drug treatments is required. The aim of this research project is to analyse the function of DNMT2 during dengue virus serotype 2 (DENV2) infection in *Aedes aegypti* mosquitoes genetically edited with the CRIPR-Cas9 system. A line of *Aedes aegypti* $\Delta dnmt2-53$ mosquitoes infected with DENV2 will be generated and analysed for: (1) the amount of 5mC-RNA; (2) the transcriptional response of *dnmt2*; (3) the prevalence of infection; (4) viral load; (5) the extrinsic incubation period; (6) the duration of the gonotrophic cycle; and (7) mosquito survival.

TRANSCRIPTIONAL CONTRIBUTION OF A HUMAN HETERODIMERIC BASIC HELIX-LOOP-HELIX TRANSCRIPTION FACTOR WHEN ISOLATED FROM THE HUMAN CELL CONTEXT

Ana Lilia Torres-Machorro¹ and Karla Miranda López-DeLucio^{1,2}

¹Laboratorio de Biología Celular, Departamento de Investigación en Fibrosis Pulmonar, Instituto Nacional de Enfermedades Respiratorias Ismael Cosío Villegas. Calzada de Tlalpan 4502, Belisario Domínguez Sección 16, Tlalpan, 14080, CDMX.

²Posgrado en Ciencias Biológicas, Universidad Nacional Autónoma de México. Posgrado en Ciencias Biológicas, Unidad de Posgrado, Edificio D, 1º Piso, Circuito de Posgrados, Ciudad Universitaria, Coyoacán, C.P. 04510, CDMX, México.

E-mail: ana.torres@iner.gob.mx

The basic Helix-Loop-Helix (bHLH) Transcription Factors (TFs) are a complex protein family that regulates a wide range of cellular processes, from cell differentiation to the circadian clock. This family primarily forms dimeric TFs through interactions among at least 100 bHLH proteins. This intricate interaction network poses a challenge in interpreting experiments conducted in human cells, as the results are likely influenced by the combined effect of altering the complex protein-protein interaction system and the direct transcriptional outcomes.

To solve this issue, and because the yeast conserves eukaryotic transcriptional regulatory mechanisms, we expressed human bHLH TFs in *Saccharomyces cerevisiae* to test their transcriptional role without other interfering endogenous tissue-specific TFs. We specifically studied the transcriptional contribution of the SCX-E47 bHLH heterodimer in regulating an original human regulatory region (core promoter and proximal enhancer) or the same region engineered with different types of core promoters.

Our findings revealed a significant transcriptional effect of the SCX-E47 heterodimer. It was positive in highly transcribed genes and negative in a low-expression gene. Importantly, this effect was influenced by the core promoter context and the basal transcription levels of the reporter gene. The study also compares the SCX-E47 data with the SCX or E47 homodimers, offering valuable insight into their functional differences.

DETECTION OF FUSION GENES IN PATIENTS DIAGNOSED WITH ACUTE LYMPHOBLASTIC LEUKEMIA

Ruth Trejo Martínez, Edgar Alejandro Turrubiarres Martínez, Sofía Bernal Silva,
Roberto Fidencio González Amaro, Perla del Carmen Niño Moreno

Centro de Investigación en Ciencias de la Salud y Biomedicina, Facultad de Medicina, UASLP. Av. Sierra Leona 550,
Lomas de San Luis, 78210. San Luis Potosí, S.L.P., México.
Cel. 332 286 8218. A210042@alumnos.uaslp.mx

Acute lymphoblastic leukemia (ALL) is the most prevalent oncological disorder among children and adolescents, characterized by the uncontrolled proliferation of lymphoid hematopoietic cells. In Mexico, ALL remains a major public health challenge, with an average survival rate ranging from 40% to 60%. This can be attributable to multifaceted factors such as the limited access to molecular diagnostic methods.

Studies have reported that the expression of fusion genes, such as BCR-ABL (Philadelphia chromosome), ETV6-RUNX1, and MLL-AF4, can correlate with prognosis and therapeutic response in pediatric patients with ALL. Objective: This study aimed to develop a standardized molecular diagnostic protocol for the identification of critical translocations and to preliminarily assess the prevalence of fusion genes among pediatric ALL patients in the State of San Luis Potosí. Methods: Bone marrow samples were obtained through aspiration and biopsy from 18 pediatric patients with confirmed ALL diagnoses (based on morphology and immunophenotype) at Hospital Central "Dr. Ignacio Morones Prieto." Total RNA was extracted from the bone marrow samples, and cDNA synthesis was performed via PCR amplification. The detection of fusion genes (BCR-ABL, ETV6-RUNX1, and MLL-AF4) was standardized using endpoint PCR techniques. Validation was conducted through the amplification of endogenous ABL gene and positive controls for the three fusion genes. Results: The standardized PCR protocols demonstrated high accuracy. Immunophenotyping revealed a predominance of common B-cell ALL (n=15), with pro-B (n=1) and pre-B (n=2) subtypes also identified. Fusion gene analysis showed no BCR-ABL translocations in the 18 samples, whereas ETV6-RUNX1 was detected in 5 out of 18 samples, and MLL-AF4 in 11 out of 18 samples. No significant association was observed between the presence of these mutations and the clinical characteristics of the patients, likely due to the limited sample size. Conclusions: The results underscore the rigorous standardization of the proposed PCR methodologies. Importantly, they highlight the critical need to integrate fusion gene detection into pediatric care settings. Such integration has the potential to enhance risk stratification and prognostic assessment, thereby informing more precise therapeutic interventions and improving patient outcomes.

References

1. Velasco, P., & Murillo, L. Leucemia aguda en Pediatría |Pediatría Integral, (2021). <https://www.pediatriaintegral.es/publicacion-2021-09/leucemia-aguda-en-pediatria/>
2. Jiménez, M. S., Hidalgo, M. A., & Ramírez, B. J. Acute lymphoblastic leukemia: a genomic perspective. Boletín médico del Hospital Infantil de México, (2017). <https://doi.org/10.1016/j.bmhmx.2016.07.007>
3. Pui, C. H., Roberts, K. G., Yang, J. J., & Mullyghan, C. G. Philadelphia Chromosome-like Acute Lymphoblastic Leukemia. Clinical lymphoma, myeloma & leukemia, (2017). <https://doi.org/10.1016/j.clml.2017.03.299>
4. Britten, O., Ragusa, D., Tosi, S., & Kamel, M. Y. MLL-Rearranged Acute Leukemia with t (4;11) (q21; q23)-Current Treatment Options. Is There a Role for CAR-T Cell Therapy?. Cells, (2019). <https://doi.org/10.3390/cells8111341>

AUTOPHAGY CHARACTERIZATION DURING AGING IN HUMAN DERMAL FIBROBLASTS

Juan Unzueta Mendoza¹, Mina Königsberg², Adriana Alarcón Aguilar²,
Ian García Aguirre³, Bulmaro Cisneros Vega³

¹Doctorado en Biología Experimental, División de Ciencias Biológicas y de la Salud, Universidad Autónoma Metropolitana Unidad-Iztapalapa, Mexico. ²Laboratorio de Bioenergética y envejecimiento celular, Departamento de Ciencias de la Salud, Universidad Autónoma Metropolitana Unidad-Iztapalapa, Mexico. ³Departamento de Genética y Biología Molecular, Centro de Investigación y Estudios Avanzados, Mexico.
C.P. 06720; Tel.: 5619190777; jjum@xanum.uam.mx

Autophagy is a process that mediates the turnover and recycling of macromolecules and organelles. This process allows the maintenance of cellular homeostasis, functioning as a control mechanism that preserves the quality of macromolecules, mobilizes nutrients and maintains immunity. Interestingly, autophagy is altered with aging; there are ambiguous reports that have observed that autophagic markers increase or decrease with aging depending on the tissue or cell type. However, it is necessary to study autophagic flux to determine which step of the process may or may not be affected. The purpose of this work is to characterize the autophagy process, from the induction of the process to the degradation of lysosomal content during aging in different cultures of dermal fibroblasts derived from individuals of different ages.

We have found that the master regulator of autophagy and transcription factor lysosomal and autophagy network (TFEB) appears to be located in the nucleus and responds appropriately to autophagy-inducing stimuli such as starvation in primary fibroblasts of skin derived from older adults. Likewise, aged fibroblasts seem to have increased autophagic markers such as LC3 and p62, which may be the reason they present a high production of autophagosomes. However, autophagic flux appears to be disrupted in skin fibroblasts derived from older adults, where the maturation-fusion step appears to be impaired. This increase in autophagic markers found could be a consequence of the successive accumulation of autophagosomes.

Preliminary conclusions show that the autophagic process appears to be exacerbated in skin fibroblasts derived from older adults, however the autophagic flux cannot be completed, compromising the maturation-fusion steps by accumulating damaged organelles and proteins.

MECHANOSENSING AND INFLAMMATION IN PREMATURE RUPTURE OF MEMBRANES

Vadillo-Pérez L., Retana-Arellano I. A., Flores Pliego A., Estrada Gutiérrez G., Gochar-Mora A., Ortega-Gómez K., Pérez-Carreón J. I., Vadillo Ortega F.

Instituto Nacional de Medicina Genómica

Premature rupture of membranes (PROM) affects approximately 10% of all pregnancies and is the leading cause of preterm birth. Mechanisms mediating the physiological rupture of the chorionic amniotic membranes (CAM) have been initially characterized, involving the abnormal activation of matrix metalloproteinases in association to the moment of the rupture of the membranes.

The mechanical resistance of CAM is dependent of the abundance and the specific molecular organization of the extracellular matrix components (ECM). No information is available to understand the role of each local cell population in ECM homeostasis in these tissues, a critical process that results in the adaptation of the structure and function of the membranes to the changing needs of pregnancy progression.

ECM homeostasis is dependent of the capacity of cells to respond to mechanical stimuli, a function known as mechanosensing, that is the result of the interaction of a family of cell receptors, mainly integrins, and their ligands in the extracellular matrix.

We studied samples of membranes from three groups of women, including normal labor, absence of labor and PROM. An altered transcriptomic profile associated to PROM was found, characterized by the under-expression of genes encoding ECM structural proteins, including collagens and laminins and cell receptors of the integrin family. These transcriptional findings were validated with protein expression in tissues, supporting the hypothesis that mechanosensing is altered in membranes obtained from PROM.

In silico analysis of gene networks revealed that all under-expressed genes in PROM tissues are controlled by NF- κ B, a transcription factor that modulates the inflammatory response. Intrauterine infection and the resulting local inflammation are a well identified clinical factor associated to PROM and this antecedent allows us to expand the initial hypothesis to involve inflammation as the modulator of mechanosensing alterations in membranes developing PROM.

This study provides evidence that CAM with PROM expresses a different pattern of ECM molecules as well as several integrins, that may be linked to changes in the composition and arrangement of the ECM resulting in altered mechanical stability in these tissues.

ANALYSIS OF THE DNA METHYLATION PATTERN OF THE SFN GENE PROMOTER IN EARLY STAGES OF THE K14E7 MURINE MODEL

¹M.Sc. Luz del Carmen Valerio Jácome, ¹Dr. Patricio Gariglio Vidal and ²Dr. Alejandro García Carranca

¹Center for Research and Advanced Studies of the National Polytechnic Institute, Av Instituto Politécnico Nacional 2508, San Pedro Zacatenco, Gustavo A. Madero, C.P. 07360, Ciudad de México., ²Biomedical Research Institute-UNAM, Circuito, Mario de La Cueva s/n, C.U., Coyoacán, C.P. 70228, Ciudad de México.
E-mail: ¹vidal@cinvestav.mx, ²carranca@iibiomedicas.unam.mx

Cervical cancer is a major public health problem. This disease is widely associated with human papillomavirus infection. It has been strongly related to the immunosuppressive microenvironment as an important factor in the progression, persistence, and severity of cancer. Epigenetics plays an important role in the initiation and development of cancer. The DNA methylation is a major form of epigenetic modification associated with target gene silencing and is correlated with cancer carcinogenesis and progression, where the promoters of genes is of great importance, such as tumor suppressor genes, which have been observed to suffer hypermethylation, which causes the expression of said genes to be affected or silenced.

On the other hand, the murine model that uses the keratin 14 promoters for the expression of the E7 oncoprotein (K14E7) is important for the study of cervical carcinogenesis under estradiol treatment, but it is also an excellent model on its own since it has a great similarity with the natural history of the disease that occurs in women because the E7 oncoprotein has been attributed the most important role in cervical carcinogenesis.

In our project, the murine model is used to study the SFN gene which encodes the 14-3-3 sigma protein (expressed in T cells and epithelial cells), this protein belongs to a family of seven highly conserved proteins. Well over 200 proteins have been shown to interact with 14-3-3. In several types of cancer SFN is a tumor suppressor, however, it is not known what its role is in cervical cancer, whether there is a decrease at the messenger level and what the molecular mechanism by which this gene is being modified.

In cell lines it is known that SFN can regulate cervical cancer cell proliferation, apoptosis, cytoskeletal remodeling and metastasis through LIMK2/Cofilin signaling. Further, in a study it is observed that 14-3-3 σ promoter methylation may be associated with the carcinogenesis of breast cancer and might represent a useful blood-based biomarker for the clinical diagnosis of breast cancer. For this reason, the present work aims to elucidate the molecular mechanism by which the E7 oncoprotein of HPV16 in the K14E7 murine model is contributing to the changes in the methylation pattern of the SFN gene promoter.

USE OF NEURAL NETWORKS FOR THE ANALYSIS AND CLASSIFICATION OF GLIOMAS THROUGH THEIR EPIGENETIC PATTERNS

David Valle-Garcia^{1*}, Clara Garcia-Chavez¹

¹Laboratorio de Neuroinmunología, Instituto Nacional de Neurología y Neurocirugía "Manuel Velasco Suárez".
Av. Insurgentes Sur 3877, La Fama, Tlalpan, 14269, Ciudad de México

*Correspondence: david.valle.edu@gmail.com

Gliomas, the most common type of primary brain tumor, exhibit complex molecular profiles that hinder their accurate classification and prognosis. Although information from different epigenetic profiles can be used to make better classifications, its application in the clinical setting remains restricted due to its high cost and limited applicability for therapeutic decision-making. Therefore, it is necessary to develop analysis tools that allow condensing different types of molecular information and associating it with relevant clinical factors. In this work, we develop a neural network algorithm for the analysis of epigenetic data derived from gliomas that allows their classification and distinction from healthy tissue.

Public databases with epigenetic information from patients with low and high-grade gliomas were used to train a convolutional neural network. This data was converted into images by mapping its values to a Space Filling Curve (SFC). After training different network architectures, we tested their predictive power using a testing dataset. We choose the optimal network/SFC configuration and made further adjustments to maximize its efficiency. Finally, the model was tested with an independent validation dataset to determine its specificity, sensitivity, and efficiency.

As a proof of concept, the study was conducted with DNA methylation data in samples derived from tumors and liquid biopsies. In tumor-derived samples, it was observed that SFCs serve as a good normalizing element that surpasses in terms of efficiency the use of feed-forward networks that analyze data directly. The study of liquid biopsies is being carried out at the time of submitting this work, but it shows promising results and suggests that SFCs can help us detect patterns that other methods cannot.

In conclusion, the conversion of epigenetic data to SFCs is a novel method that allows efficient training of neural networks and the generation of a specific, sensitive, and efficient classifier.

Keywords: Gliomas, glioblastoma, artificial intelligence, epigenetics, deep neural networks

CAN THE BIOCHEMICAL INTERACTION BETWEEN KILLER STRAINS AND NON-TOXIN-PRODUCING STRAINS OF *SACCHAROMYCES CEREVISIAE* RESULT IN RESISTANCE?

Azul Vázquez López de la Fuente¹, Verónica Morales-Tlalpan^{1,2}, Carlos Saldaña^{1,2,*}.

¹Laboratorio de Biofísica de Membranas y Nanotecnología. ²Laboratorio Nacional de Visualización Científica Avanzada, Facultad de Ciencias Naturales (LAVIS-FCN-UAQ) Universidad Autónoma de Querétaro.

*carlos.saldana@uaq.mx Tel. 442 192 1200, ext 65332

The toxin-producing *Killer* strains of *Saccharomyces cerevisiae* possess the ability to impede the growth of sensitive strains lacking the toxin or having a different mating type. This phenomenon piqued significant interest when colony growth was observed within the inhibition halo following the interaction between strain 42300, whose mating type is α , and sensitive yeast from strain 5X47 (Bevan & Makower, 1963). Firstly, it is plausible that both strains underwent genetic recombination, resulting in the emergence of strains resistant to the *Killer* toxin. Alternatively, the biochemical interplay between yeasts might have triggered resistance mechanisms via induced metabolic repression in response to environmental changes (Sesti *et al*, 2001). To delve into the questions stemming from this observation, the genetic and molecular traits of the colonies thriving within the inhibition halo will be scrutinized through techniques such as PCR, establishing parameters to elucidate this behavior. Notably, special emphasis will be placed on molecular markers associated with the *Killer* and mating types, such as the STE (Specific Transducer of Erf1) genes, responsible for encoding proteins engaged in pheromone detection, signal transduction, and gene expression regulation, as well as the FUS1 and FUS2 genes, encoding transcription factors that activate the expression of genes specific to mating types a and α , respectively (Lee & Haber, 2015).

We extend our sincere gratitude to the funding institutions: FONDEC-UAQ, FONFIVE 2024 and CONAHCyT AI-S-26966 for their generous support and financial backing provided to Carlos Saldaña.

References

1. Bevan, E. A. & Makower, M. (1963). The Physiological Basis of the Killer Character in Yeast.
2. Sesti, F., Shih, T. M., Nikolaev, N., & Goldstein, S. A. N. (2001). Immunity to K1 Killer Toxin: Internal TOK1 Blockade membrane.
3. Lee, C. S., & Haber, J. E. (2015). Mating-type gene switching in *Saccharomyces cerevisiae*.

GENETIC VARIANT RS2074192 OF THE ACE2 GENE AND ITS ASSOCIATION WITH COVID-19 SEVERITY

Jason Enrique Vazquez-Navarro¹, Marcelo Barraza-Salas¹, Ingrid Cuevas-Cárdenas¹, Samuel Arroyo Carranza¹, José Manuel Salas Pacheco², Daniel Francisco Ramos-Rosales²

¹Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango, Unidad Dgo. Av. Veterinaria S/N, Circuito Universitario, Col. Valle del Sur C.P. 34120, ²Instituto de Investigación Científica, Universidad Juárez del Estado de Durango, Av. Universidad y Fanny Anitua S/N, Zona Centro, C.P. 34000.
E-mail: 1053515@alumnos.ujed.mx

The COVID-19 disease, caused by SARS-CoV-2, was first identified in China in December 2019 and was declared a pandemic by the WHO in March 2020¹. At the beginning of 2024, was estimated to have caused more than 335,000 deaths². The ACE2 protein, involved in the regulation of the renin-angiotensin system and mediating blood pressure, has been linked to the virus internalization into the host, suggesting that polymorphisms in the coding gene may play a significant role in the pathogenesis and severity of COVID-19³.

The aim of this study is to determine the genetic variant rs2074192 of the *ACE2* gene is associated with COVID-19 severity in outpatient cases from the state of Durango. Genomic DNA was isolated from SARS-CoV-2 positive patients, who were categorized into two groups based on symptom severity (asymptomatic-mild and moderate-severe). Genotyping was performed using a TaqMan probe on the QuantStudio 5 Real-Time PCR system. Statistical analyses were conducted using SPSS v.22, with a *p*-value < 0.05.

Differences in allelic and genotypic frequencies of the genetic variant were found, as well as an association of the polymorphism with severity in women under a dominant inheritance model (OR = 0.42 (0.20-0.87)). In conclusion, the rs2074192 variant of the *ACE2* gene is associated with symptomatic severity of COVID-19, acting as a protective factor.

References

1. Habas, K., Nganwuchu, C., Shahzad, F., Gopalan, R., Haque, M., Rahman, S., Majumder, A. A., & Nasim, T. (2020). Resolution of coronavirus disease 2019 (COVID-19). Expert review of anti-infective therapy, 18(12), 1201-1211.
2. COVID-19 cases, WHO COVID-19 dashboard. 2024 (s. f.). Datadot. <https://data.who.int/dashboards/covid19/cases?n=c>
3. Faustine, I., Marteka, D., Malik, A., Supriyanto, E., & Syafhan, N. F. (2023). Genotype variation of ACE and ACE2 genes affects the severity of COVID-19 patients. BMC research notes, 16(1), 194.

ABF1 IS A DNA BINDING PROTEIN INVOLVED IN DIFFERENT DNA PROCESSES IN *CANDIDA GLABRATA*

Laura-Angélica Vera-Salazar¹, Grecia Hernández-Hernández,
Alejandro De Las Peñas¹, Irene Castaño-Navarro¹

¹Instituto Potosino de Investigación Científica y Tecnológica, Camino a la presa San José #2055 CP:78216.
E-mail: icastano@ipicyt.edu.mx

Abf1 or ARS Binding Factor 1 is a DNA binding protein present in different fungal species. In *Candida glabrata*, an opportunistic pathogen yeast we have determined that is an essential gene for viability. And participates in subtelomeric silencing of several *EPA* genes responsible of adhesion to epithelial cells and contributes to the virulence factors of the yeast. Recently we showed that the overexpression and depletion of Abf1 cause loss of viability and aberrant nuclei segregation (Hernández-Hernández et al. 2021). Furthermore, we demonstrate the negative autoregulation and its ability to form homodimers with itself by Bimolecular Fluorescence Complementation (BiFC), likewise its nuclei localization. In this work we show its participation in UV damage response, as the heterologous complementation of Abf1 with its orthologous from *Saccharomyces cerevisiae* (Yu et al. 2009; Yarragudi, Parfrey, and Morse 2007; Loo et al. 1995), a non-pathogenic yeast close related to *C. glabrata*, we show the phenotypes complemented with the orthologous from *S. cerevisiae*.

References

1. Hernández-Hernández, G., L.A. Vera-Salazar, L. Castanedo, E. López-Fuentes, G. Gutiérrez-Escobedo, A. De Las Peñas, and I. Castaño. 2021. "Abf1 Is an Essential Protein That Participates in Cell Cycle Progression and Subtelomeric Silencing in *Candida Glabrata*." *Journal of Fungi* 7 (12). <https://doi.org/10.3390/jof7121005>.
2. Loo, S., P. Laurenson, M. Foss, A. Dillin, and J. Rine. 1995. "Roles of ABF1, NPL3, and YCL54 in Silencing in *Saccharomyces Cerevisiae*." *Genetics* 141 (3). <https://doi.org/10.1093/genetics/141.3.889>.
3. Yarragudi, Arunadevi, Laura Wegener Parfrey, and Randall H. Morse. 2007. "Genome-Wide Analysis of Transcriptional Dependence and Probable Target Sites for Abf1 and Rap1 in *Saccharomyces Cerevisiae*." *Nucleic Acids Research* 35 (1). <https://doi.org/10.1093/nar/gkl1059>.
4. Yu, Shirong, Julia B. Smirnova, Errol C. Friedberg, Bruce Stillman, Masahiro Akiyama, Tom Owen-Hughes, Raymond Waters, and Simon H. Reed. 2009. "ABF1-Binding Sites Promote Efficient Global Genome Nucleotide Excision Repair." *Journal of Biological Chemistry* 284 (2): 966-73. <https://doi.org/10.1074/jbc.M806830200>.

IRF4 REGULATES TNFAIP3 EXPRESSION IN CD4+ T CELLS OF CUTANEOUS T CELL LYMPHOMA AND T CELL ACUTE LYMPHOBLASTIC LEUKEMIA

Georgina Victoria Acosta¹, Magnolia del Carmen Ramírez Hernández¹,
Miranda Desireé López Olivera¹, Laura C. Bonifaz Alfonso^{2,3}

¹División de Investigación, Hospital Juárez de México, Av. Instituto Politécnico Nacional No. 5160 Col. Magdalena de las Salina C.P 07760, ²Unidad de Investigación Médica en Inmunoquímica, Unidad Médica de Alta Especialidad (UMAE) Hospital de Especialidades, ³Centro Médico Nacional Siglo XXI, Instituto Mexicano del Seguro Social, Mexico City, Mexico. Coordinación de Investigación en Salud, Centro Médico Nacional Siglo XXI, Instituto Mexicano del Seguro Social, Mexico City, Mexico.
E-mail: giviac@gmail.com

T cells coordinate adaptive immunity, specifically, CD4+ T are the central cells of adaptive immunity. The neoplasms that arise from the transformation of CD4+ T cells are cutaneous T cell lymphoma (CTCL) and T cell acute lymphoblastic leukemia (T-ALL). CTCL comprises a group of primary lymphomas characterized by malignant skin homing T cells, among which Mycosis fungoides (MF) is the most common variant. Early CTCL is characterized by skin patches and plaques that may progress into tumors. On the other hand, T-ALL is an aggressive immature T cell neoplasm caused by the malignant transformation of T cells immature precursors. T-ALL is characterized by the uncontrolled production of malignant precursor cells in the bone marrow and thymus. In both neoplasms, the NFκB signaling pathway is constitutive active. One of the endogenous negative regulators of NFκB is the protein TNFAIP3, a dual ubiquitin editing enzyme. The expression of TNFAIP3 is decreased in several lymphomas and autoimmune diseases such as systemic lupus erythematosus, suggesting that this could contribute to their pathogenesis. To analyze the mechanism that leads to TNFAIP3 gene silencing in CTCL and T-ALL, we carried out a bioinformatic search to identify probable binding sites for transcription factors involved in the regulation of the gene. We found a probable binding site for the transcription factor IRF4. The site was validated by chromatin immunoprecipitation assays using the HBT-176 and Jurkat cell lines as model systems of CTCL and T-ALL respectively, and peripheral blood mononuclear cells (PBMCs) from healthy donors as controls.

Objective. To analyze repressive and open histone marks and validate if the probable binding site for IRF4 is functional in the promoter region of the TNFAIP3 gene.

Results. RT-PCR was used to determine TNFAIP3 mRNA expression in PBMC, HTB-176 and Jurkat cells. TNFAIP3 expression was decreased in HTB-176 and Jurkat cells compared with PBMCs. EPD and JASPAR databases for the bioinformatic search for the probable IRF4 binding site were used. IRF4 bound to the TNFAIP3 promoter in HTB-176 and Jurkat cells, but not in PBMCs from healthy controls. We detected greater enrichment of H3K27me3 in the promoter of TNFAIP3 gene in HTB-176 and Jurkat cells compared with PBMCs from healthy controls. In the case of H3K4me3, we found a more intense mark in PBMCs than in HTB-176 and Jurkat cells.

Conclusions. The results obtained suggest that the low expression of TNFAIP3 gen in the HTB-176 and Jurkat cell lines is due to the enrichment of histone mark H3K27me3 as well as the binding of IRF4, since this transcription factor can act as a transcriptional repressor.

EPIGENETIC MECHANISMS ASSOCIATED WITH THE EXPRESSION OF PROTOCADHERIN 18 IN A MODEL OF EPITHELIAL-MESENCHYMAL TRANSITION IN MCF10A, MCF7, AND MDA-MB-231 CELLS

Pablo Sinuhé Gómez González, Ángel Salmerón Hernández, Ana Cecilia Sánchez Trujillo, Gabriela Rodríguez Rodríguez, Ángel Zarain Herzberg*

Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México, UNAM. Av. Universidad 3000, 04510, Mexico City.

*Correspondence: zarain@unam.mx

Background. Recently, protocadherins have emerged as intriguing players in cancer biology¹. Protocadherin 18 (PCDH18) has shown a decrease in its expression mainly associated with hypermethylation of its promoter in colorectal cancer, and more recently², by *in silico* analysis, it was observed that the expression decreases both in cell lines and in breast cancer tumor samples³. The low expression of some protocadherins has been implicated in the progression of Epithelial-Mesenchymal Transition (EMT), for example, PCDH9 in hepatocellular carcinoma⁴ and protocadherin alpha 3 in squamous cell lung carcinoma⁵.

Objective. To analyze the expression of PCDH18 and the effect of inhibitors of DNA methyltransferases (DNMTi) and histone deacetylases (HDACi) in the EMT model in MCF10A, MCF7 and MDA-MB-231 cells.

Methods. We used MCF10A, MCF7, and MDA-MB-231 cells cultured according to ATCC recommendations. We treated the cells with 5ng/ml of TGF- β and 50ng/ml of EGF, a well-established EMT induction model (EMTi). We then performed treatments with DNMTi (5-azacytidine, 5-aza-2-deoxycytidine) and HDACi (Resveratrol (RSV) and LBH589). After the treatments, we conducted RNA extraction and RT-qPCR to analyze mRNA expression. We also evaluated the morphology of the cells using microscopy under different treatments.

Results. The expression of PCDH18 decreases in response to the effect of the EMTi model, especially in MCF10A and MCF7 cells. The expression was higher in the three cell lines treated with DNMTi and HDACi. The combined treatment of the EMTi plus LBH589, and EMTi plus RSV prevents the expression of PCDH18, mainly in MCF10A. The results suggest that restoring PCDH18 expression in MCF10A and MCF7 attenuates the progression of EMT.

Conclusions. We observed decreased PCDH18 expression in response to the EMTi model in MCF10A and MCF7 cells. However, the use of 5-azacytidine, 5-aza-2-deoxycytidine, LBH589, and RSV effectively increased the expression of PCDH18 in non-tumor breast cells and luminal A subtype (MCF7) and triple-negative (MDA-MB-231) breast cancer cells, offering a potential therapeutic strategy. Overall, these results open new avenues for further research and developing novel interventions for EMT-related diseases.

References

1. Barros-Dutra *et al.*, 2020. Asian Pac J Cancer Prev. 21 (12): 3667-3688.
2. Zhou *et al.*, 2017. Scientific Reports. 7: 2819.
3. Hernández-Oliveras *et al.*, 2021. J Cell Commun Signal. 16(3): 461-474.
4. Zhu *et al.*, 2014. Biochem Biophys Res Commun. 452(3). 567-74.
5. Tao *et al.*, 2022. Genes Genomics. 44 (2): 211:218.

PURIFICATION AND VALORIZATION OF URBAN EFFLUENTS USING PHOTOBIOREACTORS OPERATED WITH *ANABAENA INAEQUALIS*

Wendy Zárate-Hernández¹, Roger Emmanuel Sales-Pérez², Norma Alejandra Vallejo-Cantú³,
Juan Manuel Méndez-Contreras⁴

Instituto Tecnológico de Orizaba¹, Ote. 9, Emiliano Zapata, 94320 Orizaba, Ver.
juan.mc@orizaba.tecnm.mx

In recent years, the global generation of wastewater has increased significantly. This rise is mainly attributed to population growth and the expansion of industrial processes. Such wastewater typically contains chemical and microbiological agents, with high levels of organic matter, nitrogen, and phosphorus being particularly notable. These components can cause environmental damage, the destruction of ecosystems, intestinal diseases in humans, and other serious issues¹. A biotechnologically viable alternative for wastewater treatment is the use of microalgae. This unconventional alternative is characterized by high efficiency, the absence of greenhouse gas emissions, low costs, and the production of biomass².

In this study, the microalga *Anabaena inaequalis*³ was used for the treatment of municipal wastewater. Before the experiments, the species was propagated in Soil-Water Medium⁴, a synthetic medium suitable for its growth. After 15 days of propagation, a concentration of 3.16×10^5 cells/mL was achieved. The wastewater treatment was evaluated over 21 days in batch photobioreactors, resulting in a reduction of organic matter concentration, analyzed through COD levels. Among the evaluated inoculum concentrations (5%, 15%, and 25%), the 25% inoculum achieved the highest COD removal (95%) and the greatest cell growth, reaching concentrations higher than those in the synthetic medium (1.82×10^6 cells/mL). Finally, the process successfully removed inorganic contaminants (nitrogen and phosphorus), which were utilized by the microalga for growth and the generation of microalgal biomass with potential clinical and energy applications, representing promising biotechnological alternatives for the future.

References

1. Ramírez, L. (2020). Agrobiología: Una visión general y sus aplicaciones.
2. Medina Mojica, I. Y. (2020). Las microalgas una alternativa para la remoción de materia orgánica de aguas residuales domésticas.
3. Cyanobacteria, and other aquatic objects. University of New Hampshire Center for
4. Freshwater Biology. <http://cfb.unh.edu/phycokey/phycokey.htm> 18 May 2022

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly a protein or a complex of atoms, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

IMMUNOLOGY & PARASITOLOGY

ROLE OF METALLOPROTEINASES IN THE MIDGUT OF *Aedes Aegypti* MOSQUITOES INFECTED WITH DENGUE VIRUS

Sofía Alanís-Muñoz¹, Edgar Quezada-Ruiz², Ángel T. Tello-López¹, Grecia Díaz¹, Jorge Cime-Castillo¹, Juan Ludert², Humberto Lanz-Mendoza¹

¹Centro de Investigaciones Sobre Enfermedades Infecciosas, Instituto Nacional de Salud Pública, Secretaría de Salud, Cuernavaca, Morelos, México. ²Departamento de Infectómica y Patogénesis Molecular, Centro de Investigación y de Estudios Avanzados del IPN, Ciudad de México, CDMX.
E-mail: sofisam16@gmail.com

Dengue virus (DV) is the causative agent of Dengue disease and is transmitted mainly by the bite of female mosquitoes of the *Aedes aegypti* species. This virus presents different tropisms for some tissues¹ and must overcome a series of physical and immunological barriers to be released within the vector's hemocoel and, thus, subsequently infect other tissues. The main barrier that can prevent the establishment of the pathogen inside the mosquito is the intestinal epithelium². The most impermeable barrier this organ presents is the basal lamina, an acellular structure made up of different proteins with associated carbohydrates, rich mainly in laminin and type IV collagen³. Blood ingestion causes an increase in the size of the intestine, causing microfractures in the basal lamina that must be rapidly repaired⁴. CHIKV may delay the repair of these microfractures through modulation of MMPs, increasing collagen degradation and allowing further dissemination of CHIKV in the insect hemocele^{5,6}. However, what may occur in the Dengue Virus model is unknown. This study aims to associate the participation of metalloproteinases with disseminating the virus in the *Ae. aegypti* mosquito. So the change in gene expression of *Aemmp1* and *Aemmp2* was analysed in the presence of DV. RT-qPCR performed an expression analysis of these 2 genes, where a higher relative expression of *Aemmp1* mRNA was observed at 24 hr post-infection. The enzyme encoding this protein is of the gelatinase type. Therefore, an enzyme activity analysis was performed using a zymogram assay, where a band of higher lytic activity was observed in the intestines of mosquitoes that ingested the virus. Interestingly, by RT-qPCR, we detected viral genomes in the abdomens of mosquitoes that were evaluated at 24 hr post-infection, with a prevalence of 80%. Batimastat⁵, a specific inhibitor for MMPs, decreased the prevalence of infection. With these results, we suggest that MMPs could be involved in virus dissemination early in the infection of *Aedes aegypti* mosquitoes.

References

1. Guzman, M. G., & Harris, E. (2015). Dengue. *Lancet* (London, England), 385(9966), 453–465.
2. Franz, A. W., A. M. Kantor, L. Passarelli y R. Clem. 2015. Tissue Barriers to Arbovirus Infection in Mosquitoes. *Viruses*. 7: 3741-3767.
3. Yurchenco P. y J. O'Rear. 1994. Basal lamina assembly. *Current Opinion in Cell Biology*. 6:674-681.
4. Armstrong, Philip M.; Ehrlich, Hanna Y.; Magalhaes, Tereza; Miller, Megan R.; Conway, Patrick J.; Bransfield, Angela; Misencik, Michael J.; Gloria-Soria, Andrea; Warren, Joshua L.; Andreadis, Theodore G.; Shepard, John J.; Foy, Brian D.; Pitzer, Virginia E.; Brackney, Doug E. (2019). Successive blood meals enhance virus dissemination within mosquitoes and increase transmission potential. *Nature Microbiology*
5. Dong S. V. Balaraman, A. M. Kantor, J. Lin, D. Grant, N. Held, A. Franz. (2017). Chikungunya virus dissemination from the midgut of *Aedes aegypti* is associated with temporal basal lamina degradation during bloodmeal digestion. *PLoS Negl Trop Dis*. 11(9).
6. Kantor A., S. Dong, N. Held, E. Ishimwe, A. Passarelli, R. Clem y A. Franz. (2017). Identification and Initial Characterization of Matrix Metalloproteinases in the Yellow Fever Mosquito, *Aedes aegypti*. *Insect Mol Biol*. 26(1):113-126.
7. Cui Y. D. Grant, J. Lin, X. Yu y A. Franz. (2019). Zika Virus Dissemination from the Midgut of *Aedes aegypti* is Facilitated by Bloodmeal-Mediated Structural Modification of the Midgut Basal Lamina. *Viruses*. 11:1056.

INVOLVEMENT OF THE ASMASA6 FROM *E. HISTOLYTICA* IN THE REPAIR OF PLASMA MEMBRANE DAMAGE INDUCED BY PORE-FORMING PROTEINS

Sairy Yarely Andrade-Guillen, Fátima Berenice Ramírez-Montiel, Fátima Tornero-Gutiérrez, Ángeles Rangel-Serrano, Itzel Páramo-Pérez, José Martínez-Álvarez, Fernando Anaya-Velázquez, Bernardo Franco Bárcenas, Luis Felipe Padilla-Vaca

Departamento de Biología, División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Noria Alta s/n. Guanajuato, Gto., C.P. 36050, México. 4731217354
E-mail: padillaf@ugto.mx; sy.andradeguillen@ugto.mx

Entamoeba histolytica is a pathogenic protozoan that infects humans, mostly affecting people living in poverty, lacking basic sanitation, and having limited access to healthcare facilities. During the infective process, this parasite is confronted by different elements of the host's immune system, where the main mechanism of action is damage to the parasite's plasma membrane (PM). In our research group, the involvement of acid sphingomyelinases (aSMases) in repairing the damage to *E. histolytica*'s PM was described. It was demonstrated that trophozoites overexpressing the *EhaSM6* gene secrete twice as much aSMase activity compared to the control strain, and when exposed to pore-forming molecules (SLO, Magainin, and β -Defensin), they show an increase of 6 to 25 fold in secreted aSMase activity associated with higher trophozoite viability. In response to damage, lysosomes undergo exocytosis, forming patches at the site of injury and releasing their contents into the extracellular medium, including aSMase6, which hydrolyzes sphingomyelin and generates ceramide. The *EhaSM6* gene encodes a 48.8 kDa protein with a predicted signal peptide for protein secretion and a processing site in the C-terminal region. It is suggested that this mechanism of repairing damage to the plasma membrane mediated by aSMases could be functional and favor the parasite's survival under conditions that amoebic trophozoites may face in the host.

References

1. Ramírez-Montiel, F., Mendoza-Macías, C., Andrade-Guillen, S., Rangel-Serrano, Á., Páramo-Pérez, I., Rivera-Cuéllar, P. E., y Padilla-Vaca, F. 2019. Plasma membrane damage repair is mediated by an acid sphingomyelinase in *Entamoeba histolytica*. *PLoS pathogens*, 15(8).
2. Zeidan YH, Hannun YA. Activation of acid sphingomyelinase by protein kinase C δ -mediated phosphorylation. *Journal of Biological Chemistry* 2007;282:11549– 11561

MACROPHAGE MIGRATION INHIBITORY FACTOR (MIF) CONTRIBUTES TO MACROPHAGE-M1 POLARIZATION BY REGULATING MITOCHONDRIAL FUNCTION

Antonio Andrade-Meza^{1,2}, Christian Daniela Ortiz-Robles^{1,4}, Imelda Juárez-Avelar^{1,3},
Emma S. Calderón-Aranda⁴, Miriam Rodríguez-Sosa¹

¹Laboratorio de Inmunidad Innata, Unidad de Investigación en Biomedicina (UBIMED), Facultad de Estudios Superiores Iztacala (FES-I), Universidad Nacional Autónoma de México (UNAM); Tlanepantla, Estado de México, 54090. México. ²Programa de Doctorado en Ciencias Biomédicas, UNAM, Coyoacán, Mexico City, 04510, Mexico. ³Programa de Doctorado en Ciencias Biológicas, UNAM, Coyoacán, Mexico City, 04510, Mexico. ⁴ Departamento de Toxicología, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV), Mexico City, 07360, Mexico. Antonio Andrade Meza. Av de los Barrios #1, Unidad de Investigación en Biomedicina (UBIMED) Lab5, planta alta. Tlanepantla, Edo. Méx, 54090, México. jnksaam@gmail.com, +525585971608

Macrophage migration inhibitory factor (MIF) is a proinflammatory cytokine produced in immune cells in response to inflammatory stimuli and is involved in several diseases in which macrophage polarization is crucial. However, it is not known whether MIF influences macrophage polarization. Our study aimed to assess whether MIF plays a role in macrophage polarization. Bone marrow-derived macrophages (BMDMs) from MIF knockout (*Mif*^{-/-}) mice were polarized toward M1 with lipopolysaccharide and interferon- γ (LPS+IFN- γ), M2 with interleukin (IL)-4+IL-13 or treated with recombinant MIF and compared to WT macrophages treated in the same way. We analyzed some phenotypic markers for M1 (reactive oxygen species, ROS; and nitric oxide, NO), M2 (arginase activity), and mitochondrial function. MIF deficiency decreased the production of ROS, NO and mitochondrial membrane potential (MMP) and impaired oxidative phosphorylation (OXPHOS) in M1-*Mif*^{-/-} macrophages, supporting that MIF contributes to M1 polarization. Arginase activity, OXPHOS and MMP were reduced in M2-*Mif*^{-/-} macrophages. These findings demonstrate that MIF plays a role in the maintenance of mitochondrial function with slight differences depending on macrophage polarization type and contributes to macrophage reprogramming by manipulating metabolic pathways with a potential for therapeutic purposes.

EXOSOMES SECRETED BY PBMCS INFECTED WITH *HELICOBACTER PYLORI* TRANSPORT TNF- α MODULATES EMT, MIGRATION AND INVASION IN AGS CELLS

Josefina Atrisco-Morales^{1,2}, Juan Carlos Juárez-Cruz³, Miguel Ángel Mendoza-Catalán⁴,
Dinorah Nashely Martínez-Carrillo^{1,2}, Mónica Ramírez⁵, Napoleón Navarro Tito³,
María Angélica Santana-Calderón⁶ and Gloria Fernández-Tilapa^{1,2}

¹Laboratorio de Investigación Clínica, Laboratorio de Investigación en Biomoléculas². ³Laboratorio de Biología Celular del Cáncer, ⁴Laboratorio de Biomedicina Molecular, ⁵CONACYT-UAGro, Chilpancingo de los Bravo, Gro, ⁶Laboratorio de Inmunología Celular, CIDC, IICBA-UAEM, Cuernavaca, Morelos.
E-mail: gfernandezt@uagro.mx

Background. *Helicobacter pylori* (*H. pylori*), release of cytokines such as TNF- α , contribute to the development and progression of gastric cancer (GC). In the gastric tumor microenvironment (TME), the interaction between immune cells and tumor cells through exosomes contributes to tumor progression. Previously, our group reported an increase in soluble and exosome-contained cytokines (IL-1 β , IL-6, TNF- α , IL-10, IL-17A, IL-21, IL-22) secreted by PBMCs infected by *H. pylori*¹. **Objective.** The objective of this study was to evaluate the effect of exosomes and TNF- α , secreted by peripheral blood mononuclear cells (PBMCs) infected with *H. pylori* on EMT, migration, invasion, activation of Akt, NF- κ B, and levels of E-cadherin, vimentin, and Twist1, in AGS cells. **Material and Methods.** Exosomes released by PBMCs infected with *H. pylori* (PBMCs-*Hp*) and uninfected PBMCs (PBMCs-C) were obtained by ultracentrifugation. The effect of exosomes on the EMT, migratory and invasive processes of AGS cells was evaluated through Western blot (WB) or wound closure and invasion assays with Transwell chambers, respectively. AGS cells were treated with recombinant TNF- α (rTNF- α), PBMCs-*Hp* exosomes, chemical Akt inhibitor (perifosine), neutralizing antibody for TNF- α (anti-TNF- α), or with combined treatment, to determine the cell migration and invasion. The level of Akt, Akt-pSer473, NF- κ B p65, NF- κ B p65-pSer536, Twist1, E-cadherin, and vimentin was determined by WB. **Results.** Exosomes secreted by PBMCs-*Hp* induced an increase in the migration and invasion of AGS cells. It was confirmed that rTNF- α promotes the migration and activation of Akt, and treatment with anti-TNF- α reduced this effect. Inhibition of Akt phosphorylation decreased migration. On the other hand, in AGS cells, exosomal TNF- α from PBMCs-*Hp* promoted EMT, migration, invasion, NF- κ B activation, Twist1 and E-cadherin overexpression, without changes in Akt activation or levels of vimentin. The changes promoted by exosomal TNF- α were reversed by anti-TNF- α and perifosine. **Conclusion.** The present study demonstrates that exosomal TNF- α secreted by PBMCs-*Hp* contributes to the progression of gastric cancer, through the promotion of EMT, migration and invasion of AGS cells.

References

1. Atrisco-Morales, J., Ramírez, M., Castañón-Sánchez, C.A., Román-Román, A., Román-Fernández, I.V., Martínez-Carrillo, D.N., García-Arellano, S., Muñoz-Valle, J.F., Rodríguez-Ruiz, H.A., Fernández-Tilapa, G., (2022). International Journal of Molecular Sciences, 23, 8801.

INDUCTION OF TRAINED IMMUNITY BY BOVINE RESPIRATORY DISEASE VACCINE IN CATTLE

Marisol Báez-Magaña, Juventino García-Vilchis¹, Gladys Romero-Corona¹, Cristian Eduardo Vázquez-Anguiano¹, Alejandra Ochoa-Zarzosa¹ and Joel Edmundo López-Meza¹

¹Centro Multidisciplinario de Estudios en Biotecnología, Facultad de Medicina Veterinaria y Zootecnia, Universidad Michoacana de San Nicolás de Hidalgo, Km 9.5 Carretera Morelia-Zinapécuaro, Posta Veterinaria. 58893, Morelia, Michoacán. mbaez@umich.mx

Trained immunity (TI) is a form of memory present in cells of the innate immune system¹. TI enables these cells to mount a strong response to heterologous stimuli. This training induces metabolic and epigenetic changes in the innate cells, resulting in increased production of proinflammatory mediators. The study of TI in dairy cattle is an innovative approach to treating different animal health problems through the research and enhancement of vaccines. Bovine respiratory disease (BRD) is relevant to cattle because it causes 50% of deaths in young animals despite vaccination. This project aims to analyze the BRD vaccine's effects on cattle's TI response. For this, peripheral blood samples were collected from twenty-two Holstein cows from 1 month to 2 years old. The animals were categorized into four groups: 1) calves, 2) heifers, 3) pregnant cows vaccinated once, and 4) pregnant cows vaccinated twice. Sampling was conducted 15 days before vaccination and, 14 and 30 days after vaccination (first challenge). The cytokines secretion (TNF- α and IL-6) and ROS production by peripheral blood mononuclear cells (PBMCs) challenged with pathogenic microorganisms were analyzed. Results showed that the vaccine did not affect the PBMC populations, and TNF- α and IL-6 at the systemic level were found at basal levels. The ROS production in PBMCs significantly increased (~ 2,000 and ~10,000-foldss) 14 and 30 days after vaccination. These findings were consistent across all four groups. However, in PBMCs challenged with *Escherichia coli*, *Klebsiella pneumoniae*, *Candida albicans*, and *Pseudomonas aeruginosa* (second challenge), the secretion of IL-6 increased approximately 3-fold in the calves and heifers groups 14 days after vaccination. In contrast, TNF- α production showed an upward trend 30 days after vaccination in the calves, heifers, and pregnant cows vaccinated once groups. These results suggest that the BRD vaccine can induce TI in the early stages of cattle development.

MITOCHONDRIAL DYNAMICS IN B CELL RESPONSE AGAINST LIPIDIC ANTIGENS

Giovanna Barrera Avela, Iván Galarce Sosa, Elías Isidro Magallán, José Rundquist Sánchez, Rodrigo Monteagudo Ramos, Gabriel Trejo Sánchez, Claudia Albany Reséndiz Mora, Isabel Baeza Ramírez and Carlos Wong Baeza

¹Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Laboratorio de Biomembranas, Prolongación Carpio y Plan de Ayala s/n, Colonia Santo Tomás, Alcaldía Miguel Hidalgo, Ciudad de México, CP.11340.
E-mail: aveleida@gmail.com

Mitochondria has a crucial role in B cell response through changes in mitochondrial dynamics and mitochondrial reactive oxygen species (mt-ROS) production, which directly influence the mitochondrial membrane potential ($\Delta\Psi_m$)¹. Cell membrane lipids can present antigenic properties when they form stable non-bilayer phospholipid arrangements (NPAs), which can be induced by some drugs such as chlorpromazine. This response against NPAs can induce the germinal center reaction where B cells undergo class-switch recombination and somatic hypermutation, leading to the production of high-affinity anti-NPAs IgG antibodies and B cell differentiation into memory B cells or plasma cells. Anti-NPA antibodies can generate autoimmunity and have been detected in diseases such as Systemic Lupus Erythematosus². We analyzed the mitochondrial dynamics, mt-ROS production, and $\Delta\Psi_m$ by flow cytometry in a murine model that resembles human lupus induced by the administration of liposomes bearing NPAs. We found that mitochondrial fusion increased the $\Delta\Psi_m$ and decreased mt-ROS production, favoring the differentiation of germinal center B cells into memory B cells in mice administered with lipidic particles compared to the control. In contrast, mitochondrial fission increased mt-ROS and decreased $\Delta\Psi_m$, which promotes plasma cell differentiation. Therefore, it was established that mt-ROS contributes to the modulation of mitochondrial dynamics and $\Delta\Psi_m$ in B cell response.

References

1. Xie, J. H., Li, Y. Y., & Jin, J. (2020). The essential functions of mitochondrial dynamics in immune cells. *Cellular & molecular immunology*, 17(7), 712–721.
2. Reséndiz-Mora, A., Tescucano, A., Barrera-Aveleida, G., Sotelo-Rodríguez, A., Nevárez-Lechuga, C., Galarce-Sosa, I., Wong-Baeza, I. & Baeza, I.W.-B., C. (2022) Anti-Non-Bilayer Phospholipid Arrangement Antibodies Trigger an Autoimmune Disease Similar to Systemic Lupus Erythematosus in Mice. In Lionaki, M.S., Karagiannis, M. (eds) *Systemic Lupus Erythematosus–Pathogenesis and Management*. IntechOpen, London, United Kingdom

ANALYSIS OF ENTOMOPATHOGENIC PROPERTIES OF *P. SYRINGAE* CDBB-B1293 AGAINST *Aedes Aegypti*, THE MAIN VECTOR OF THE DENGUE VIRUS

¹Valeria Calderón Frontana, ¹Jose Ángel Rubio Miranda, ²Daniel Talamás Lara, ³Daniel A. Estrada Bárcenas, ³Juan Carlos Estrada Mora, ¹Carlos Vázquez Calzada, ¹Leticia Cortés Martínez, ¹Febe Elena Cazares Raga, ¹Fidel de la Cruz Hernández-Hernández

¹Departamento de Infectómica y Patogénesis Molecular. ²Laboratorio Nacional de Servicios Experimentales.

³Colección Nacional de Cepas Microbianas y Cultivos Celulares. Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Av. Instituto Politécnico Nacional No. 2508. Col. San Pedro Zacatenco. C.P. 07360 México, Ciudad de México. Tel: (55) 57473800 ext.5646
jose.rubio@cinvestav.mx; cruzcruz@cinvestav.mx

Mosquitoes of *Aedes* genus are the vectors of the dengue virus and represent a major public health problem. Currently, the main mosquito control strategies are based on the use of chemical insecticides and alternatives as biocontrol are under development, including the use of entomopathogenic microorganisms such as *Bacillus thuringiensis* (Bt). It has been shown that bacterial strains of the genus *Pseudomonas* including *P. aeruginosa* and *P. fluorescens* have entomopathogenic activity and can also be considered as biological controllers of mosquito populations and suggest the possibility that other strains of this genus could be amenable for mosquito biocontrol. **Objective:** Analyze the larvicidal activity of the *Pseudomonas syringae* CDBB-B1293 strain against *Ae. aegypti* and characterize its entomopathogenic mechanisms. **Material and methods:** To analyze larvicidal activity, first instar larvae of *Ae. aegypti* were infected with 10^{10} colony forming units (CFU) and maintained at 28°C and their viability was monitored up to 6 days after infection. For the analysis of biofilm production, *P. syringae* strain was cultured in 5 ml of nutrient broth and incubated at 28°C for 24 hours. Subsequently, 100µl of bacterial culture was inoculated in 24-well culture plates and biofilm production was quantified after 24 hours using crystal violet. For biofilm deposition on larval surface, third instar *Ae. aegypti* larvae were infected with 10^{10} CFU and after 24 hours infection, scanning electron microscopy (SEM) were conducted under a JSM-6510LV microscope. **Results:** Larvae infected with *P. syringae* had a mortality rate of 90% after 6 days of infection and developing only to the third instar. *P. syringae* was able to develop biofilm both in culture plates and on the surface of *Ae. aegypti* larvae. **Discussion and Conclusion:** The present work demonstrated that the *P. syringae* strain CDBB-B1293, a plant pathogenic bacterium, has larvicidal activity against *Ae. aegypti*, an effect that had only been reported for *P. aeruginosa*, *P. protegens*, *P. fluorescens* and *P. putida* against larvae of mosquitoes of the genus *Culex*, *Aedes* and *Anopheles*. Furthermore, it was demonstrated that *P. syringae* CDBB-B1293 can produce biofilm on the larval surface of *Ae. aegypti*. It remains to be analyzed what other virulence factors *P. syringae* CDBB-B1293 produce during its interaction with *Ae. aegypti* larvae.

EFFECT OF SUPPLEMENTATION WITH L-ARGININE AND L-SERINE ON THE CYTOKINE PROFILE OF NEONATES BORN BY CESAREAN SECTION AND VAGINAL DELIVERY

Ingrid Yaritzit Carreon¹, María Angélica Santana-Calderón² and Otoniel Rodríguez Jorge³

¹Universidad Autonoma del Estado de Morelos (UAEM), ²Centro de Investigacion en Dinamica Celular (CIDC).

^{1,2}Av. Universidad 1001, Chamilpa, 62210 Cuernavaca, Mor. Tel: +52 (777)3297000 ext. 3666/3667.

E-mail: ingrid.carreon@uaem.edu.mx

Birth is a critical moment in immune system regulation. The adaptation from the nearly sterile intrauterine environment to an extrauterine one rich in antigens presents a significant immunological challenge. The process involves changes in immune cell metabolism, which are part of this challenge, as meeting bioenergetic demands entails obtaining metabolites from a microenvironment not fully understood immunologically. In the postnatal stage, the immune system slowly transition from an innate response towards the characteristics necessary for fine selectivity that is displayed by the adaptive immune system. The innate immune system leads neonatal protection, while the adaptive one requires extended time to respond fully. However, this doesn't mean adaptive system functions are absent; in fact, it also have an innate-like response and is biased towards tolerance. This innate an tolerant profile allows for the establishment of the baby's microbiota and the sudden encounter with an antigen full world. Later, the neonatal immune system predominantly builds Th2 immune responses rather than Th1 responses. This imbalance is believed to explain neonates' susceptibility to virus and bacteria infections. In 2019, 47% of total deaths in children under 5 were during the neonatal period, with 24% attributed to infections. Birth, as seen so far, is a determining process in immune system regulation. Alterations could interfere with necessary attributes in early life. Without these, neonates would not only be susceptible to infections but could also present immunodeficiencies or autoimmune conditions. Cesarean section, compared to vaginal birth, alters the neonatal immune response. While it's a surgical intervention ensuring the health of both mother and baby, it lacks the same cellular and metabolic signals coordinating the neonatal immune response in vaginal birth. These include physical and oxidative stress, vaginal microbial colonization, among others. The absence of these cellular processes may result in predisposition to short- and long-term conditions (allergies, asthma, inflammatory problems). The current panorama demands seeking ways to regulate the neonatal immune system response. Supplementation with amino acids, such as L-arginine and L-serine, has been proved important for the function and activation of CD4+ T lymphocytes. In this study, we compared the cytokine profiles in CD4+ T cells from newborns delivered vaginally and by cesarean section, focusing on the role of supplementation with L-arginine and serine as possible regulators of the neonatal immune system. Our preliminary results indicate that supplementation with Arg and Ser reduces the production of all the Th cytokines tested. Intriguingly, the supplementation augments T cell viability. Our results could have an impact in the culture of T cells for immune interventions.

References

1. Pieren DK, Boer MC, Wit J. (2022). The adaptive immune system in early life: The shift makes it count. Doi: 10.3389/fimmu.2022.1031924.
2. Gurdeniz G, Ernest M, Rago D, Kim M. (2022). Neonatal metabolome of caesarean section and risk of childhood asthma. Doi: 10.1183/13993003.02406-2021.
3. Developed, E., Group, IN., Mortality, C. & Estimation, M. (2019) Child Mortality 2019.
4. Longkun D, Lu G. (2023). Factors of Fetal Origin in the Regulation of Labor Initiation and Preterm Birth, Maternal-Fetal. Doi: 10.15252/ebr.202256352.
5. Calanis K, Rodríguez O, et al. (2023). Unique Characteristics of Neonatal T Cells, Medical Research Archives.
6. Raynor J, Chi H. (2024). Nutrients: Signal 4 in T cell immunity. doi: 10.1084/jem.20221839.

ASSOCIATION OF THE METABOLIC STATE AND ACTIVATION RESPONSE OF NEONATAL AND ADULT CD4⁺ AND CD8⁺ T LYMPHOCYTES

Francisco Castañeda-Gómez¹, María Angélica Santana-Calderón¹, Otoniel Rodríguez Jorge¹

¹Laboratorio de inmunología celular y de sistemas, Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México.
e-mail: orj@uaem.mx, cel: +52 (777)3297000 ext. 3666/3667

The activation of T lymphocytes is crucial for the immune response, and this process is linked to highly regulated metabolic changes. Naïve T lymphocytes rely on catabolic oxidative metabolism (β -oxidation) and oxidative phosphorylation (OXPHOS). They undergo metabolic reprogramming towards glycolytic metabolism upon activation. This reprogramming allows them to meet the energy demands associated with clonal expansion and effector functions. Different subsets of T cells have distinct metabolic profiles. A byproduct of mitochondrial metabolism is the generation of reactive oxygen species (ROS), which can act as signaling molecules to trigger cellular responses but on excess leads to cellular stress. Of notice, most of what we know about metabolism during T cell activation has been described in adult T cells. Newborns are highly susceptible to infections by intracellular pathogens and have a characteristic immune response adapted to the challenges of birth, leaving them vulnerable to pathogens. Neonatal T cells are highly glycolytic in basal conditions and have low effector functions. Evaluating the metabolic state of neonatal CD4⁺ and CD8⁺ T lymphocytes in basal and stimulated conditions will allow us to contribute knowledge on the relationship between cellular metabolism and the activation of these cells. These data will feed computational models to explain the mechanism behind the limited response of neonatal T lymphocytes and propose strategies to achieve proper activation of these cells. In this work, lipid consumption, glucose consumption, mitochondrial ROS production, and mitochondrial mass and membrane potential were assessed through flow cytometry using fluorescent probes. Our preliminary results indicate that neonatal CD4⁺ and CD8⁺ T cells consume glucose and lipids in basal conditions, but much more after TCR/CD28 activation.

STIMULATING THE IMMUNE FUNCTION OF NEONATAL CD4⁺ T CELLS WITH CYTOKINES

Alejandra Cedillo Baños¹, María Angélica Santana-Calderón¹ and Otoniel Rodríguez Jorge¹

¹Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos. Av. Universidad 1001, Colonia Chamilpa, Cuernavaca, Morelos, México. Tel (777) 3297000 ext. 3666/3667, e-mail: alejandra.cedilloban@uaem.edu.mx

Newborns are a population highly susceptible to infections, exhibiting a high morbidity and mortality rate. Their immune system differs from that of adults, due to adaptations at birth and the transition from a sterile environment to one filled with foreign antigens. These adaptations make the newborns tolerant to many antigens but leave them vulnerable to infections.

Neonatal CD4⁺ T cells lack immunologic memory and have an immune bias toward tolerance and barrier immunity (Th2 type). In response to stimulation, they show a limited production of effector cytokines; however, they respond with a higher proliferation rate than adult cells.

Notably, neonatal T cells have demonstrated the capacity to mount responses like those of their adult counterpart in specific conditions. For instance, in a murine model of influenza virus infection in the respiratory tract, neonatal CD4⁺ T cells responded similarly or even more robustly than adult cells. Additionally, the BCG vaccine in neonates has been shown to induce a Th1-type response.

Efficient effector response relies on antigen-presenting cells (APCs), such as dendritic cells. A higher antigen presentation capacity by APCs is associated with increased expression of co-stimulatory molecules and cytokines. We hypothesize that neonatal T-cell responses could show a more robust effector capacity in the presence of relevant cytokines, such as IL-1 β , IL-6, IL-12, and IFN- β .

Therefore, in this work, we aim to characterize the activation response of neonatal CD4⁺ T cells to stimulation through the TCR/CD28 in the presence of IL-12, IFN- β , IL-1 β , or IL-6 cytokines. Cellular activation and phenotypes will be assessed based on the expression of activation markers like CD69 and CD25, effector cytokine production, expression of transcription factors, and cell proliferation as compared to the response of adult CD4⁺ T cells, all of which will be evaluated through flow cytometry.

Our preliminary results suggest that, while in adult cells the supplementation with cytokines generally does not affect effector cytokine production, except for IL-1B; in neonates, cytokine supplementation results in an increase in effector cytokine production compared to the control stimulated through the TCR/CD28, with IL-6 supplementation having a greater effect. These data suggest that there is a differential response between neonatal and adult CD4⁺ T cells to the same stimulus, highlighting the importance of further characterization of neonatal CD4⁺ T cell responses.

NUCLEOLAR FACTOR PESCADILLO IS REQUIRED FOR CELL PROLIFERATION IN THE HUMAN PARASITE *TRYPANOSOMA BRUCEI*

Laila Espinosa Renteria, Luis Enrique Florencio Martínez,
Santiago Calvillo Martínez and Tomás Nepomuceno Mejía

Unidad de Biomedicina, Facultad de Estudios Superiores Iztacala, UNAM. Av. de los Barrios 1,
Col. Los Reyes Iztacala, Tlalnepantla, Edo. de México, CP 54090, México
lailaespino233@gmail.com; tnepomuceno@unam.mx

Human African trypanosomiasis, also called sleeping sickness, is a vector-borne disease that is endemic in 36 African countries. *Trypanosoma brucei* is the etiological agent of the disease. This flagellated protozoan is an attractive biological model because it has unconventional genetic characteristics that are poorly represented in other eukaryotes. The fragmentation of the 28S-type ribosomal RNA chain into two large and four small and independent molecules suggest that synthesis of the ribosome subunit 60S is a very intricate process in this parasite. To date, little is known about the identity and function of factors involved in rRNA processing in the early-diverging eukaryote *T. brucei*. Pescadillo is an essential, nucleolar factor closely related with cell viability and synthesis of the 60S ribosomal subunit in human and yeast cells. To determine if TbPes is essential for the proliferation of procyclic forms of *T. brucei* in culture, we generated two knockdown cell lines where we can induce the degradation of the TbPes mRNA with doxycycline. The growth curves show that the absence of TbPes causes a reduction close to 50% in the parasite viability. We are currently performing RT-PCR assays to confirm the reduction of the TbPes transcript, and indirect immunofluorescence experiments to look for nucleolar morphology changes. Moreover, we will analyze variations in rRNA processing by Northern blot.

This work was supported by PAPIIT-UNAM (grant IA200623) and CONAHCYT (grant CF-2023-I-547).

MOLECULAR STUDY OF THE PSEUDOURIDINE SYNTHASE CBF5 IN THE HUMAN PATHOGEN *LEISHMANIA MAJOR*

Luis Enrique Florencio Martínez, Santiago Martínez Calvillo, Tomás Nepomuceno Mejía

Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México,
Av. de los Barrios 1, Col. Los Reyes Iztacala, Tlalnepantla, Estado de México CP 54090, México.
luiseff@unam.mx; tnepomuceno@unam.mx

Cbf5, also known as dyskerin, is the enzymatic subunit of box H/ACA snoRNP complexes, and catalyzes the pseudouridylation of ribosomal RNA, U2 snRNA and some messenger RNAs. Pseudouridine residues are the most abundant chemical modification in noncoding RNAs. However, little is known about Cbf5 in the ancestral protozoan parasites of the genus *Leishmania*, microorganisms that cause different debilitating to fatal diseases. Here, we studied the Cbf5 orthologue of *L. major* (ID: LmjF.21.1760; LmCbf5). Our *in silico* analysis showed that LmCbf5 possesses the four canonical domains and its predicted three-dimensional architecture is similar to that reported in *Saccharomyces cerevisiae*. Furthermore, we generated a promastigote cell line of *L. major* that expresses a PTP-tagged version of LmCbf5 to identify factors that interact with it *in vivo*. After the molecular characterization of the transgenic parasites, tandem affinity purifications, mass spectrometry and bioinformatic analyses were carried out. Notably, dozens of proteins were identified, including RNA helicases, GTPases, methyltransferases, chaperons, and ribosomal proteins, among others. Interestingly, we also purified several *Leishmania*-specific factors, whose function has not been described and that could represent therapeutic targets for leishmaniasis. This work was supported by PAPIIT-UNAM (grant IA200623) and CONAHCYT (grant CF-2023-I-547).

IN VIVO ROLE OF $\gamma\delta$ CELLS IN A LUPUS MOUSE MODEL INDUCED BY NPA STABILIZATION

Edgar Iván Galarce Sosa, Giovanna Berenice Barrera Aveleida, Jesús Elías Isidro Magallán, Edgar José Rundquist Sánchez, Rodrigo Ramos Monteagudo, María Isabel Baeza Ramírez, Carlos Wong Baeza and Claudia Albany Reséndiz Mora

Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Laboratorios de Biomembranas y de Enzimología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, CDMX, México. Unidad Profesional Lázaro Cárdenas, Prolongación de Carpio y Plan de Ayala s/n, Col. Santo Tomás C.P. 11340, CDMX. México.
E-mail: ivan.gal27@hotmail.com; Tel: +52 55 57296000, ext. 62326

Systemic lupus erythematosus (SLE) is a chronic multisystem autoimmune disease with highly varied clinical manifestations, with a complex pathogenesis driven by immune activation against self-antigens caused by a deficiency in self-tolerance¹. Our research group has identified that changes in the cell bilayer, due to stable non-bilayer phospholipid arrangements (NPA), promote autoantibodies formation against these lipids arrangements². Since $\gamma\delta$ cells have a functional plasticity that has been linked to autoantibody formation, they can be activated by lipids and participate in the pathogenesis of SLE through the secretion of various cytokines³. In the present work, $\gamma\delta$ cells from the spleen and mesenteric lymph nodes from BALB/c mice with lupus induced by stable NPA were evaluated by flow cytometry to assess their activation, proliferation, cell cycle, mitochondria type, and cytokine production. A significant increase in the absolute number and the activation of $\gamma\delta$ cells of lupus mice was detected throughout the study in the spleen and mesenteric nodes in contrast to the control group. These $\gamma\delta$ cells of lupus mice were proliferating and were mainly found in the S and G2 stages of the cell cycle. Furthermore, they showed metabolic reprogramming by fission of their mitochondria and mainly produced IL-4 and IFN γ , cytokines involved in the production of IgG class antibodies. These results suggest that $\gamma\delta$ cells are involved in the processes that enable the adaptive response establishment and possibly also participate in producing high-affinity anti-NPA antibodies.

References

1. Kirkpatrick, K. V., & Nocton, J. J. (2024). Unusual Presentations of Systemic Lupus Erythematosus. *Medical Clinics of North America*, 108(1), 43–57. <https://doi.org/10.1016/J.MCNA.2023.05.015>
2. Resendiz-Mora, A., Wong-Baeza, C., Nevarez-Lechuga, I., Landa-Saldivar, C., Molina-Gomez, E., Hernandez-Pando, R., & Baeza, I. (2021). Interleukin 4 deficiency limits the development of a lupus-like disease in mice triggered by phospholipids in a non-bilayer arrangement. *Scand J Immunol*, 93(3), e13002. doi:10.1111/sji.13002
3. Hu, Y., Hu, Q., Li, Y., Lu, L., Xiang, Z., Yin, Z., Kabelitz, D., & Wu, Y. (2023). $\gamma\delta$ T cells: origin and fate, subsets, diseases and immunotherapy. *Signal Transduction and Targeted Therapy*, 8(1). <https://doi.org/10.1038/S41392-023-01653-8>

THE DELETION OF SMAD7 ENHANCES THE INHIBITORY EFFECT OF TGF- β ON THE EFFECTOR FUNCTIONS OF CD8+ T LYMPHOCYTES

Verónica-Yutsil García-Rasilla¹; Jesús-Daniel Zambrano-Romero¹; Eugenio Contreras-Castillo¹; José Luis Ramos-Balderas¹; Paula Licona-Limón¹

¹Departamento de Biología Celular y del Desarrollo, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Mexico City, Mexico.

E-mail: veronicag@ifc.unam.mx plicona@ifc.unam.mx

CD8+ T cells are very important for immune defense against intracellular pathogens, including viruses and bacteria, and for tumor surveillance. Transforming growth factor β (TGF β) is a potent immunosuppressive cytokine known to regulate several CD8+ T cell functions. It restrains the expression of IFN γ , granzyme B, and perforin, which are essential for CD8+ T cell effector functions¹. Additionally, TGF β elicits cell cycle arrest and apoptosis in effector CD8+ T cells. Smad proteins are the main intracellular mediators of the canonical TGF β signaling pathway. This pathway is initiated when one of the three isoforms of TGF β binds to TGF β R2, which recruits and phosphorylates TGF β R1. TGF β R1 then phosphorylates Smad2 and Smad3. Subsequently, Smad4 is recruited, and together with Smad2 and Smad3, translocate into the nucleus to regulate the transcription of TGF β target genes. When TGF β binds to its receptor and activates the signaling pathway, Smad7, which serves as a negative regulator of canonical TGF β signaling, is released into the cytoplasm, where it acts by inhibiting the phosphorylation of Smad2/3 and promoting the degradation of TGF β receptor I and Smad2/3 by facilitating their ubiquitination.

In this study, our objective was to elucidate the role of Smad7 in the effector functions of CD8+ T cells. Using animals with conditional deletion of Smad7 in the T lymphocyte compartment (CD4 Cre), we discovered that Smad7-deficient CD8 lymphocytes are more sensitive to TGF β inhibition. Interestingly, *in vitro* experiments showed that in the presence of TGF β , there is a alteration in proliferation capacity and apoptosis of CD8 T lymphocytes lacking Smad7 (Smad7KO) and a reduction in the expression of effector molecules such as IFN γ , granzyme B, and perforin. *In vivo* experiments, using a melanoma mouse model, it was observed that Smad7 deficiency accelerates tumor growth, reduces the frequency and number of CTLs and reduces the cytotoxic capacity of CD8+ T cells.

References

1. Thomas, D. A., & Massagué, J. (2005). TGF- β directly targets cytotoxic T cell functions during tumor evasion of immune surveillance. *Cancer cell*, 8 (5), 369-380.
2. Shi, X., Yang, J., Deng, S., Xu, H., Wu, D., Zeng, Q., ... & Zhou, H. (2022). TGF- β signaling in the tumor metabolic microenvironment and targeted therapies. *Journal of Hematology & Oncology*, 15 (1), 135

PLASMODIUM VIVAX MITOCHONDRIAL DNA POLYMORPHISM, AND ITS RELATIONSHIP TO GENES ENCODING SEXUAL PROTEINS AND THE INFECTION PATTERN IN MOSQUITOES NYSSORHYNCHUS ALBIMANUS AND ANOPHELES PSEUDOPUNCTIPENNIS, CHIAPAS, MEXICO

Lilia González Cerón, Frida Santillán Valenzuela, Clemente Mosso González, Cuauhtémoc Villarreal Treviño
Centro Regional de Investigación en Salud Pública, Instituto Nacional de Salud Pública, Tapachula, Chiapas, México.
E-mail: lgonzal@insp.mx

Ny. albimanus and *An. pseudopunctipennis* are the main malaria vectors in Mexico. Mitochondrial (MIT) DNA of *P. vivax* has aided to understand their evolutionary relationships, geographical origin and dispersal¹. In southern Mexico, by experimental feedings with *P. vivax* infected bloods, we discovered different oocyst infection level in those mosquito species², and genes encoding sexual markers suggested different *P. vivax* subpopulations³. Here, the MIT gene polymorphism, and its relationships with *P. vivax* sexual markers and vector susceptibility was explored.

P. vivax DNA sequences were extracted from database plamodb.org⁴. MIT genes and Pvs25-130/Pvs47-27 SNPs (sexual markers) of 15 Mexican isolates were obtained. Experimental infectivity data was retrospectively searched in our databases. To make comparisons, similar information of 159 parasites from outside Mexico was included, and the genetic relationships were analyzed using phylogenetic trees in MEGA v11.

DNA sequences of Sal-I strain were used as reference⁴. Three isolates from Mexico were genetically like Sal-I strain (Pvs25-130Ile/Pvs47-27Lys) with high oocyst infection in *Ny. albimanus*. While other isolates had a variant phenotype Pvs25-130Thr/Pvs47-27Glu, and 11 isolates had three nucleotide changes in *coxI* (1,425pb); G1040A (exclusive to Mexican parasites), G1196C and T1440A, with high oocyst infection in *An. pseudopunctipennis*. While one isolate had G1196C and T1440A changes in *coxI*, and change A382G in *coxIII* (792pb), and with similar oocyst infection in both vector species. *Cytb* (1,123pb) was conserved among Mexican parasites. The ML phylogenetic tree clustered parasites genetically similar to Sal I and infectious to *Ny. albimanus* with other Latin American parasites, while variant parasites were clustered in a different branch. Those clusters were separated from parasites of other continents.

These results highlight the mitochondrial DNA in explaining genetic and biological differences of *P. vivax*, and the importance of analysis MIT genomes in a higher number of parasites will be discussed. This work is supported by CONAHCyT CF-2023-I-2685.

References

1. Taylor JE, Pacheco MA, Bacon DJ, et al. (2013). *Mol Biol Evol*, 30, 2050–2064.
2. Gonzalez.ceron L, Rodriguez MH, Nettel J, et al. (1999). *Infect Immun*, 67, 410-412.
3. Gonzalez-ceron L, Rodriguez MH, Ovilla MT, et al. (2019.) *Front Genet*, 2019, 10:1362.
4. Hupaldo DN, Luo Z, Melnikov A, et al. (2016). *Nat Genet*, 48, 953–958.

OBTENTION OF HUMAN NEUTRALIZING ANTIBODIES AGAINST DENGUE VIRUS THROUGH THE SELECTION OF B-CELLS

Dulce Desireé Izar Marín¹, Juan Carlos Muñoz Herrera¹,
Sonia Mayra Pérez-Tapia^{1,2}, María Martha Pedraza Escalona^{*3}

¹Unidad de Desarrollo e Investigación en Bioterapéuticos (UDIBI), Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Ciudad de México, C. P. 11340, México; ²Laboratorio Nacional para Servicios Especializados de Investigación, Desarrollo e Innovación (I+D+i) para Farmoquímicos y Biotecnológicos, LANSEIDI-FarBiotec-CONACyT; ³CONAHcyT-Unidad de Desarrollo e Investigación en Bioterapéuticos (UDIBI), Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional,

Ciudad de México, 11340, México.

*E-mail: desireeizar@gmail.com

Dengue virus (DENV), a National Health problem, belongs to the arboviruses (short for “arthropod-borne virus”), as it is transmitted by a hematophagous mosquito of the genus *Aedes*¹. It is an enveloped single positive-stranded RNA virus and possesses two open reading frames that code for structural and nonstructural proteins. The second open reading frame codifies to three associated structural proteins: membrane (M) and envelope (E) and capsid (C). The E protein of DENV is one of the main targets of neutralizing antibodies².

The participation of volunteers convalescing from DENV infection was carried out by filling out an informed consent form previously approved by the CEI-ENCB. Subsequently, the recognition of DENV-E by the serum of the patients was evaluated by using an indirect ELISA. A serum obtained previously was used as positive control (antibody titer 1:28,897 against the E protein), while a serum that does not show recognition was used as negative control. Once the patients had been selected, a second whole blood collection was performed to isolate PBMCs and from these, B cells were selected using microbeads anti-CD22 antibodies. Cell separation was performed in a flow cytometer, where negative selection includes surface markers, such as CD3, CD14 and CD16, while positive selection of plasma and memory B cells includes CD20, CD27 and CD38 markers. Finally, infection with Epstein Barr virus was performed to transform B cells into antibody-producing lymphoblastoid lines, which were grown in 96-well plates with irradiated fibroblasts and IMDM medium.

Forty-four serum samples were evaluated (38 from patients from the state of Guerrero and 6 from Veracruz), of which 33 sera recognized the E protein of DENV1 by presenting a radius greater than 2 compared to the negative control. The antibody titer was determined, and the sera were divided into 4 groups according to the value obtained. A serum, with the highest antibody titer (1:2317) was selected to isolate and to immortalize memory B cells (CD20⁺CD27⁺) and plasmablast cells (CD20⁺CD38⁺). The antibodies produced by these cells were analyzed and selected by affinity and neutralizing activity.

References:

1. Uno, N., & Ross, T. M. (2018). Dengue virus and the host innate immune response. *Emerging microbes & infections*, 7(1), 167.
2. Ávila-Ramírez, M. L., Reyes-Reyes, A. L., Avila-Bonilla, R. G., Salas-Benito, M., Cerecedo, D., Ramírez-Moreno, M. E., Villagrán-Herrera, M. E., Mercado-Curiel, R. F., & Salas-Benito, J. S. (2023). Differential Gene Expression Pattern of Importin β 3 and NS5 in C6/36 Cells Acutely and Persistently Infected with Dengue Virus 2. *Pathogens*, 12(2), 191.

STUDY OF NFAT ACTIVATION IN PORCINE ALVEOLAR MACROPHAGES VIA HRGFP REPORTER GENE EXPRESSION

Eunice López-Ahumada¹, Ilane Hernández-Morales¹, Aarón Iván Vélez Ramírez¹

¹Laboratorio de Investigación Interdisciplinaria, Escuela Nacional de Estudios Superiores Unidad León, Universidad Nacional Autónoma de México, Blvd., UNAM #2011, predio El Saucillo y Comunidad de los Tepetates, El Potrero, 37684 León, Gto.
E-mail: eunice.ahumada@comunidad.unam.mx

Background: The first line of host responses to pathogen invasion is the innate immune defenses¹. Innate immunity cells, particularly macrophages, possess pattern recognition receptors (PRRs) that detect damage-associated molecular patterns (DAMPs) and pathogen-associated molecular patterns (PAMPs), triggering cellular responses such as the production of pro-inflammatory molecules^{2,3}. Toll-like receptor 4 (TLR4) interacts with lipopolysaccharide (LPS) and CD14 in macrophages activating the transcription factor NFAT (Nuclear Factor of Activated T-cells), which regulates the expression of pro-inflammatory cytokines like IL-12 and TNF- α ⁴⁻⁷. Studies suggest that viral glycoproteins can induce an immune response through TLR4 activation, acting as a PAMPs⁸. **Objective:** To develop models for studying NFAT activation in macrophages stimulated with viral glycoproteins by quantifying hrGFP expression as a reporter gene. **Methodology:** The pNFAT-hrGFP plasmid (Agilent Technologies, No.240053) was amplified and purified (LPS free). The 3D4/31 cell line was cultured in RPMI medium with standard supplements. Macrophages were transfected using Lipofectamine™ 2000 (Thermo Fisher, No.11668019), following the manufacturer's specifications, and transfected clones were selected. NFAT activation was estimated based on the mRNA expression of hrGFP and fluorescence, using LPS and dexamethasone as stimulation controls. Additionally, a low-cost CO₂ incubator was designed to maintain optimal conditions of 37°C and 5% CO₂ for macrophage culture and as a proof-of-concept tool to measure hrGFP fluorescence. **Results:** 3D4/31 macrophages express hrGFP in a transient transfection model using the plasmid pNFAT-hrGFP. The expression of hrGFP was determined by RT-PCR using total mRNA from transfected cells. Additionally, a customized CO₂ incubator was constructed to further develop an instrument for GFP fluorescence quantification. **Conclusion:** These results validate the use of the pNFAT-hrGFP plasmid for studying immune signaling pathways in porcine macrophages. Moreover, we offer a low-cost alternative for the cellular culture to be used as a tool to measure hrGFP fluorescence. This project was carried out under the grant PAPIIT-IA208322 from DGAPA-UNAM.

References

1. Pancer Z, Cooper MD (2006). *Annu Rev Immunol*, **24**, 497-518.
2. Diamond MS, Kanneganti TD (2022). *Nat Immunol*, **23**, 165-176.
3. Abbas AK, Lichtman AH, Pillai Shiv (2022). *Cellular and Molecular Immunology*, **10**, 64-72.
4. Dasu MR, Jialal I (2013). *J Diabetes Complications*, **27**, 417-421.
5. Ostuni R, Zanoni I, Granucci F (2010). *Cellular and Molecular Life Sciences*, **24**, 4109-4134.
6. Zanoni I, Bodio C, Broggi A (2011). *Immunol Lett*, **142**, 41-47.
7. Fric J, Zelante T, Wong AYW, Mertes A, Yu HB, Ricciardi-Castagnoli P (2012). *Blood*, **7**, 1380-1389.

B55 β AS A PIVOTAL MOLECULE IN ARTHRITIS DEVELOPMENT AND ANTI-TNF- α THERAPY

Dulce Malvaez-Luis^{1,2}, Jesús E. Gutiérrez-Hernández², Laura Q. López-Hernández², Perla A. Bejar², Karla P. Guzmán-Barrenechea², Florencia Rosetti², José C. Crispín², Iris K. Madera-Salcedo^{2*}

¹Faculty of Chemistry, Universidad Autónoma del Estado de México, ²Department of Immunology and Rheumatology, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán. *Vasco de Quiroga 15, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México, México. Tel. 55-5487-0900 Ext 3401. E-mail: iris.maderas@incmnsz.mx

Rheumatoid arthritis (RA) is an autoimmune disease characterized by chronic inflammation in the joints, which can lead to tissue destruction. TNF- α is the major cytokine involved in the pathogenesis of AR. Although anti-TNF therapy represents a great therapeutic breakthrough, its usefulness is limited by an elevated percentage of non-responder patients (40%) in RA. Despite advances in understanding the pathogenesis of RA, the processes that initiate and perpetuate the disease and the reasons why some patients exhibit resistance to TNF inhibition are not fully understood. B55 β , a molecule required for the termination of the immune response, is aberrant expressed in T cells from RA patients, and TNF affects B55 β function in these cells. Based on this, we pose the following questions: What is the role of B55b in the pathogenesis of RA? Does the expression of B55b determine the efficacy of treatment with TNF- α inhibitors? To address this, we induced collagen-induced arthritis (CIA) in CD4.Cre^{Ppp2r2bfl/fl} (T cells-specific B55b KO) and CD4.Cre^{Ppp2r2b+/+} (WT) mice, treated or untreated with adalimumab (TNF- α inhibitor), and analyzed the proportion of CD4 T helper cells in the synovium. We found that B55b KO mice develop arthritis earlier and more robustly following immunization, characterized by synovial hyperplasia and cellular infiltration, in contrast to WT mice. In addition, B55b KO mice have a consistent higher proportion of follicular T cells (Tfh), but lower levels of regulatory T cells (Tregs) in the inflamed joints compared to WT mice, suggesting that Tfh cells could exert their inflammatory effect locally in the joint. Importantly, when anti-TNF- α treatment was initiated after onset of arthritis, both experimental groups of mice reduced the severity of arthritis, however, B55b KO mice demonstrated a superior response. Furthermore, anti-TNF- α treatment normalized the populations of Tfh and Treg cells in B55 β KO mice to levels comparable to those in WT mice. Collectively, our results highlight B55 β as a pivotal molecule in arthritis development and the effector function of infiltrated CD4 T cells, crucially impacting the effective response to anti-TNF- α therapy.

CF-2019/1564468 (IKMS)

IDENTIFICATION OF PROTEIN FACTORS INVOLVED IN THE ATYPICAL BIOGENESIS OF THE 60S RIBOSOME SUBUNIT IN THE HUMAN PATHOGEN *LEISHMANIA MAJOR*

Aldo Yael Miranda Muñoz, Luis E. Florencio Martínez, Santiago Martínez Calvillo, Tomás Nepomuceno Mejía

Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México,
Av. de los Barrios 1, Col. Los Reyes Iztacala, Tlalnepantla, Estado de México CP 54090, México.
aldo.yael2602@gmail.com; tnepomuceno@unam.mx

The nucleolus is the nuclear body where the ribosome biogenesis occurs. Cytoplasmic ribosomes are ribonucleoprotein complexes responsible for translation in all kingdoms of life. However, little is known about this process in the early-branching protozoan parasites of the *Leishmania* genus. These microorganisms affect millions of people worldwide. In contrast to other eukaryotes, *Leishmania major* possess only 12 copies of the ribosomal gene repeat. As occurs in other trypanosomatids, in *L. major* ribosomes are uncommon structures due to fragmentation of the 28S-type rRNA chain into two large and four small and independent rRNA molecules. Thus, the synthesis of ribosomes in *L. major* seems to be a very complex process that could require unique factors. To identify elements involved in the generation of the 60S subunit of the ribosome in *L. major*, we generated a promastigote cell line that expresses a PTP-tagged version of Nop7, a nucleolar element essential for cell viability. After determining the correct expression and nucleolar localization of LmNop7-PTP, we purified the protein complexes that in vivo interact with Nop7 by tandem affinity chromatography. A representative fraction of affinity elute material was fractioned in SDS-PAGE and Sypro Ruby stained. We observed several bands with molecular masses ranging from 25 to 150 kDa. The nature of these factors will be established by mass spectrometry assays coupled with bioinformatics tools. We anticipate the identification of some *Leishmania*-specific proteins, which would represent potential therapeutic targets for the control of leishmaniasis, an important neglected tropical disease. This work was supported by PAPIIT-UNAM (grant IA200623) and CONAHCYT (grant CF-2023-I-547).

POPULATION REGULATION OF *SCHINUS MOLLE* ON TROPHOZOITES OF *ENTAMOEBIA HISTOLYTICA* IN AXENIC CULTURE

Arely Morales-Vaca¹, López-Vancell Rosario¹, Karla María Morales-Zamora¹, Ivette Andrea Martínez-Santiago¹, Mario Néquiz-Avedaño¹, Norma Lilia Salaiza-Suazo¹, Pérez-Hernández Jesús¹, Augusto José González Canto¹

¹Experimental Medicine Research Unit "Dr. Ruy Pérez Tamayo", Faculty of Medicine, UNAM.
General Hospital of México "Dr. Eduardo Liceaga". C.P: 06726. Tel. +52 5543815536.
E-mail: morales_arely@ciencias.unam.com

Amoebiasis is a gastrointestinal infection caused by *Entamoeba histolytica* and it is responsible for high mortality and morbidity in children and young adults [1] it becomes in a health problem worldwide [2]. *E. histolytica* is still endemic in tropical and subtropical regions. The symptoms are diarrhea, dysentery and in some specific cases liver abscesses caused by amoeba migration to other organs resulting in death. Currently the treatment of amoebiasis is the use of metronidazole but in the pharmacognosy use different plants to treatments for gastrointestinal infection. In the bibliography we found that *S. molle* has antiprotozoal and antibacterial proprieties in methanolic extract [3]. The objective is to determine the effect of two conditionings of *S. molle* on the growing and viability of trophozoites from *E. histolytica* in axenic culture at different concentrations.

S. molle were obtained from Xochimilco's market. Only the leaves were dried and ground to powder. Thirty grams of dried and powdered material form *S. molle* was suspended in 300 mL boiled deionized H₂O, centrifuge and collected the supernatant in 50 mL tubes after that we lyophilized. The concentrations used were 15, 30, 60 and 90 mg of each sample in vials was sterilized at 121° for 30 minutes Each treatment assay was performed in triplicate and included a negative control with an initial population of 2x10⁵ *E. Histolytica* stain HM-1: IMSS on 6 mL of TYI-S-33 medium. The tubes were incubated at 37°C for 72 hours. The number of dead trophozoites per milliliter was determined using a hemocytometer and Trypan blue. For the pulverized of 15 mg there was no population decrease, while in 60 and 90 mg a decrease of 61.06% and 93.80% was observed compared to the control. Viability remained above 90% and only in the 60 mg had a decrease of 61%. In the first two concentrations of the lyophilized aqueous extract of 15 and 30 mg there was no population decrease greater than 50%, while in 60 and 90 mg a decrease of 67.4 to 93.4% was observed compared to the control. Viability remained above 80% with 15, 30 and 60 mg; However, at 90 mg it had a decrease of 52%.The observed effect includes a population decrease depending on the increase in the concentration of the sample used, thus observing a significant inhibition and death effect at 60 and 90 mg.

References

1. Bansal, D., Sehgal, R., Chawla, Y., Malla, N., & Mahajan, R. C. (2006). Multidrug resistance in amoebiasis patients. Indian Journal of Medical Research, 124(2), 189-194.
2. Morán, P., Serrano-Vázquez, A., Rojas-Velázquez, L., González, E., Pérez-Juárez, H., Hernández, E. G., ... & Ximénez, C. (2023). Amoebiasis: Advances in Diagnosis, Treatment, Immunology Features and the Interaction with the Intestinal Ecosystem. International Journal of Molecular Sciences, 24(14), 11755.
3. Quintanilla-Licea, R., Mata-Cárdenas, B. D., Vargas-Villarreal, J., Bazaldúa-Rodríguez, A. F., Ángeles-Hernández, I. K., Garza-González, J. N., & Hernández-García, M. E. (2014). Antiprotozoal activity against *Entamoeba histolytica* of plants used in northeast Mexican traditional medicine. Bioactive compounds from *Lippia graveolens* and *Ruta chalepensis*. Molecules, 19(12), 21044-21065.

VARIANTE RS10109853C/T DE *IDO2* Y SU RELACIÓN CON ARTRITIS REUMATOIDE

Betsabe Guadalupe Najera Ruiz¹, Isela Montufar Robles¹, Julián Ramírez-Bello³, Rosa Elda Barbosa Cobos², Cecilia Gonzales Calixto⁴, José Bonilla Delgado⁵, Enoc Mariano Cortés Malagón¹

¹División de Investigación, Hospital Juárez de México (HJM), Ciudad de México, México. ²Servicio de Reumatología, Hospital Juárez de México (HJM), Ciudad de México, México. ³Subdirección de investigación clínica, Instituto Nacional de Cardiología "Ignacio Chávez", Ciudad de México. ⁴Facultad de enfermería 2, Universidad Autónoma de Guerrero (UAGro), Acapulco de Juárez, México.

⁵Unidad de Investigación, Hospital Regional de Alta Especialidad Ixtapaluca IMSS-BIENESTAR, Ciudad de México.

Introducción. La artritis reumatoide es una enfermedad autoinmune, crónica y progresiva. Se caracteriza por afectar las articulaciones y sus tejidos circundantes; causa dolor, rigidez, hinchazón y movimiento limitado. En México, esta enfermedad se reporta con una prevalencia de 1.6%. El factor genético como son las variantes de un solo nucleótido (SNVs) contribuyen en gran medida para el desarrollo de esta enfermedad. Diversos estudios en modelos murinos de artritis reumatoide han reportado que la enzima *IDO2* es un factor importante en el desarrollo de la enfermedad. En estos modelos, se ha estudiado el papel de la SNV en *IDO2* (rs10109853C/T), siendo el alelo T un factor de susceptibilidad para artritis reumatoide. En pacientes con AR, el papel de esta SNV no ha sido investigado en población mexicana.

Objetivo. Evaluar la variante rs10109853C/T del gen *IDO2* en controles y pacientes con AR.

Material y métodos. El DNA genómico se aisló de sangre total en 288 controles y 376 pacientes con AR del Hospital Juárez de México. La genotipificación fue realizada mediante RT-PCR usando sondas TaqMan (Applied Biosystems), el equilibrio de Hardy-Weinberg y el análisis de asociación fueron realizados con el programa SNPstats.

Resultados. No se encontró diferencias estadísticamente significativas bajo el modelo alélico. El genotipo C/T mostró una asociación con AR (1.68, p=0.016) bajo el modelo codominante.

Conclusiones. Nuestros resultados muestran que la presencia del genotipo C/T de la variante rs10109853 en el gen de *IDO2* está asociado con susceptibilidad para AR, en una muestra de población mexicana.

PREVALENCE OF VIRULENCE GENES IN STAPHYLOCOCCUS AUREUS ISOLATED FROM PATIENTS WITH ATOPIC DERMATITIS AND THEIR EFFECT ON TSLP PRODUCTION BY HUMAN KERATINOCYTES

Vanegas-Flores Claudia Leticia¹, González-Zacarías Jacqueline Carolina¹, Valeria Fernanda Avila-Mendoza¹, Velazquez-Sámano Guillermo², Rangel-Castañeda Itzia Azucena¹, Garza-Velasco Raúl¹, Moreno-Eutimio Mario Adán¹, Pastelin-Palacios Rodolfo^{1*}

¹Departamento de Biología. Facultad de Química, Universidad Nacional Autónoma de México (UNAM). Cd. Universitaria, 04510. Mexico City, Mexico. ² Servicio de Alergia e Inmunología, Hospital General de México. "Dr. Eduardo Liceaga", México City, Mexico.

*Corresponding author: rodolfop@unam.mx

Atopic dermatitis (AD) is the most prevalent chronic inflammatory skin disease, facilitating colonization by *Staphylococcus aureus*. Keratinocytes can produce a cytokine called TSLP (thymic stromal lymphopoietin) in response to various stimuli, triggering a Th2-type immune response characteristic of AD. This study assesses the impact of the presence of *S. aureus* with high levels of virulence on TSLP production in keratinocytes. To determine the prevalence of virulence genes in *S. aureus* strains from the skin of patients with AD compared to healthy subjects, and to analyze how strains with a greater number of virulence factors affect TSLP production in human keratinocytes. Twenty-four strains of *S. aureus* were isolated from patients with AD and eight from healthy individuals. The strains were identified using MALDI-TOF mass spectrometry, verifying the presence of 16s and NUC genes. Virulence genes were detected by PCR. HACAT cells line were incubated with supernatants from the strains, and TSLP production was measured by ELISA. *S. aureus* strains from patients with AD showed a differential pattern of virulence factors compared to those from the skin of healthy subjects. Supernatants from the more virulent strains were associated with increased production of TSLP in human keratinocyte lines, suggesting a relationship between the virulence of *S. aureus* and the exacerbation of the immune response in AD. The presence of virulence genes in *S. aureus* associated with AD correlates with elevated TSLP production in keratinocytes, which could contribute to the severity and chronicity of the disease. This finding highlights the importance of studying bacterial virulence factors as potential therapeutic targets in the treatment of AD.

Funding. Research and Technological Innovation Projects, UNAM. PAPIIT 2023 No. IA208423 and PAPIIT 2024 No. IN221024.

THE SIALOMUCIN CD43 INDUCES AUTOPHAGY-DEPENDENT CELL DEATH IN LYMPHOID TUMORS DURING STARVATION

Erick Israel Pérez-García^{1,2}; Erika Melchy-Pérez;
Ángel Flores-Alcántar¹ & Yvonne Rosenstein¹

¹Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología., Universidad Nacional Autónoma de México (UNAM), ²Posgrado en Ciencias Bioquímicas–UNAM.
E-mail: yvonne.rosenstein@ibt.unam.mx

CD43 (leukosialin) is a transmembrane glycoprotein expressed in a variety of hematopoietic cells, including B and T lymphocytes. In addition to being a pan T cell marker, CD43 is a marker for a variety of malignancies, particularly myeloid (granulocytic) sarcoma, B and T cell lymphomas, leukemias, and solid tumors, with CD43 expression associated with poor prognosis. We have shown that in solid tumors such as NSCLC cells, CD43 contributes to bypassing the HIPPO pathway and favors the secretion of pro-angiogenic and extracellular matrix remodeling molecules. Furthermore, due to its ability to sense and transduce signals from the outside to the inside of the cell, CD43 also contributes to the cellular transformation process by increasing survival and decreasing apoptosis.

With these observations and based on preliminary laboratory data that indicate that a decrease in CD43 levels promotes an increase in apoptosis of cells subjected to nutritional stress conditions, we set out to investigate the molecular mechanisms by which CD43 promotes survival in a lymphoid cancer context, in Jurkat cells (acute lymphoblastic leukemia).

CD43 expression was diminished by transfecting Jurkat cells with specific small interfering RNAs (siRNAs). Jurkat cells that expressed normal levels of CD43 (CD43Hi) and cells with decreased levels of CD43 (CD43Low) were further cultured in a serum-free medium, mimicking the nutritional stress presented in the tumor microenvironment. By measuring annexin V binding and cleaved caspase 3 staining by flow cytometry, we observed that the CD43Low cells died by an apoptosis-mediated mechanism. However, we also observed that, when cultured in the absence of serum for 72 hours, the CD43Hi cells died by an apoptosis-independent mechanism.

A correlation analysis using the GEPIA2 platform revealed a positive correlation between the CD43 levels and the levels of ULK1 and ATG13, two proteins that form the autophagy-initiating complex. We will show data evaluating whether the absence of nutrients triggers a cell death mechanism due to autophagy in the CD43Hi cells, revealing that under starvation conditions and depending on the expression level of CD43, cells die by apoptosis or autophagy-dependent cell death.

Funded by CONAHCYT and DGAPA-PAPIIT/UNAM

THE NS1 PROTEIN OF THE DENGUE VIRUS PROMOTES EARLY SECONDARY TISSUE INFECTION OF *Aedes aegypti* MOSQUITOES

Edgar Quezada-Ruiz^{1,2}, Angélica Silva-Olivares¹, Abigail Betanzos Fernández¹, Daniel Talamás Lara¹, Salvador Hernández-Martínez², Grecia Díaz², Jose Raymundo Cruz-Perez¹, Jorge Cime-Castillo², Juan Ludert², Humberto Lanz-Mendoza²

¹Departamento de Infectómica y Patogénesis Molecular, Centro de Investigación y de Estudios Avanzados del IPN, Ciudad de México, CDMX. ²Centro de Investigaciones sobre Enfermedades Infecciosas, Instituto Nacional de Salud Pública, Secretaría de Salud, Cuernavaca, Morelos, México.
E-mail: edgar.quezada@cinvestav.mx

Dengue is a viral infection transmitted by the bite of infected female mosquitoes of the species *Aedes aegypti* and *Ae. albopictus*. The dengue virus (DENV) genome is a positive polarity RNA encoding three structural proteins (C, prM, and E) and seven non-structural proteins (NS1, NS2A, NS2B, NS3, NS4A, NS4B and NS5), these last ones involved in the replicative cycle within the cell. The non-structural protein 1 (NS1) has gained relevance in dengue research due to its crucial role in the pathogenesis and transmission of the disease. It is the only non-structural protein that is actively and continuously secreted by infected cells. In humans, NS1 is associated with the pathogenesis of dengue, inducing alterations in vascular endothelial and facilitating the spread of the virus to other tissues¹. When a mosquito feeds on an infected person, it ingests both the virion and the NS1 protein. In the mosquito, the DENV must infect different tissues until it reaches the salivary glands²; Recent studies have shown that NS1 can negatively immunoregulate the vector's antiviral response, favoring virus replication within the mosquito³; however, it is unknown whether this protein may also alters barrier components of the mosquito that could facilitate more rapid dissemination of the virus. Mosquitoes that ingested NS1 showed increased intestinal permeability, as assayed by the presence of brilliant blue dye FCF used as a marker, in the hemocoel. This effect was reversed by inactivating the protein by heat or blocking it with antibodies. Changes in intestinal permeability were characterized by structural analyses using conventional histology and transmission electron microscopy. In addition, delocalization of two key proteins for the septate junctions of the intestinal epithelium was observed in the presence of NS1. Finally, by qRT-PCR and IFA assays, we confirmed the presence of viral particles and genomes in secondary mosquito tissues, which were significantly decreased when NS1 was blocked with antibodies or denatured. The results of this study demonstrate that ingestion of NS1 protein by *Aedes aegypti* mosquitoes alters the architecture of the intestinal epithelium and facilitates DENV dissemination to secondary organs. These findings uncover new functions of NS1 within the mosquito vector and highlight the importance of NS1 as a potential therapeutic target for dengue control.

References

1. Puerta-Guardo H., D. Glasner, D. Espinosa, C. Wang, R. Beatty y E. Harris. (2019). Flavivirus NS1 triggers Tissue-Specific Vascular Endothelial Dysfunction Reflecting Disease Tropism, *Cell Reports*. 26: 1598–1613
2. Salazar MI, Richardson JH, Sánchez-Vargas I, Olson KE, Beaty BJ. (2007). Dengue virus type 2: replication and tropisms in orally infected *Aedes aegypti* mosquitoes. *BMC Microbiol* 7, 9.
3. Liu J, Liu Y, Nie K, Du S, Qiu J, Pang X, Wang P, Cheng G. (2016). Flavivirus NS1 protein in infected host sera enhances viral acquisition by mosquitoes. *Nat Microbiol*. 1(9): 16087.

STUDY ON THE ANTI-INFLAMMATORY POTENTIAL OF DL-3-HYDROXY-3-ETHYL-3-PHENYLPROPIONAMIDE, AN ANTICONVULSANT DRUG, IN MICROGLIA DERIVED FROM MICE WITH EPILEPSY

*Claudia Albany Reséndiz Mora, Edgar José Rundquist Sánchez, Giovanna Berenice Barrera Aveleida, Edgar Iván Galarce Sosa, Gabriel Trejo Sánchez, Jesús Elías Isidro Magallán, Rodrigo Ramos Monteagudo, Isabel Baeza Ramírez and Carlos Wong Baeza

Laboratorios de Biomembranas y de Enzimología, Departamento de Bioquímica, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional. Prolongación de Carpio y Plan de Ayala s/n, Colonia Santo Tomás, Alcaldía Miguel Hidalgo, Ciudad de México, CP.11340. Tel +52 55 57296000 Ext 62326.

*E-mail: cbanyrm@icloud.com

Epilepsy is a chronic neurological disorder characterized by recurrent seizures due to an imbalance between inhibitory and excitatory signalling. It affects around 70 million people around the world and is characterized by a long-lasting predisposition to generate seizures with different effects and with an increased and persistent inflammatory state in the microenvironment of the neural tissue¹. Despite the increasing availability of new pharmacological options, approximately 30% of cases are drug-resistant. Currently, there is no drug that can prevent, stop, or cure epilepsy. In this study, we are examining the effects of the drug DL-3-hydroxy-3-ethyl-3-phenylpropionamide (HEPP) on the regulation of inflammation and the production of reactive oxygen species by microglial cells. Microglial cells are considered the resident macrophages of the central nervous system and are often categorized into two main polarization states: a classically activated M1-like phenotype, associated with proinflammatory and neurotoxic responses, and an alternatively activated M2-like phenotype, associated with anti-inflammatory and neuroprotective functions². To determine by flow cytometry and confocal microscopy whether the drug HEPP modulates pro- and anti-inflammatory cytokine production as well as reactive oxygen species production in microglia cells from mice with epilepsy. Four groups with five male C57BL/6 mice will be used. The first group will be administered with Tris-saline solution, while the other three will be induced in a chronic model of status epilepticus by administration of pentylenetetrazole. The second group will not be treated; while the third and fourth groups will be treated with valproic acid (reference anticonvulsant drug) or HEPP, respectively. According to the Racine behavioural scoring scale, at characteristic seizure stages, mice will be euthanized, and brains will be removed. Microglia cells from the different study groups will be purified using a Percoll gradient and stained for analysis of variation, activation, and cytokine production via flow cytometry and confocal microscopy.

References

1. Kośmider K. et. al. Second generation of antiepileptic drugs and oxidative stress. *International Journal of Molecular Sciences*. 2023 15;24(4):3873.
2. Yu C. et. al. Microglia in epilepsy. *Neurobiology of Disease*. 2023 1;185:106249.

ACTIVATION OF NKT CELLS AND T γ δ LYMPHOCYTES IN RESPONSE TO MYCOBACTERIAL LIPIDS

*Claudia Albany Reséndiz Mora, Jesús Elías Isidro Magallán, Edgar José Rundquist Sánchez, Giovanna Berenice Barrera Aveleida, Edgar Iván Galarce Sosa, Gabriel Trejo Sánchez, Rodrigo Ramos Monteagudo, Isabel Baeza Ramírez and Carlos Wong Baeza

Laboratorios de Biomembranas y de Enzimología, Departamento de Bioquímica, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional. Prolongación de Carpio y Plan de Ayala s/n, Colonia Santo Tomás, Alcaldía Miguel Hidalgo, Ciudad de México, CP.11340. Tel +52 55 57296000 Ext 62326.

*E-mail: cbanyrm@icloud.com

When protein antigens are presented to CD4 T cells, they activate, proliferate, and cooperate with B cells, leading to germinal center formation and high-affinity IgG antibody production¹. In some infectious diseases, such as malaria, tuberculosis, and leprosy, and some autoimmune diseases, like Systemic Lupus Erythematosus, IgG antibodies against lipid antigens have been found². However, only some studies investigate the cells and mechanisms involved in responses against lipid antigens³. In this work, we demonstrate that T γ δ and NKT cells from mice administered with *Mycobacterium tuberculosis* lipids and mice with lupus induced by the stabilization of lipidic particles are activated and express CD69 and CD25. These cells also produce IL-4 and IFN- γ , which could induce B cells to produce IgG class antibodies, IL-17, which could lead to chronic inflammation, and granzymes and perforins, which could cause cytotoxicity. Furthermore, both cells were mainly in the G1 stage of the cell cycle, which suggests that they are mainly participating in the adaptative response against lipid antigens.

References

1. Tonti E., Fedeli M., Napolitano A., Iannacone M., Von Adrian U., Guidotti L., Abrignani S., Casorati G. and Dellabona P. (2012) Follicular Helper NKT Cells Induce Limited B Cell Responses and Germinal Center Formation in the Absence of CD4⁺T Cell Help. *J Immunol*, 188:3217-3222.
2. Wong-Baeza C, Reséndiz-Mora A, Donis-Maturano L, Wong-Baeza I, Zárate-Neira L, Yam-Puc JC, Calderón-Amador J, Medina Y, Wong C, Baeza I. and Flores-Romo L. (2016) Anti-Lipid IgG Antibodies Are Produced via Germinal Centers in a Murine Model Resembling Human Lupus. *Front. Immunol.* 7:396.
3. De Libero G. & Mori L. (2010) How the immune system detects lipid antigens. *Progress Lipid Research*, 49:120-127.

IMMUNOMETABOLIC CONTROL OF NEONATAL CD4⁺ T CELL RESPONSES

Jonathan Ricardo Robles Reyes¹, Angelica Santana Calderón¹, Otoniel Rodríguez Jorge¹

¹Laboratorio de inmunología celular y de sistemas, Centro de Investigación en Dinámica Celular, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México.

Av. Universidad No. 1001, Col Chamilpa, Cuernavaca, Morelos, México. C.P. 62209; 777 329 7900 ext 3666
orj@uaem.mx

Neonates have a high mortality and morbidity rate. One of the leading causes of neonatal death is their high susceptibility to infections by intracellular pathogens. CD4⁺ T cells are one of the main arms of the Immune system against this type of pathogen. However, the immune response of neonatal CD4⁺ T cells is characterized by a tolerogenic and Th2 bias, highlighting the need to study the neonatal immune response and seek strategies to enhance the response. CD4⁺ T cells enter the circulation as naïve T cells, with a mainly catabolic program based on the oxidation of available nutrients and OXPHOS for ATP synthesis. Naïve T cells are activated when they recognize their antigen through the TCR along with costimulatory signals, such as those from CD28. CD28 activates several signaling pathways, including PI3K/Akt/mTOR, leading to enhanced TCR-mediated signaling and increased metabolic rate. T cell activation induces metabolic reprogramming, changing to a glycolytic and anabolic metabolism, a metabolism necessary for the synthesis of the biomolecules necessary to fulfill its functions as an effector CD4⁺ T lymphocyte. In this new program, some metabolites present in the tissue microenvironment or produced by metabolic reprogramming, like lactate, have immunoregulatory activity, which can affect T cell activation at different levels (metabolism, signaling, transcription, etc). Previously, we reported that neonatal T cells metabolism is characterized by high glycolytic activity and proliferation in basal conditions, which could induce high lactate levels. In this work, we employ logical modeling of the influence of metabolism on CD4⁺ T cell activation to address the influence of the neonatal metabolic state on CD4⁺ T cell activation. Through computational simulations and experiments, we explore the use of amino acid supplementation and inhibition of lactate internalization as possible strategies to enhance neonatal T cell immune responses. Our preliminary results point to an inhibitory role of Arg and Ser on the activation of neonatal T cells as well as an improvement in their viability.

6-PENTADECYL SALICYLIC ACID MODIFIES THE SECRETION OF CYTOKINES OF HUMAN NATURAL KILLER CELLS *IN VITRO*

C. Rosales-González, E. Estrada-Muñiz and L. Vega

Center for Research and Advanced Studies of the National Polytechnic Institute, Toxicology, Mexico City, Mexico.

E-mail: lvega@cinvestav.mx

The 6-pentadecyl salicylic acid (6SA) is a chemical compound from the anacardic acids group that has been related to immunostimulatory. Natural killer (NK) cells are innate immune cells that are activated by damaged cells. Previous studies have shown an increase in NK population and in pJNK and p-p38 of these cells in *in vivo* models. Currently, it is unknown whether 6SA directly activates NKs and in this study we examined the effects of 6SA exposure on the activation of the NK-92 MI human cell line.

The cells were exposed for 24 h to 6SA (0-100 μ M) and we evaluated cell viability by neutral red incorporation assay (NRa) and annexin V/propidium iodide (apoptosis assay). The inhibitory concentration at 50% of the cell viability (IC_{50}) was 40.93 and 73.39 μ M of 6SA using the NRa and the apoptosis assay, respectively. Subsequently, cell proliferation was quantified by the tritiated thymidine incorporation assay, determining that exposure to 6SA does not modify the proliferative activity of NK proliferation. NKs were exposed to non-cytotoxic concentrations of 6SA for 24 h and then they were co-cultured for 4 h with K562 cells in different ratios to evaluate their cytolytic activity against the target cells by flow cytometry. The results showed that the exposure to 6SA did not alter the cytolytic capacity of NKs.

Although this particular function of NK cells is not modified, there is still the possibility that 6SA can modify the secretion of cytokines by these cells as it has been reported that the exposure to 6SA modifies the phosphorylation of kinases relevant to the secretion of cytokines that can alter the response of other immune cells. Currently, tests are being carried out to quantify the levels of TNF- α , IFN- γ , IL-10, GM-CSF, perforin and granzyme B with the aim to determine whether 6SA can influence the ability of NK cells to recruit other immune cells.

The preliminary results show that 6SA has no direct effects on the cytolytic activity of NK-92 MI cells, suggesting that the increase in this population in murine models is probably due to the effect of 6SA on other immune cells that triggers the increase in this population, probably by changing the profile of secreted cytokines.

Acknowledgments: NK cells were donated by Dr. Erika Rendón from UNAM. Funding was provided by CONAHCyT's grant number 21067 and scholarship 833615.

NEW LIPOSOMAL AMPHOTERICIN B DERIVATIVE: A POTENTIAL CHAGAS DISEASE TREATMENT

Lisset Torres Martínez¹, Ignacio Martínez Martínez¹, Jaime Zamora Chimal², Arturo Galván Hernández³, Iván Ortega Blake³, Bertha Espinoza Gutiérrez*

Department of Immunology, Biomedical Research Institute, National Autonomous University of Mexico, Mexico City 04510, Mexico; Experimental Medicine Research Unit, Faculty of Medicine, National Autonomous University of Mexico, Mexico City 06720, Mexico²; Institute of Physical Sciences, National Autonomous University of Mexico, Cuernavaca 6251, Morelos, Mexico³; Biomedical Research Institute, National Autonomous University of Mexico, Mexico City 04510, Mexico, 5556228943, besgu@iibiomedicas.unam.mx*

Amphotericin B (AmpB) is a polyene antibiotic and antifungal, characterized by its mycosamine chain, which is responsible for pore formation in the membrane of pathogens which contain ergosterol, such as *Trypanosoma cruzi*. Despite its trypanocidal effect, the administration of AmpB in mammals is highly toxic due to its interaction with cholesterol present in these cells. However, AmpB analogs and their liposomal formulations decrease its toxicity, have high selectivity for ergosterol, and maintain the biological activity of the original AmpB molecule. A21, an AmpB analog, has garnered significant interest for the treatment against *T. cruzi* thanks to its high selectivity for ergosterol and low toxicity towards mammalian cells. Recent studies have demonstrated that the combination of A21 with Benznidazole (Bzn) rescued 100% of mice infected with a highly virulent strain of *T. cruzi*, by significantly reducing parasitemia in the blood and decreasing inflammatory infiltrate characteristic of the disease exacerbation. Nevertheless, there is limited knowledge about the activity of liposomal formulation of A21 (L-A21). Therefore, this work compared the effects of A21 and L-A21 in vitro and a murine model infected with *T. cruzi*. To compare the cytotoxic effect between A21 and L-A21, H9C2 cells were incubated with 12.5 – 200 μ M of A21 and L-A21 for 6, 12, and 24 h. Cell viability was quantified using the MTT reduction assay. BALB/c mice were infected with a highly virulent strain of *T. cruzi*, followed by administration of A21 or L-A21, alone or in combination with Bzn, at doses of 20 mg/kg/d for 22 days. Throughout the experiment, survival percentage and parasitemia in the blood were quantified, and histopathological and liver function analyses were performed at the end of the administration period and 76 days post-treatment. Additionally, the concentrations of anti-inflammatory and proinflammatory cytokines in serum were evaluated before, during, and after treatment. The results showed that A21 and L-A21 do not have a cytotoxic effect on mammalian cells. L-A21 rescued 100% of the infected mice from death, showed a significant reduction in parasitemia in blood, did not cause hepatic damage, and reduced inflammatory infiltrate and pro-inflammatory cytokines production more effectively than A21. In conclusion, we suggest L-A21 as a new candidate for Chagas disease treatment.

OPTIMIZING HUMAN SERUM PRETREATMENT BLOOD SERUM PRETREATMENT FOR ACCURATE TOTAL ENDOTOXIN QUANTIFICATION USING LAL ASSAY

Keren Tovar-Presas¹, Dalia Flores-Molina¹, Diana Pérez-Barrios¹, Jesús Ramírez-Mercado¹,
Katya Carrasco-Urritia¹, Ana Lidia Arellano-Ortiz², Alejandra Vargas-Caraveo^{1*}

¹Universidad Autónoma de Ciudad Juárez, Instituto de Ciencias Biomédicas, Departamento de Ciencias Químico Biológicas, Av. Benjamin Franklin no. 4650 Zona PRONAF CP 32315, Cd. Juárez, Chih.; ²Universidad Autónoma de Ciudad Juárez, Ciudad Universitaria, Av. Jose Jesús Macias Delgado 18100, Ciudad Universitaria, 32000, Juárez, Chih.
E-mail: alejandra.vargas@uacj.mx

Metabolic endotoxemia is a condition resulting from the translocation of endotoxin or lipopolysaccharide (LPS) from Gram-negative bacteria in the gut into the bloodstream, triggering an inflammatory response. Increased LPS in the blood has been linked to fat absorption processes and increased intestinal barrier permeability. The most conventional method for measuring LPS is the Limulus Amebocyte Lysate (LAL) assay. However, this technique is inaccurate for quantifying LPS in blood serum because it only detects these molecules in their free form, while most are bound to serum proteins, causing masking. Therefore, this project proposed developing a proteolysis pretreatment of blood serum with proteinase K to unmask LPS so it can be quantified by the LAL assay. Blood samples were obtained from healthy adults and those with metabolic alterations in both preprandial and postprandial states. LPS unmasking by proteolysis was confirmed by SDS-PAGE and electrotransfer, and its concentration was measured using the LAL assay. This new method preliminarily determined that the total serum LPS concentration in proteolyzed blood serum was five times greater than in whole blood serum. This increase is associated with postprandial masking, which is linked to fat consumption and various health conditions such as obesity and type 2 diabetes.

IDENTIFICATION OF PERCHLORIC ACID-SOLUBLE PROTEINS FROM *TRICHOMONAS VAGINALIS*

Laura Isabel Vázquez-Carrillo¹, Daniel Agustin Bravata Alcantara¹, Héctor Quezada Pablo², Ana Laura Guzmán Ortiz², Marcos Morales Reyna¹, María Elizabeth Álvarez-Sánchez^{1*}

¹Posgrado en Ciencias Genómicas, Universidad Autónoma de la Ciudad de México (UACM), San Lorenzo 290, Col. Del Valle, CP 03100 Ciudad de México, México. ²Laboratorio de Investigación en Inmunología y Proteómica, Hospital Infantil de México, Dr. Márquez 162 Col. Doctores, CP 06720 Ciudad de México, México.

*corresponding author: maria.alvarez@uacm.edu.mx; elizabeth2@gmail.com & Co-first authors

Trichomonas vaginalis is a flagellated parasite that causes the most prevalent non-viral sexually transmitted infection. This parasite has a perchloric acid-soluble protein (PSP), tv-psp1. It is expressed under normal culture conditions and it has a possible putative ribonuclease function. This work aimed to find other PSPs in *T. vaginalis* and their possible functions. First, we obtained PSP extracts and analyzed them with SDS-PAGE One and Two-Dimensional electrophoresis. Then, the PSPs present in the observed bands and spots were identified by mass spectrometry (HPLC-MS/MS with an ion trap). Finally, we searched for their possible functions through an *in silico* analysis. By 2D gel electrophoresis results revealed the presence of 11 PSPs with molecular weights between 11- 93 kDa. The MS analysis revealed the presence of 77 PSPs, however only 45 were further studied after stringent filtering criteria. The Gene Ontology and String analysis showed that 17 proteins were involved in the ribosome synthesis pathway. The TrichDB analysis showed that the KEGG pathways purine metabolism, glycolysis metabolism, methane metabolism, and thiamine metabolism were also represented. Lastly, the interactome analysis with Cytoscape indicated that the function of 5 proteins were related to terms like Thermic shock protein, homeostasis, and elongation factor.

IMPACT OF THE FORM OF BIRTH AND EFFECT OF FLAGELLIN ON THE ACTIVATION OF NEONATAL CD4⁺ AND CD8⁺ T CELLS

Carlos Jesús Ventura-Martínez¹, Linda Aimara Kempis-Calanis¹,
María Angélica Santana-Calderón¹, Otoniel Rodríguez Jorge¹

¹Laboratorio de inmunología celular y de sistemas, Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México.

E-mail: orj@uaem.mx, cel: +52 (777)3297000 ext. 3666/3667

Neonates constitute a highly vulnerable demographic, representing more than 40% of deaths among children under 5 years old, with infections being a primary cause. Neonatal T cells exhibit low activation and cytotoxicity in CD8⁺ T cells. Various factors, including maternal nutrition, smoking, and delivery method, influence neonatal health. Research indicates that neonates delivered via cesarean section have a higher likelihood of developing childhood asthma, as well as metabolic and immunological chronic inflammatory conditions later in life. Our study focused on examining the response of neonatal CD4⁺ and CD8⁺ T cells obtained from cord blood, comparing those from cesarean section births to vaginal deliveries. We employed two stimulation methods: 1) anti-CD3+anti-CD28, and 2) anti-CD3+Flagellin, the latter based on previous findings suggesting TLR5 signals could offer co-stimulatory effects. We assessed proliferation (both homeostatic and in response to stimulation), activation of NFAT, NF-κB, and AP-1 transcription factors, and quantified key cytokines produced by T cells (IL-2, IL-4, IL-5, IL-6, IL-9, IL-10, IL-13, IL-17, IL-22, TNF-α, and IFN-γ, TGF-β1), along with a set of innate cytokines commonly produced by neonatal T cells (IL-1β, IL-7, IL-8), as well as the effector mediators of CD8⁺ T cells (Granzyme, and Perforin). Our findings revealed disparities in transcription factor stimulation and cell proliferation between T cells from neonates delivered via cesarean section versus vaginal delivery. Notably, cells from cesarean section births exhibited heightened proliferation following T cell stimulation. Moreover, cytokine production varied based on the stimulation method. Notably, CD3+Flagellin stimulation induced a higher proliferation rate in cells from both delivery methods, underscoring neonatal cells' sensitivity to specific stimulatory conditions. In conclusion, the method of birth leads to variations in neonatal T cell responses. This could be attributed to the disruptions experienced during vaginal delivery, such as mechanical forces, alterations in stress hormones, and transitions to hypoxic states, among others, which establish a tolerogenic program. On the other hand, Flagellin promotes a distinct immune response, suggesting that the TLR5 signaling pathway may serve as a co-stimulator in CD4⁺ T cells.

EVALUATION OF THE IMMUNE RESPONSE IN RATS DURING PHYSIOLOGICAL CARDIAC HYPERTROPHY INDUCED BY PREGNANCY

José Alberto Verduzco Palafox¹, Lilian Karem Flores Mendoza¹, Guadalupe González Ochoa¹, José Francisco Muñoz Valle², Samuel García Arellano², José Guadalupe Soñanez Organist¹

¹Universidad de Sonora. Blvd. Lázaro Cárdenas No.100, Col. Francisco Villa, Navojoa, Sonora. CP. 85880. Tel. 6421161558; a216211464@unison.mx

²Universidad de Guadalajara. Sierra Mojada 950, Independencia Oriente, 44340 Guadalajara, Jal.

Pregnancy causes physiological cardiac hypertrophy, as well as a systemic inflammatory process. Physiological cardiac hypertrophy is an adaptive and reversible process induced by increased blood volume overload¹, while pathological cardiac hypertrophy is an irreversible process caused by pressure overload caused by infarction or heart failure. During pathological cardiac hypertrophy, an uncontrolled inflammatory process is observed, triggering deregulation in the production of cytokines² such as growth factors³, chemokines⁴ and cytokines of the Th1 and Th2 profile⁵. This deregulated increase in cytokine production triggers damage to cardiomyocytes. In the case of physiological cardiac hypertrophy induced by pregnancy, the genetic and soluble levels of the main cytokines that coordinate the immune response before, during and after pregnancy are unknown. Its quantification will give us an overview of the hearts systemic and specific immune response is induced during and after pregnancy. Therefore, this work aimed to quantify the levels of gene expression in the heart and the serum-soluble levels of growth factors, chemokines, and cytokines of the Th1 and Th2 profile before, during and after pregnancy. The gene expression results indicate an increase in the expression of cytokines of the Th1 (TNF- α , IL-1 β , IL-6, IL-18) and Th2 (IL-10) profile, during the first stage of pregnancy with predominance in the production of Th2 profile cytokines. While, in the postpartum, only TNF- α showed an increase in its expression. Soluble GM-CSF, GRO KC, MCP-1, MIP-1 α , and RANTES increase during early pregnancy. RANTES also showed an increase in the postpartum period. On the other hand, MIP-3 α showed a decrease at the beginning of pregnancy, IL-2 a decrease at the end of pregnancy and postpartum, and IL-4 a decrease in the postpartum. According to the results, an increase in the differentiation of blood cells and migration to inflamed tissues is suggested during the first stage of pregnancy. In the heart, predominance in the production of cytokines of the Th2 profile, compared to Th1, is monitored; However, this increase did not correlate with soluble cytokine levels.

Keywords. *Physiological cardiac hypertrophy, Pregnancy, Cytokines.*

References

1. Eghbali M, Deva R, Alioua A, Minosyan TY, Ruan H, Wang Y, Toro L, Stefani E. Molecular and functional signature of heart hypertrophy during pregnancy. *Circ Res.* 2005 Jun 10;96(11):1208-16. doi: 10.1161/01.RES.0000170652.71414.16. Epub 2005 May 19. PMID: 15905459.
2. Bartekova, M., Radosinska, J., Jelemensky, M., & Dhalla, N. S. (2018). Role of cytokines and inflammation in heart function during health and disease. *Heart Failure Reviews*, 23(5), 733–758. <https://doi.org/10.1007/s10741-018-9716-x>
3. Orlic, D., Kajstura, J., Chimenti, S., Limana, F., Jakoniuk, I., Quaini, F., Nadal-Ginard, B., Bodine, D. M., Leri, A., & Anversa, P. (2001). Mobilized bone marrow cells repair the infarcted heart, improving function and survival. *Proceedings of the National Academy of Sciences*, 98(18), 10344–10349. <https://doi.org/10.1073/pnas.181177898>
4. Cavalera, M., & Frangogiannis, N. (2014). Targeting the Chemokines in Cardiac Repair. *Current Pharmaceutical Design*, 20(12), 1971–1979. <https://doi.org/10.2174/13816128113199990449>
5. Mosmann, T. R., & Coffman, R. L. (1989). TH1 and TH2 Cells: Different Patterns of Lymphokine Secretion Lead to Different Functional Properties. *Annual Review of Immunology*, 7(1), 145–173. <https://doi.org/10.1146/annurev.iy.07.040189.001045>

MACROPHAGE POLARIZATION ANALYSIS IN A MOUSE MODEL OF SYSTEMIC LUPUS ERYTHEMATOSUS TRIGGERED BY LIPID ANTIGENS

Carlos Wong Baeza*, Rodrigo Ramos Monteagudo, Giovanna Berenice Barrera Avela, Edgar Iván Galarce Sosa, Jesús Elías Isidro Magallán, Gabriel Trejo Sánchez, Edgar José Rundquist Sánchez, Claudia Albany Reséndiz Mora, María Isabel Baeza Ramírez

Laboratorios de Biomembranas y de Enzimología, Departamento de Bioquímica, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional. Prolongación de Carpio y Plan de Ayala s/n, Colonia Santo Tomás, Alcaldía Miguel Hidalgo, Ciudad de México, CP.11340. Tel +52 55 57296000 Ext 62326.

*E-mail: charlywong@icloud.com

Systemic lupus erythematosus (SLE) is a chronic and diverse autoimmune disorder characterized by the body's loss of tolerance to nuclear and cytoplasmic antigens. This loss results in the production of various autoantibodies, lymphoproliferation, and the deposition of immune complexes, which can lead to inflammation and failure of multiple organs¹. This aberrant immune response originates from innate and adaptive immunity, generating the distinctive components of the disease. Macrophages are now known to play a crucial role in the development of lupus. Their capacity for efferocytosis is diminished, and they promote tissue inflammation through their polarization. This polarization, determined by intrinsic and extrinsic factors, classifies macrophages into M1 (proinflammatory) and M2 (anti-inflammatory)². In SLE, abnormal polarization toward M1 has been observed to contribute to inflammation, although M2 also plays complex roles in disease development³. In this study, we will evaluate the polarization of macrophages into M1 and M2 phenotypes. We will use antibodies labeled with fluorochromes to identify the overall macrophage population using markers CD68 and F4/80.

Additionally, we will utilize CD80 and CD40 to detect M1 macrophages and CD206 and MGL to identify M2 macrophages. To determine the presence of cytokines, intracellular staining will be done using the GolgiStop™ kit to stop protein transport (cytokines are not released from the cell) and permeabilize the cells for the entry of antibodies. The pro-inflammatory cytokines IL-6 and IL-12 will be measured for M1 and for the M2 IL-10 and TGF- β , along with Arg-1. Samples will be acquired on a Cytex® Aurora flow cytometer, and the results will be analyzed with the FlowJo™ and Prism programs to determine the type of population and cytokine production.

References

1. Xiao, Z. X., Miller, J. S., & Zheng, S. G. (2021). *Autoimmunity Reviews*, 20(2), 102743.
2. Yang, S., Zhao, M., & Jia, S. (2023). *Frontiers In Immunology*, 14, 1-15.
3. Ma, C., Xia, Y., Yang, Q., & Zhao, Y. (2019). *Clinical Immunology*, 207, 1-9.

IMMUNOLOGICAL ENDOTYPES OF ATOPIC DERMATITIS IN THE MEXICAN POPULATION: DIFFERENTIAL EXPRESSION OF CLA⁺ T CELLS AND CCR4/CCR10 RECEPTORS

Zumaya-Pérez Luis Carlos¹, Velazquez-Sámano Guillermo², Garza-Velasco Raúl, Pastelin-Palacios Rodolfo¹ and Moreno-Eutimio Mario Adán¹

¹Department of Biology, Faculty of Chemistry, National Autonomous University of Mexico (UNAM), Mexico City.

²Allergy and Immunology Service, General Hospital of Mexico "Dr. Eduardo Liceaga", Mexico.

E-mail: marioadan@inmunoquimica.com

Introduction. Atopic dermatitis (AD) is a chronic inflammatory skin disease with highly heterogeneous clinical manifestations. CLA⁺ T cells expressing CCR4 and CCR10 receptors play a central role in the immunopathogenesis of AD. While AD endotypes have been described in various populations, they remain unexplored in the Mexican population.

Objective. To propose AD endotypes in the Mexican population based on the frequencies of CLA⁺ T cells differentially expressing CCR4 and CCR10 receptors.

Methods. A total of 47 patients with mild, moderate, and severe AD were enrolled based on the Severity Scoring of Atopic Dermatitis (SCORAD), along with 16 allergic patients and 15 healthy controls. The frequency of CLA⁺ T cells differentially expressing CCR4 and CCR10 receptors was determined by flow cytometry. Additionally, the production of IFN- γ , IL-4, and IL-17 was measured under in vitro stimulation. Clinical data were extracted from patients' electronic medical records. The obtained results were analyzed using principal component analysis (PCA) followed by unsupervised K-means cluster analysis.

Results. AD patients showed an increased frequency of CLA⁺ T cells, suggesting greater migration of these cells to the skin. PCA of cellular subpopulations revealed the presence of two clusters. Twenty principal components accounted for approximately 96% of the variance. Unsupervised K-means cluster analysis of these components identified two distinct clusters in AD patients. Cluster 1 showed an increased frequency of CD4⁺CLA⁺ T cells, a higher prevalence of allergic diseases, and reduced quality of life. Cluster 2 was characterized by an increased frequency of CD8⁺CLA⁺ T cells, the presence of *S. aureus*, and elevated IgE levels.

Conclusion. Two distinct immunological endotypes of AD were identified in the Mexican population, distinguished by differences in cellular populations and clinical manifestations.

References

1. Weidinger, S., et al., Atopic dermatitis. *Nat Rev Dis Primers*, 2018. 4(1): p. 1
2. Czarnowicki, T., et al., Early pediatric atopic dermatitis shows only a cutaneous lymphocyte antigen (CLA)(+) TH2/TH1 cell imbalance, whereas adults acquire CLA(+) TH22/TC22 cell subsets. *J Allergy Clin Immunol*, 2015. 136(4): p. 941-951 e3.
3. Czarnowicki, T., L.F. Santamaria-Babi, and E. Guttman-Yassky, Circulating CLA(+) T cells in atopic dermatitis and their possible role as peripheral biomarkers. *Allergy*, 2017. 72(3): p. 366-372.
4. Kobayashi, T., et al., Dysbiosis and Staphylococcus aureus Colonization Drives Inflammation in Atopic Dermatitis. *Immunity*, 2015. 42(4): p. 756-66.

CHARACTERIZATION OF TAU138 AND ITS ROLE IN RNA POL III TRANSCRIPTION AND CHROMATIN ORGANIZATION IN TRYPANOSOMATID PARASITES

Eduardo García Huerta, Luis E. Florencio Martínez, Tomás Nepomuceno Mejía and Santiago Martínez Calvillo

Facultad de Estudios Superiores Iztacala, UNAM,
Av. de los Barrios 1, Col. Los Reyes Iztacala, Tlalnepantla, Edo. de México, CP 54090
scalv@unam.mx

The Trypanosomatidae family includes the pathogens responsible for sleeping sickness (*Trypanosoma brucei*), Chagas disease (*Trypanosoma cruzi*), and leishmaniasis (*Leishmania* spp.). These protozoan parasites have complex life cycles that involve multiple morphological stages in the vertebrate host and transmitting insects. RNA polymerase III (RNAP III) plays an important role in the synthesis of small essential non-coding RNA molecules such as tRNAs, U6 snRNA and 5S rRNA. In yeast and vertebrates, RNAP III needs transcription factors TFIIIA, TFIIIB, and TFIIIC to initiate transcription. TFIIIC is a six-subunit complex composed of two subcomplexes, called α A and α B. In some organisms, TFIIIC is also involved in chromatin organization. Nowadays, limited information is available about RNAP III transcription and chromatin organization in trypanosomatids. Here we characterize the Tau138 subunit in *T. brucei* (TbTau138). *In silico* analyses showed that TbTau138 possesses the typical winged helix domains (WH) of Tau138 orthologs, whose predicted three-dimensional structure is conserved. As expected, TbTau138 was localized to the nucleus of the parasite by indirect immunofluorescence assays. To identify Tau138 interacting partners, we are currently performing tandem affinity purifications. Inducible RNA interference (RNAi) experiments suggest that TbTau138 is essential in procyclic forms of *T. brucei*. Eduardo García received a postdoctoral fellowship from DGAPA, UNAM. This work was supported by grants IN208224 (PAPIIT, UNAM) and CF-2023-I-820 (CONAHCYT).

EFFECT OF TWO CONDITIONINGS OF *CITRUS AURANTIUM* ON THE PROLIFERATION OF *ENTAMOEBIA HISTOLYTICA* TROPHOZOITES CULTURED *IN VITRO*

Ivette Andrea Martínez-Santiago^{1*}, Rosario López-Vancell¹, Karla María Morales-Zamora¹, Mario Néquiz-Avendaño¹, Norma Lilia Salaiza-Suazo¹, Arely Morales-Vaca¹, Jesús Pérez-Hernández¹, Augusto José González Canto¹

¹Unidad de Investigación Medicina Experimental, Facultad de Medicina, UNAM.
Campus Hospital General de México "Dr. Eduardo Liceaga" C.P: 06726
Tel: 5547723762 E-mail: ivetemtzs@ciencias.unam.mx

Amebiasis is an infection caused by the intestinal protozoan, *Entamoeba histolytica*. Globally, it is considered the second cause of death in parasitic diseases, affecting developing regions with poor sanitation and hygiene systems. Among clinical cases, asymptomatic colonization occurs in 90% and symptomatic (invasive) amebiasis in the remaining 10%. Invasive amebiasis frequently causes dysentery or diarrhea, but can also cause extraintestinal amebiasis, mainly forming amoebic liver abscesses (ALA). In Mexico, amoebiasis is one of the five main causes of gastrointestinal disease. Currently, there are antiparasitic treatments that help treat the disease, however, they do not eradicate it, and so it is important to evaluate new pharmacological alternatives focused on the properties of medicinal plants. *Citrus aurantium*, also known as "bitter orange", is a fruit tree corresponding to the *Rutaceae* family. Its main use in traditional medicine is based on its digestive, antioxidant, anti-inflammatory, antispasmodic and antihemorrhagic properties, which is why it is important in pharmacognosy where we seek to know the antiparasitic effect on *E. histolytica*. The major objective of our work is to determine the effect of *C. aurantium* leaves on the growth and viability of *E. histolytica* trophozoites in axenic culture at different concentrations.

Two preparations of the sample were carried out, starting from the leaves of *C. aurantium*, which were dried and crushed to obtain the powder. The first step involved using the powdered leaves. Furthermore, an infusion was made with a part of the powder to prepare an aqueous extract, yielding two phases: the solid residue from filtration and the lyophilized liquid. Subsequently, in both conditions, 15, 30, 60 and 90 mg were weighed in test tubes and sterilized. Each tube was then filled with 6 ml of amoebic culture medium (TYI-S-33) and an initial population of 1.2×10^5 *E. histolytica* trophozoites. The tubes were incubated at 37°C for 72 hours, and at the end, the population and viability were determined using the Trypan blue exclusion technique and a hemocytometer.

The results showed that the powdered treatment at a concentration of 60 mg induced a 136% increase in population compared to the control. For the aqueous extract phase with the solid residue from filtration, a 15 mg concentration resulted in a 71% growth increase over the control. Finally, in the lyophilized liquid phase, the cell population increased by 7% with 30 mg. The assays with *C. aurantium* indicated a delayed inductive effect on amoebic population growth.

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex molecule, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

MEDICINE, HEALTH & NUTRITION

GENOMIC ANALYSIS OF MUTATIONS IN THE BRUTON TYROSINE KINASE GENE IN PATIENT WITH X-LINKED AGAMMAGLOBULINEMIA AND GASTRIC CANCER

Dulce Jazmín Alvarez Olvera¹, Laura Itzel Quintas Granados¹, Gabriela López Herrera², Norma Estela Herrera González³ and Eduardo Carrillo Tapia¹

¹Autonomous University of Mexico City, College of Sciences and Humanities, Mexico City, Mexico, ²The Immunodeficiencies Research Unit, National Institute of Pediatrics, Mexico City, Mexico, ³Posgraduate area of the IPN Medical School, Mexico City, Mexico.

E-mail: eduardo.carrillo@uacm.edu.mx

X-linked agammaglobulinemia (XLA) is a primary immunodeficiency disorder that involves mutations in the Bruton Tyrosine kinase (*BTK*) gene located on X-chromosome (Xq22.1). *BTK* has a crucial role in the early stages of B cell development and in the maintaining the number of peripheral B cell and their response to antigen receptor crosslinking, whereby mutations in *BTK* affect signaling pathways and lead to recurrent bacterial infections in the upper and lower respiratory tract after loss of maternal IgG around 6 months of age. In XLA patients 90-95% have mutations in *BTK* and the rest of patients have defects in other genes. A recent study that evaluated 783 patients from 40 centers around the world showed that the most frequent complications in XLA patients are infectious diseases or autoimmune conditions, orchitis, and arthritis, suggesting that inflammatory, infectious, and autoimmune conditions can occur in XLA patients. A previous study reported that in 16 XLA patients with a mean age of 29 ± 11 years were diagnosed with gastric cancer (GC). Since gastric cancer ranks as the fourth highest cause of mortality in Mexico, examining occurrences in XLA patients can enhance comprehension regarding the distinct needs, obstacles, and features of Mexican XLA patients. Here we report the case of a Mexican male diagnosed with XLA at the age of 1. Following diagnosis, the patient underwent immunoglobulin replacement therapy. However, there is no record of the health history from the time of diagnosis until the age of 24. Nevertheless, laboratory routinary tests indicate that levels of IgA, IgG and IgM levels were consistently below normal ranges. In 2020, at the age of 29, the patient was diagnosed with diffuse carcinoma affecting the stomach and pylorus, along with bronchiectasis. Unfortunately, the patient passed away in October 2021. Genetic analysis revealed deletions in exons 3 and 4 of the *BTK* gene, suggesting potential implications for the development of GC as a complication of XLA.

BORON DERIVATIVES AS ACE/ACHE INHIBITORS FOR THE CONTROL OF HYPERTENSION: DESIGN, SYNTHESIS AND IN SILICO EVALUATION

Erik Andrade-Jorge¹, Alina Barquet-Nieto¹, José G. Trujillo-Ferrara¹, Roberto I. Cuevas-Hernandez¹, Jessica E. Rodríguez-Rodríguez², Rafael Villalobos-Molina², Itzell A. Gallardo-Ortíz²

¹Lab de Bioquímica, Sección de Estudios de Posgrado e Investigación, Escuela Superior de Medicina, Instituto Politécnico Nacional, Plan de San Luis y Díaz Mirón SN, CDMX, 11340, cel 55-18135062. ²Unidad de Investigación en Biomedicina y Carrera de Enfermería, Facultad de Estudios Superiores-Iztacala, Universidad Nacional Autónoma de México, Estado de México, 54090. E-mail: andrade136@hotmail.com

High blood pressure (HTN) is a global challenge, characterized by persistent elevation of blood pressure and a significant risk factor for cardiovascular diseases. In Mexico and around the world, its prevalence contributes to the need for effective treatments. In this context, boron derivatives have emerged as a class of compounds with promising pharmacological properties¹⁻⁴. The aim of the work was to evaluate in-silico and in-vitro a series of boroxazolidones as inhibitors of angiotensin II converting enzyme (ACE) and acetylcholinesterase (AChE), relevant therapeutic objectives in the control of hypertension. The results revealed a notable affinity of the compounds for ACE and AChE, even better than the reference compounds in many cases. Specifically, the BXZ-Lys compound demonstrated high binding capacity to both ACE and AChE, suggesting significant therapeutic potential. The synthesis and chemical characterization of the compounds confirmed their structure and provided a solid basis for their further study. In the enzyme inhibition assays, inhibition of ACE activity was observed, especially with the compound BXZ-Lys. Regarding AChE, all compounds showed a higher affinity than ACE, again highlighting BXZ-Lys as the best candidate, with a significant competitive inhibition and a high interaction affinity with the enzyme. In summary, boroxazolidone amino acid derivatives show promising therapeutic potential as ACE and AChE inhibitors, especially the compound BXZ-Lys. These findings offer new perspectives in the development of more effective and better-tolerated treatments for high blood pressure and other conditions related to blood pressure regulation.

References

1. Romero-Aguilar, K. S, et al, 2019. Expert Opinion on Therapeutic Patents, 29(5), 339–351.
2. Campos-Nonato, et al, 2018. Salud Publica Mex, 60 (3), 233–243.
3. Moncloa, Alfonso Bryce, et al, 2015, A. T. S. Diagnóstico Perú, 54(4).
4. Alcazar JM, Oliveras A, et al, 2016, Lancet, 387 (10022), 957–967.

DIFFERENTIAL EFFECT OF MSCS ON THE IMMUNOMODULATION CAPACITY OF BREAST CANCER CELLS

V. M. Arenas-Luna^{2,1}, A. Cortes-Morales³, J. Montesinos³, B. Cisneros², S. Hernandez¹

Panamerican University School of Medicine¹, Mexico City, Mexico; Center for Research and Advanced Studies, Mexico City, Mexico²; Oncology Research Unit, Oncology Hospital, National Medical Center, IMSS, Mexico City, Mexico³.
varenas@up.edu.mx; shernand@up.edu.mx

The therapeutic interest in mesenchymal stem cells (MSC) has progressively grown as they participate in various physiological processes such as homeostasis, regeneration, tissue repair, and modulation of the immune response. Currently, there are important challenges and questions that must be addressed, as increasing evidence implicates them in the development and progression of cancer. It has been observed that MSC, through soluble molecules, can promote processes in cancer cells such as survival, proliferation, epithelial-mesenchymal transition, among others. However, the effect that MSC may have on the immunoregulatory properties of breast cancer has not been fully studied.

Therefore, the aim of this work was to study the effect that bone marrow-derived MSCs (hBM-MS) through their conditioned medium (hBM-MS-cm) can have on the immunoregulatory capacity of breast cancer cells MDA-MB-231 and BT-474 lines.

Methods, Results & Conclusion. By qRT-PCR, we evaluated the gene expression of TGF- β , IDO, IL-4, and IL-10 levels in MDA-MB-231 and BT-474 breast cancer cells in the presence of hBM-MS-cm. Additionally, coculture assays were performed with peripheral blood-derived mononuclear cells (MNC) previously activated with phytohemagglutinin, to evaluate the proliferation of MNC. The differentiation of MNC towards regulatory T cells in the presence of breast cancer cells with and without hBM-MS-cm was also evaluated. Finally, in the supernatant of the co-culture assays, the concentration of TGF- β , IL-4, and IL-10 proteins and the expression of intracellular IDO in the tumor cells were determined.

hBM-MS-cm promoted the overexpression of the immunosuppressive genes TGF- β , IDO, and IL-10 in MDA-MB-231 cells. The immunoregulation assays with MNC in coculture with MDA-MB-231 cells and hBM-MS-cm showed a reduction in lymphocyte proliferation and an increase in the levels of IL-10 and TGF- β proteins from the supernatant, and intracellular IDO in MDA-MB-231 cells. However, TNF levels were reduced, while an increase in the proportion of regulatory T cells was observed. In contrast, hBM-MS-cm did not affect the immunomodulatory capacity of BT-474 tumor cells. In this way, a differential immunoregulatory effect was observed between these two breast cancer cell lines, representative of different subtypes: MDA-MB-231 (triple negative) and BT-474 (luminal). Understanding the immune response in a broader tumor context could help design therapeutic strategies based on the aggressive behavior of breast cancer.

SEARCH FOR GENETIC BIOMARKERS OF RISK FOR SECONDARY FAILURE TO SULFONYLUREAS

Mónica Vianney Ayala Martínez, Diana Barrera Oviedo, Alejandra Figueroa Rios, Nidia Samara Rodríguez Rivera

Universidad Nacional Autónoma de México, Departamento de Farmacología, Facultad de Medicina
Av. Universidad 3000, Copilco Universidad, CDMX, CP 04510. 55 5623 2165.
nidia.rodriguez@comunidad.unam.mx

Background. Diabetes is a highly prevalent pathology nationally and worldwide. In Mexico is the second leading cause of death and the first cause of disability. Nearly half of patients with type 2 diabetes (T2D) cannot reach glycemic goals, even after years of treatment. Despite the wide availability of drugs, the same hypoglycemic agent produces different therapeutic responses in everyone, along with severe side effects, suggesting that interindividual response is genetically determined. Appropriate treatment selection has become vital, and international associations recommend precision medicine as a tool to choose and individually guide the proper use of oral hypoglycemics. This makes the search and selection of drug response biomarkers of paramount importance.

Objective. Describe the frequencies of the rs757110 (S1369A), rs1799854 (-3C/T), and rs5219 (E23K) polymorphisms in *ABCC8* and *KCNJ11* genes in a Mexican mestizo population of T2D patients receiving treatment with sulfonylureas (SU) and compare them with other populations.

Methods. Allele discrimination genotyping with real-time PCR was performed on the three polymorphisms, using genomic DNA extracted from the peripheral blood of patients with T2D.

Results. The frequencies of all three study polymorphisms were found to be in Hardy-Weinberg equilibrium. In the rs757110 (S1369A) polymorphism in *ABCC8* and rs5219 (E23K) in *KCNJ11*, the behavior of genotype frequencies reflects the history of ancestry in the Mexican population and the tendency of these polymorphisms to be inherited jointly.

The rs1799854 (-3C/T) polymorphism in the *ABCC8* gene had an atypical distribution compared to the expected behavior, even when compared to another population of Mexican mestizos. This polymorphism may be related to an increased risk of secondary failure to sulfonylurea treatment.

Conclusions. The rs1799854 (-3C/T) polymorphism in the *ABCC8* gene showed an atypical distribution in Mexican patients with type 2 diabetes mellitus, and its possible relationship with secondary failure to sulfonylureas needs further investigation to determine whether it can be used as a biomarker for personalized medicine, especially in Mexico.

ANTIOBESOGENIC EFFECT OF D-LIMONENE, ELLAGIC ACID, GALLIC ACID, AND P-COUMARIC ACID IN THE LIVER OF RATS FED WITH HIGH-FAT DIET

¹Luis Alberto Ayala-Ruiz, ¹Luis Gerardo Ortega-Pérez, ²Joel Edmundo López-Meza, ¹Patricia Ríos-Chávez

¹Facultad de Biología, Universidad Michoacana de San Nicolás de Hidalgo. Cd. Universitaria, C.P. 58030, Morelia, Michoacán, México. ²Centro Multidisciplinario de Estudios en Biotecnología, Facultad de Medicina Veterinaria y Zootecnia Km. 9.5 Carretera Morelia-Zinapécuaro, Posta Veterinaria. C.P. 58893.
E-mail: 1232816g@umich.mx

Intake of high fat and sugar diets leads to an accumulation of adipose tissue and is linked to obesity, insulin resistance, cardiovascular diseases, and oxidative stress. Diets rich in saturated fatty acids triggers various signaling pathways that increase reactive oxygen species and inflammation in different cells, thereby impacting cellular function¹. Terpenes and phenols are natural products found in plants, and these compounds have many biological properties. *Callistemon citrinus* belongs to the Myrtaceae family, which harbors a high concentration of such compounds. This study examined the antiobesogenic effect of d-limonene, ellagic acid, gallic acid, and p-coumaric acid in rats fed with a high-fat diet (HFD). 54 male albino Wistar rats were randomly divided into nine groups (n=6). Group I was the control (Purina® Rodent Chow food); group II was HFD (63% of the Purina® Rodent Chow food, 41.66% of INCA® vegetable fat, 41.66% of lard, and 16.66% of sucrose). Groups III to IX were fed with HFD and administered orally with different compounds, group III (metformin at 20 mg/kg), group IV (*C. citrinus* at 200 mg/kg), group V (d-limonene at 0.43 mg/kg), group VI (ellagic acid at 74.3 µg/kg), group VII (gallic acid at 6.94 µg/kg), group VIII (p-coumaric acid at 0.47 µg/kg), and group IX (d-limonene at 0.43 mg/kg, ellagic acid at 74.3 µg/kg, gallic acid at 6.94 µg/kg, and p-coumaric acid at 0.47 µg/kg) for 23 weeks. Ellagic acid, d-limonene, gallic acid, and p-coumaric acid reduced weight gain, Lee index, adiposity index, liver weight, glucose levels, and triacyl glycerides levels. Our study demonstrates that terpenes and phenols have anti-obesity and hypolipidemic functions, suggesting that they might be effective for treatment of obesity.

References

1. Li, Q., Liu, Z., Huang, J., Luo, G., Liang, Q., Wang, D. & Hu, J. (2013). Journal of the Science of Food and Agriculture, 93(6), 1310-1316.

ESTIMATE OF THE CONSUMPTION OF NON-CALORIC SWEETENERS IN ADULTS AND THEIR ASSOCIATION WITH THE RISK OF METABOLIC DISEASES

Edgar Daniel Barreras Duran¹, Norma Patricia Adán Bante²,
Jesús Alfredo Rosas Rodríguez² and Norma Aurora Stephens Camacho^{1*}

¹Universidad Estatal de Sonora, Blvd. Manlio Fabio Beltrones 810, Colonia Bugambillas, CP 85875 Navojoa, Sonora,

²Universidad de Sonora, Blvd. Lázaro Cárdenas No. 100, Colonia Francisco Villa. CP85880, Navojoa, Sonora.

*E-mail: aurora.stephens@ues.mx

Non-caloric sweeteners (NCS) are natural or synthetic additives that provide the sweet flavor of foods without the energy load of sugar¹. Its consumption is acceptable as a measure of glycemic control for diabetic people². Currently, they are used as a preventive measure against obesity by controlling caloric intake without discriminating against age groups or the nutritional status of the consumer³⁻⁶. There is controversy about the effects they can generate in the long term since multiple studies indicate that their consumption may be related to the development of chronic pathologies such as insulin resistance, obesity, and metabolic syndrome⁷⁻⁹. Our work aims to estimate the daily amount of ENC consumed by adults from Navojoa, Sonora, assessing their nutritional and metabolic status and inferring whether the intake of ENC is related to the metabolic disorders presented. The population evaluated was 51 adults, 37 women and 14 men. Our results show that 76% of the participants are unaware of the effects of ENC but are aware of its consumption (73%). The average intake of these sweeteners in the participants showed that sucralose has the highest daily consumption at 33 mg, followed by Acesulfame potassium at 20.2 mg, Aspartame at 17.5 mg, and *Steviol glycosides* at 15 mg. Determining nutritional status indicates that 53% are categorized in some stage of obesity, 31.3% as overweight and the rest as average weight. The risk of developing metabolic complications was categorized as substantially increased with 52.9% of the population evaluated; 25.5% were considered without risk, and the rest increased. The statistical analysis shows that the consumption of ENCs is associated with the development of metabolic risk in the study volunteers with a Chi² value of 7.37 and a Cramer's V of 0.76, p-value > 0.05, and 2 degrees of freedom. Normal glucose values were observed in 45 volunteers with an average of 90.19±4.9 mg/dL; five volunteers showed values of 109.2±6.9 mg/dL, being considered prediabetic, and a single volunteer was categorized as diabetic by showing values of 135.7 mg/dL. Finally, the insulin resistance values indicated that of the population evaluated, 60.8% are insulin resistant with a HOMA index of 3.62±2.3; the rest were considered normal with a value of 1.3±0.32. In conclusion, the average consumption of ENC in older adults is below the recommended daily intake; however, the rate of obesity and overweight prevails in the sample evaluated, and there is a relationship between consumption and the probability of generating a metabolic alteration.

Reference

1. Azad, M. B., Archibald, A., Tomczyk, M. M., Head, A., Cheung, K. G., de Souza, R. J., Becker, A. B., Mandhane, P. J., Turvey, S. E., Moraes, T. J., Sears, M. R., Subbarao, P., & Dolinsky, V. W. (2020). *International Journal of Obesity*, 44(10), 2137–2148.
2. Gómez-Arauz, A. Y., Bueno-Hernández, N., Palomera, L. F., Alcántara-Suárez, R., De León, K. L., Méndez-García, L. A., Carrero-Aguirre, M., Manjarrez-Reyna, A. N., Martínez-
3. Reyes, C. P., Esquivel-Velázquez, M., Ruiz-Barranco, A., Baltazar-López, N., Islas- Andrade, S., Escobedo, G., & Meléndez, G. (2019). *Journal of Immunology Research*, 2019, 1–10.
4. Grembecka, M. (2015). Natural sweeteners in a human diet. *Roczniki Państwowego Zakładu Higieny*, 66(3), 195–202.
5. Liauchonak, I., Qorri, B., Dawoud, F., Riat, Y., & Szewczuk, M. R. (2019). *MDPI AG (Vol. 11, Issue 3)*.
6. Paredes-Serrano, P., Alemán-Castillo, S., Castillo-Ruiz, O., & Perales-Torres, A. L. (2016). *BIOTecnica*, 18(1), 55.
7. Romo-Romo, A., Almeda-Valdés, P., Brito-Córdova, G. X., & Gómez-Pérez, F. J. (2017). *Gaceta Medica de Mexico*, 153(1), 61–74.
8. Sylvetsky, A. C., Bauman, V., Blau, J. E., Martin Garraffo, H., Walter, P. J., Rother, K. I., & Author, C. (2017). *Toxicol Environ Chem*, 99(3), 535–542.
9. Sylvetsky, A. C., Brown, R. J., Blau, J. E., Walter, M., & Rother, K. I. (2016). *Nutrition & Metabolism*, 13(1), 71
10. Sylvetsky, A. C., Sen, S., Merkel, P., Dore, F., Stern, D. B., Henry, C. J., Cai, H., Walter, P. J., Crandall, K. A., Rother, K. I., & Hubal, M. J. (2020). *Molecular Nutrition & Food Research*, 64(11), 1–10.

ANALYSIS OF THE EXPRESSION OF ENZYMATIC ONCOLOGICAL CLINICAL BIOMARKERS IN HUMAN SALIVA

Silvia Barrios Aguilar¹, Cesar A. Esquivel Chirino², Mario A. Bolaños Carrillo³,
Martín Salas Paniagua⁴, Delina G. Montes Sanchez⁴

Alumna de Licenciatura en Biología, Universidad Autónoma de Sinaloa. Profesor-investigador, Facultad de Odontología, Universidad Nacional Autónoma de México. Profesor-investigador, Área de Ciencias Naturales, Universidad del Valle de México, Campus Guadalajara Sur. Profesor-investigador, Licenciatura en Estomatología, Complejo Regional Sur, Benemérita Universidad Autónoma de Puebla. Carretera Libramiento Tecnológico San Marcos Necoxtla Km 7.5, Tehuacán, Puebla, México.
Cellphone: +52 (1) 22 24 38 78 79
Email: delina.montessanchez@viep.com.mx

Introduction. Saliva plays an important role in the oral cavity due to lubrication, repair, autolysis, antimicrobial and balancing properties. It is considered one of the most important body fluids for the diagnosis of oral and systemic diseases. Cancer is a multifactorial pathological process that is characterized by a high rate of cell duplication, damaging nearby tissues, and causing the formation of aggregates, which often migrate to distant places in the body, to invade them and continue proliferating (metastasis), causing the death of affected individuals. **Objective.** To analyze the main salivary enzymes (amylase, lysozyme and lipase) as clinical oncological biomarkers in the development of head and neck cancers, promoting the potential future use of saliva as a non-invasive, economical and easily accessible diagnostic method. **Methodology.** Saliva was collected from patients diagnosed with some type of head or neck cancer, the protein concentration was quantified, and the extracts were analyzed by mass spectrometry, to identify, evaluate, interpret and synthesize the information obtained, focusing on the main salivary enzymes: amylase, lysozyme and lipase, as potential markers in oncological processes. **Results.** Enzymes are among the proteins with the greatest diversity of functions in the body, they are involved in multiple metabolic processes and, therefore, are also key players in health-disease processes. Cancer is a pathology that is characterized by the increase and acceleration of all cellular functions and activities, which leads to changes in the expression of many proteins, including enzymes. Total saliva samples analyzed by mass spectrometry highlight the presence of peaks that, in terms of molecular weight, resemble the molecular weights of salivary enzymes. **Conclusion.** There's not enough data about the role with which salivary enzymes could be participating in the oncological process. Lysozyme is the most reported in the literature, and it is suspected that it has a relationship with the pathophysiological process of carcinogenesis in the digestive system. It is even mentioned in some articles that lysozyme could function effectively as a clinical biomarker in digestive oncological processes. The other two enzymes need to be analyzed in more detail.

DEVELOPMENT OF AN ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA) FOR THE RAPID DETECTION OF THE NON-STRUCTURAL PROTEIN 1 (NS1) OF THE DENGUE VIRUS

Maidoly Batista Fernández¹, Julio García Cordero¹, Gabriela Mellado Sánchez²,
David Andrés Benavides Fernández³, Edgar Morales Ríos¹, Leticia Cedillo Barrón¹

¹Center for Research and Advanced Studies (CINVESTAV-IPN), Av. IPN #2508 Col. San Pedro Zacatenco 07360 Mexico City, Mexico, ²Unidad de Desarrollo e investigación de Bioprocesos (UDIBI), Prolongación de Carpio and Plan de Ayala 11340 Mexico City, Mexico, ³Center for Engineering and Industrial Development, Av. Pie de la Cuesta #702, Desarrollo San Pablo 76125 Querétaro, Mexico.
E-mail: maidoly.batista@cinvestav.mx

Dengue disease is caused by the Dengue Virus (DENV), and is considered as a re-emergent disease due to the significant increase in incidence over recent decades. Since early 2023, an unprecedented peak of cases has been recorded, with over five million cases across 80 countries. The Americas are among the most affected regions, reporting over 80% of the cases and the presence of all four viral serotypes (DENV-1, DENV-2, DENV-3, and DENV-4)¹. There are multiple strategies for DENV diagnosis, one common approach is the development of sandwich ELISAs to detect the NS1 viral antigen², a marker of early infection stages³.

This study aims to design, develop, and evaluate a sandwich ELISA to detect NS1 protein in different samples, from the recombinant protein to DENV-infected patient serum, potentially contributing to better viral diagnosis kits in the future. Initially, the NS1 protein of DENV-2 was expressed and purified from bacteria transformed with a plasmid encoding for NS1 protein. This recombinant protein was used for rat immunization and to standardize sandwich ELISA conditions.

We produced and purified specific anti-NS1 monoclonal murine antibodies (mAbs) and rat polyclonal antibodies (pAbs) for assay development. Both mAbs and pAbs recognized and detected the NS1 protein by an indirect ELISA. Optimal concentrations were set at 1 µg/mL for mAbs and 4 µg/mL for pAbs in the home sandwich sandwich ELISA. To do this, we established mAb as the capture antibody and pAb as the detection antibody.

The assay developed in this work is being validated through the detection of the native NS1 protein from supernatants of cells infected with DENV as well as the evaluation of sera from patients infected with the DENV. At the same time and with the appropriate sample size of human sera we will determine the sensitivity and specificity of the assay we propose.

References

1. PAHO. (2023).
2. Raafat N, Blacksell SD, Maude RJ. (2019). *Trans R Soc Trop Med Hyg.* 113(11)
3. Fisher R, Lustig Y, Sklan EH, Schwartz E. (2023). *Viruses.* 15(2).

WOOD SMOKE EXTRACT INDUCES ALTERATIONS IN MITOCHONDRIAL BIOGENESIS AND INCREASED PRODUCTION OF REACTIVE OXYGEN SPECIES IN NORMAL HUMAN FIBROBLASTS

Carina Becerril Berrocal, José Pedraza Chaverri, Marisol Orozco Ibarra, Emiliano Aparicio Trejo, Alfonso Salgado Aguayo, Erika Rubí Luis García

Instituto Nacional de Enfermedades Respiratorias "Ismael Cosío Villegas" Calz. de Tlalpan 4502, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México, CDMX.
Teléfono: 55 1008 86 03; email: cabe5568@gmail.com

Abstract. Several studies have shown that mitochondrial quality control deregulation is associated with the pathogenesis of various lung diseases, including chronic obstructive pulmonary disease (COPD). Inhalation of wood smoke induces long-term pathological changes in the lung, its main effect being the development of COPD, which is associated with an increase in oxidative stress and apoptosis of lung cells. The accumulation of dysfunctional mitochondria induces an increase in the generation of reactive oxygen species and cell death. This work aims to analyze the effect induced by wood smoke extract (WSE) *in vitro* on mitochondrial dynamics in normal human lung fibroblasts and whether these changes are associated with the deregulation of mitochondrial biogenesis processes. **Method.** Human lung fibroblast cell lines CCD-16 and CCD-19, obtained from ATCC, were used. The fibroblasts were incubated with dilutions of WSE (1% and 2.5%) for 24 hours. Subsequently, morphological changes were visualized by bright-field microscopy. The mitochondrial network was visualized by confocal microscopy using Mito Tracker Red. Protein expression analysis was performed by Western Blot (WB). Mitochondrial membrane potential ($\Delta\Psi_m$) was evaluated using the lipophilic cationic dye JC-1. ATP production was measured using a total ATP determination kit. **Results.** Our results showed that WSE at 2.5% induces morphological changes and fragmentation in the mitochondrial network with perinuclear accumulation of mitochondria in fibroblasts. We observed that administration of different doses of WSE (1% and 2.5%) significantly decreases $\Delta\Psi_m$. WSE alters the expression levels of fusion proteins such as Mfn1 and fission proteins such as Fis1. Additionally, stimulation with WSE (2.5%) decreases total ATP levels compared to the control group ($p < 0.05$). On the other hand, mitochondrial superoxide production was determined using the MitoSOX probe. Our results showed an increase in O_2^- production when stimulated with WSE at 2.5% compared to the control group ($p < 0.05$).

EFFECT OF GLYCINE BETAINE ON THE PROLIFERATION OF HUMAN COLON ADENOCARCINOMA CELLS HT-29

Eiby Grisell Benitez Ibarra¹, Lizeth Anahí López Castro², Norma Aurora Stephens Camacho³, Jesús Alfredo Rosas Rodríguez¹, Ramona Icedo García¹ and Guadalupe González Ochoa¹

¹Universidad de Sonora, Lázaro Cárdenas del Río 100, Francisco Villa, CP 85880, Navojoa, Son., ²Universidad Estatal de Sonora, Blvd. Manlio Fabio Beltrones 810, Bugambillas, CP 85875, Navojoa, Sonora. E-mail: guadalupe.gonzalezchoa@unison.mx, Tel: 6424259950, CP 85880.

Cancer is the abnormal and accelerated growth of cells that confer malignancy and the capacity to affect other organs¹. Three of the most frequent types of cancer are lung cancer, breast cancer and colon cancer worldwide². Colon cancer is a malignant neoplasm that is generated in the normal mucosa of the colon, and can develop in the different segments of the large intestine³. In recent years, research has shown that some natural compounds can act adjuvantly in the treatment of colon cancer. In this regard, Glicine Betaine (GB) and products with high concentrations of GB (beets, wheat bran and others) have shown beneficial effects against cancer cells⁴⁻⁷. To determine the anti-proliferative effect of GB against cancerogenic cells, this study aimed to analyze the effect of GB in human colon adenocarcinoma cells HT-29. The cells HT-29 were treated with several concentrations of GB (7.8, 15.6, 31.2, 62.5 y 125 mg/ml) incubated 24, 48 or 72 h at 37°C and 5%CO₂. After the respective incubation time, an MTT assay was done on cells treated with GB or without GB (control). Cells' viability dropped to 50% in assays where cells were incubated with GB 24 and 48 h; whereas it dropped to 39% in cells incubated with GB 72h. Our results indicate that GB might have a dose-dependent antiproliferative effect on human colon adenocarcinoma cells HT-29

References

1. Cuevas, R. (2017). CHARACTERIZATION OF COLON CANCER. *Cirugía Paraguaya*, 41(1), 8-13. <https://doi.org/10.18004/sopaci.abril.8-13>
2. Dekker, E., Tanis, P. J., Vleugels, J. L. A., Kasi, P. M., & Wallace, M. B. (2019). Colorectal cancer. *The Lancet*, 394(10207), 1467-1480. [https://doi.org/10.1016/s0140-6736\(19\)32319-0](https://doi.org/10.1016/s0140-6736(19)32319-0)
3. Leal-Omar, J., Zarate-Guzmán, J., Peniche-Moguel, P., Gil-Rojas, N., Briones-Fraga, S., Gómez-García, T., Morones-Navarrete, G., & Mena-Contreras, A. A. (s. f.). Demografía del cáncer colorrectal en los últimos 5 años de un hospital oncológico. *www.scielo.org.mx*. <https://doi.org/10.24875/end.m19000125>
4. Hurtado, A. (2021). La Betaína: un Osmoprotector natural. *Badajoz Veterinaria*, 23, 12-19. <https://dialnet.unirioja.es/descarga/articulo/8153834.pdf>
5. Zhao, G., He, F., Wu, C., Li, P., Li, N., Deng, J., Zhu, G., Ren, W., & Peng, Y. (2018). Betaine in Inflammation: Mechanistic Aspects and Applications. *Frontiers In Immunology*, 9. <https://doi.org/10.3389/fimmu.2018.01070>
6. Guo, Y., Xu, L., Ding, Z., Liao, Y., Wang, H., Lan, Z., Guan, W., & Liu, C. (2015). Betaine Effects on Morphology, Proliferation, and p53-induced Apoptosis of HeLa Cervical Carcinoma Cells in Vitro. *Asian Pacific Journal Of Cancer Prevention*, 16(8), 3195-3201. <https://doi.org/10.7314/apjcp.2015.16.8.3195>
7. Kar, F., Hacıoğlu, C., Kaçar, S., Şahintürk, V., & Kanbak, G. (2019). Betaine suppresses cell proliferation by increasing oxidative stress-mediated apoptosis and inflammation in DU-145 human prostate cancer cell line. *Cell Stress & Chaperones*, 24(5), 871-881. <https://doi.org/10.1007/s12192-019-01022-x>

EFFECT OF EMPAGLIFLOZIN TREATMENT ON MAGNESEMIA IN METABOLIC SYNDROME

Juan C. Cabrera-Ángeles^{1,2*}, Zambrano-Vásquez Oscar R.², Hernández-Alba Ana L.^{1,2}, Fernando Cortes Camacho², Valle-Velázquez Estefania², Gonzaga-Sánchez José G.², Sánchez-Lozada Laura Gabriela², Juárez Rojas Juan G.⁵, M. Carmen Castillo Hernández¹, Osorio-Alonso Horacio²

¹Sección de Estudios de Posgrado e Investigación, Escuela Superior de Medicina-IPN, México City, México.

²Departamento de Fisiopatología Cardio-Renal, Instituto Nacional de Cardiología Ignacio Chávez, Mexico City, Mexico.

⁵Departamento de Farmacología, Instituto Nacional de Cardiología Ignacio Chávez, Mexico City, Mexico.
cabrera97juancarlos@gmail.com*

Background. Magnesium (Mg²⁺) is an essential ion that plays a key role in all energy-dependent transport systems, maintenance of vascular tone, and secretion and action of insulin, thus depletion impairs glucose homeostasis and insulin sensitivity resulting in illnesses including diabetes, arterial atherosclerosis, and nephropathy. On the other hand, metabolic syndrome (MS) clustered risk factors (RF) that has been related to hypomagnesemia. It has been reported that strict metabolic control reduces urinary Mg wasting and leads to normomagnesemia. Sodium-glucose cotransporter type 2 inhibitors (SGLT2i) have shown beneficial effects on RF, but its effects on magnesium homeostasis it is unknown.

Aim. To study the effect of empagliflozin on magnesium homeostasis in metabolic syndrome.

Methodology. MS was induced in male Wistar rats (200-220g) with a type Paygen diet. Once established and validated MS, rats were randomly assigned to MS untreated, Empagliflozin (12.5 mg/kg/day). We validate the establishment of metabolic syndrome through body weight, blood pressure (BP), fasting blood glucose, glucose tolerance test (GTT), and lipid profile measurements. Also were analysed serum magnesium, diuresis and magnesuria.

Results. Previous to empagliflozin treatment, the results showed an increase in blood pressure, fasting glucose, and impaired GTT and lipid profile, which demonstrated the establishment of metabolic syndrome. Also, in MS group increased serum magnesium, magnesuria and diuresis compared with the control group, which demonstrated alterations in the body status of magnesium during MS. Empagliflozin treatment was successful in controlling blood pressure and improving fasting blood glucose, GTT, and lipid profile. The serum levels of magnesium were restored with the treatment of empagliflozin, and no significant changes were found in magnesuria and diuresis between the treated group when compared with the untreated group. However, the urinary excretion of magnesium was higher in the MS group of 60 days compared with 30 days.

Conclusion. The control of risk factors with empagliflozin restores magnesemia in metabolic syndrome, suggesting enhanced magnesium transporters function in the kidney. Therefore, empagliflozin could delay the progression to diabetes and cardiovascular disease induced by MS.

CONAHCYT: B230044

SEQUESTOSOME1 NUCLEAR TRANSLOCATION AFTER DNA DAMAGE IN LUNG EPITHELIAL CELLS

Sandra Cabrera, Iliana Herrera, Pamela Gutiérrez, Gabriel Muciño, Annie Pardo, Moisés Selman

¹Laboratorio de Fibrosis, Unidad de Biopatología Pulmonar, Ciencias-INER, Universidad Nacional Autónoma de México.

²Instituto Nacional de Enfermedades Respiratorias "Ismael Cosío Villegas", México.

DNA damage is known to play an important role in pulmonary fibrosis and DNA repair systems are crucial for the survival of lung cells. Exogenous agents that cause DNA damage are mainly environmental factors, including chemical attacks, ultraviolet rays, and X-rays. But also cellular metabolism acts as a source of endogenous ROS that cause DNA damage. The protein sequestosome1 (SQSTM1), also known as p62 is a multidomain protein induced by various forms of cellular stress and has been implicated in several signal transduction pathways. p62, is degraded by autophagy, and acts as a cargo receptor for autophagic degradation of ubiquitinated targets. It is also suggested to shuttle ubiquitinated proteins for proteasomal degradation. In this study, we decided to explore the role of p62 after UV-DNA damage in lung epithelial cells in vitro and in vivo using 2 animal models of lung fibrotic diseases. To induce lung fibrosis, C57BL6 mice were intratracheally instilled with bleomycin, sacrificed after 28 days and lungs were obtained to perform immunohistochemistry and immunofluorescence. To induce hypersensitivity pneumonitis, C57BL6 mice were intranasally instilled with 50 µg SR in 25 µL of sterile saline for three weeks (three times per week) and were euthanized three days after the last exposure.

We have found increased immunoreactivity for p62 in lungs from bleomycin treated and *Saccaropolyspora rectivirgula* exposed mice compared to saline treated control mice. Interestingly, we have found nuclear p62 subcellular localization in epithelial cells and macrophages in damaged lungs compared to control lungs. These findings indicate that cellular stress induces p62 expression, but also nuclear relocalization in the lung. In vitro, MLE12 lung epithelial cells were exposed to UV irradiation by 3, 6, 12 and 30 minutes. After UV irradiation we observed chromatin condensation and micronuclei formation DNA damage was corroborated by p53 and H2A positive nuclear signals by immunofluorescence. Additionally, we observed p62 nuclear translocation after UV- irradiation DNA damage in MLE12 cells. In summary, our results indicate that DNA damage could induce p62 nuclear shuttling in epithelial cells and this phenomenon could be part of DNA repair mechanisms in the lung.

EFFECT OF COMPOUNDS FROM THE AQUEOUS EXTRACT OF *ERYTHRAEA TETRAMERA SCHIEDE* ON PANCREATIC LIPASE ACTIVITY

Abelardo Camacho Luis¹, Kimberly Quezada Cardenas¹, Alejandra Vázquez Raygoza¹, Graciela Zambrano Galván¹, Laura Barragán Ledezma¹, Maricela Esteban Méndez²

¹Medicine and Nutrition Faculty, Juarez University of Durango State, University Avenue and Fanny Anitua, Durango, Durango. México, C.P. 34000. ²Interdisciplinary Research Center for the Integral Regional Development – Durango, National Polytechnic Institute. Sigma 119 Fracc. 20 de noviembre II. Durango, Dgo. México. C. P. 34220
E-mail: abelardo.camacho@ujed.mx

Currently, obesity is recognized as a global health problem of growing importance and an important risk factor for the development of chronic diseases such as diabetes mellitus, high blood pressure, dyslipidemia, coronary heart disease, cancer, among others (1). There are few medications to treat it; reducing lipid absorption through inhibition of pancreatic lipase has become the most favorable strategy. The only drug approved by the Food and Drug Administration (FDA) as a pancreatic lipase inhibitor is Orlistat, which can prevent the absorption of approximately 30% of lipids in the blood. Compounds from *Erythraea tetramera Schiede* could become promising alternatives for the development of safe drugs for the treatment of obesity (2). The purpose of the present work was to evaluate the effect of these compounds on the activity of pancreatic lipase. The inhibitory activity on pancreatic lipase of the compounds daidzein, quercetagenin, apigenin, 4-methoxycinnamic acid, p-coumaric acid and ferulic acid was quantified using a colorimetric assay that measures the release of p-nitrophenol described by Bustanji et al, 2010 with modifications (3). The 4-methoxycinnamic acid and ferulic acid showed highest percentage of inhibition (92.09% and 76.49%, respectively), the IC₅₀ of the compounds was determined as: 96 µM for ferulic acid and 101 µM for 4-methoxycinnamic acid. Finally, the type of inhibition of the compounds was determined, resulting in uncompetitive results for both.

References

1. Stevens, G.A., Singh, G.M., Lu, Y. et al. (2012). *Popul Health Metrics* 10, 22 (2012). <https://doi.org/10.1186/1478-7954-10-22>
2. Saucedo-Tamayo, M. del S. et al. (2014) *Biocencia*. 16, 2 (ago. 2014), 8–14. DOI:<https://doi.org/10.18633/bt.v16i2.39>.
3. Bustanji, Y., Issa, A., Mohammad, M., Hudaib, M., Tawah, K., Alkhatib, H., ... & Al-Khalidi, B. (2010). *J Med Plants Res*, 4(21), 2235-42.

ANALYSIS OF PULMONARY SURFACTANT PROTEIN C DURING AGING

Uriel Camacho-Silverio¹, Tania Valdivia-Herrera¹, Mariana Río de la Loza¹, David A. Lizcano¹, Remedios Ramírez¹, Fernanda Toscano-Marquez², Moisés Selman², Annie Pardo¹, Yair Romero¹

¹Facultad de Ciencias, UNAM. Laboratorio de biopatología pulmonar. ²Instituto Nacional de Enfermedades Respiratorias, Laboratorio de biopatología Pulmonar
E-mail: urielcs3143@ciencias.unam.mx

Introduction. Aging is characterized by a decline in the activity of various biological mechanisms and an increased susceptibility to diseases¹. Alveolar Epithelial Cells Type II (AECII) are responsible for the biosynthesis and secretion of Pulmonary Surfactant (PS), as well as in the regenerative capacity of alveolar epithelium. PS is a dynamic system that reduces surface tension within the alveoli. It is composed of phospholipids and hydrophobic proteins (SP-B, SP-C) that play a role in the surfactant's biophysical activity, along with hydrophilic proteins (SP-A, SP-D) associated with the lungs' innate immunity. Pulmonary surfactant proteins SP-B and SP-C are stored in Multilamellar Bodies (MLB) within AECII.² Their suppression is implicated in the disruption of pulmonary homeostasis^{3,4}, a characteristic often linked to aging in the literature but supported by limited evidence. Therefore, it is crucial to study the changes in surfactant during aging to understand how these changes might contribute to aberrant changes in the alveolar epithelium. **Methods.** Immunodetection assays and relative gene expression analysis were performed to determine the expression status of SP-C in whole lungs and AECII from mice with accelerated aging (*Zmpste24*^{-/-}) and natural aging (wild type, C57: 18-28 months). Ultrastructural analysis using Transmission Electron Microscopy (TEM) was conducted to identify changes in MLB. In organoid models, immunostaining assays for SPC and cell senescence markers were performed. **Results.** SPC expression was increased in AECII and lungs in both natural and accelerated aging models. Moreover, cells in culture from both models demonstrated the ability to proliferate and regenerate. Contrary to some studies, natural aging is associated with an increase in SP-C expression. The isolated cells retained proliferative capacity, suggesting that in diseases where SP-C activity is reported to be altered with aging, other initiating factors may be required. The knockout model also appears to successfully mimic what occurs with this system during natural aging. The presence and number of MLBs suggest the maintenance of surfactant activity; however, the presence of AM may indicate a more active role of the surfactant surveillance system. **Conclusions.** In summary, our study highlights an increase in SP-C expression during both natural and accelerated aging in the lung, contrary to previous findings. This suggests ongoing cellular regeneration despite aging.

References

1. López-Otín, C., Blasco, M. A., Partridge, L., Serrano, M. & Kroemer, G. The hallmarks of aging. *Cell* 153, 1194–1217 (2013).
2. Mason, R. J. & Dobbs, L. G. Alveolar Epithelium and Pulmonary Surfactant. in Murray and Nadel's Textbook of Respiratory Medicine 134-149.e5 (Elsevier, 2016). doi:10.1016/B978-1-4557-3383-5.00008-7.
3. Foster, C. D., Zhang, P. X., Gonzales, L. W. & Guttentag, S. H. In vitro surfactant protein B deficiency inhibits lamellar body formation. *Am J Respir Cell Mol Biol* 29, 259–266 (2003).
4. Ruwisch, J. et al. Air Space Distension Precedes Spontaneous Fibrotic Remodeling and Impaired Cholesterol Metabolism in the Absence of Surfactant Protein C. *Am J Respir Cell Mol Biol* 62, 466–478 (2020).

POLAR FRACTION OF *LOPHOCEREUS SCHOTTII* DECREASES *TGFB1* GENE EXPRESSION IN THE PROGRESS OF CHEMICALLY INDUCED HEPATOCARCINOGENESIS

Marina Campos-Valdez¹, Jaime Sánchez-Meza¹, Arturo Orozco-Barocio², José A. Domínguez-Rosales¹, Juliana Marisol Godínez-Rubí³, Sarai C. Rodríguez-Reyes⁴, Erika Martínez-López⁴, Miriam R. Bueno-Topete¹, Manuel A. Castro-García, Guillermo M. Zúñiga-González⁵, Laura V. Sánchez Orozco¹

¹Instituto de Enfermedades Crónicas Degenerativas, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara, 44340 Guadalajara, Jalisco, México, ²Laboratorio de Inmunobiología, Departamento de Biología Celular y Molecular, Centro Universitario de Ciencias Biológicas y Agropecuarias, Universidad de Guadalajara, 44600 Zapopan, Jalisco, México. ³Laboratorio de Patología Diagnóstica e Inmunohistoquímica, Departamento de Microbiología y Patología, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara, Guadalajara, Jalisco, México, ⁴Instituto de Nutrigenética y Nutrigenómica Traslacional, Centro Universitario de Ciencias de la Salud, Guadalajara, Jalisco, México, ⁵Laboratorio de Mutagénesis, Centro de Investigación Biomédica de Occidente, Instituto Mexicano del Seguro Social, Guadalajara, Jalisco, México.
E-mail: laura.sorozco@academicos.udg.mx; Phone number: +52 33 3954 5677

Introduction. There is evidence of the anticancer action of the polar fraction of *Lophocereus schottii* (LsPF) in experimental models of lymphoma, but its effect on hepatocellular carcinoma (HCC) models has not been studied.

Objective. To evaluate the effect of LsPF on the progress of liver damage induced by chronic administration of diethylnitrosamine (DEN) and N-2-Fluorenylacetamide (2-AAF).

Materials and Methods. Four groups of Male Wistar rats (180-200 g) were formed: 1) Control (Ctl); 2) LsPF, treated 3 times a week with LsPF (50 mg/Kg, i.g.); 3) Damage (Dmg), weekly treated with DEN (50 mg/Kg, i.p) on day one and with 2-AAF (25 mg/Kg, i.g.) on day three; 4) Damage+LsPF (Dmg+LsPF), received the same treatment (Tx) as Dmg group but since the seventh week LsPF (50 mg/Kg) was administrated along with the Dmg Tx. Treatments were maintained for 13 weeks (wks) and the animal's liver and serum were collected. Hematoxylin & Eosin and Masson's Trichrome stains were performed, in addition to serum biochemistry analysis. Parametric Student's t-tests or nonparametric Kruskal-Wallis and Mann-Whitney U tests were performed.

Results. The mean final weights of the Dmg and Dmg+LsPF groups were significantly lower compared to the Ctl and LsPF groups. Furthermore, the livers of these Dmg groups presented tumors, discoloration, and significant hepatomegaly (increase in the ratio between the liver and animal weight). The Dmg Tx increased the levels ALT, AST, ALKP, total bilirubin, total protein, and GGT, and there were no significant differences between the Dmg and the Dmg+LsPF group. However, the gene expression analysis showed that in the Dmg group, the expression of *CAT*, *SOD*, *COL1A* and *TGFB1* significantly increased compared to the Ctl group; when the expression of the same genes was compared between the Ctl and Dmg+LsPF, there were no significant differences. Additionally, *TGFB1* gene expression was reduced statistically in Dmg+LsPF compared to Dmg. The administration of LsPF by itself increased the serum levels of ALT and total proteins, and the expression of *CAT* and *COL1A*. However, it is important to note that histologic analysis did not show any alteration in the liver following administration of the LsPF.

Conclusions. Gene expression analysis provided evidence that LsPF may act on genes associated with HCC development; nevertheless, this treatment did not significantly revert liver damage induced by the Dmg Tx. **Acknowledgments.** Grant PIN 2021 CUCS, UdG, P3E 271879-2023 and PRO SNI.

OBESITY LEADS TO TRANSCRIPTOMIC ALTERATIONS IN ONE-CARBON METABOLISM-RELATED GENES: IN SILICO ANALYSIS OF GTEX STUDY

Daniel Cantú-Ruiz, Erika Castaño-Moreno, Víctor Treviño, Rocío Díaz de la Garza*

Tecnológico de Monterrey, Institute for Obesity Research, Ave. Eugenio Garza Sada 2501, Monterrey 64849, NL, México.

*rociodiaz@tec.mx

B complex vitamins, such as folate, are involved in one-carbon metabolism (1-CM), which participates in the anabolism of methionine, purines, thymidylate, methylations, and reducing power in the cell. Lower folate status has been found in people with obesity; however, the mechanism behind this condition and its significance are unknown. Here, we report the changes in the transcriptomes of 1-CM-related genes regarding the BMI increments in human tissues. We used the Genotype-Tissue Expression (GTEx) database to analyze the expression of 77 1-CM-related genes in 42 human tissues normalized by TPM. The BMI, continuous and categorical ($BMI >29.99$ and <25) was used as a predictor variable. Linear regression models adjusted by sex and age were performed per gene. Wilcoxon comparisons between sexes within BMI groups were used for visual representation and validation. Differential expressions are estimated as log₂-fold-change (log₂-FC). A p-value <0.05 for significance statistics was used. The analyses were performed in the R language. Out of the 42 human tissues, significant changes in the expression of several genes were found in adipose (subcutaneous and visceral), and skeletal muscle tissues (SAT, VAT, SMT, correspondingly). 11 genes of the folate cycle were differentially expressed in obesity of both sexes (3 genes up regulated and 8 down regulated $p < 0.001$). On SAT, two folate transporter genes (*FOLR2* & *ABCC3*) were upregulated (log₂-FC: 0.39, and 0.4, respectively), whereas 4 genes involved in folate forms interconversion (*FPGS*, *ALDH1L1*, *MTHFD2L*), and *GART*, necessary for the purine synthesis, were down regulated (log₂-FC: -0.16, -0.42, -0.11, and -0.14, respectively). On VAT, the reduced folate carrier (*SLC19A1*) was down regulated (log₂-FC: -0.21), and 4 genes of the folate cycle, (*FPGS*, *SHMT1*, *SHMT2*, and *MTHFS*) were also downregulated in individuals with BMI >30 (log₂-FC: -0.2, -0.44, -0.18, and -0.18, respectively). On SMT, the *DHFR*, which is necessary for folic acid and dihydrofolate reduction to active folate, was upregulated (log₂-FC: 0.16), while *SHMT2*, responsible for the interconversion of Ser-Gly was downregulated (log₂-FC: -0.08). In obesity, among 42 tissues, only SAT, VAT, and SMT exhibit alterations in several 1-CM-related genes. This suggests that obesity plays a role in the modulation of the expression of folate pathway genes in tissues with high metabolic activity. We postulate that the generalized lower gene expression of the enzymes related to 1-CM and folate forms conversion could contribute to lower cell methylation capacity, lower or altered DNA/RNA synthesis and higher homocysteine with possible consequences in the plasma folate levels. Further experimental confirmation will shed light on this important aspect of the primary metabolism that may guide nutritional recommendations for individuals with obesity.

EFFECT OF *EGREGIA MENZIESII* ALGAE EXTRACTS ON VIABILITY AND MIGRATION OF RAT INTESTINAL IEC-6 CELL LINE

G. Carrión Hernández^{1,2}, Molina-Jiménez T.³, López-Franco O.⁴, Muñoz-Flores M.⁴, T. Bañuelos-Olivares⁵, Medina-Sánchez A.⁶ and Zepeda R.C.²

¹Programa de Maestría en Ciencias Biológicas, Universidad Veracruzana. ²Centro de Investigaciones Biomédicas, Universidad Veracruzana. ³Facultad de Química Farmacéutica Biológica, Universidad Veracruzana. ⁴Laboratorio de medicina traslacional- Instituto de ciencias de la salud, Universidad Veracruzana. ⁵Investigaciones Oceanológicas, Universidad Autónoma de Baja California. ⁶Instituto de Química aplicada, Unidad Veracruzana.
Email: cgchdez@gmail.com, zS23024960@estudiantes.uv.mx

Colorectal cancer is a chronic, non-communicable disease characterized by the formation of polyps in the inner lining of the colon. In 2022, it ranked third in mortality rate globally and in Mexico, making it a significant health concern. Common treatments for this cancer include surgery, chemotherapy, and radiotherapy. However, these treatments can be invasive, lead to adverse reactions, and may not be specific to the cancer cells. As a result, alternative therapies are being explored, such as natural products containing active biomolecules. Algae, for example, have been found to contain secondary metabolites with various biological activities that can potentially combat cancer selectively because they do not cause cytotoxicity in non-tumor cells. Our study evaluated the effect of hexane (HX), dichloromethane (DCM), and methanol (MeOH) extracts of the brown algae *E. menziesii* on cell viability and migration in non-tumor cells of the intestine. The extracts were tested at different concentrations and time points on the IEC-6 cell line (non-tumor cell from mouse intestine). The results indicated that the Hx extract showed moderate toxicity at 24h, while the DCM extract exhibited moderate toxicity at 24h and 48h. However, both extracts showed no effect at 72h.

On the other hand, the MeOH extract did not demonstrate any cytotoxic effect on the IEC-6 line. The cell migration assay also showed that the extracts did not affect cells at concentrations $\leq 100 \mu\text{g/ml}$. The DCM extract had the most significant impact, followed by Hx and MeOH. Overall, the extracts did not negatively affect the viability and migration of the IEC-6 cell line at concentrations $\leq 100 \mu\text{g/ml}$.

Our results open the possibility of testing the *E. menziesii's* extracts on tumor cells using innovative methodologies like metabolomics and chemometrics to identify seaweed-derived compounds that could potentially reduce cancer progression.

EXPLORING THE CONNECTION BETWEEN VITAMIN B12 DEFICIENCY AND OBESITY

Erika Castaño-Moreno, Daniel Cantú-Ruiz, Víctor Treviño, Rocío Díaz de la Garza*

Tecnológico de Monterrey, Institute for Obesity Research, Ave. Eugenio Garza Sada 2501, Monterrey 64849, NL, México.

*rociodiaz@tec.mx

One-carbon metabolism (1-CM) is an essential metabolic pathway in all cells, with folate and vitamin B12 playing crucial roles. Vitamin B12 deficiency has been observed in individuals with obesity across the life cycle. In Mexico, 70% of the population is affected by overweight and obesity—furthermore, 34% of women in Mexico experience vitamin B12 deficiency. Obesity with dysfunctional adipose tissue is characterized by proinflammatory and oxidative stress cycles. However, the impact of these stages on vitamin metabolism remains unknown. To learn about this possible metabolic connection, we used the Genotype-Tissue Expression (GTEx) database to analyze the expression of 77 1-CM-related genes in 42 human tissues from 948 individuals normalized by TPM. The BMI, continuous and categorical (BMI > 29.99 & < 25) was used as a predictor variable. Linear regression models adjusted by sex and age were performed per gene. Wilcoxon comparisons between sexes within BMI groups were used for visual representation and validation. Differential expressions were estimated as log₂-fold-change (log₂-FC) with a p-value <0.05 for significance. All analyses were performed in the R language. Intracellular vitamin B12 metabolism requires the activity of 10 main genes. The expression of eight of those genes was altered in subcutaneous and visceral adipose tissues (SAT and VAT) and skeletal muscle tissues (SMT) of individuals with a BMI >30 Kg/m² compared to those with a BMI <25. Genes coding for enzymes that use vitamin B12 as a cofactor (*MTR* and *MUT*), the transcobalamin receptor (*CD320*), and those responsible for generating the vitamin B12 active forms (*MMACHC*, *MMADHC*, *MTRR*, *MMAB*, *MMAA*) in VAT and SAT were downregulated with a log₂-FC between -0.1 to -0.3, p<0.05. On the contrary in SMT, the *MMADHC* and *MMAA* were upregulated with a log₂-FC between 0.1 to 0.2, p<0.05. These results suggest that the fatty tissues of subjects with obesity decrease vitamin B12 active form availability. These results prompt us to hypothesize that tissues would have a local vitamin deficiency, thereby impacting the metabolic pathways in which vitamin B12 plays a role, such as methionine synthesis and methylmalonic acid catabolism. These two reactions are crucial for folate metabolism, methyl donor production, and the substrate's availability for the Krebs cycle. We, therefore, postulate that due to the reduced availability of the active forms of vitamin B12, folate metabolism, and energy metabolism are affected by obesity in specific tissues in humans. How this affects 1-CM, global vitamin B12 individual status and metabolic health requires further exploration.

EVALUATION OF THE EFFECT OF THE GGXN POLYMER ON THE BEHAVIOR OF INDUCED HEMIPARKINSONISM IN RAT

Joshua Imanol Cervantes Andraca¹, Ehekatzin García-Valdés², Patricia Vergara-Aragón¹, Verónica Edith Gallegos-Hernández², Vannia Daniela Merced-Canales¹, Rosalba Desiré Romero-Palma¹, Mónica Michele Hernández-Cruz¹, Maximiliano Loredo-Rosales¹, Nazarett Tabita Mendoza-Centeno¹

¹Universidad Nacional Autónoma de México, Facultad de Medicina, Fisiología, México. ²Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada, Legaria, IPN, México.
E-mail: 09ehekatzin94@gmail.com

Introduction. Parkinson's disease (PD) is a neuro-degenerative, progressive and chronic condition. Caused by a destruction of the dopaminergic neurons of the substantia nigra in the pars compacta. Causing motor, cognitive, and behavioral alterations in the induced hemiparkinsonism model.

Objective. To design and characterize a functional food based on amaranth and a derivative of guar gum (GgXN) to establish the effect of its administration on the Forced Swimming and Open Field test which will allow us to evaluate the behavior of rats with induced hemiparkinsonism.

Methodology. In this project, the methodology was divided into two stages:

Stage 1. Elaboration and characterization of the functional food.

Stage 2. A total of 24 male rats of the Wistar strain with an average weight of 350±50 g were taken, randomly divided into 3 groups: Control (SS) n=8, Hemi n=8 and Hemi+functional food (Hemi*FF) n=8.

Results. The behaviour tests were performed (Forced Swim Test and Open Field Test).

Forced Swimming Test. For the Hemi group, a 125% increase in immobility time was found compared to the control group. On the other hand, the Hemi+FF group presents greater mobility (23.68%) compared to the Hemi group.

Open Field Test. The Hemi and Hemi*FF groups presented a decrease in the number of frames traveled of 27.95% and 17.04% respectively, compared to the control group.

Conclusions. The functional food developed for the present study and administered orally for 14 days managed to reverse the depression and anxiety behaviors observed in the induced hemiparkinsonism model.

References.

1. Pedrosa Carrasco, A., Timmermann, L., & Pedrosa, D. (2018). Management of constipation in patients with Parkinson's disease. *Npj Parkinson's Disease*, 4(1). doi:10.1038/s41531-018-0042-8
2. Harrison., & Jameson, L. (2018). *Harrison Principios de medicina interna*. McGraw-Hill.

TRANSCRIPTOMIC ANALYSIS OF BONE NEOFORMATION IN THE DBA/1 MOUSE MODEL OF SPONDYLOARTHRITIS: ROLE OF MULTIPLE OSTEOGENIC MECHANISMS

Eduardo Chaparro-Barrera¹, Susana Aideé González-Chávez¹, Mercedes Bermúdez-Cortés², José Pablo Romero-López³ and César Francisco Pacheco-Tena¹

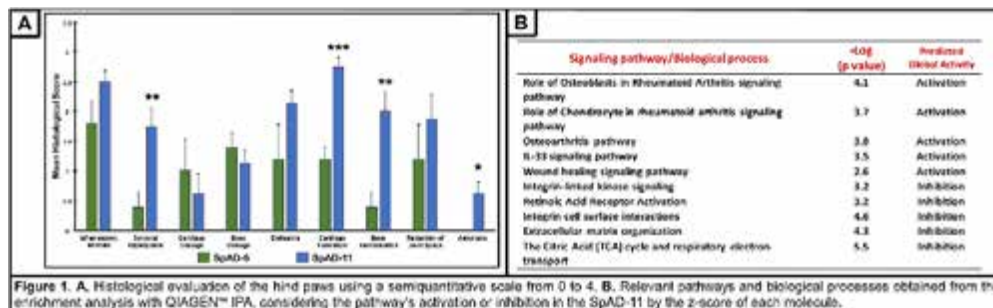
¹Facultad de Medicina y Ciencias Biomédicas, UACH, 31109, Chihuahua, Chih., México., ²Facultad de Odontología, UACH, 31110, Chihuahua, Chih., México., ³Facultad de Estudios Superiores Iztacala, UNAM, 54090 Tlalnepantla, Edo. de Méx., Méx. Dirección Postal: 31136 E-mail: a311388@uach.mx Tel: 614-338-51-85

Introduction. The molecular mechanisms of bone neoformation in spondyloarthritis (SpA) have not been fully elucidated; therefore, current therapy is still insufficient to halt the progression of ossification¹.

Objective. To identify differentially expressed mediators in articular structures with bone neoformation in a murine model of SpA through transcriptomic and histopathologic analysis.

Material and Methods. Two groups of DBA/1 mice with spontaneous arthritis (SpAD) were studied using the model described by Braem et al.,² and were compared at two times: 6 weeks (SpAD-6) and 11 weeks (SpAD-11) after starting the model. H&E staining was employed to compare histological differences between groups. Whole-genome microarrays of the tarsal joint were performed to identify differentially expressed genes (DEGs) in the SpAD-11 vs SpAD-6. Bioinformatic analysis was applied to associate the DEGs to their link with normal and abnormal bone biology. The microarray was validated using qRT-PCR relative quantification by the $\Delta\Delta CT$ method. IHQ analysis was conducted to confirm the differential expression of proteins in the tissue.

Results. Histological analysis showed that SpAD-11 had greater chondral and bone neoformation compared to SpAD-6 (Figure 1A). Whole genome microarrays identified 4,019 GDEs, with 2,348 under-regulated and 1,671 over-regulated. Bioinformatic analysis of the GDEs revealed in the SpAD-11, abnormal musculoskeletal phenotypes, and identified key down-regulated genes: *ank*, *dmp1*, *enpp1*, *sost*, *ebf1*, *dkk1*, and *spp1*. Enrichment analysis predicted relevant signaling pathways and biological processes (Figure 1B). Microarray validation confirmed statistically significant under-regulation of *mepe*, *sost*, and *phex* genes in the SpAD-11. Protein analysis through IHQ showed statistical relevance in the under-regulation of sclerostin and collagen 10 and an up-regulation of wnt-2 in SpAD-11.



Conclusion. The transcriptome analysis of the SpA model in the DBA/1 mouse shows intense ossification in younger age and dysregulation of various signaling pathways, indicating potential therapeutic targets.

References

1. Klavdianou K, Kanellou A, Daoussis D. (2022). *Mediterr J Rheumatol*, 33, 115-125.
2. Braem K, Carter S, Lories RJ (2012). *Biol Proced Online*, 14, 1-5

ONE-CARBON METABOLISM AND FETAL GROWTH IN THE PRINCESA COHORT

Carolina Chetirquen-Silva¹, Gabriela Alemán-Escondrillas², Daniel Eduardo Sandoval-Colin¹,
Marisol Castillo-Castrejón⁴, Marie S. O'Neill³, Felipe Vadillo-Ortega^{1,6}, Erika Chavira-Suárez^{1,6}

¹Instituto Nacional de Medicina Genómica. Periférico Sur 4809, Arenal Tepepan, Tlalpan, 14610 Ciudad de México, México.

²Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán. Vasco de Quiroga 15, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México, México. ⁴Universidad de Oklahoma. Parrington Oval 660, Norman, 73019 Oklahoma, EE. UU. ⁵Universidad de Michigan. S State St 500, Ann Arbor, 48109 Michigan, EE. UU. ⁶Facultad de Medicina UNAM. Escolar 411A, Copilco Universidad, Coyoacán, 04360 Ciudad de México, México.

Teléfono +52 55 5904 2181

Email carochetirquen@gmail.com

Maternal diet is a major source of nutrients that participate in one-carbon metabolism (1CM) such as folate, choline, serine, methionine, vitamin B2, B6, and B12. The synthesis of nucleic acids, proteins, and their regulatory products by gene expression is modulated by 1CM. Thus, 1CM plays an essential role in proliferation and differentiation of organ development. Currently, the global intake of 1CM nutrients during pregnancy and their impact on fetal growth remain unknown. This study aims to examine the correlation between dietary intake of 1CM nutrients and fetal growth during normal pregnancy. We conducted nutritional and clinical data analysis, using five-step multiple-pass 24-hour dietary recalls (24HR) to assess dietary intake and intrauterine ultrasound measurements for intrauterine growth assessment, obtained during prenatal care visits of women included in the Pregnancy Research on Inflammation, Nutrition and City Environments: Systematic Analyses (PRINCESA) Cohort (n=935). Uncomplicated term pregnancies were selected (n=594) to examine the 1CM nutrients trajectories and fetal growth indicators on a monthly basis. We used the lme4 package in Rstudio for statistical analysis. Spearman correlation tests were performed to evaluate associations between 1CM nutrient trajectories and fetal growth indicators. Our results suggest that the progression of pregnancy was associated with an increase in vitamin B2, B6, and B12 intake, which exceeded dietary guidelines. By contrast, folic acid intake decreased ($p < 0.001$), while serine, methionine, choline, and folate intake remained unchanged ($p < 0.05$) throughout gestation. The recommended folate dietary allowance was met through the combined folate and folic acid intake, whereas the adequate choline intake was not met. We observed positive correlations between vitamin B2, B6, and B12 and intrauterine growth ($r = 0.14$, $p < 0.05$), however the correlation between folic acid and intrauterine growth remained negative ($r = -0.14$, $p < 0.05$). Overall, intrauterine growth assessment by ultrasound showed similar patterns as INTERGROWTH-21st standards. In conclusion, maternal intake of B complex vitamins increases, while folic acid decreases in relationship to intrauterine growth. The low-magnitude correlations observed in our analysis may be attributable to the complex metabolic process, the influence of methodology, and possible limitations in the available data. Future studies will be conducted to examine the effects and interactions of 1CM status in the maternal diet on fetal growth.

INTERMITTENT FASTING REDUCES MITOCHONDRIAL FUNCTION IN THE COLON TO REVERT OBESITY-DRIVEN OXIDATIVE STRESS BY MODULATION OF THE GUT MICROBIOTA AND THE METABOLOME PROFILE

Citlally Condado-Huerta^{1,4}, Adriana López-Barradas¹, Alan Hernández-Melgar², Rommel A. Carballo-Castañeda², Raúl De la Torre-Baez³, María Encarnación-García³, Mónica Sánchez-Tapia¹, Omar Granados Portillo¹, Nimbe Torres¹, Armando R. Tovar¹, Porfirio Nava³, Aldo Moreno-Ulloa², Francisco Flores-Murrieta⁴, Laura A. Velázquez-Villegas¹

¹Depto. de Fisiología de la Nutrición, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán, 14080, CDMX.

²Depto. de Innovación Biomédica, Centro de Investigación Científica y de Educación Superior de Ensenada, 22860,

Ensenada, BC. ³Depto. de Fisiología, Biofísica y Neurociencias, Centro de Investigación y de Estudios Avanzados

del Instituto Politécnico Nacional, 07360, CDMX, 4Sección de Estudios de Posgrado, Escuela Superior de Medicina,

Instituto Politécnico Nacional, 11340, CDMX. Vasco de Quiroga 15, Tlalpan 14080 CDMX,

5554-890900 ext. 2802, laura.velazquezv@incmnsz.mx

Oxidative stress in the colon caused by impaired mitochondrial function (MF) during obesity is associated with damage to the epithelial barrier and development of intestinal diseases. Intermittent fasting (IF) improves MF in metabolic tissues and positively modifies gut microbiota composition, suggesting that this intervention may exert a beneficial effect in colon homeostasis. The aim was to evaluate the effect of IF on MF in the colon and its modulation by the gut microbiota in a diet-induced obesity mouse model. C57Bl/6 mice were fed with control of high-fat/high-sucrose diet (HFSD) for 12 weeks, followed by 4 weeks of IF intervention or HFSD, with or without antibiotic treatment. We evaluated MF in colonic mitochondria determined by oxygen consumption rate (OCR), together with metabolome and gut microbiota composition from the feces. Mice who underwent IF intervention had a significant decrease in colonic mitochondria OCR compared to the HFSD group, a parameter that was sharply increased by antibiotic administration. Moreover, changes in gut microbiota composition and metabolome profile caused by IF intervention in obese mice improved colon morphology and epithelial barrier integrity, effects that were worsened after antibiotic treatment. We conclude that IF reduces MF in the colon via changes in bacterial taxonomy and metabolites abundance to decrease oxidative stress caused by obesity, establishing a close connection between host MF and gut microbiota.

FUNGAL METABOLITES AS INHIBITORS OF SHIKIMATE KINASE FROM METHICILLIN-RESISTANT *STAPHYLOCOCCUS AUREUS*

Azhiel Contreras-Chávez, Alfredo Téllez Valencia, Jorge Cisneros Martínez, Claudia Avitia Domínguez

Facultad de Medicina y Nutrición. Universidad Juárez del Estado de Durango. Av. Universidad y Fanny Anitúa s/n.
C.P. 34,000. Durango, Dgo., México.
E-mail: claudia.avitia@ujed.mx

Nowadays, *Staphylococcus aureus* is one of the leading causes of infections worldwide, affecting developing and developed countries. Infections caused by this pathogen consist of skin and soft tissue infections, pneumonia, osteoarticular infections, toxic shock syndrome, and bacteremia. Despite the implementation of active surveillance efforts and prevention programs, *Staphylococcus aureus* continues to be a pathogen with a high mortality rate due to its ability to develop antibiotic resistance, such as methicillin-resistant *Staphylococcus aureus* (MRSA) strains. As a response to this, different research groups are focusing on metabolic pathways present in bacteria and absent in humans, such as the Shikimate Pathway (SP). Therefore, the enzymes within this pathway have been identified as promising targets for the design of new antibiotics. Shikimate Kinase (SK) is the fifth enzyme in SP, it catalyzes the conversion of shikimate to shikimate-3-phosphate. In the present work, a chemical library of fungal metabolites was studied to find inhibitors of SK from MRSA (SaSK). The data showed that fusicin, alternariol, and asperphenamate inhibited SaSK 50, 73, and 55 % when were assessed at a concentration of 100 μ M. Furthermore, molecular docking studies suggested that these compounds interacted with residues from the catalytic and ATP binding sites. Additionally, in silico prediction of their ADMETox properties indicated that these compounds support the structural characteristics to be considered potential drug candidates. In conclusion, the natural products reported here can serve as leads to design more potent SaSK inhibitors and encourage the search for a new drug against MRSA.

HUMAN SPERM DYSFUNCTION IS ASSOCIATED WITH OVERWEIGHT, OBESITY AND OXIDATIVE STRESS

Aída Guadalupe Díaz Martell¹, Ana Alicia Sánchez Tusie²

Advanced Biomedicine Research Center. Autonomus University of Querétaro. Ejido Bolaños SN CP. 76140. Querétaro, Qro.
E-mail: anatusie@gmail.com

Introduction. Overweight and obesity favor the development of male infertility problems. The increase in adipose tissue characteristic of obesity promotes an increased production of reactive oxygen species (ROS) and leads to a state of oxidative stress (OE) that is harmful to sperm cells (1). The effects of OE on spermatozoa are known through data obtained from spermograms, which do not provide information on cell functionality. The acrosomal reaction (AR) test is categorized as a functional test, since it allows us to know the state of the acrosome, a vesicle located in the head of the spermatozoon that contains hydrolytic enzymes that, when released, degrade the outermost layers of the egg, facilitating fertilization (2). AR is characterized by the fusion of the plasma membrane with the acrosomal membrane and is considered a fundamental event for fertilization, since only spermatozoa with AR achieve fertilization (3). Studying the effects of OS on RA allows us to know if it interferes with a fundamental event for male fertility such as RA.

Objective. Determining the effects of OS on functional parameters of spermatozoa from men with normal weight, overweight and obesity.

Materials and methods. A cross-sectional correlation study, with 15 participants, were divided into 3 groups according to BMI: the group with normal weight, overweight and obesity. The ejaculate was obtained by masturbation. The semen sample was analyzed according to World Health Organization criteria. AR was induced in spermatozoa using progesterone as an inducer; ROS in spermatozoa and antioxidant enzyme activity in seminal plasma were evaluated.

Results. The obese group presented higher percentages of spermatozoa with ROS (39 %) compared to the normal weight group (30.5 %) $p = 0.016$. The increase of ROS in spermatozoa correlates with a higher spontaneous AR ($R^2 = .380$, $p = 0.014$). A decrease in the percentages of normal sperm morphology was found with increasing BMI ($R^2 = .504$, $p = 0.001$). **Discussion.** Now it is not possible to attribute the decrease in sperm functionality to a state of EO. **Conclusion.** The data we obtained suggest that ROS increases the spontaneous RA of spermatozoa from obese male.

References

1. Almabhouh, F. A., Md Mokhtar, A. H., Malik, I. A., Aziz, N. A. A., Durairajanayagam, D., & Singh, H. J. (2020). *Andrologia*, 52(1),
2. Oehninger, S., Blackmore, P., Morshedi, M., Sueldo, C., Acosta, A. A., & Alexander, N. J. (1994). *Fertility and Sterility*, 61(2), 349–354.
3. Darszon A, Nishigaki T, Beltrán C, Treviño CL. (2011). *Physiological Reviews*, 91(4):1305-55.

TUMOR-EDUCATED PLATELETS IN HEPATOCELLULAR CARCINOMA: A MULTICENTRIC STUDY

Mariela Judith Domínguez Domínguez¹, Vilma Araceli Maldonado Lagunas², María Teresa Álvarez Bañuelos¹, Clara Luz Sampieri Ramírez¹, Jonathan García-Román³, Hilda Montero L. de Guevara¹, Alma Delia Campos Parra¹, Diana Gabriela Domínguez Lazcano¹, Ángela Rodríguez Padrón⁴, María Cristina Ortiz León¹, José Manuel Hurtado Capetillo⁴, José Luis Morales Velázquez⁵, Nubia Leticia López García⁴, Emmanuel Severino Quintana⁶, Ingrid Simón Lara¹ and Rebeca García Román¹

¹Instituto de Salud Pública, Universidad Veracruzana; ²Laboratorio de Epigenética Instituto Nacional de Medicina Genómica (INMEGEN); ³Facultad de Medicina, Zona Poza-Rica-Tuxpan, Universidad Veracruzana; ⁴Hospital Regional de Alta Especialidad ISSSTE Veracruz; ⁵Centro Estatal de Cancerología "Dr. Miguel Dorantes Mesa", Xalapa, Veracruz; ⁶Clínica-Hospital ISSSTE Xalapa.

Background. Hepatocellular carcinoma (HCC) is the most common primary liver cancer and a major cause of cancer-related deaths worldwide¹. It has been demonstrated that tumor cells are capable of "educating" platelets, when these platelets come into direct contact with the tumor, they sequester tumor RNA, which leads to an altered function. It's because of that it has been proposed that Tumor-Educated Platelets (TEP) can be used in monitoring treatments and as diagnosis in cancer. Likewise, the potential of the platelet-to-lymphocyte ratio (PLR) has been demonstrated for the detection of residual disease and as a prognostic method for cancer, as well as its ability of platelets to influence the process of cell migration²⁻⁴. **Methods.** It was a longitudinal, exploratory, and multicenter study. The participating institutions were: Clínica-Hospital ISSSTE Xalapa, Hospital Regional ISSSTE Veracruz, and Centro Estatal de Cancerología "Dr. Miguel Dorantes Mesa" (CECan). Data collection and the experimental part were conducted over a period of 18 months. An immunofluorescence assay was carried out to observe platelet size and morphology. The PLR calculation was performed to make comparisons between groups, and a wound healing assay was conducted to analyze cell migration using the HEPG2 cell line derived from hepatocellular carcinoma. Descriptive statistics were used to summarize the characteristics of the study population. Kaplan-Meier curves were constructed to estimate the survival probability over time. Data were processed, analyzed, and graphed using the statistical software R Studio version 4.0.2, SPSS version 27, the software ImageJ with its wound healing tool and CellProfiler. **Results.** A total of 95 patient records diagnosed with HCC were analyzed. The average age of the patients was 67.2 years, with females being predominant at 52.6% of the total. Additionally, 32.6% of the patients were in the terminal stage of the disease (BCLC D) at the time of diagnosis, and 51.5% were also diagnosed with some type of diabetes. The platelets from both groups (comparison and control) showed no differences in terms of size and shape. Patients with PLR ≥ 125 had significantly shorter median OS compared to those with PLR < 125 (5.7 months vs. 10.5 months). Moreover, we found that patients in stages B and D had higher PLR and worse survival (mean PLR value of 191.37 and an OS of 8.76 months and a mean PLR value of 208.41 and an OS of 4.8 months respectively) compared to stages A and C patients. Finally, concerning the studies on cell migration, we were able to observe an increase in migration in all tested conditions, especially at the concentration of 1 platelet:1 HepG2 cell. **Conclusions:** This study provides valuable information on the prognosis of hepatocellular carcinoma, as well as on the influence of platelets on cancer progression. It was observed that these platelets can influence the migration of hepatic tumor cells, suggesting a crucial role in cancer progression. These findings highlight the importance of understanding the interaction between tumor cells and platelets in cancer processes. Also, these findings suggest that a higher PLR (PLR ≥ 125 , may be indicative of a worse prognosis in HCC Hispanic (Mexican) patients. Further investigation and validation of PLR as a prognostic marker in larger Hispanic cohorts is warranted.

References

1. Njei B, Rotman Y, Ditch I, Lim JK. Emerging trends in hepatocellular carcinoma incidence and mortality. *Hepatology* [Internet]. 2015 Jan 1 [cited 2023 Dec 12];61(1):191-9. Available from: https://journals.lww.com/hep/fulltext/2015/01000/emerging_trends_in_hepatocellular_carcinoma.26.aspx
2. Trinh H, Dzul SP, Hyder J, Jang H, Kim S, Flowers J, et al. Prognostic value of changes in neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR) and lymphocyte-to-monocyte ratio (LMR) for patients with cervical cancer undergoing definitive chemoradiotherapy (dCRT). *Clinica Chimica Acta*. 2020 Nov 1;510:711-6.
3. Smith RA, Bosonnet L, Raraty M, Sutton R, Neoptolemos JP, Campbell F, et al. Preoperative platelet-lymphocyte ratio is an independent significant prognostic marker in resected pancreatic ductal adenocarcinoma. *The American Journal of Surgery*. 2009 Apr 1;197(4):466-72.
4. Orellana R, Kato S, Erices R, Bravo ML, Gonzalez P, Oliva B, et al. Platelets enhance tissue factor protein and metastasis initiating cell markers, and act as chemoattractants increasing the migration of ovarian cancer cells. *BMC Cancer* [Internet]. 2015 Dec 12 [cited 2024 Jan 14];15(1):1-16. Available from: <https://bmccancer.biomedcentral.com/articles/10.1186/s12885-015-1304-z>

CURCUMINOIDS EFFECTS ON ANTIOXIDANT SYSTEM IN PLASMA AND LIVER OF RATS

Emilio Romero Rangel, María Teresa Espinosa García, Raúl Enríquez Habib¹,
Marco Antonio Juárez Oropeza y Patricia Victoria Torres Durán

Depto. de Bioquímica, Facultad de Medicina UNAM.¹Instituto de Química,
UNAM Avenida Universidad 3000, Coyoacán, Cd. México 04710
pavitodu@yahoo.com.mx

Introduction. Curcumin is a polyphenol found in turmeric and has been reported to have antioxidant, anti-inflammatory, among others. Polyphenols rich diet would be beneficial for the treatment of metabolic diseases, such as, metabolic syndrome, and dyslipidemia diseases. This work summarizes the influence of oral supplementation with curcumin or diacetyl curcumin on antioxidant system in plasma and liver metabolites biochemist profile.

Methods. Twenty-four male Wistar rats (180-200g) were treat 8 weeks with curcumin (C) and diacetyl curcumin (A) suspended in soybean oil, with rodent-appropriate orogastric tube.

The groups were the follow:

- I. Control group, (C)
- II. Control soybean oil (CO) 100 mg/0.5mL soybean oil.
- III. Curcummin M, (M) 50 mg/0.5 mL soybean oil.
- IV. Diacetyl curcumin (DM), 50 mg/0.5 mL soybean oil

At the end of the experimental time, the rats were euthanized, obtaining blood, liver and retroperitoneal fat. In both blood and liver were measured: triacylglycerols, total cholesterol, glucose and thiobarbituric acid reactive substances (TBARS).

Results. The increase in body weight at the end of the experimental time was not different among the groups. The control with oil soy bean didn't differences statistics differences in all the parameters. In plasma, there were changes among the TBARS concentration in groups treated.

In addition, in liver, the results have shown an antioxidant effect in rats treated with curcuminoids with an increase in glutathione.

Acknowledgments. CONACyT, FOINS No. 307152, "Complejos metálicos novedosos de curcumina y curcuminoides en la batalla contra el cancer".

References

1. Róź´an´ ski, G.; Kujawski, S.; Newton, J.L.; Zalewski, P.; Słomko, J. Curcumin and Biochemical Parameters in Metabolic-Associated Fatty Liver Disease (MAFLD)- A review, *Nutrients* 2021, 13, 2654.
2. Lini Alappat and Atif B Awad. Curcumin and obesity: evidence and mechanisms. *Nutrition Reviews* 2010, Vol. 68(12):729-738.
3. Rahmani S, Asgary S, Askari G, Keshvari M, Hatamipour M, Feizi A. and Sahebka A. Treatment of Non-alcoholic Fatty Liver Disease with Curcumin: A Randomized Placebocontrolled Trial. *Phytother. Res.* 2016, 30: 1540-1548.

ANTIOXIDANT CHARGE REVERTS HIGH-DENSITY LIPOPROTEIN (HDL)-INDUCED ENDOTHELIAL DYSFUNCTION IN WOMEN WITH ACUTE CORONARY SYNDROME

Diego Estrada Luna¹, Elizabeth Carreón Torres², Angélica Saraf Jiménez Osorio¹, Gabriel Betanzos Cabrera¹, Natalia Martínez González¹ y Óscar Pérez Méndez²

¹ Universidad Autónoma del Estado de Hidalgo, Circuito Ex Hacienda La Concepción S/N, Carretera Pachuca-Actopan, San Agustín Tlaxiaca 42160, México. ² Instituto Nacional de Cardiología "Ignacio Chávez" Juan Badiano 1, Sección XVI, Tlalpan, Ciudad de México 14080, México.

Telefono: +52 771-705-1399

Correo electrónico: destrada_luna@uaeh.edu.mx

Background. the composition of high-density lipoproteins (HDL) becomes altered during the postprandial state, probably affecting their functionality vis-à-vis the endothelium. Since acute coronary syndrome (ACS) in women is frequently associated with endothelial dysfunction, it is likely that HDL are unable to improve artery vasodilation in these patients. Therefore, we characterized HDL from women with ACS in fasting and postprandial conditions. We also determined whether microencapsulated pomegranate (MiPo) reverts the HDL abnormalities, since previous studies have suggested that this fruit improves HDL functionality. **Methods.** Eleven women with a history of ACS were supplemented daily with 20 g of MiPo, for 30 days. Plasma samples were obtained during fasting and at different times, after a lipid load test to determine the lipid profile and paraoxonase-1 (PON1) activity. HDL were isolated by sequential ultracentrifugation to determine their size distribution and to assess their effect on endothelial function, by using an in vitro model of rat aorta rings. **Results.** MiPo improved the lipid profile and increased PON1 activity, as previously reported, with fresh pomegranate juice. After supplementation with MiPo, the incremental area under the curve of triglycerides decreased to half of the initial values. The HDL distribution shifted from large HDL to intermediate and small-size particles during the postprandial period in the basal conditions, whereas such a shift was no longer observed after MiPo supplementation. Consistently, HDL isolated from postprandial plasma samples hindered the vasodilation of aorta rings, and this endothelial dysfunction was reverted after MiPo consumption. **Conclusions.** MiPo exhibited the same beneficial effects on the lipid profile and PON1 activity as the previously reported fresh pomegranate. In addition, MiPo supplementation reverted the negative effects of HDL on endothelial function generated during the postprandial period in women with ACS.

References

1. Lee WC, Chen JB, Moi SH, Yang CH. Association of proportion of the HDL-cholesterol subclasses HDL-2b and HDL-3 and macrovascular events among patients undergoing hemodialysis. *Sci Rep.* 2021 Jan 21;11(1):1871.
2. Cattaneo, M.G.; Vanetti, C.; Decimo, I.; Di Chio, M.; Martano, G.; Garrone, G.; Bifari, F.; Vicentini, L.M. Sex-specific eNOS activity and function in human endothelial cells. *Sci. Rep.* 2017;7,9612.

RESIDUAL GLYCOSIDASE ACTIVITY DISGUISE THE EFFECT OF COMMERCIAL DIGESTIVE ENZYMES ON IN VITRO DIGESTION OF PHENOLIC GLYCOSIDES

César Femat-Castañeda, José Luis González-Alfonso, Azucena Herrera-González, Francisco J. Plou-Gasca, Cecilia Guerrero-Siancas, Lorena Amaya-Delgado, Javier Placido Arrizon-Gaviño, Marisela González-Ávila*

Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco. A.C. (CIATEJ) Av. Normalistas 800.
Colinas de la Normal. 44270 Guadalajara, Jalisco. México.
E-mail: mgavila@ciatej.mx

Dietary phenolic compounds mitigate oxidative stress in various illnesses, offering therapeutic benefits such as improved insulin sensitivity, reduced hepatic glucose output, inhibition of key carbohydrate digestive enzymes, and modulation of glucose absorption, thereby enhancing post-prandial glycemic control (Sarkar *et. al.*, 2022). However, their hydrophobic nature results in low solubility and stability in aqueous conditions, which limits their bioavailability and biological activity when ingested orally (Nurzyńska-Wierdak, 2023, Andreu *et. al.*, 2023). Glycosylation is proposed as a solution to enhance the stability, solubility, bioavailability, and bioactivity of these compounds, leading to the development of new bioactive molecules with pharmacological potential (Li *et. al.*, 2019, Herrera-González *et. al.*, 2017). The present study investigated the digestibility and bioavailability of different phenolic compounds, including phlorizin, phlorizin monofructoside, phloretin, and phloretin 4'-O- α -D-glucopyranoside, under simulated gastrointestinal conditions without microbiota or food presence. The study aimed to understand how human digestive conditions affect the hydrolysis or degradation of these compounds during stomach and small intestine phases. Results showed that phlorizin and phlorizin monofructoside undergo hydrolysis during the stomach phase, which remains unchanged in the small intestine phase. Phloretin monoglucoside hydrolyzes until the small intestine phase, while phloretin remains stable throughout digestion. These findings indicate the greater stability of β -glycosidic linkages compared to α -glycosidic bonds under gastric-like conditions. Further tests revealed that acidic pH affects only the glycosidic bond of phlorizin monofructoside, while small intestine pH values did not induce hydrolysis in any compounds. The study also evaluated the residual glycosylated activity of gastric and intestinal enzymes, showing significant activity levels: Pepsin from porcine gastric mucosa (190.937 U/g), Lipase from porcine pancreas (14.452 U/g), Pancreatin (52.39 U/g), and Bile salts (Oxgall) (84.05 U/g). These results suggest the probable influence of these digestive components on the hydrolysis of glycosylated phenolic compounds. Overall, the study provides valuable insights into the digestion of glycosylated phenolic compounds and the role of various digestive components in in vitro simulations. These findings could enhance the understanding of the digestibility and bioavailability of these compounds, potentially benefiting human health.

References

- Herrera-González, A., Núñez-López, G., Morel, S., Amaya-Delgado, L., Sandoval, G., Gschaedler, A., Remaud-Simeon, M., & Arrizon, J. (2017). In *Applied Microbiology and Biotechnology* (Vol. 101, Issue 13, pp. 5223–5234).
- Li, B., Chang, S., Jin, D., Zhang, S., Chen, T., Pan, X., Fan, B., Lv, K., & He, X. (2019). *International Journal of Biological Macromolecules*, (Vol. 135, pp 373–378).
- Nurzyńska-Wierdak, R. (2023). In *Molecules* (Vol. 28, Issue 4).
- Sarkar, D., Christopher, A., & Shetty, K. (2022). In *Frontiers in Endocrinology* (Vol. 12).

EVALUATION OF HBB PROTEIN EXPRESSION IN EXTRACELLULAR VESICLES ISOLATED FROM PLASMAS OF WOMEN WITH BI-RADS 4 CLASSIFICATION

Juan Carlos Fernández Hernández¹, Mónica Sierra Martínez², Gustavo Acosta Altamirano³
Pedro Cortés Reynosa⁴, Araceli Posadas Mondragón¹, Eduardo Pérez Salazar⁴

¹Escuela Superior de Medicina, Salvador Díaz Mirón esq. Plan de San Luis S/N, Miguel Hidalgo, Casco de Santo Tomas, 11340 Ciudad de México, CDMX #1, ²Unidad de investigación en salud Hospital Regional de Alta Especialidad de Ixtapaluca del IMSS-Bienestar, Carr. Federal México-Puebla Km. 34.5, 56530 Ixtapaluca, Méx, ³Dirección de Investigación, Hospital General de México, Dr. Balmis 148, Doctores, Cuauhtémoc, 06720 Ciudad de México, CDMX, ⁴Departamento de Biología Celular Cinvestav, Av Instituto Politécnico Nacional 2508, San Pedro Zacatenco, Gustavo A. Madero, 07360 Ciudad de México, CDMX#3.
E-mail: eduardo.perez@cinvestav.mx

Breast cancer is a public health problem of international relevance, is the most common global malignancy and the leading cause of cancer deaths. Extracellular vesicles (EVs) consist of lipid bilayers that encapsulate a complex cargo, including proteins, nucleic acids, and metabolites. EV cargo plays pivotal roles in converting mammary cells to carcinogenic cells and metastatic foci by extensively inducing proliferation, angiogenesis, pre-metastatic niche formation, migration, and chemoresistance. Recent research has observed a significantly higher relative expression of the hemoglobin β chain protein (HBB) in invasive breast cancer tissues with respect to carcinoma *in situ* and a positive classification between HBB and the proliferation marker Ki-67. Therefore, the objective of this research was to evaluate the expression of the HBB protein in extracellular vesicles of patients with breast cancer and in healthy women. 2 mL of blood samples were taken with sodium citrate as an anticoagulant from women in the process of being diagnosed with breast cancer, subsequently serial centrifugations and ultracentrifugation were performed to obtain the enriched fraction of EVs. To characterize the EVs, the nanoparticle tracking assay (NanoSight NS300), transmission electron microscopy, and flow cytometry were performed. The EVs obtained were lysed with RIPA buffer, subsequently western blots were performed to evaluate the expression of the HBB protein in the EVs and flotillin-2 as a marker of EVs.

Likewise, immunohistochemistry and immunofluorescence techniques were performed to identify the expression of the HBB protein in tissue from patients with breast cancer. With the previous methodology 5 micrometer sections were made from biopsies of patients with different stages of breast cancer, subsequently they were incubated with HBB primary antibody and secondary antibodies. We evidenced the presence of the HBB protein in EVs of healthy women and in women diagnosed with breast cancer, as well as a higher relative expression through immunohistochemistry and immunofluorescence with respect to the stage and molecular classification of breast cancer. With the results obtained, we can infer the participation of the HBB protein in EVs in neoplastic processes, however, is necessary do research to clarify its role in neoplastic processes.

ANOSMIA, ODYNOPHAGIA, AND AGEUSIA IN COVID-19 POSITIVE PATIENTS

Karen Elizabeth Franco Reyes¹, Sergio Manuel Salas Pacheco¹, Omar Tremillo Maldonado¹, Leslie Gamboa Guerrero¹, Oscar Almeda Ojeda¹, Gamaliel Ortiz Sarabia¹, Rogelio González González¹, Luis Javier Solís Martínez¹, José Manuel Salas Pacheco², Adda Agustina Sandoval Carrillo²

¹Facultad de Odontología, Universidad Juárez del Estado de Durango; ²Instituto de Investigación Científica. elifrancoory@gmail.com

Introduction. SARS-CoV-2, the virus responsible for COVID-19, emerged in Wuhan, China in 2019 and became a pandemic in March 2020, challenging the healthcare system. The transmission of this virus occurs mainly through saliva droplets expelled while speaking, coughing, or sneezing, causing typical symptoms such as fever, cough, and headaches, as well as atypical symptoms like anosmia, odynophagia, and dysgeusia. Since its appearance, it has been extensively studied due to its infectious profile, rapid progression, and the relationship between comorbidities, lifestyle, age, and gender of the patient with the progression of the disease.

Objective. To determine the relationship between odynophagia, dysgeusia, and anosmia with mortality caused by COVID-19 in patients from the city of Durango.

Materials and Methods. This study was conducted at the State Public Health Laboratory of Durango from December 2020 to December 2021, analyzing data from 16,345 individuals diagnosed with COVID-19. The symptomatology was obtained from the National Epidemiological Surveillance System of the Mexican Ministry of Health.

Results. Male gender (7.5%). The most common comorbidities were hypertension (18.7%) and diabetes (19.5%). The comorbidity with the highest risk of mortality was hypertension.

People aged 60 years or older were at higher risk of dying from COVID-19. The most common occupation was employee.

Discussion and Conclusions. From the results obtained in this study, we observed that for the three symptoms, mortality increased in patients over 60 years old, individuals with one or more comorbidities, and males. Additionally, occupations requiring significant physical effort, such as farming, jobs involving greater contact with people (drivers), retirees, and household workers had a significant incidence in mortality. Further more, our results support other studies showing that being a healthcare worker or having asthma does not present a higher risk as initially believed during the SARS-CoV2 lockdown.

FORMULATION OF A COOKIE WITH THE ADDITION OF FLOUR FROM WINE INDUSTRY RESIDUES

Carlos Emilio Galindo Corona^{1,3}, Nathiely Ramírez Guzmán^{1,3} and Cristian Torres León^{2,3}

¹Faculty of Biological Sciences, Universidad Autónoma de Coahuila, Torreón, ²Research Center and Ethnobiological Garden (CIJE), Universidad Autónoma de Coahuila, ³Agri-food and Agro-industrial Bioeconomy Research Group. Universidad Autónoma de Coahuila, Torreón, México.
corona-carlos@uadec.edu.mx

The main by-product of winemaking is grape pomace, constituting about 25% of the total weight of grapes¹. The main nutrients present in grape residue include proteins, fats, fiber, along with micronutrients, vitamins and phenolic compounds that exhibit antioxidant activity against free radicals². The objective of this project was: to elaborate a cookie with the residues of the wine industry. For the methodological section, the residue from Parras Coahuila was processed. It was dried, milled and passed through a 100 mesh sieve, since this is the particle size of commercial wheat flour. After obtaining the flour, it was subjected to techno-functional tests, which were the following: water and oil retention capacity, swelling, foaming capacity, then it was incorporated replacing 20% of the conventional flour by pomace flour in the production of cookies, which were subjected to functionality tests. The results of these tests show similarities with wheat flour and the cookies are quite similar to the control group, being crunchy and with the ability to crumble in the mouth as well as having a higher amount of fiber and the presence of antioxidant capacity. It is concluded that the flour presents good capacities to be used in the elaboration of cookies besides giving them characteristics of interest and functionality.

References

1. Bordiga, M., Travaglia, F. & Locatelli, M. Valorisation of grape pomace: an approach that is increasingly reaching its maturity – a review. *International Journal of Food Science and Technology* vol. 54 933–942 Preprint at <https://doi.org/10.1111/ijfs.14118> (2019).
2. Taşeri, L. *et al.* Determination of drying kinetics and quality parameters of grape pomace dried with a heat pump dryer. *Food Chem* **260**, 152–159 (2018).

ETIOLOGY, TREATMENT AND SURVIVAL OF PATIENTS WITH HEPATOCELLULAR CARCINOMA IN VERACRUZ, MÉXICO

Mariela Judith Domínguez Domínguez¹, Diana Gabriela Domínguez Lazcano¹, Jaime Morales Romero¹, Verónica Rocío Vásquez Garzón², Omar Elind Arroyo Helguera¹, Alma Delia Campos Parra¹, Bryan Adrián Priego Parra³, Julieta López Vazquez¹, Ingrid Simón Lara¹, Noelia de Yaira López Maya¹, Monserrat Trejo Valencia⁷, Ángela Rodríguez Padrón⁴, José Manuel Hurtado Capetillo⁴, José Luis Morales Velázquez⁵, Jacqueline Ayza Ríos Garces⁵, Nubia Leticia López García⁴, Emmanuel Severino Quintana⁶ and Rebeca García Román

¹Instituto de Salud Pública, Universidad Veracruzana, México ²Laboratorio de fibrosis y cáncer, Universidad Autónoma Benito Juárez de Oaxaca (UABJO), México, ³Centro de Investigaciones Biomédicas, Universidad Veracruzana, México, ⁴Hospital Regional de Alta Especialidad ISSSTE Veracruz; ⁵Centro Estatal de Cancerología "Dr. Miguel Dorantes Mesa", Xalapa, Veracruz; ⁶Clínica-Hospital ISSSTE Xalapa, ⁷Facultad de Biología, Universidad Veracruzana. Av. Luis Castelazo Ayala S/N, Col. Industrial Ánimas, Xalapa, Ver. 2288418900, rebgarcia@uv.mx

Hepatocellular carcinoma (HCC) is the most important liver tumor, accounting for 90% of liver cancer cases. It ranks fourth in cancer mortality and seventh in incidence worldwide; the risk factors are hepatitis B and C viruses, steatotic disease associated with metabolic dysfunction (MASLD), and liver cirrhosis. This study aimed to describe the relationship between etiology and treatment with survival of patients with hepatocellular carcinoma in the state of Veracruz. It was a retrospective, multicenter study at the Centro Estatal de Cancerología (CECan), the Hospital Regional ISSSTE Veracruz, and the Clínica Hospital ISSSTE Xalapa. The data were obtained through the patient's clinical records, the total number of subjects included was 153 people diagnosed with HCC from January 2012 to December 2023. Descriptive statistics and Kaplan-Meier curves were used. Liver cirrhosis was the main etiology of HCC and the most used treatment was sorafenib. 16.1% of subjects with HCC were in the BCLC A stage (best survival), 23.9% in the BCLC B stage, 20% in the BCLC C stage, and 27.1% in the BCLC D stage (highest prevalence). The overall survival time of these patients was 7.5 ± 1.0 months. Patients with MASLD had better survival than other etiological factors, 17.9 ± 5.9 months. Liver resection at 8.4 ± 2.4 months and sorafenib at 8.3 ± 1.3 months were the treatments with the best survival. In this study, a relationship was observed between etiology and patient survival. A statistically significant increase in cirrhosis-related mortality was observed. The type of treatment did not present any significant relationship with death.

THE EFFECT OF OXLDL ON A CELLULAR MODEL OF HER2+ BREAST CANCER

Jeshua Johayra Garcia-Soberanes, Octavio Galindo-Hernández, Nicolás Addiel Serafín Higuera, Víctor Guadalupe García González*

Departamento de Bioquímica, Facultad de Medicina Mexicali, Universidad Autónoma de Baja California. Dr. Humberto Torres Sanginés S/N, Centro Cívico, Mexicali B.C., C.P. 21000; Tel. (686)557-1622 ext.45309; E-mail: vgarcia62@uabc.edu.mx ; jeshua.garcia@uabc.edu.mx

Breast cancer is the neoplasm with the highest incidence in women around the world¹. HER2-positive breast cancer is a subtype of cancer characterized by overexpression or amplification of the HER2 gene, which is involved in regulating cell growth. HER2 overexpression is associated with cancer development aggressively. Although the development of cancer is multifactorial, reports indicate that high concentrations of low-density lipoproteins (LDL) in plasma are related to a greater risk of its development. High concentrations of LDL in the bloodstream make them more susceptible to modifications such as oxidation, due to contact with reactive oxygen species in the endothelial intima. oxLDL promotes a state of chronic inflammation that activates MAPK signaling pathways, which will induce greater cell proliferation and suppression of apoptosis². **Aim.** Elucidate the molecular mechanisms which LDL and oxLDL uses to promote the activation of oncogenes in a cellular model of HER2+ breast cancer. **Materials and methods.** Characterization of LDL oxidation was carried out. By using BT-474 cell line, the effect of LDL and oxLDL in cell viability was evaluated with MTT cell viability assay; protein expression was characterized by Western blot after 24h treatments. The data obtained was analyzed with one-way ANOVA followed by Dunnett's Multiple Comparison test in GraphPad Prism program (8.0.2 version), statistical probability $P < 0.05$ was considered significant. **Results.** oxLDL have a higher expression of HER2, therefore the cell viability and proliferation were increased.

References

1. Globocan. (n/d). <https://gco.iarc.who.int/today/>
2. Zingg, J.M.; Vlad, A.; Ricciarelli, R. Oxidized LDLs as Signaling Molecules. *Antioxidants* 2021, 10, 1184.

EVALUATION OF THE EFFECT OF A GUAR GUM DERIVATIVE IN A MURINE MODEL OF OBESITY

Ehekatzin García-Valdés¹, Verónica Edith Gallegos-Hernández¹, Stephania Galaviz- Hernandez³, Joshua Imanol Cervantes Andraca², Vannia Daniela Merced-Canales², Rosalba Desiré Romero-Palma², Mónica Michele Hernández-Cruz², Maximiliano Loredó-Rosales², Nazarett Tabita Mendoza-Centeno², Frida Azucena Valentín García², Mónica Rosalía Jaime Fonseca¹, Patricia Vergara-Aragón^{2*}

¹Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada, Legaria, IPN, México. ²Universidad Nacional Autónoma de México, Facultad de Medicina, Departamento de Fisiología, México. ³Hospital Infantil de México "Federico Gómez".
Corresponding Autor: Patricia Vergara Aragón. Universidad Nacional Autónoma de México, Facultad de Medicina, Departamento de Fisiología, México. 5522530813. Paty_ver@hotmail.com

Guar gum (GG) is a high molecular weight polymer that has been used for weight control, blood glucose control, and hyperlipidemia treatment. However, due to its high viscosity, it is difficult to add it to foods in order to obtain its physiological benefits, so modifications to the polymer are made to reduce its viscosity and molecular weight. The purpose of this study is to obtain a derivative of guar gum polymer and evaluate its effect on weight, glucose, and lipid profile in a murine model.

Materials and Methods. The GG derivative was obtained by microwave hydrolysis, and the differences between guar gum (GG) and the obtained polymer (GGd) were evaluated by FTIR and bromatological analysis. For the murine model, 18 male Wistar rats were randomly divided into 3 groups: Control, HFD, and HFD+GGd. HFD and HFD+GGd groups were fed a high-fat diet ad libitum for 9 days. After this period, the HFD+GGd group was orally administered the GGd polymer for 30 days. Their weights, food consumption, postprandial glucose, and blood samples for blood chemistry were recorded.

Results No significant differences were found in the FTIR spectrum between GG and GGd. Bromatological analysis showed that GGd has a high dietary fiber content and a significant reduction in viscosity. In murine models, it was found that the administration of GGd caused a decrease in weight (7%), food consumption (18.1%), cholesterol (13.39%), and LDL (14.17%), as well as a lower glucose response compared to the HFD group.

Conclusion. GGd has a high concentration of dietary fiber as well as reduced viscosity and molecular weight after microwave treatment, and its consumption in rats decreases weight, glucose, and lipid profile.

“ALACHES”, QUELITES THAT BESIDES BEING NUTRITIOUS, HAVE EFFECT ON *HELICOBACTER PYLORI*

Erika Gomez-Chang¹, Amanda Gálvez-Mariscal², Irma Romero¹

¹Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México, Cd. Universitaria, C.P. 04510, CDMX, México ²Departamento de Alimentos y Biotecnología, Facultad de Química, Universidad Nacional Autónoma de México, Cd. Universitaria, C.P. 04510, CDMX, México.
E-mail: ergoc@bq.unam.mx

Quelites comprise a great variety of edible plants that grow up around other major crops. *Anoda Cristata* (Alache), belongs to this group of more than 500 species that include leaves, stems, flower buds, guides, shoots and inflorescences of young and tender annual green plants that are traditionally consumed in Mexico since pre-Hispanic times; however, nowadays they are considered neglected and underutilized species. Particularly, Alaches are eaten steamed or cooked like a soup in boiling water. In addition to their great nutritional value, Alaches are traditionally used for the treatment of different ailments such as gastrointestinal diseases.

Approximately 10% of the world's population is affected by peptic ulcers, whose main etiological factors are the chronic consumption of NSAID's and the infection by *H. pylori*. Current treatments, which are directed towards *H. pylori* eradication or to the inhibition of gastric acid secretion, fail in ~20%.

In order to promote the consumption of quelites in the daily diet, by demonstrating and added health-value, the *in vitro* anti-*H. pylori* properties [effect on the bacteria growth and upon two of its colonization factors (adherence to AGS cells and urease activity)] of the Aqueous and Dichloromethane-methanol extracts of Alache and the *in vivo* gastroprotective activity of its mucilage were investigated.

The results showed that the Dichloromethane-methanol extract exert a good inhibitory effect with a minimal inhibitory concentration of 250 µg/ml. Both of the extracts and the mucilage inhibit bacterial adhesion by 30 to 50%. None of the samples affect the urease activity. The mucilage has a gastroprotective effect >50%.

Since the consumption of quelites is relevant as part of the local traditions of only a few ethnic groups, this works provides important information to promote the inclusion of quelites in the population's daily diet, as they may have the potential to prevent or even to control diseases related to *H. pylori* infection.

Acknowledgements. To DGAPA PAPIIT, UNAM IN207622, and CONACyT 214286. Authors thank to QAs M. Martínez, V. Uribe, and A. Miranda.

EFFECT OF TWO O-GLCNACTRANSFERASE INHIBITOR DRUGS ON VPH-POSITIVE CERVICAL CANCER CELLS

Ulises González González¹, Arturo Edgar Zenteno Galindo¹, María Cristina Castañeda Patlán¹, Beatriz Xóchitl Ávila Curiel¹, Saira Karina Ramírez Thomé¹, Risk Díaz Castillejos¹, Carlos Josué Solórzano Mata^{1*}

¹Facultad de Medicina y Cirugía, Centro de Investigación Facultad de Medicina UNAM-UABJO, Universidad Autónoma "Benito Juárez" de Oaxaca, Col. Ex hacienda de Aguilera s/n Carretera a San Felipe del Agua, 68020, Oaxaca, México.

*E-mail: universidad99@cecad.uabjo.mx

Cervical cancer (CaCu) is the fourth most common malignant neoplasm in women worldwide and the second most common in underdeveloped countries; histopathologically, the epidermoid type is the most frequent¹. CaSki cell line belongs to an epidermoid carcinoma VPH-16² and expresses the E6 and E7 oncoproteins^{3,4}. O-GlcNAcylation is a dynamic UDP-GlcNAc mediated post-translational modification derived from the hexosamine biosynthetic pathway. The GlcNAc residue is added to the Ser/Thr of nucleocytoplasmic and mitochondrial proteins^{5,6}. This modification is carried out by two enzymes: O-glycosyl-N-acetylglucosamine transferase (OGT), which adds O-GlcNAc, and β -N-acetylglucosaminidase (OGA), which removes this glycosylation^{7,8}. Increased O-GlcNAcylation is a characteristic feature in cancer cells, regulating metabolic and signaling pathways in the process of carcinogenesis and tumor progression^{9,10}.

In the present work, we evaluated the effect of two drugs that inhibit OGT activity: OSMI-1¹¹ and Alloxan¹², in CaSki cells. Cells were cultured in culture plates and OSMI-1 and Alloxan were added separately at different concentrations. Proliferation was assessed by MTT, their IC₅₀ was calculated and cellular expression of O-GlcNAc and OGT was evaluated.

Our results showed that OSMI-1 decreased CaSki proliferation from 1 μ M with a significance from 5 μ M and an IC₅₀ of 12.11 μ M; in immunocytochemistry, a significant decrease in O-GlcNAc expression was observed from 10 μ M, however, OGT expression was maintained. With Alloxan, cell proliferation was significantly decreased at 10 mM and an IC₅₀ of 5.8 mM was obtained. In immunocytochemistry, a significant decrease of O-GlcNAc was observed from 1 mM to practically nonexistent, but OGT expression remained elevated.

In conclusion, OSMI-1 and Alloxan are effective in decreasing O-GlcNAcylation and decreasing CaSki cell proliferation, but the changes evident at the cellular level in O-GlcNAc ratio are better with Alloxan.

References

1. Bedford S. Cervical cancer: physiology, risk factors, vaccination and treatment. *Br J Nurs*. 2009 Jan 22-Feb 11;18(2):80-4.
2. Wu TC, Hsieh ST, Purow BW, Kurman RJ. Demonstration of human papillomavirus (HPV) genomic amplification and viral-like particles from CaSki cell line in SCID mice. *J Virol Methods*. 1997 May;65(2):287-98.
3. Choo CK, Rorke EA, Eckert RL. Differentiation-independent constitutive expression of the human papillomavirus type 16 E6 and E7 oncogenes in the CaSki cervical tumour cell line. *J Gen Virol*. 1994 May;75 (Pt 5):1139-47.
4. Rasi F, Zarredar H, Amini M, Onsoni H, Dadashzadeh K, Khanmohammadi M, Vahedi L, Mokhtarzadeh A, Baradaran B, Bannazadeh Baghi H. Suppression of E6 Oncogene Induces Apoptosis in CaSki Cervical Cancer Cells. *Asian Pac J Cancer Prev*. 2023 Oct 1;24(10):3389-3395.
5. Hart GW, Slawson C, Ramirez-Correa G, Lagerlof O. Cross talk between O-GlcNAcylation and phosphorylation: roles in signaling, transcription, and chronic disease. *Annu Rev Biochem*. 2011;80:825-58.
6. Yang X, Qian K. Protein O-GlcNAcylation: emerging mechanisms and functions. *Nat Rev Mol Cell Biol*. 2017 Jul;18(7):452-465. doi:10.1038/nrm.2017.22. Epub 2017 May 10.
7. He XF, Hu X, Wen GJ, Wang Z, Lin WJ. O-GlcNAcylation in cancer development and immunotherapy. *Cancer Lett*. 2023 Jul 10;566:216258.
8. Ajit Varki, et. al., *Essentials of Glycobiology*, 4th edition, Cold Spring Harbor (NY): Cold Spring Harbor Laboratory Press; 2022.
9. Tan, E. P., Duncan, F. E. & Slawson, C. The sweet side of the cell cycle. *Biochem. Soc. Trans.* 45, 313-322 (2017).
10. Ferrer CM, Sodi VL, Reginato MJ. O-GlcNAcylation in Cancer Biology: Linking Metabolism and Signaling. *J Mol Biol*. 2016 Aug 14;428(16):3282-3294.
11. He XF, Hu X, Wen GJ, Wang Z, Lin WJ. O-GlcNAcylation in cancer development and immunotherapy. *Cancer Lett*. 2023 Jul 10;566:216258.
12. Liu Y, Ren Y, Cao Y, Huang H, Wu Q, Li W, Wu S, Zhang J. Discovery of a Low Toxicity O-GlcNAc Transferase (OGT) Inhibitor by Structure-based Virtual Screening of Natural Products. *Sci Rep*. 2017 Sep 26;7(1):12334.
- 13.12. Konrad RJ, Zhang F, Hale JE, Knierman MD, Becker GW, Kudlow JE. Alloxan is an inhibitor of the enzyme O-linked N-acetylglucosamine transferase. *Biochem Biophys Res Commun*. 2002 Apr 26;293(1):207-12.

THE MICROBIOTA AND THE PRODUCTION OF SECONDARY AND TERTIARY BILE ACIDS DEPEND ON THE TYPE OF PROTEIN CONSUMED

Omar Granados Portillo¹, Berenice Palacios González², Rocio Guizar Heredia¹,
Adriana López-Barradas¹ and Sandra Tobon Cornejo¹

¹Nutrition Physiology Laboratory, National Institute of Medical Sciences and Nutrition "Salvador Zubirán",

²Scientific Linkage Unit, Faculty of Medicine, UNAM at the National Institute of Genomic Medicine.

omar.granadosp@incmnsz.mx

Obesity, a major public health problem around the world¹ is associated with hypertension, hypercholesterolemia, hypertriglyceridemia and insulin resistance², and lead to type 2 diabetes and cardiovascular disease³. In the last decade, animal and human studies have demonstrated that during obesity, there is a dysbiosis of the gut microbiota, and it has been associated with the appearance of metabolic endotoxemia⁴, glucose intolerance, and insulin resistance⁵. Thus, it could be possible that the metabolic health benefits observed during the consumption of a S diet could be mediated by preventing the gut microbiota dysbiosis, even after the consumption of a high-fat diet although it has been suggested that these effects occur through an increase in bile acid (BA) excretion⁸. This work tries to elucidate whether the beneficial effects described by the consumption of S are due to its isoflavones or to the type of protein itself. For this purpose, mice of the C57BL/6 strain were fed with S or casein (C) in the presence or absence of cholesterol (Ch). The intestinal microbiota of the different experimental groups was characterized. Interestingly, the type of protein determined the biodiversity of microorganisms in the animals (Shanon), where the S and SC groups showed a significant increase in richness and diversity with respect to the groups of animals fed with casein. The pCoA analysis revealed that the intestinal microbiota is differentially modified by the type of protein. The relative abundance in the main phyla of animals fed casein was around 50% Verrucomicrobia, 25% Bacteroidetes and 15% Firmicutes compared to animals fed S where Verrucomicrobia decreased to 15%, Bacteroidetes remained at 25% and Firmicutes increased to 25%, especially in those fed with SC. At the family level, 70% belonged to Akkermansiaceae and Murobaculaceae compared to the animals that consumed S where this 60% belonged to Akkermansiaceae, Murobaculaceae, Ruminococcaceae and Lichnospiraceae. The linear discriminant analysis effect size (LefSe) indicated a clear difference in fecal microbiota between animals fed casein or soy protein. Interestingly, the number of bacterial genera had greater abundance in the groups of animals fed with S compared to those fed with C. The presence of Ch and genistein, independent of the type of protein, led to greater production of primary BAs by the liver. However, animals fed S and Ch had greater production of secondary bile acids. This may be due to the greater bacterial abundance and richness as demonstrated in fecal microbiota analyses.

References

1. Houston DK, Nicklas BJ, Zizza CA (2009). *J Am Diet Assoc* 109:1886–1895
2. Despres JP, Lemieux I (2006). *Nature* 444:881–887
3. Doria A, Patti ME, Kahn CR (2008). *Cell Metab* 8:186–200
4. N. Fuke, N. Nagata, H. Suganuma, T. Ota, *Nutrients* 2019, 11, 2277.
5. P. D. Cani, J. Amar, M. A. Iglesias, M. Poggi, C. Knauf, D. Bastelica, A. M. Neyrinck, F. Fava, K. M. Tuohy, C. Chabo, A. Waget, E. Delmee, B. Cousin, T. Sulpice, B. Chamontin, J. Ferrieres, J.-F. Tanti, G. R. Gibson, L. Casteilla, N. M. Delzenne, M. C. Alessi, R. Burcelin, *Diabetes* 2007, 56, 1761.
6. Torres N, Torre-Villalvazo I & Tovar AR (2006) Regulation of lipid metabolism by soy protein and its implication in diseases mediated by lipid disorders. *J Nutr Biochem* 17, 365 – 373.

NOPAL AND PIRFENIDONE IMPROVE BODY AND LIVER WEIGHTS, AND INSULIN RESISTANCE IN A MOUSE OBESITY MODEL WITH DIETHYLNITROSAMINE

Jorge Gutiérrez-Cuevas, Daniel López-Cifuentes, Ana S. Sandoval-Rodríguez, Juan Armendariz-Borunda

Department of Molecular Biology and Genomics, Institute for Molecular Biology in Medicine and Gene Therapy, University of Guadalajara, CUCS, Guadalajara 44340, Jalisco, Mexico
E-mail: gutierrezcj05@gmail.com

Introduction. Obesity is an epidemic in the world, associated with insulin resistance and liver diseases. Mexico has a high prevalence of obesity in the adult population (36%). Nearly half of the Mexican population has metabolic dysfunction-associated steatotic liver disease (MASLD). Pirfenidone (PFD) has anti-inflammatory and anti-fibrotic effects, and improves the body weight, steatosis and insulin signaling in a mouse model of Metabolic dysfunction-associated steatohepatitis (MASH). Nopal (*Opuntia ficus indica*) increases insulin signaling and decreases hepatic steatosis by increasing fatty acid oxidation and decreasing oxidative stress. The aim of this study was to investigate the effects of freeze dried nopal (NOP) and PFD on anthropometric parameters and insulin sensitivity in high fat diet (HFD)-induced obese male C57BL/6J mice and with diethylnitrosamine (DEN).

Methods. Five-six-week-old mice were in each group (n=5) and were treated with DEN (25 mg/kg) and fed with normal diet (ND), ND plus DEN (ND+DEN), HFD (60 kcal% Fat, D12492), HFD plus DEN (HFD+DEN), HFD plus DEN plus supplements (cellulose, maltodextrin, and casein, HFD+DEN+SUPPL), HFD plus DEN plus NOP (HFD+DEN+NOP), HFD plus DEN plus PFD (HFD-DEN+PFD), and HFD plus DEN plus NOP plus PFD (HFD+DEN+NOP+PFD) for 16 weeks. Freeze dried nopal in fine powder (7%) were mixed with HFD and PFD (300 mg/kg/day) also were mixed with HFD. PFD dosage was adjusted according to body weight and mixed with the diets three times a week. Food intake was measured three times a week, and measurement of body weight each week. Anthropometric and ITT data were analyzed in SPSS.

Results. All HFD mice developed obesity ($P \leq 0.05$). PFD and NOP plus PFD reduced body weight ($P \leq 0.05$). Liver weight and glucose levels were increased in HFD, HFD+DEN, HFD-DEN-SUPPL groups ($P \leq 0.05$), but NOP ($P \leq 0.05$), PFD ($P \leq 0.01$), and NOP plus PFD ($P \leq 0.001$) reduced liver weight; and NOP ($P \leq 0.05$), PFD ($P \leq 0.01$), and NOP plus PFD ($P \leq 0.05$) reduced glucose levels.

Conclusions. We showed that intervention with nopal and pirfenidone improved insulin resistance and decreased body and liver weights in obese mice with diethylnitrosamine.

Funding. This work was supported by CONAHCYT, Mexico, under grant CF-2023-I-473 to JGC.

References

1. Sofía Morán-Ramos, et al. (2012). *J Nutr*, 142(11), 1956-1963.
2. Ana Sandoval-Rodríguez, et al. (2020). *Hepatol Commun*, 4(3), 434-449.
3. Jorge Gutiérrez-Cuevas, et al. (2021). *Cardiovasc Drugs Ther*, 35(5), 927-938.
4. Paulina Vidal-Cevallos, et al. (2022). *Clin Liver Dis (Hoboken)*, 19(2), 68-72.

MITOCHONDRIAL EXPRESSION OF KIR5.1 AND KIR6.2 CHANNELS IN THE PROGRESSION OF TRIPLE-NEGATIVE BREAST CARCINOMA

María del Carmen Gutiérrez Galván, Linley Pammely Prado Celis, Rodrigo Zamora Cárdenas,
Tania Ferrer Villada y Ricardo A. Navarro Polanco

Centro Universitario de Investigaciones Biomédicas, Universidad de Colima
Av 25 de Julio 965 Col. Villas San Sebastián, C.P. 28045 Colima, Colima, México.
312-3161000 ext 47507 E-mail: magdal@ucol.mx

Triple-negative breast cancer (TNBC) is the subtype of breast carcinoma with fewer therapeutic options, high recurrence, and that mostly appears in women under 40 years of age. These characteristics are, in part, responsible for the high lethality of this type of cancer. Therefore, searching for new therapeutic alternatives and early diagnosis are priorities in treating this pathology. Various research in recent years has shown that K⁺ channels play a crucial role in different types of cancer, including breast cancer. These works have shown how alterations in the activity of K⁺ channels can affect the invasion and migration of cancer cells, both in vitro and in vivo. However, the specific types of K⁺ channels, their subcellular location, and their functional role in TNBC remain unknown. Previously, in our laboratory, we found an important expression of two Kir family channels (Kir6.2 and Kir5.1) in TNBC cells. In this work, we compared the expression of these channels subcellular localization, and colocalization in non-cancerous mammary epithelium cells (MCF-10A) and stage VI TNBC (HCC-1428). We used the combination of two techniques, immunocytochemistry, and confocal microscopy, to generate images that were subsequently analyzed with the *ImageJ* software. Our results show that both K⁺ channels increase their expression in cancerous cells compared to non-cancerous cells. Additionally, we established that Kir5.1 has an almost exclusive localization in cellular mitochondria, while Kir6.2 (KATP) is expressed both in mitochondria and in other non-mitochondrial subcellular structures. These results suggest that these channels (Kir6.2 and Kir5.1) have a functional role in this cellular organelle and may be potential molecular targets in this pathology, which will have to be determined in future experiments

KINETIC AND STRUCTURAL CHARACTERIZATION OF SMALL MOLECULES CAPABLE OF INHIBITING SRC HOMOLOGY-2-CONTAINING PROTEIN TYROSINE PHOSPHATASE 2

Brenda Paola Gutiérrez Soto¹, Claudia Isela Avitia Domínguez¹, Adriana Castillo Villanueva², Jesús Antonio Oria Hernández² and Alfredo Téllez Valencia¹

¹Facultad de Medicina y Nutrición, Universidad Juárez del Estado de Durango, Av. Universidad y Fanny Anitua S/N, Durango 34000, ²Instituto Nacional de Pediatría, Insurgentes Sur 3700, Letra C, Av. Insurgentes Sur, Insurgentes Cuicuilco, Coyoacán, 04530, Ciudad de México.
E-mail: atellez@ujed.mx

The SHP2 homology domain-containing protein tyrosine phosphatase 2 (SHP2) is a non-receptor phosphotyrosine phosphatase that participates in several signaling pathways promoting cell survival and proliferation. The overexpression or presence of mutations in SHP2 has been implicated in the development, progression and metastasis of many types of cancer such as juvenile myelomonocytic leukemia, acute myeloid leukemia, colon and lung cancer, among others. Therefore, SHP2 is considered an excellent therapeutic target for the design of a new drug for the treatment of cancer. Therefore, in this work, a series of 108 benzimidazole-derived compounds was studied to determine their SHP2 inhibition capability. The results showed that compounds **CSHG2**, **CSHG3** and **CSHG4** showed the highest inhibition potential with an IC₅₀ value of 30, 10, and 11 μ M, respectively. Molecular docking analysis indicated that these compounds were able to interact with residues important for enzyme catalysis. Finally, in silico prediction of their ADMETox properties suggested that the three inhibitors support the structural characteristics to be considered as potential drug candidates. Therefore, these compounds represent a god hit in the search of new drugs for cancer treatment.

EFFECT OF *IBERVILLEA SONORAE* (WEREKE) ON OXIDATIVE STRESS IN THE KIDNEY DURING DIABETES

Ana Lilia Hernández Alba^{1,2}, Vanessa Wendi Suarez Barrios^{1,2}, Juan Carlos Cabrera Angeles^{1,2}, Estefania Valle Velasquez^{1,2}, Fernando Cortes Camacho², Oscar René Zambrano Vásquez², José Guillermo Gonzaga Sánchez^{1,2}, Laura Gabriela Sánchez Lozada², Juan Gabriel Juárez Rojas², Horacio Osorio Alonso², Abraham Said Arellano Buendia²

¹Sección de Estudios de Posgrado e Investigación, Escuela Superior de Medicina-IPN, 11340 Ciudad de México, CDMX,

²Lab. Fisiopatología Renal, Depto. de Nefrología, Instituto Nacional de Cardiología Ignacio Chávez,

14080 Ciudad de México, CDMX 14080, México.

(52) 5572278477 E-mail: anliliahz@gmail.com

Introduction. Hyperglycemia is a primary factor in the development and progression of diabetic nephropathy (DN), the leading cause of chronic kidney disease. Inflammation, oxidative stress, and metabolic alterations play a prominent role in the pathogenesis and progression of DN. Therefore, inhibiting or blocking these mechanisms is crucial for delaying disease progression. The aim of this study was to evaluate the effect of *Ibervillea sonorae* (*I.s*) on diabetes-induced renal oxidative stress.

Methodology. Male Wistar rats (280-310 g) were allocated to three experimental groups: control (C, n=7), diabetic (D, n=7) [55mg/kg BW streptozotocin, IP] and D + *I.s* (D+*I.s*, n=7) [400mg/Kg BW/day, orally]. After confirming diabetes (blood glucose >200 mg/DL) the three experimental groups were maintained for 1 month with water and food on demand. Blood and urine were collected before the sacrifice, and the kidneys were removed and snap-frozen until further processing.

Results. At the end of the study, group D showed lower body weight (294 ± 14.66 vs 457.1 ± 12.51 gr), hyperglycemia (464 ± 12 vs 108.1 ± 3.7 mg/dL), higher diuresis (79 ± 3 ml/24h) and polyuria compared to group C. Treatment with *I.s* significantly reduced hyperglycemia (374 ± 26.7 mg/dL) and diuresis (55.4 ± 9.2 ml/24h), but these values were higher than those recorded in the group of animals C. In serum creatinine (0.6 ± 0.03 vs 0.31 ± 0.01 mg/dL.), urea was evaluated (11.6 ± 0.9 vs 7.8 ± 0.3 mmol/L); there was an increase in D concentrations compared to C, treatment with *I.s* improved these parameters. In renal cortex, the oxidizing effect (ROS (110268 \pm 15589 vs 55373 \pm 5470 UAF/mg prot), complex I and II, GSH activity) was evaluated, in which group D increased compared to C and treatment attenuated these markers. On the other hand, GPx activity (0.07 ± 0.007 vs 0.09 ± 0.005 U/mg) and SOD expression, Kim-1 at the renal level decreased in group D compared to C, treatment increased the expression of these enzymes. These results were associated with decreased urinary excretion of NAG.

Conclusion. *I.s* delays the progression of diabetic nephropathy through an antioxidant mechanism independent of its effects on blood glucose levels.

TEAR PROTEINS AS POTENTIAL BIOMARKERS IN RETINOBLASTOMA

Jesús Hernández Monge¹, Andrés Rodríguez Rodríguez², Leonardo Lozoya Lozoya¹,
Juan Pedro Luna Arias³, Karla Grisel Calderón González² and Vanesa Olivares Illana²

¹Investigador por México-Laboratorio de Biomarcadores Moleculares, Instituto de Física de la Universidad Autónoma de San Luis Potosí, Av. Chapultepec # 1570, 78295 San Luis Potosí- Mexico. ²Laboratorio de Interacciones Bimoleculares y Cáncer, Instituto de Física de la Universidad Autónoma de San Luis Potosí, Av. Chapultepec # 1570 San Luis Potosí- Mexico, , ³Departamento de Biología Celular-CINVESTAV, Av Instituto Politécnico Nacional 2508, San Pedro Zacatenco, CDMX.
E-mail: jesus.hernandez@conahcyt.mx, phone +525514293615

The cancer of the retina or retinoblastoma is the leading cause of cancer among paediatric ocular malignancies. One case for every 15,000 to 20,000 live births and 9,000 new cases are estimated per year around the world (1). Despite this cancer has been described for ages, the predominant treatment is enucleation that was established since 1800's (2). The reason for this is mainly due to its late diagnosis, therefore children are treated when tumors have reached an advanced stage, overall, in middle and low-income countries (3). Today the only way to approach a diagnosis of retinoblastoma is through clinical signs, such as leukocoria that is a white reflection of the pupil and strabismus or misaligned eyes.

Therefore, it is clear that there is a necessity of a support such as biomarkers for the early diagnosis so the doctors can bring an opportune and proper treatment. In this regard we put hands to work using tears as a non-invasive way to obtain biological material. We recruited a group of healthy children as well as a group of children with retinoblastoma confirmed diagnostic. The tear proteins from both groups were used to compare their proteomes through mass spectrometry. After statistic filters, we propose a list of proteins that exhibit differential expression among controls vs retinoblastoma cases, which hopefully may serve as early diagnostic biomarkers.

References

1. Pandey AN. 2014. Saudi J Ophthalmol 28:310-5
2. Martinez-Sanchez M, Hernandez-Monge J, Rangel M, Olivares-Illana V. 2022. FEBS J 289:4371-82
3. Martinez-Sanchez M, Moctezuma-Davila M, Hernandez-Monge J, Rangel-Charqueno M, Olivares-Illana V. 2020. PLoS One 15:e0234337

EFFECT OF CANCER CELLS-DERIVED CONDITIONED MEDIUM ON CARDIOMYOCYTE ENERGY METABOLISM

^{1,2}Fernando Emiliano Jiménez Mondragón, ¹Álvaro Marín Hernández

¹Departamento de Bioquímica, Instituto Nacional de Cardiología, Juan Badiano #. 1, Colonia Sección XVI, México, 14080, México.

²Programa de Maestría y Doctorado en Ciencias Bioquímicas, Circuito de Posgrado, Ciudad Universitaria, Alcaldía Coyoacán, C.P. 04510, México, CDMX.

E-mail: emicetes@gmail.com

Recent reports reveal a close relationship between the heart damage and cancer, where cancer cells may negatively affect the cardiac function by releasing factors (cytokines and metabolites), as has been observed in experimental murine models¹. Our research group evaluated the effect of AS-30D hepatoma growth in rats on the heart function; after seven days of cell implantation, an adverse effect was observed on the heart function. In this sense, it has also been reported that exposure of skeletal and cardiac muscle cells to conditioned culture medium from cancer cells decreased mitochondrial activity. These results suggest that tumors may induce alterations in the energy metabolism of heart cells². Due to little experimental evidence on this topic, we are interested in analyzing the modifications induced by a conditioned medium from cancer cell culture on protein content, enzyme activity, and fluxes of energy pathways (oxidative phosphorylation (OxPhos) and glycolysis) of cardiomyocytes.

In this study, the experimental models were rat C6 glioma cells and H9c2 rat cardiomyocytes. C6 glioma cells at 100% confluence were incubated in DMEM without fetal bovine serum in normoxia and hypoxia for 24 hours to obtain the conditioned medium (CM). Afterward, H9c2 cells at 80% confluence were incubated with cancer cell CM for 24 hours. H9c2 cells CM was used as control CM. The oxygen consumption associated to ATP synthesis (OxPhos) was measured to determine alterations in the energy metabolism of cardiomyocytes. The Oxphos decreased by 35% in H9c2 cells exposed to CM generated in normoxia and 65% in cells exposed to CM generated in hypoxia, compared to control cells, while in both conditions, the cellular viability was more than 85%. These results suggest that the CM obtained in hypoxia may contain higher levels of factors that can alter the energy metabolism of heart cells.

References

1. Finke, D., Heckmann, M. B., Frey, N., & Lehmann, L. H. (2021). *Frontiers in physiology*, 12, 729713.
2. Wiggs, M. P., Beaudry, A. G., & Law, M. L. (2022). *Cells*, 11(12).

BLACKBERRY JUICE FERMENTED WITH TWO CONSORTIUMS OF LACTIC ACID BACTERIA: PHYSICOCHEMICAL AND ANTIOXIDANT PROPERTIES DURING STORAGE

Liliana Lugo-Zárate^{1,2}, Luis Delgado-Olivares², Nelly del Socorro Cruz-Casino²,
Nayeli Shantal Castrejón-Jiménez³, Diego Estrada-Luna¹ and Angélica Saraí Jiménez Osorio^{*}

¹Área Académica de Enfermería, Instituto de Ciencias de la Salud, Universidad Autónoma del Estado de Hidalgo, Circuito Actopan Tilcuautla s/n. Ex Hacienda La Concepción, Pachuca 42160, Hidalgo, México; lu279379@uaeh.edu.mx (L. L.-Z.); destrada_luna@uaeh.edu.mx (D. E.- L.). ²Área Académica de Nutrición, Instituto de Ciencias de la Salud, Universidad Autónoma del Estado de Hidalgo, o Actopan Tilcuautla s/n. Ex Hacienda La Concepción, Pachuca 42160, Hidalgo, México; ldelgado@uaeh.edu.mx (L. D.-O.); ncruz@uaeh.edu.mx (N. S. C.-C.). ³Área Académica de Medicina Veterinaria y Zootecnia, Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo, Av. Universidad km 1 Ex-hacienda de Aquetzalpa A.P. 32, Tulancingo 43600, México; nayeli_castrejon@uaeh.edu.mx (N. S. C.-J.).

*Correspondencia: angelica_jimenez@uaeh.edu.mx (A. S. J.-O.); Tel.: +55 7717172000 extensión 4323.

Abstract. Fermentation of fruit juices with lactic acid bacteria (LAB) is a sustained way to increase fruit exploitation and storage lifespan. Blackberries are a source of antioxidants with proven health benefits. We investigated the effect of fermenting a blackberry juice (BJ) supplemented with whey and two LAB mixtures on physicochemical and antioxidant characteristics during storage. Two consortiums were inoculated (9 log CFU/mL) in BJ supplemented with whey (WH, 1:1) over 48 hours at 37° C. Consortium 1 (BJWH/C1) includes autochthonous *Levilactobacillus brevis*, *Lactiplantibacillus plantarum*, and *Pediococcus acidilactici*. Consortium 2 (BJWH/C2) includes *Lacticaseibacillus casei* and *Lactobacillus rhamnosus*, previously isolated from agave. The pH, lactic acid production, viscosity, stability, reduced sugars, color, total phenolic content, anthocyanins, and antioxidant capacity were evaluated during fermentation and storage for 28 days. After 16 h, the two consortiums observed an increase in LAB content of 29-38%. Although fluctuations were observed during storage, the minimum LAB count was 9.8 log CFU/mL at 28 days. Lactic acid production increased up to 95% with good storage stability. The BJWH/C2 sample increased total phenolic content during storage. We found that adding whey increases biomass, and its physicochemical properties are preserved during storage.

MATERNAL POLYCYSTIC OVARY SYNDROME CHANGES THE ARCHITECTURE AND CELL FUNCTION OF PANCREATIC ISLETS IN MOUSE DESCENDANTS

Rosa López-Castillero¹, Emilio Espinoza², David Barrera³, Rosario Ortiz¹, Nayeli Torres-Ramírez¹, Paola del Valle¹, Israel Muñoz¹, Erick López¹, Silvia Juárez¹, Luisa Escobar¹ and Olga Echeverría¹

¹Departamento de Biología Celular, Facultad de Ciencias, UNAM. ²Departamento de Bioquímica y Biología Estructural, Instituto de Fisiología Celular, UNAM. ³Departamento de Biología de la Reproducción, INCMNSZ. nayeli.torres@ciencias.unam.mx

Hyperandrogenism is one of the main features in women with polycystic ovary syndrome (PCOS), and it is generally accompanied by insulin resistance and hyperinsulinemia, which can also be present in the descendants. In prenatal models of maternal hyperandrogenism, changes in the weight and size of the pancreas have been reported, as well as an increase in apoptosis. However, the effects of hyperandrogenism are not fully understood. Therefore, this work aims to evaluate insulin production, pancreas morphology and islet architecture in the offspring of a postnatal mouse model of PCOS. Dehydroepiandrosterone (DHEA) was administered to Balb/c mice for 20 consecutive days, subsequently the mice were mated with control males. The adult offspring of the group treated with DHEA (O-DHEA) presented similar insulin concentrations as the control group, however, the insulin tolerance test showed insulin resistance in the O-DHEA group. Morphological analysis of the pancreas showed a higher number of islets in the O-DHEA group, as well as an increase in islet area. Immunolocalization of insulin, glucagon and somatostatin to identify α , β and δ cells, respectively, showed loss of the normal pancreatic architecture, increase in the proportion of glucagon-positive cells. Moreover, polyhormonal cells positive for both insulin and glucagon, which are present during metabolic dysfunction, were observed in animals of O-DHEA group. These results suggest an association between maternal PCOS and changes in the organization and function of the pancreatic islets, thus increasing the risk of metabolic diseases in the offspring.

Reference

1. Sanchez-Garrido MA, Tena-Sempere M. (2020). *Mol Metab*, 35, 100937.

EFFECT OF GLYCINE BETAINE ON P53 AND CAS3 EXPRESSION IN HUMAN COLON ADENOCARCINOMA CELLS HT-29

Lizeth Anahí López Castro¹, Norma Aurora Stephens Camacho², Jesús Alfredo Rosas Rodríguez¹,
Ramona Icedo García and Guadalupe González Ochoa¹

¹ Universidad de Sonora-Campus Navojoa, Blvd. Lázaro Cárdenas No. 100 Col. Francisco Villa, Navojoa, Sonora, CP 85880.

² Universidad Estatal de Sonora, Blvd. Manlio Fabio Beltrones 810, Bugambillas, CP 85875, Navojoa, Sonora.

Cell phone: +1 52 6424259969. E-mail: guadalupe.gonzalezchoa@unison.mx

Cancer is a pathology defined as accelerated cell proliferation, a process that occurs due to genetic alterations that affect normal cell growth, resulting in the generation of tumor cells. Colon or colorectal cancer (CRC) ranks third in oncological deaths in Mexico, with an incidence of 14,900 and 1.2 million cases in Mexico and worldwide, respectively^{1,2,3}. Studies have reported that the frequency of consumption of foods rich in betaine and choline has a beneficial effect against some types of cancer^{4,5}. Nevertheless, the effect of Glycine betaine (GB) on the HT-29 colorectal cancer tumor line is still unknown. For all of the above, our objective is to evaluate the effect of GB on the genetic expression and synthesis of regulatory proteins of cellular processes such as cell proliferation (P53) and apoptosis (CAS3) in HT-29 cancer cells. Methodology: HT-29 cells were treated with GB with the following concentrations: 5, 15.6, 20, 31.2, 62.5 mg/mL, HT-29 cells without GB were used as control. After 24 hours in incubation (37°C, 5% CO₂) cells were lysated. RNA purification was followed by cDNA synthesis and qPCR to detect p53 and CAS3 relative expression using GAPDH as a reference gene. Additionally, cell's supernatants or lysates were used to determine protein levels of P53 and CAS3 by ELISA. The results indicated that in cells treated with GB (62.5 mg/mL) the levels of relative mRNA and protein of p53 and CAS3 were up-regulated in comparison with non-treated cells. GB might have an antiproliferative and apoptotic effect in the human colon adenocarcinoma cell line HT-29.

References

1. OMS. (2022). *Cáncer*. <https://www.who.int/es/news-room/fact-sheets/detail/cancer>
2. Ma, Y., Hu, M., Zhou, L., Ling, S., Li, Y., Kong, B., & Huang, P. (2018). Dietary fiber intake and risks of proximal and distal colon cancers: A meta-analysis. *Medicine (United States)*, 97(36). <https://doi.org/10.1097/MD.00000000000011678>
3. Valdés, A., García-Cañas, V., Simó, C., Cifuentes, A., & Ibáñez, C. (2012). Compuestos de la dieta y su efecto en cáncer de colon : Evaluación Foodómica mediante técnicas de Transcriptómica y Metabolómica . *Cromatografía y Técnicas Afines*, 33(2), 51–66.
4. Carbajo Hurtado, A. (2021). La Betaína: un Osmoprotector natural. 23, 1–19.
5. Lu, M. S., Fang, Y. J., Pan, Z. Z., Zhong, X., Zheng, M. C., Chen, Y. M., & Zhang, C. X. (2015). Choline and betaine intake and colorectal cancer risk in chinese population: A case-control study. *PLoS ONE*, 10(3), 1–14. <https://doi.org/10.1371/journal.pone.0118661>

METABOLIC AND ULTRASTRUCTURAL MODIFICATIONS IN LIVER OF OFFSPRING BORN OF POLYCYSTIC OVARY SYNDROME MOUSE MODEL

Erick López-Cruz¹, Emilio Espinoza², David Barrera³, Rosario Ortiz¹, Rosa López-Castillero¹, Paola del Valle¹, Luisa Escobar¹, Olga Echeverría¹, Nayeli Torres-Ramírez¹

¹Departamento de Biología Celular, Facultad de Ciencias, UNAM. ²Departamento de Bioquímica y Biología Estructural, Instituto de Fisiología Celular, UNAM. ³Departamento de Biología de la Reproducción, INCNSZ. nayeli.torres@ciencias.unam.mx

Polycystic ovary syndrome (PCOS) is characterized by ovulatory dysfunction and hyperandrogenism. The prevalence of metabolic dysfunction-associated fatty liver disease is related with hyperandrogenism in PCOS. Moreover, offspring with maternal hyperandrogenism (MHA) exhibit alterations in hepatic lipid metabolism. Therefore, it is important to know whether MHA increases hepatic lipid accumulation and its impact on hepatocyte metabolism and ultrastructure. The aim of this work was to evaluate the effect of MHA on lipid and glycogen accumulation, lipogenic and gluconeogenic enzyme expression and ultrastructure in the liver of adolescent and young adult offspring in a PCOS mouse model. In this study, 25-day-old female Balb/c mice received dehydroepiandrosterone (DHEA) subcutaneously daily for 20 days, while the control group received only sesame oil. All the animals were mated with healthy males. Livers from pups of MHA (DHEA) and the control group aged 1 and 4 months were processed for optical and electron microscopy, western blot of gluconeogenic and lipogenic enzymes, as well as for the quantification of total lipids, cholesterol and triacylglycerols. Greater accumulation of total lipids, cholesterol and triacylglycerols was found in the MHA group, which correlated with an increase in the number and size of lipid droplets. Furthermore, a rise in fatty acid synthase in 1-month-old pups and PPAR-gamma were observed in adults with MHA. Besides, adults of MHA group showed higher expression of the gluconeogenic enzymes, pyruvate carboxylase and phosphoenolpyruvate carboxykinase, as well as lower glycogen accumulation. In addition, in the hepatocytes of adult mice with MHA, endoplasmic reticulum and mitochondrial swelling were observed. These results suggest that MHA stimulates hepatic lipid synthesis and storage, gluconeogenesis and mitochondrial and endoplasmic reticulum stress.

References

1. Aburto C, Barrera D, Ortiz R, Espinoza E, Parra L, González J, Escobar ML, Vázquez GH, Echeverría O, Torres N. (2022). *Metabolites*, 12(12), 1182.

EVALUATION ELECTROACUPUNCTURE EFFECT IN PATIENTS WITH NEUROPATHY DIABETIC BY GENETIC MARKERS OF CD4 POLARIZATION

Carina López Leyva^{1,2,3}, Dr. Rodrigo Romero Nava², Martha Itzel Ramírez Alvarado^{1,4}, Diana Reyes Martínez^{1,4}, Alejandra Contreras Valladares¹, Flores Tizapa Hazel Rayel^{1,4}, González Torres Luis Fernando^{1,4}, Berenice Mondragón Zúñiga^{1,4}, Marco Antonio Sánchez Villa^{1,4}, Charbel Giuseppe Solano García^{1,4}, Guadalupe Montserrat Leyva Olvera^{1,4}, Rodrigo López Valor^{1,4}, Jorge Enrique Palomino Carrillo^{1,4}, María Fernanda Pérez Hernández^{1,3} and Dr. José de Jesús Peralta Romero^{*,1,2}

¹Unidad de Investigación médica en Bioquímica, UMAE Hospital de Especialidades "Dr. Bernardo Sepúlveda, CMN Siglo XXI, IMSS. ²Sección de Estudios de Posgrado e Investigación, de la Maestría en Ciencias de la Salud, Escuela Superior de Medicina del Instituto Politécnico Nacional. ³Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México. ⁴Escuela Nacional de Medicina y Homeopatía, Instituto Politécnico Nacional.

^{*}Investigador Responsable, Unidad de Investigación médica en Bioquímica, UMAE Hospital de Especialidades "Dr. Bernardo Sepúlveda, CMN Siglo XXI, IMSS. Av. Cuauhtémoc 330, Doctores, Cuauhtémoc, 06720 Ciudad de México, CDMX. Cel: 5532318563, email drjperalta@hotmail.com y drjjperalta@gmail.com

The American Diabetes Association (ADA) mentions four kinds of diabetes, and the diabetes type 2 (T2DM) is the most prevalent, it is characterized by poor insulin secretion by beta pancreatic cells, insulin resistance and in consequence an insufficient answer compensative. To long term, the constant hyperglycaemia can cause chronic complications, the symptomatologic most commonly is the Diabetic Neuropathy (DN). It is estimated 30-50% of diabetics patients irremediably will have this disease, and the Neuropathy diabetic peripheral symmetric (NDPS) is responsible for 25% of traumatic amputations of the country. It includes different factors that impact the nervous system, which has many clinical manifestations. These factors can be genetics, the hyperactivity of polyols pathway, the oxidative stress potential, and the decrease of the Na/K/ATPase, the microvascular damage resulting in ischemia and lastly, but not more important, the inflammation after microglial activation that occurs next to nervous peripheral damage, especially de Schwann in distal level.

Furthermore, the dysfunctional innate immunity is enhanced by NF-KB activation is secondary to the presence of oxygen reactive species, producing more amount of TNF alfa, IL1B, IL6 and adhesion molecules interstitial 1 and selectin E. Also, the lymphocytes T are key cells in the inflammation regulation that are induced by Schwann cells stimulated leading chemokines as CXCL9, CXCL10, CXCL11 and more, these induces cells aggregation of T CD4+, Th1. When the naïve lymphocytes are activated, the T CD4 cells can polarize to Th1, Th2, Th17 and T regulators (Tregs). These have a specifically activity, Th1 and Th17 are proinflammatory, while Th2 and Tregs anti-inflammatory activity. CD4 subpopulations require polarizing cytokines come from antigen presenting cells that will produce transcriptions genes for the expression of other genes including effectors cytokines for executing specifically actions in DN.

The electroacupuncture (EA) consists of the use of electric stimulation connected to acupuncture needles in specific places for pain management, it is better, and more effective than acupuncture. The EA acts in different pathways for eases the pain. Information has been collected that mentions how the EA decrease Th1 and Th17 concentrations and the CD4 cells with anti-inflammatory function increases, achieving an equilibrium in different diseases. NDPS doesn't have a treatment with the intention of improving pathophysiology

mechanisms, these are focussed to ease the pain. That's the reason to search for other alternative that not only relieves pain, also acts as an anti-inflammatory tool, improving the sensibility and motor function.

Hypothesis. EA therapy will change the transcription regulators responsible for CD4 polarization decreasing the proinflammatory and increasing the anti-inflammatory ones.

General objective. Evaluate the electroacupuncture effect in patients with diabetic neuropathy compared to sham electroacupuncture group measuring the expression of transcription factors of T CD4 cells polarization. **Particular.** Measure and compare the proinflammatory and anti-inflammatory transcription factors in patients with diabetic neuropathy in those who received electroacupuncture and sham therapy.

Methodology. It is a double-blind randomized clinical trial, with simple random assignment, longitudinal, experimental, prospective, analytical, and comparative. Protocol registration number: 2020-785-070 and approval CONBIOÉTICA-09-CEI-009-20160601.

Acknowledgment for financial support. Instituto Mexicano del Seguro Social. Temas Prioritarios de Salud, Población vulnerable y Temas emergentes. R-2020-785-070. 2. CONAHCYT "Ciencia de Frontera 2023". CF-2023-G-1314. 3. SECTEI problemas específicos SECTEI/132/2023

ESTROGEN MODULATION OF CAMKII AND CALCIUM HANDLING PROTEINS IN H9C2 HYPERTROPHIED MYOTUBES

Silvia Araceli López Morán¹, Julieta Palomeque¹, Julio Altamirano Barrera¹,
Christian Silva-Platas¹ and Gerardo García Rivas^{1*}

¹Tecnologico de Monterrey, Centro de Investigación Biomédica and The Institute for Obesity Research,
Hospital Zambrano Hellion, TecSalud, 66278 San Pedro Garza García, Mexico. Tel: 81 8888 0000.
E-mail: *gdejesus@tec.mx

Introduction. CaMKII (Calcium (Ca²⁺) calmodulin-dependent protein kinase II) is a key regulator of intracellular Ca²⁺ dynamics. Moreover, it is a recognized signal involved in cardiovascular diseases (CVD) such as pathological cardiac hypertrophy (CH). CH is characterized by structural changes and alterations in Ca²⁺ handling. CVD are less prevalent in young females, and drastically increase at post menopause, suggesting that female sex hormones play an important cardioprotective role. CVD can be studied in H9c2 myoblasts, which are able to differentiate to myotubes, presenting characteristics closer to a cardiac phenotype.

Methodology. We developed a model of hypertrophied H9c2-derived myotubes using angiotensin II (Ang). Cells were pre-treated with 17- β estradiol (E2) and estrogen receptor antagonist ICI-182,780 (ICI). Cell hypertrophy was determined by cell area using calcein AM under confocal microscopy. Changes in expression of the main Ca²⁺ handling proteins were evaluated by real-time PCR and Western Blot. Two-way ANOVA was used, p<0.05 (*) was considered significant difference.

Results. It was observed that Ang II treatment generated an increase in cell area (63.7 \pm 9.6* % vs control), which was prevented with an E2 pre-treatment (14.2 \pm 2.1 % vs control), while ICI prevented the antihypertrophic properties of E2 (48.3 \pm 7.3 % vs control). Ang II significantly increased BNP gene expression with a two-fold change, which was significantly reduced with E2. Moreover, CaMKII phosphorylation at T287 increases in hypertrophied myotubes 1.3-fold change, which is prevented with E2. Furthermore, no difference was observed in SERCA2a or PLB at mRNA between groups.

Conclusions. Pretreatment with E2 in hypertrophied myotubes prevents structural and molecular damage induced by Ang II, mediated by genomic estradiol receptor pathway. E2 modulates CaMKII phosphorylation indicating that CaMKII pathway could mediate Ang II-driven hypertrophy.

ANTI-FIBROGENIC ROLE OF IFC-305 IN FIBROBLASTS FROM PATIENTS WITH IDIOPATHIC PULMONARY FIBROSIS AND ITS EFFECTS ON MITOCHONDRIAL FUNCTION

Erika Rubí Luis García, Victoria Chagoya y Hazas, Carina Becerril Berrocal, Nora Gabriela Velasco Loyden, Enrique Chávez Jiménez, Mariel Maldonado, Andrea Barajas Aguilar, Liliana Montes Aguilar, José Cisneros-Lira

Instituto Nacional de Enfermedades Respiratorias, Ismael Cosío Villegas (INER). Calz. de Tlalpan 4502 Del. Tlalpan Col. Sección XVI 14080 Ciudad de México. Teléfono:5526552175; e-mail: erikarubi.84@gmail.com.

Abstract. Idiopathic pulmonary fibrosis is an age-associated disease characterized by alterations in mitochondrial function and metabolic dysregulation. Although some drugs help to slow the progression of the disease, they have not been able to impact patient survival. The compound IFC-305, an adenosine derivative, has demonstrated a protective effect against liver fibrosis in both *in vivo* and *in vitro* models, which has been associated with a reduction in the expression levels of profibrotic proteins, including collagen 1. Furthermore, it has been reported that the administration of IFC-305 restores mitochondrial dysfunction and cellular redox state in liver cirrhosis. Therefore, the administration of IFC-305, which can decrease the expression of profibrotic proteins and improve mitochondrial oxidative metabolism, could represent a potential therapeutic strategy against pulmonary fibrosis. **Method.** Fibroblasts from patients with IPF obtained from biopsies for diagnostic purposes were used. These were obtained at INER with the approval of the Science and Bioethics Committee (assigned code: B07-24) and with the informed consent of the patients. Commercial lines of normal human lung fibroblasts were used as controls. Fibroblasts from patients with IPF were stimulated with different doses of IFC-305 (1, 2.5, and 5 mM) for 24 hours for subsequent analysis. **Results.** Stimulation of control fibroblasts with TGF- β (5 ng/ml) for 24 hours induced an increase in α -SMA production, which was reduced by the administration of 5 mM IFC-305, determined through immunocytochemical analysis. Administration of IFC-305 (2.5 mM) for 7 days in control fibroblasts stimulated with TGF- β resulted in a decrease in the expression levels of the α -SMA protein. Additionally, it was assessed whether stimulation with IFC-305 (2.5 and 5 mM) induces alterations in the expression levels of the pro-fibrosing proteins Colla1 and α -SMA, using qRT-PCR in control fibroblasts and fibroblasts obtained from patients with IPF. The findings revealed a significant decrease ($p < 0.05$) in the expression levels of Colla1 and α -SMA in fibroblasts extracted from patients with IPF compared to control fibroblasts.

EFFECT OF *CALLISTEMON CITRINUS* PHYTOSOMES ON BRAIN AND GUT MICROBIOTA OF HIGH FAT DIET RAT MODEL

Oliver Rafid Magaña-Rodríguez¹, Patricia Ríos-Chávez², José Armando Hernández Soto²

¹Facultad de Químico Farmacobiología, Universidad Michoacana de San Nicolás de Hidalgo. Tzintzuntzan 173, Matamoros, C.P. 58240, Morelia, Michoacán, México. ²Facultad de Biología, Universidad Michoacana de San Nicolás de Hidalgo. Cd. Universitaria, C.P. 58030 Morelia, Michoacán, México.
oliver.rodriguez@umich.mx

High fat diets (HFD) are commonly consumed around the world, mainly in America. This diet produces an increase in reactive oxygen species therefore, leads to a state of oxidative stress. HFDs promote the change in abundance and richness in the gut microbiota. Currently, several studies confirm the close link between the Central Nervous System (CNS) and the intestinal microbiota called the “gut-brain microbiota axis”; understanding that modifying the microbiota promotes CNS disorders and neurodegenerative diseases¹. The extract of *C. citrinus* has proven to have anticancer, anti-inflammatory, and antioxidant properties²; however, its action in the brain and its behavior at the level of the intestinal microbiota-brain axis is unknown. The objective of this study was to investigate the effectiveness of *Callistemon citrinus* phytosomes over the gut-brain microbiota axis in male Wistar rats fed with a HFD. An obesity model was used through a high-calorie diet. The rats were fed with a diet containing 63% normal chow (Rodent diet® brand rat chow), 18.5% lard, and 18.5% vegetable fat, with 25% fructose. The animals were randomly divided into nine groups (n = 6 per group): Control, High Fat Diet (HFD), Vehicle, HFD plus Orlistat (5mg/kg), *C. citrinus* leaf extract (200mg/kg), HFD plus *C. citrinus* leaf extract (200 mg/kg), and three HFD groups administered with *C. citrinus* leaf phytosomes (50, 100 y 200 mg/kg), all groups had free access to water. The weight of all groups was measured weekly to perform morphometric analyses. At the end of the 16 weeks of the experimental procedure, the whole brain was extracted, as well as samples fecal content, in order to analyze the antioxidant capacity in the brain through super oxide dismutase activity and the glutathione levels. Subsequently, the analysis of the richness of the gut microbiome will be carried out, using molecular methods in order to determine if *C. citrinus* can avoid the dysbiosis caused by HFD.

References

1. Ortega-Pérez, L., Ayala-Ruiz, L., Magaña-Rodríguez, O., Piñón, J., Aguilera-Méndez, A., Godínez-Hernández, D., Ríos-Chávez, P. (2023). *Pharmaceutics*, 15.
2. Wang, B., Kong, Q., Li, X., Zhao, J., Zhang, H., Chen, W., & Wang, G. (2020). *Nutrients*, 12(10), 3197.

IDENTIFICATION OF ENDOMETRIAL STEM CELL MARKERS IN MENSTRUAL BLOOD

Mejia-Carmona G. E.¹, Dickens Terrazas D.², V.J. Carrasco Urrutia³, Robles Escajeda E.⁴,
Alday Montañez F.D.¹, Martínez Martínez A.*¹

¹Universidad Autónoma de Ciudad Juárez, Instituto de Ciencias Biomédicas, Departamento de Ciencias Químico Biológicas, Av. Benjamin Franklin no. 4650 Zona PRONAF CP 32315; ²Quiescente Molecular, C. Rio Amazonas 4080, Los Nogales C.P. 32350; ³Endofem, Av. Campos Eliseos 9371, Campos Eliseos Consultorio 565, C.P. 32472; ⁴ University of Texas at El Paso, 500 W University Ave, El Paso, Tx, C.P. 79968.
alejandro.martinez@uacj.mx

Menstruation is the vaginal bleeding that occurs as part of the menstrual cycle. Each month, a woman's body prepares for possible fertilization; if this does not happen, the uterus sheds its lining, which exits the body through the vagina. Menstrual blood contains various components such as water, dead endometrial cells, lipids, proteins, hormones, and endometrial stem cells (MenSCs). MenSCs are responsible for regenerating the endometrium after each menstruation, possessing proliferative capacity and immunological properties that help the endometrium regain the necessary size and conditions for the implantation of a fertilized egg. The study and therapeutic application of stem cells are growing, but the methods of obtaining them involve some risk. MenSCs offer several advantages over stem cells derived from bone marrow, including easy isolation, abundant proliferation, and no need for invasive collection methods, thus posing no risk. Additionally, these cells have properties and characteristics that make them suitable for future diagnostic or therapeutic applications. Therefore, we have isolated and identified stem cells from menstrual blood, identifying their morphological and molecular characteristics in fresh samples to create a method for identifying these cells, facilitating the diagnosis of uterine diseases. So far, MenSCs have been found to express DJ1, but are negative for CD45, and further analysis of other markers such as Sox2 is expected.

RESVERATROL AND QUERCETIN IMPROVE SARCOPENIC OBESITY BY REGULATING THE ANGIOTENSIN II/AT1 PATHWAY AND MYOSTATIN CONCENTRATIONS IN A METABOLIC SYNDROME RAT MODEL

Jimena Alejandra Méndez Castro¹, Viviana Estefanía García Vázquez¹, Vicente Castrejón Téllez¹, Agustina Cano Martínez¹, Elizabeth Carreón Torres², Leonardo del Valle Mondragón³, María Esther Rubio Ruiz¹

Departments of: ¹Physiology, ²Molecular Biology, ³Pharmacology, Instituto Nacional de Cardiología Ignacio Chávez, Juan Badiano 1, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México, CDMX. Tel. 5555732911 ext. 25400
E-mail: esther_rubio_ruiz@yahoo.com

Introduction. Sarcopenic obesity (SO) is described as 'reduced lean mass disproportionate to adipose tissue'. On the other hand, Metabolic Syndrome (MS) is a clinical condition characterized by the association of diseases such as hypertension, central obesity, insulin resistance and dyslipidemia. There are different pathways involved in SO, among which the Renin-Angiotensin System (RAS) stands out, which has been little explored. Myostatin is a negative regulator of skeletal muscle mass and it is proposed as a target molecule for the treatment of muscle wasting¹. However, it has been shown that a therapy with natural compounds such as Resveratrol (RSV) and Quercetin (QRC) is effective for the treatment of MS², but its effect on the Angiotensin II (Ang II) and its receptor AT1 and their association with SO is unknown. **Objective.** Determine the role of the Ang II/AT1 pathway and the expression of myostatin in a murine model of MS treated with RSV+QRC **Methodology.** 24 male Wistar rats were used and divided into groups. Group I: controls (C), Group II: MS animals, Group III: C-RSV+QRC, and Group IV: MS-RSV+QRC. The C group received water, and the MS group received 30% commercial sugar in the drinking water for 6 months. The C-RSV+QRC and MS-RSV+QRC groups received RSV at a dose of 50 mg/kg/day and QRC at 0.95 mg/kg/day for 1 month. **Results.** The signs of MS in the rats decreased, and the amount of muscle mass increased with the treatment of RSV+QRC and the concentration of Ang II and myostatin decreased in MS rats treated with RSV+QRC **Conclusion.** Treatment with RSV and QRC is effective for treating SO by reducing myostatin levels and regulating Ang II/AT1 pathway.

References

1. Rubio-Ruiz, Guarner-Lans, Pérez-Torres (2019). International Journal of Molecular Sciences, 20, 2-20.
2. Rubio-Ruiz, Guarner-Lans, Cano-Martínez, Díaz-Díaz (2019). Molecules, 24, 2-15.

PARTICIPATION OF CALRETICULIN IN HUMAN FETAL MEMBRANES

Montserrat Mijares Rodríguez, Charlotte Olivares Yañez, Felipe Vadillo-Ortega, Noemí Meraz Cruz

Unidad de Vinculación Científica, Facultad de Medicina, UNAM en el INMEGEN,
Periférico Sur N°4809, Arenal Tepepan, Tlalpan, 14610, CDMX, México.
E-mail: nmc@unam.mx

Calreticulin (CRT) is a 46 kDa calcium-binding chaperone protein, located in different intracellular compartments, as well as the extracellular environment. The presence of this protein in placenta¹, amniotic² and cervical fluid³ has been reported, relating it to the development of complications during pregnancy, including preterm birth², influencing the expression and activity of extracellular matrix metalloproteinases (MMPs)⁴. The objective of this study was to identify the presence of calreticulin in fetal membranes (FM) at term and its relationship with MMPs involved in membrane rupture (MR). For this, human FM were collected from 37 to 40 weeks of gestation with and without labor, extracts and tissue sections were prepared to be analyzed using SDS-PAGE, zymography, western blot and immunohistochemistry to reveal the presence of CRT and MMPs.

Results: The secretion of CRT, MMP-2 and MMP-9 was identified in both membranes by SDS-PAGE, zymography and western blot. The tissue distribution of both metalloproteinases and CRT in these tissues was determined. CRT was immunolocalized in the compact layer of amnion cells with labor, and in chorion cells without labor.

Conclusions: The rupture of fetal membranes is related to the expression and local activity of MMP-2 and MMP-9. This study reveals for the first time that these membranes secrete CRT, in addition to these MMPs. CRT was identified in the amnion of FM with labor. This protein is added to the biochemical process of membrane rupture, because CRT is associated with MMP-9 activity. It is possible that the physiological function of this protein is mainly involved in the process of fetal membranes rupture during childbirth.

References

1. Shi, Z., Hou, W., Hua, X., Zhang, X., et al. (2012). *Cell Biochem. Biophys.*, 63(2), 183–189.
2. Musilova, I., Andrys, C., Drahosova, M., Soucek, O., et al. (2016). *J. Matern.-Fetal Neonatal Med.*, 29(24), 3921–3929.
3. Andrys, C., Musilova, I., Drahosova, M., Soucek, O., et al. (2018). *J. Matern.-Fetal Neonatal Med.*, 31(4), 481–488.
4. Xu, P., Alfaidy, N., & Challis, J. R. (2002). *J Clin Endocrinol Metab.*, 87(3), 1353–1361.

CHARACTERIZATION OF CARDIOVASCULAR RISK PHENOTYPES AND THEIR ASSOCIATION WITH CLINICAL ACTIVITY AND METABOLIC ENDOTOXEMIA IN SYSTEMIC LUPUS ERYTHEMATOSUS PATIENTS

Paulina Esmeralda Mora-García^{1,2}; Bertha Campos-López^{1,2}; Isela Parra-Rojas^{1,3}; Karen Pesqueda-Cendejas^{1,2}; Sonia Luquin²; Margarita Montoya-Buelna^{1,4}; Sergio Cerpa-Cruz⁵ y Ulises De la Cruz-Mosso^{1,2}

¹Red de Inmunonutrición y Genómica Nutricional en Las Enfermedades Autoinmunes, Centro Universitario de Ciencias de La Salud, Universidad de Guadalajara, 44340 Guadalajara, Jalisco, Mexico. ²Instituto de Neurociencias Traslacionales, Departamento de Neurociencias, Centro Universitario de Ciencias de La Salud, Universidad de Guadalajara, 44340 Guadalajara, Jalisco, Mexico. ³Laboratorio de Investigación en Obesidad y Diabetes, Facultad de Ciencias Químico-Biológicas, Universidad Autónoma de Guerrero, 39087 Chilpancingo de los Bravo, Guerrero, Mexico. ⁴Laboratorio de Inmunología, Departamento de Fisiología, Centro Universitario de Ciencias de La Salud, Universidad de Guadalajara, 44340 Guadalajara, Jalisco, Mexico. ⁵Departamento de Reumatología, O.P.D. Hospital Civil de Guadalajara Fray Antonio Alcalde, 44280 Guadalajara, Jalisco, Mexico. E-mail: ulises.mosso@academicos.udg.mx

SLE is the prototypical autoimmune disease, their main cause of death are cardiovascular diseases (CVDs), where CVDs risk factors such as serum and dietary vitamin D status, C-reactive protein, uric acid, dyslipidemia, and metabolic endotoxemia could be related. The aim of this study was to characterize CVDs risk phenotypes and their association with clinical activity and metabolic endotoxemia in SLE patients. A cross-sectional study was carried out in 121 SLE and 189 women control subjects (CS). Lipopolysaccharide (LPS) serum levels were evaluated using a commercial ELISA and CVDs risk phenotypes were determined as low risk if they had <3 factors and high CVDs risk if they had ≥3 CVDs factors. We observed that SLE patients presented a high frequency of the high CVDs phenotype 61% (74) compared to the CS. Subsequently, we observed that SLE patients with a high CVDs phenotype have a higher percentage of renal activity than patients with low-risk phenotype (44 vs. 19%, $p=0.008$), a higher Mex-SLEDAI index score (1 vs. 0, $p=0.003$) where 49% of these patients presented disease activity. According to the biochemical variables, patients with a high CVDs phenotype presented higher levels of total cholesterol (169.27 vs. 152.11 mg/dL, $p=0.03$), triglycerides (118.09 vs. 66.38 mg/dL, $p<0.001$) in comparison with patients with low CVDs phenotype. Also, we observed that patients with a high CVDs phenotype presented lower levels of calcidiol (20.90 vs. 27.27 ng/mL, $p=0.008$) and albumin (3.87 vs. 4.08 g/dL, $p=0.03$) compared to patients with low CVDs phenotype. When evaluating the characteristics of nutritional and consumption status according to the CVDS phenotype in patients with SLE, we observed some significant differences ($p<0.001$). Serum LPS levels were significantly higher levels ($p=0.007$) and in high CVDs risk phenotypes ($p=0.04$). We observed that 41% of SLE patients presented metabolic endotoxemia compared to 25% of CS ($p=0.02$). Furthermore, patients with endotoxemia had a trend of lower serum levels of calcidiol ($p=0.05$). In conclusion, SLE patients had a CVDs risk phenotype, characterized by greater clinical disease activity and metabolic endotoxemia.

EVALUATION OF A POTENTIAL SIGNALING PATHWAY INVOLVED IN THE ANTIHYPERTROPHIC EFFECT OF CANNABIDIOL (CBD) IN CARDIAC MYOBLASTS

Carolina Morales-Ochoa¹, Christian Silva-Platas¹, Omar Lozano García^{1,2}, Eduardo Vázquez-Garza¹, Judith Bernal-Ramírez¹, Karla Carvajal³, Guillermo Torre-Amione^{1,2}, Gerardo García-Rivas^{*1,2}

¹Tecnológico de Monterrey, Escuela de Medicina y Ciencias de la Salud; ²Tecnológico de Monterrey. Institute for Obesity Research, San Pedro Garza García, Mexico; ³Instituto Nacional de Pediatría. Nutrición Experimental. Ciudad de México, Mexico. Tel: 81 8888 0000. *gdejesus@tec.mx

Background. Cardiac hypertrophy is a crucial component in heart failure, hence its wide attention as a molecular target and interest in studying its molecular mechanism. On the other hand, CBD, a phytocannabinoid, has been reported to activate various receptors, including peroxisome proliferator-activated receptors (PPARs), which are also implicated in the development of pathological hypertrophy. In this study, we investigate if CBD anti-hypertrophic activity depends on PPAR γ activation.

Methods. We used rat H9c2 myoblasts to investigate the effects of CBD on cardiomyocyte hypertrophy. To induce hypertrophy, the cells were treated with Angiotensin II (Ang. II), and CBD was administered to the hypertrophied cells. Mitochondrial Ca²⁺ handling was evaluated using Calcium green dye, and mROS were evaluated using mitoSOX. Moreover, to elucidate the signaling pathway involved in the CBD anti-hypertrophic effect, we measured the expression levels of hypertrophy biomarkers, PPAR isoforms and AMPK using qPCR. Also, the phosphorylation of AMPK was evaluated via Western Blot.

Results. Our findings indicate that the administration of CBD to hypertrophied cardiac myoblasts treated with Ang. II leads to a reduction in cell size and other biomarkers associated with hypertrophy and also reduced mROS production. Moreover, hypertrophy on cardiac cells revealed a 3-fold increase in MCU expression. Under this condition, the respiratory control ratio was reduced by 30%, concomitant with a reduction of calcium retention capacity and mitochondrial dysfunction. Notably, the antihypertrophic effect of CBD was abolished in the presence of GW9662, a specific PPAR γ antagonist, confirming that PPAR γ activation is crucial for the effect of CBD. Additionally, a possible interaction between PPAR α and the AMPK signaling pathway was observed, as evidenced by a significant increase in PPAR α expression (2.32-fold) and AMPK phosphorylation (60 \pm 0.%) following CBD treatment.

Conclusion. These findings offer a promising avenue for the use of CBD and modulation of PPAR γ receptors as a therapeutic strategy to prevent or reverse cardiac hypertrophy. The potential involvement of mitochondrial calcium handling in this process further enhances the hope for innovative therapeutic interventions. However, a deeper understanding of the hypertrophic inhibition mechanisms is needed to fully realize these possibilities.

PROTEOMIC ANALYSIS OF TARGETS DEREGULATED BY THE MIRNAS MIR-221, MIR-145 AND LET-7C INVOLVED IN PROSTATE CANCER

Gabriela Carolina Morales Sandoval, Adriana Contreras Paredes, David Guillermo Pérez Ishiwara, María del Consuelo Gómez García, María Olivia Medel Flores

Escuela Nacional de Medicina y Homeopatía-IPN, Av. Guillermo Massieu Helguera 239, La Purísima Ticomán, Gustavo A. Madero, 07320 Ciudad de México, CDMX, 5540888630
E-mail: medelflores@yahoo.com

Prostate cancer (CaP) is the sixth cause of death in the male population worldwide. Mexico occupies the first place¹. Currently, there is no single method to diagnose this pathology, and it is generally diagnosed in advanced stages, where treatment options and the probability of success decrease². There is evidence that inflammation and infection by the Human Papillomavirus high risk (HPV) are involved in CaP³ through the deregulation of different molecules, among which miRNAs stand out⁴.

Methodology. To analyze the targets deregulated by miRNAs miR-221, miR-145, and Let-7c, which are reported to be deregulated in CaP. The targets of miRNAs were analyzed through the databases obtained from miRbase, which contain two different algorithms: Target Scan and miRDB. The database Open Targets was used to determine the deregulated targets in CaP.

Results. The analysis of the targets of our miRNAs of interest was done through validation in the two bases contained in miRbase, and the analysis carried out in Open Targets was done through the experimental and bibliographic validation that the target has in this platform. For the research, clinical evidence, the status of the target in the pathology, and whether they are considered biomarkers were taken into account. Having independently filtered these two databases, we realized the data overlap to observe the association between the targets of the miRNAs and their deregulation in PCa.

Following this, we exhaustively reviewed the literature to identify which of these targets are associated with HPV infection, resulting in a total of 10 protein targets.

References

1. GLOBOCAN, 2020
2. Carlsson, S., & Vickers, A. (2020). *Medical Clinics of North America*, 104(6), 1051–1062.
3. Medel-Flores, et al. (2018). *Genetics and Molecular Biology*, 41(4), 781–789.
4. Khatami et al. (2022) *Microbial Pathogenesis*, 166, 105503–105503

EFFECTIVIDAD DEL ÓXIDO DE ALUMINIO SOBRE LA ADHESIÓN DE BRACKET EN ESMALTE FLUORÓTICO

Anareli Mota Morales¹, Omar Tremillo Maldonado¹, Luis Javier Solís Martínez¹,
Nohé Vargas Chávez¹, Ismael Gonzales Anaya¹

¹Facultad de Odontología, Universidad Juárez del Estado de Durango.
Email: anareli98@hotmail.com

Dental fluorosis (DF) is an endemic pathology whose pathogenesis is associated with alterations in the development of dental enamel caused by chronic exposure to high amounts of fluoride ion during the development of dental organs (DO). Clinically, it manifests as hypocalcified areas that can present a whitish opaque coloration and that resemble horizontal lines or defects that can become pigmented and acquire shades ranging from yellow to dark brown. The tooth surface may also present hypoplastic areas, characterized by loss of adamantine structure, which may compromise large areas of the tooth surface, as well as the esthetics and function of the affected ODs. For its treatment, there are several alternatives, which vary from invasive to conservative, according to the degree of involvement. However, in each of the treatments, the main limitation is the poor adhesion provided by the adhesive systems on fluorosis enamel, since phosphoric acid fails to demineralize the dental fluorapatite, resulting in poor etching patterns on the enamel surface ²; Due to the decreased effectiveness of the bonding protocols, other treatments are affected, such as orthodontic treatments, being necessary to use different enamel conditioning techniques, such as micro abrasion with aluminum oxide and phosphoric acid, which will provide conditions to improve adhesion.

To compare the effectiveness of enamel conditioning techniques, using the conventional method with 37% phosphoric acid and air abrasive with 53 micron aluminum oxide with the aqueous solution mixture (Ethanol 17%) (using "AquaCare" as the main tool).

An experimental, cross-sectional study was carried out in the postgraduate laboratory of the Faculty of Dentistry at the Universidad Juárez del Estado de Durango, from June 2022 to July 2022. The compression force that supported the adhesion of the brackets in 34 dental organs was analyzed with a testing machine.

The compression test results were collected, classified according to their group and recorded. A Student's T test was used, obtaining a significant value of 0.001..

In the present study it was proved that there is a significant difference between the enamel conditioning procedures with fluorosis, favoring the abrasive air technique, since it eliminates unfavorable oxides, contaminants and increases the roughness of the surface, compared to phosphoric acid if placed in a conventional way, it does not provide favorable conditioning patterns for micro mechanical anchorage, causing a decrease in bracket resistance on the enamel surface that is provided by chemical adhesion.

Keywords. Fluorosis dental, adhesión, bracket.

References

1. FeleJ Fejerskov, O., Larsen, M. J., Richards, A., & Baelum, V. (1994). Dental tissue effects of fluoride. *Advances in dental research*, 8(1), 15–31. (1).
2. Opinya, G. N., & Pameijer, C. H. (1986). Tensile bond strength of fluorosed Kenyan teeth using the acid etch technique. *International dental journal*, 36(4), 225–229.

DEVELOPMENT OF A BIOPOLYMER AS A SYSTEM DELIVERY FOR ANTIDIABETIC SECONDARY METABOLITES FROM PURPLE SWEET POTATO

Alethia Muñoz-Ramírez¹, Abraham Heriberto García-Campoy², Brenda Avelina López Muñoz³

¹CONAHCYT-IPICYT/División de Materiales Avanzados, Camino a la presa San José 2055, Lomas 4ta sección, CP 78216, San Luis Potosí, SLP, ²Universidad Autónoma de San Luis Potosí, Facultad de Ciencias, Av. Parque Chapultepec 1570, Privadas del Pedregal, CP 78295, San Luis Potosí, SLP, ³Universidad Autónoma de San Luis Potosí, Facultad de Ciencias Químicas, Manuel Nava 6, CP 78210, San Luis Potosí, SLP.
E-mail: alethia.muniz@ipicyt.edu.mx

Diabetes mellitus is a metabolic disorder, specifically, type 2 is characterized by deficient insulin actions results from an inadequate insulin secretion and diminished tissue responses to this enzyme. In 2021 there were 537 million people worldwide affected by this disease. However, it is anticipated that by 2045 this number will increase by 46%. Traditional medicine is currently searching for therapeutic alternatives to mitigate the side effects caused by some drugs. Additionally, there is a requirement to develop novel materials to optimize their delivery and efficacy. This research has developed a starch biopolymer with citric acid and glycerol as cross-linking agents, to which the extract of *Ipomoea batata* was added, in order to evaluate its hypoglycemic effect.

The *in vivo* model used was the zebrafish, species were separated into 10 fishes per tank, which were induced into diabetic state by immersion in 111 mM glucose solution. Later, the treatment was administered, there was one tank for biopolymer with extract (BP-CM), one for biopolymer (BP) only and another for extract (CM), additionally a pharmacological, diabetic and healthy control. Finally, the sacrificing of the fishes was performed to analysis of blood glucose and triglycerides levels.

The achieved results successfully reduced blood glucose levels, as evidenced by a diabetic control measurement of 91.66 mg/dL, compared to measurements of 37 mg/dL, 52.5 mg/dL, and 35.6 mg/dL for fishes treated with BP-CM, BP, and CM, respectively. Triglycerides were also measured, indicating a significant difference between diabetic control and treatment: 312 mg/dL for diabetic control, and 113, 164.5, and 211 mg/dL for BP-CM, BP, and CM, respectively. The starch biopolymer has been shown to be effective as a delivery system, it has also been shown to have anti-diabetic activity itself, which enhances the effects of the extract, and it is an inexpensive material and easy to produce.

CHARACTERIZATION OF NRF2 ROLE AND MODULATION EFFECT IN A CHEMORESISTANT BREAST CANCER MODEL

Andrea Muñoz Ayala¹, Brenda Chimal-Vega¹, Nicolás Serafín Higuera², Ana Gabriela Leija Montoya¹, Octavio Galindo-Hernández¹, Víctor García González¹

¹Facultad de Medicina Mexicali, Universidad Autónoma de Baja California, Humberto Torres Sanginés S/N, Centro Cívico; C.P. 21000, Mexicali, B.C., México, Tel: +52(686)114-3280, ²Facultad de Odontología Mexicali, Universidad Autónoma de Baja California, Mexicali 21000, México.
E-mail: andrea.munoz.ayala@uabc.edu.mx

Introduction. Breast cancer (BC) is the most common malignancy in women worldwide. Estrogen receptor (ER) positive BC represents about 80 % of cases, tamoxifen is the election neoadjuvant treatment. However, a large percentage of patients develop chemoresistance, leading to relapses. Previously, we generated a tamoxifen's metabolites chemoresistant variant (MCF-7^{VarH}), starting from a RE+ cell line, that acquired a Triple-Negative (TNBC) like phenotype with diminished hormonal receptors and an increased migratory capacity. Our analysis suggest Nuclear Factor Erythroid 2-Related Factor 2 (NRF2) may have a crucial role in those phenotypic changes. Nrf2 is a transcription factor with seven domains (named Neh1- Neh7) that plays an essential role in managing oxidative stress and has been related to chemoresistance and tumoral progresion. Therefore, Nrf2 modulation can contribute to BC patients' treatment, especially in those with chemoresistant tumors. **Objectives.** Characterize the role of Nrf2 in RE- TNBC transition, modulate Nrf2 expression and activity by chemical and biological strategies in MCF-7^{VarH} and evaluate their effects on cancer progression. **Methodology.** We analyzed Nrf2 expression and cellular localization, targets downstream Nrf2 and cancer associated progression processes. Nrf2 modulation was performed through siRNAs design and through the selection of small molecules using molecular docking (MD) tool. **Results.** We determine important Nrf2 implications in cancer progression related processes in MCF-7^{VarH}. We designed, synthetize and transfect two siRNAs against Nrf2 to MCF-7^{VarH}. Preliminary result suggest siRNAs transfection can effectively reduce Nrf2 expression and impact chemoresistance. Through MD we selected 10 compounds that potentially can interact with Neh2 Nrf2 domain, the critical DNA interaction domain. **Conclusion.** Nrf2 has an important role in our BC chemoresistance model phenotypic changes, and its modulation can effectively reduce cancer progression associated processes.

ANTILIPEMIC EFFECT OF THE ETHANOL FRACTION OF JUSTICIA SPICIGERA ON THE LIVER OF RATS FED WITH A HIGH-FAT DIET

Marina Murillo Villicaña¹, Cinthia Itzel Landa Moreno¹, Jenaro Lemus De La Cruz¹,
María de los Ángeles Rangel Rosales¹, Mayra Murillo Villicaña², Claudia Isabel García Berumen¹,
Rafael Salgado-Garciglia¹, Asdrúbal Aguilera Méndez¹, Alfredo Saavedra Molina¹

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Morelia 58030, México.

²Hospital General de Morelia Dr. Miguel Silva, Universidad Nacional Autónoma de México,

0850421k@umich.mx, 1419561g@umich.mx, 1315680c@umich.mx, 1642374d@umich.mx, tezcamay@gmail.com, claudiaberumen07@gmail.com, rafael.salgado@umich.mx, amendez@umich.mx, francisco.saavedra@umich.mx

Hyperlipidemia is characterized by increased levels of total lipids (total cholesterol), such as low-density lipoprotein cholesterol, total lipids, and triacylglycerides, as well as decreased levels of high-density lipoprotein cholesterol [1]. This clinical condition is mainly due to a high-fat diet, representing a risk factor that contributes to the appearance of metabolic diseases, such as fatty liver [2]. Previous studies have demonstrated the beneficial effects of *Justicia spicigera* extracts obtained with high polarity solvents on the lipid profile in rats induced to diabetes mellitus with streptozotocin. Those effects were attributed to the phenolic-type compounds of the plant [3]. According to the above, in our study the polar fraction of the *Justicia spicigera* extract was used to obtain these compounds that were administered to rats fed with a high-fat diet for 240 days. The purpose of evaluating the ability of the extract to avoid liver complications caused by increased lipids in the blood. The rats were administered with the extract during the last 60 days of the diet and the values of body weight, glucose and triacylglycerides were determined at the beginning of the diet and during the treatment. The serum lipid profile was evaluated, as well as the levels of insulin, leptin, adiponectin and the enzyme pancreatic lipase once the 60 days of treatment were completed. Liver lipid deposition was observed by the oily red staining technique and the ability of the extract to reduce lipid peroxidation in liver mitochondria was determined.

Referencias

1. Nagarthna, P. K. M., HarshaVardhini, N., Bashir, B., & Sridhar, K. M. (2020). *J. Adv. Sci. Res.*, 11(01), 1-6.
2. Wali, J. A., Jarzebska, N., Raubenheimer, D., Simpson, S. J., Rodionov, R. N., & O'Sullivan, J. F. (2020). *Nutrients*, 12(5), 1505.
3. Murillo-Villicaña, M., Noriega-Cisneros, R., Peña-Montes, D. J., Huerta-Cervantes, M., Aguilera-Méndez, A., Cortés-Rojo, C., & Saavedra-Molina, A. (2022). *Nutrients*, 14(9), 1946.

EFFECT OF CONSUMING STANDARDIZED MEALS WITH DIFFERENT MACRONUTRIENT PROPORTIONS OF METABOLIC FLEXIBILITY

Charlotte Olivares Yañez¹, Monserrat Mijares Rodríguez¹, Yazmith Briseño²,
Berenice Palacios González², Noemí Meraz Cruz¹

¹Depto. Bioquímica, Fac. Medicina, Universidad Nacional Autónoma de México. Cd. Universitaria, 04510. Coyoacán, CDMX, México. ²Laboratorio de Envejecimiento Saludable del Instituto Nacional de Medicina Genómica (INMEGEN) en el Centro de Investigación sobre Envejecimiento (CIE-CINVESTAV Sede Sur), CDMX, 14330, México.
nmc@unam.mx

Metabolic flexibility is a process in which the body is able to respond or change energy substrates in different physiological states. This flexibility plays an important role in an individual's health because losing it increases the risk of obesity, metabolic syndrome, insulin resistance and type 2 diabetes. Taking into consideration that humans spend most of their waking hours in a postprandial state, the organism's metabolic flexibility response to a standardized food may provide valuable information about the individual's current metabolic health. **Objective.** To characterize metabolic flexibility in young adults after the consumption of metabolic challenges containing different proportions of macronutrients. **Methodology.** Clinical and dietary information was collected from 70 individuals (47 women and 23 men), including anthropometric analysis, bioimpedance study, biochemical parameters and indirect calorimetry. Blood samples were taken by capillary puncture during fasting and after consuming standardized meals. **Results.** All the participants exhibited inadequate habits, with similar consumption of fruits, salads, vegetables and practice of physical activity. Insulin response was not significantly affected by differences in the body fat percentage, but the glucose response showed an increment of 0.29%. Overweight and obese individuals had higher total cholesterol values compared to those with normal weight. The fasting respiratory quotient showed values RER= 0.69-0.8. In the postprandial state, the values were RQ=0.81-0.89. In males, values higher than 1 were found. These results varied in relation to the consumed challenge. **Discussion.** In general, our study showed a significant increase in biochemical and anthropometric parameters with age. Personalized analysis was required for the data obtained by calorimetry, as age, weight, and constitution showed variability in the results. This makes the study interesting because we can find overweight or obese individuals who are metabolically healthy, from whom there is currently no consensus or single criterion to define them.

RESISTINA INDUCE MIGRACIÓN E INVASIÓN EN CÉLULAS DE CÁNCER PROSTÁTICO PC3: ROL DE LAS VESÍCULAS EXTRACELULARES

Mario Israel Oregel-Cortez^{1,2,3}, Héctor Frayde-Gómez^{1,2,4}, Tatiana Romero-García³
and Octavio Galindo-Hernández^{1,2*}

¹Departamento de Bioquímica, Facultad de Medicina, Universidad Autónoma de Baja California, Mexicali 21100, Baja California, México; mario.israel.oregel.cortez@uabc.edu.mx (M.I.O.-C.); fraydeh@uabc.edu.mx (H.F.-G.); tatiana.romero@uabc.edu.mx (T.R.-G.). ²Laboratorio Multidisciplinario de Estudios Metabólicos y Cáncer, Universidad Autónoma de Baja California, Mexicali 21100, Baja California, México. ³Facultad de Deportes, Universidad Autónoma de Baja California, Mexicali 21289, Baja California, México. ⁴Hospital Regional de Especialidad No. 30, Instituto Mexicano del Seguro Social, Mexicali 21100, Baja California, México.

*Correspondencia: octavio.galindo@uabc.edu.mx

La literatura reporta que niveles fisiológicos de resistina incrementan la agresividad tumoral en el cáncer prostático. Por otro lado, describe cómo las células son capaces de secretar vesículas extracelulares (VEs) con diversas moléculas cargo. Incluso, las VEs secretadas por células tumorales están relacionadas con un incremento en la progresión tumoral. Células de la línea PC3 fueron estimuladas con diferentes concentraciones de resistina para evaluar los procesos involucrados en la invasión celular, esto mediante ensayos de cierre de herida y zimografía respectivamente. La fracción enriquecida de VEs fue obtenida mediante ultracentrifugación diferencial de los sobrenadantes, con la finalidad de ser utilizada como estímulo en los cultivos celulares prostáticos. El proceso de invasión celular fue analizado mediante cámaras de Boyden recubiertas con matrigel. Nuestros resultados demuestran que la resistina induce un incremento en los procesos de invasión celular. Adicionalmente, las células que reciben el tratamiento con VEs obtenidas de medios condicionados que han sido tratadas con resistina, incrementan su agresividad. En resumen, nuestros hallazgos muestran que la resistina y las VEs secretadas por células estimuladas con resistina, incrementan la progresión tumoral prostática in vitro a concentraciones fisiológicamente alcanzables jugando un papel importante e innovador en la progresión tumoral.

Palabras claves: resistina; cáncer prostático; vesículas extracelulares; invasión.

EFFECT OF DIAZOXIDE AND MODERATE-INTENSITY EXERCISE ON THE FUNCTION OF MITOCHONDRIA ISOLATED FROM SKELETAL MUSCLE DURING OBESITY

Jessica Ortega Pérez¹, Koré Montserrat Moreno Calderón¹, Elizabeth Sánchez Duarte², Alfredo Saavedra Molina¹, Christian Cortés-Rojo¹, Alain R. Rodríguez Orozco³, Rocío Montoya Pérez¹

¹Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo. ²Departamento de Ciencias Aplicadas al Trabajo, Universidad de Guanajuato Campus León. ³Facultad de Ciencias Médicas y Biológicas "Dr. Ignacio Chávez", Universidad Michoacana de San Nicolás de Hidalgo.

E-mail: 1577840d@umich.mx

Obesity is a chronic disease characterized by an excessive accumulation of adipose tissue that harms health and is associated with an imbalance between energy intake and expenditure². Various factors trigger this disease, but the main ones are usually poor nutrition and a sedentary environment². It is known that during obesity, the skeletal muscle is structurally and metabolically affected, due to a decrease in its contraction force, capacity to support fatigue, and cellular damage, which are associated with inadequate mitochondrial function^{5, 4, 3}.

Therefore, alternative treatments with pharmacological and non-pharmacological importance, such as diazoxide and exercise, have been reported to have positive effects on mitochondrial functionality by improving antioxidant defense and metabolic and oxidative stress. However, the interaction between both treatments is still unknown^{1, 4, 6}.

Hence, in the present work, the respiration and swelling of mitochondria isolated from the skeletal muscle of male Wistar rats fed with a high-calorie diet (8 weeks), administered retroperitoneally with diazoxide (14 days), and exercised at a moderate intensity (8 weeks) were evaluated. The experimental groups were classified into 8 (n=6): Control, High-fat diet (HFD), Exercised (EJER), Diazoxide (DZX), HFD + EJER, HFD + DZX, DZX + EJER and HFD + EJER+ DZX. Finally, the data obtained reflect a functional decline in mitochondria during obesity and an improvement from exercise. Likewise, it is shown that the interaction between exercise and diazoxide increases mitochondrial function despite being fed a high-calorie diet.

References

1. Cade, W. T. (2018). Current opinion in clinical nutrition and metabolic care, 21(4), 267.
2. Di-Meo, S., Iossa, S., and Venditti, P. (2017). Journal of Endocrinology, 233(1), R15-R42.
3. Gómez-Barroso, M., Moreno-Calderón, K. M., Sánchez-Duarte, E., Cortés-Rojo, C., Saavedra-Molina, A., Rodríguez-Orozco, A. R., y Montoya-Pérez, R. (2020). Antioxidants, 9(12), 1232.
4. Heinonen, S., Jokinen, R., Rissanen, A., and Pietiläinen, K. H. (2020). Obesity Reviews, 21(2), e12958.
5. Heo, J. W., Yoo, S. Z., No, M. H., Park, D. H., Kang, J. H., Kim, T. W., ... and Kwak, H. B. (2020). International journal of environmental research and public health, 15(10), 2301.
6. Leduc-Gaudet, J. P., Hussain, S. N., Barreiro, E., and Gouspillou, G. (2021). International Journal of Molecular Sciences, 22(15), 8179.

ANTIOXIDANT CAPACITY OF PIGMENTED MAIZE FROM MEXICO

Eiko Osawa-Martínez^a, Yury Rodríguez-Yañez^b, Benito Minjarez^b, Salvador Mena-Munguía^b, Irais Poblete Naredo^a, Adolfo Sierra-Santoyo^a, Arnulfo Albores Medina^a

^aDepartamento de Toxicología, CINVESTAV Av. IPN 2508, Alc. G.A. Madero 07360, San Pedro Zacatenco, Cd. México.

^bUniversidad de Guadalajara, Centro Universitario de Ciencias Biológicas y Agropecuarias, Camino Ramón Padilla Sánchez No. 2100, Nextipac, Zapopan, Jalisco, México.

Corresponding autor: aalbores@cinvestav.mx

Maize is a worldwide consumed cereal. In Mexico, maize grains display a colors variety, ranging from purple to yellow depending on their anthocyanins content, like cyaninide-3-O-glucoside (C3OG), an abundant antioxidant in plants, useful in pathologies like high blood pressure, hyperglycemia and hyperlipidemia. The study aim was to quantify the anthocyanin content in colored grains and cobs, assessing their antioxidant capacity. Materials and methods: anthocyanins were identified and quantitated by HPLC in purple, blue and yellow grains and cobs, and their antioxidant capacity assessed by cellular viability of HaCaT, HepG2, HeLa and A549 cell challenged with hydrogen peroxide. The total antioxidant capacity (TAC) calibrated with Trolox for purple maize and purple maize cob extract and tested with DCFH-DA antioxidant assay in the grain and purple maize cob extract. Results: Purple and blue maize extracts showed the highest content of C3OG as compared to those observed in white and yellow. The MTT viability assays the purple grain extract in HaCat, HeLa, HepG2, and A549 cell cultures were tested with C3OG 500 μ M. A549 and HaCaT cells showed greatest proliferation. HaCaT cell showed the highest antioxidant capacity when were added with C3OG (500 μ M). The purple grain extract showed the highest TAC, 10 min the extract addition, then TAC rapidly decreased. For the DCFH-DC the highest concentration of anthocyanins in the purple grain extract showed the lowest fluorescence so there are few reactive oxygen species. Thus, maize grains and colored cobs are a good source of antioxidants, and this property is related to the color intensity. The anthocyanins obtained from colored maize grains and cobs could be useful food supplement.

GENETIC DETECTION OF DENGUE VIRUS SEROTYPES USING THE CRISPR-CAS12A SYSTEM

Marco Antonio Piñón-Chávez¹ and Armando Hernández-García*

¹Laboratory of Biomolecular Engineering and Bionanotechnology, Institute of Chemistry, Universidad Nacional Autónoma de México. Circuito exterior N/A No. Circuito de la, Investigación Científica, C.U., 04510, Mexico City, Mexico.

*E-mail: armandohg@iquimica.unam.mx

Introduction. According to WHO, Dengue is an endemic disease present in 80 countries, whose maximum peak was reached in last 2023, reporting around 6.5 million of cases.¹ Dengue is caused by four variants (serotypes) of the virus (DENV1-4) and is characterized by a biphasic febrile episode.² Timely detection is crucial for optimal patient treatment and providing statistical data on disease's distribution. Therefore, development of highly specific, sensitive, and accessible biosensors is needed. The CRISPR-Cas12a system has been proposed as an alternative model for genetic detection, especially when is coupled with a pre-isothermal amplification system³. Here, we aimed to develop a novel method based on CRISPR-Cas12a and LAMP for detecting the Dengue virus variants with high specificity and sensitivity.

Methodology. The 3' UTR region of the Dengue virus will be detected using a two-step method:

Loop-mediated isothermal amplification (LAMP), which enables the generation of multiple copies of the target region.

Detection using the ribonucleoprotein AsCas12a, which recognizes a 20 nt sequence and exhibits *trans* collateral cleavage activity of reporter DNA molecules.

Results. The 3' UTR region of DENV contains a higher number of conserved sites within the genome of each virus variant, leading to the design of LAMP primers for its simultaneous amplification. These primers have been evaluated for all DENV variants (1-4), yielding positive results within 1 h of amplification. Additionally, gRNAs have been designed for their individual detection, resulting in effective discrimination between variants.

Conclusions. CRISPR-Cas12a coupled to LAMP enabled simultaneous amplification and detection of specific sequences associated to DENV variants, which were detected fluorescently using a plate reader.

References

1. World Health Organization (2023) Dengue and severe dengue. Available in: <https://www.who.int/news-room/fact-sheets/detail/dengue-and-severe-dengue> (Apr 23, 2024).
2. Bhatt, P.; Sabeena, S. P.; Varma, M.; Arunkumar, G. (2021) *Curr. Microbiol.* 78(1), 17–32.
3. Hernandez-Garcia, A.; Morales-Moreno, M. D.; Valdés-Galindo, E. G.; Quezada, A.; Jimenez-Nieto, E. P. (2022) *Diagnostics*, 12(6).

HYPERGLYCEMIA AND OXIDANT STRESS IN AN ADULT POPULATION FROM MEXICO CITY

Irais Poblete Naredo, Audrey Bravo Prado, Mario I. Gutiérrez Velasco, Rogelio O. Corona Núñez, Diego Delfín López, Irina del Rosario López Gámez and Arnulfo Albores Medina

Cinvestav-IPN. Department of Toxicology, Av. Instituto Politécnico Nacional #2508 Col. San Pedro Zacatenco C.P. 07360. Gustavo A. Madero, Ciudad de México.
5557473308, aalbores@cinvestav.mx.

Introduction. Type 2 diabetes mellitus (T2DM) is one of the main diseases that cause disability and death worldwide, and in Mexico, it represents the second cause of death. Hyperglycemia, an early symptom of T2DM, triggers cellular oxidative stress due to a loss of balance between the individual's antioxidant and oxidant mechanisms. To reduce the morbidity and mortality of T2DM, health education programs (HEP) instruct adult populations to improve their eating and physical activity habits to reduce their hyperglycemic condition.

Objective. Determine the efficacy of a HEP by comparing different anthropometric, biochemical and oxidative stress parameters in the enrolled population.

Methodology. Eighty-six adults with hypertension, obesity and/or diabetes were enrolled in the HEP "Abriendo tu corazón", and parameters as weight, height, waist circumference, capillary glucose, and blood pressure were weekly monitored for 8 weeks. Blood samples were collected at the beginning and at the end of the program to evaluate the levels of blood glucose, glycated hemoglobin, total cholesterol, and triglycerides. Plasma samples were used to determine the total antioxidant capacity (TAC), total oxidant state (TOS), oxidant stress index (OSI) and two oxidant stress biomarkers: thiobarbituric acid reactive substances (TBARS) and advanced oxidation protein products (AOPP). The protocol was approved by the "Comité de Bioética para la Investigación en Seres Humanos (COBISH)-Cinvestav", 091/2022.

Results. From the participants, 66.3% completed the HEP and a significant reduction in glucose levels, glycated hemoglobin, cholesterol, TAC and TBARS was observed at the end of the intervention. Those changes were associated with the highly frequent participants to the program. Results denote that these interventions are effective to reduce glucose levels and other clinical and oxidative parameters associated with T2DM development.

Acknowledgements. The project is financed by the Secretaría de Educación, Ciencia, Tecnología e Innovación (SECTEI/169/2023) granted to AAM.

MITOCHONDRIAL DYSFUNCTION IN THE MODEL OF SPONDYLOARTHRITIS IN DBA/1 MICE

Rodrigo Prieto-Carrasco¹, Susana Aideé González-Chávez¹ and César Pacheco-Tena¹

¹Laboratorio PABIOM, Facultad de Medicina y Ciencias Biomédicas, Universidad Autónoma de Chihuahua, Chihuahua 31125, México.
E-mail: rprieto@uach.mx; Tel.: +52-614-2157024

Introduction. Spondyloarthritis (SpA) is a family of chronic rheumatic diseases characterized by inflammatory arthritis and abnormal bone proliferation that involve peripheral and axial joints. Dysregulation of mitochondrial function contributes to the pathophysiology of several autoimmune diseases. Although most studies have focused on the role of mitochondrial dysfunction in the context of systemic lupus erythematosus or rheumatoid arthritis, mitochondrial defects are also linked to other rheumatic diseases like SpA¹.

Objective. Determine the alterations in mitochondrial fission, fusion, biogenesis, and mitophagy in the SpA model in DBA/1 mice.

Materials and methods. DBA/1 male mice were included and confined for 6 and 11 weeks as described for the SpA model; additionally, BALB/c mice were used as controls. The RNA expression levels of mitochondrial dysfunction genes were evaluated in mice hind paws joints by RT-qPCR using specific primers and compared between the three groups using the one-way ANOVA test. Significant differences were considered when $p \leq 0.05$.

Results. Compared to BALB/c mice, all the analyzed genes were significantly lower in SpA-DBA/1 mice at 6 and 11 weeks. The expression of DNM1L (Drp1), which is involved in mitochondrial fission, was lower in the 6-week SpA group, and this decrease was accentuated at 11 weeks. The expression of the mitochondrial fusion regulator, MFN2, was lower in the 6-week and its expression levels increased by 11 weeks. PPARGC1A (PGC-1 α), the master regulator of mitochondrial biogenesis, and PINK1, which is involved in the mitophagy process, were found to be decreased after 6 weeks (Figure 1).

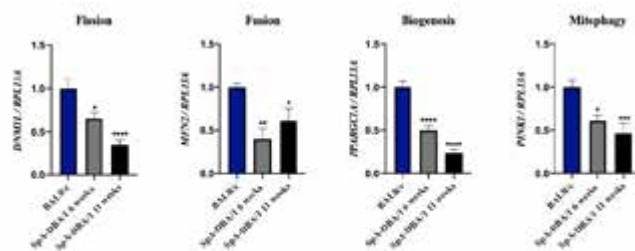


Figure 1. Alteration in RNA expression in the mitochondrial processes of fission, fusion, biogenesis and mitophagy in the Spondyloarthritis (SpA) model in DBA/1 mice. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ y **** $p < 0.0001$ vs BALB/c

Conclusions: Mitochondrial dynamics maintain the integrity and function of mitochondria; deregulation between fission, fusion, biogenesis, and mitophagy causes the accumulation of damaged and dysfunctional mitochondria. In this model, the decrease in fission prevents dysfunctional mitochondria from being correctly eliminated by mitophagy. In addition, the biogenesis process is also altered, preventing the production of new functional mitochondria.

Reference

1. Becker YLC, Duvvuri B, Fortin PR, Lood C, Boilard E. The role of mitochondria in rheumatic diseases. *Nat Rev Rheumatol.* 2022 Nov;18(11):621-640.

COMPARISON OF CHEMO CYTOTOXICITY IN A 2D VS 3D MODEL BY LDH IN PRIMARY CELL CULTURE FROM PATIENTS WITH PANCREATIC CANCER

Valeria Punzo Mora³, Eric Alejandro García López¹, José Luis Ventura Gallegos², José Luis Rodríguez Olivares¹, Yanin Chávarri Guerra¹, Ildefonso Roberto de la Peña López¹, Eucario León Rodríguez¹, Jesús Alejandro Gabutti Thomas¹, Alejandro Zentella Dehesa^{1,2}

¹National Institute of Medical Sciences and Nutrition Salvador Zubirán. ²Department of Genomic Medicine and Environmental Toxicology, Biomedical Research Institute, UNAM. ³National Technological Institute of Mexico, Morelia Campus. Vasco de Quiroga 15, Belisario Domínguez Secc. 16, Tlalpan, 14080 Ciudad de México, CDMX. 55 1333 4526. azentell@iibiomedicas.unam.mx

The evaluation of cytotoxicity in monolayer (2D) models is commonly carried out by metabolic assays such as MTT or crystal violet; otherwise there may be errors pronounced by chemical interference with the chemotherapeutic agents. For cell culture in 3D spheroids, different techniques have been emerged to evaluate cytotoxicity such as morphological image analysis; however, an ideal standard technique has not been established to evaluate cytotoxicity to chemotherapeutic agents. On clinical studies, the enzyme lactate dehydrogenase (LDH) is used to determinate if the cells of tissues have been damaged, that is why this enzyme is considered as a scape enzyme

Compare the sensitivity of measuring cytotoxicity by chemotherapeutic agents by LDH in a 2D and 3D model in primary culture of cells from patients with pancreatic cancer.

In 2D model, the cell culture was in a plate of 48 wells, and 3D in Corning Spheroid Microplate REF4515. The chemotherapeutic agents used were: Cisplatin, Carboplatin, Oxaliplatin, Paclitaxel, Doxorubicin, Gemcitabine, Irinotecan, 5-FU. The evaluation of LDH cytotoxicity was using the protocol described in the In Vitro Toxicology Assay Kit, Lactic Dehydrogenase based in Sigma-Aldrich catalog tox7-1kt, the time for the evaluation was 48 hours after the administration of the chemotherapeutic agents.

Between 2D and 3D models the chemo-sensitivity taking LDH levels had a general average sensitivity of 36% in a range of 29-45 %. In the morphological analysis, qualitative changes are observed in the treatments.

LDH quantification is a useful tool to evaluate cytotoxicity in both models. The differences in the intensity cytotoxic effect can be attributed to the junctions generated or changes in the morphology of the model. This is important, so the model 3D is closer to what happens clinically in growth-tumors.

UNLOCKING THE LITHIUM POTENTIAL: A REVIEW OF ITS ANTICANCER PROPERTIES

Laura Itzel Quintas Granados¹, Edgar Yebrán Villegas Vázquez²,
Octavio Daniel Reyes Hernández², Gabriela Figueroa González²

¹Universidad Autónoma de la Ciudad de México, Colegio de Ciencias y Humanidades, Doctor García Diego 168, Colonia Doctores, Alcaldía Cuauhtémoc, 06720. ²Laboratorio de Farmacogenética, Unidad Multidisciplinaria de Investigación Experimental Zaragoza, Facultad de Estudios Superiores Zaragoza, Universidad Nacional Autónoma de México, 09230.
E-mail: itzel.quintas@uacm.edu.mx

Lithium, a therapeutic cation primarily utilized in the management of bipolar disorders, exhibits noteworthy attributes as an anticancer agent. Here we analyzed various aspects of lithium, spanning its cellular uptake mechanisms to novel delivery methods, supported by previous reports on genomic, transcriptomic, and proteomic data. Lithium acetoacetate (LiAcAc), lithium chloride (LiCl), lithium citrate ($\text{Li}_3\text{C}_6\text{H}_5\text{O}_7$), and lithium carbonate (Li_2CO_3) induce apoptosis, trigger autophagy, and impede tumor proliferation, invasion, and metastasis, along with inducing cell cycle arrest. The transport and permeation pathways of the Li^+ ion coincide with those of other ions, notably sodium, thereby dictating extracellular and intracellular Li^+ concentrations based on its intake. However, LiAcAc necessitates specialized transporters such as monocarboxylate transporters (MCTs). Both in vivo and in vitro cancer models demonstrate lithium's capability to activate the Wnt/ β -catenin pathway. Upon binding of a secreted glycoprotein (Wnt) to its receptor, the Glycogen Synthase Kinase 3 β (GSK3 β) becomes activated. Lithium inhibits GSK3 β through Ser-9 phosphorylation, leading to the stabilization of free β -catenin in the cytoplasm. Consequently, GSK3 β may exhibit either anti- or pro-apoptotic activity depending on the cell type. Data suggest that lithium salts such as citrate, carbonate, chloride, and acetoacetate can elicit antitumor effects, including apoptosis, autophagy, cell death, inhibition of tumor growth, proliferation, invasion, metastasis, and cell cycle arrest in both in vivo and in vitro models. The selection of lithium delivery forms, whether nanosized or original, significantly influences the antitumor response, contingent upon factors such as cellular uptake, cancer type, dosage, and administration. Nevertheless, the therapeutic potential of lithium hinges on several variables, including tumor characteristics, lithium salt type, and dosage regimen. Further investigation is required to ascertain the selectivity of Li^+ ions for tumor cells, as well as optimal dosages and administration protocols.

EVALUATION OF THE APOPTOTIC EFFECT OF THE O1P4BAD PEPTIDE IN JURKAT CELLS OF ACUTE LYMPHOBLASTIC LEUKEMIA AND PBMC

Ana Mayra Reséndiz Corona¹, Edgardo Becerra Becerra¹, Guadalupe García-Alcocer¹,
Laura C. Berumen¹, Irasema Mendieta¹

¹Posgrado en Ciencias Químico Biológicas, Facultad de Química, Universidad Autónoma de Querétaro,
Las Campanas S/N, C.P. 76010, Santiago de Querétaro, Qro.
E-mail: aresendiz@alumnos.uaq.mx, alicia.mendieta@uaq.mx

Leukemia is a type of cancer from blood cells that proliferate and accumulate in the bone marrow and other hematopoietic tissues. Leukemias are one of the main causes of childhood death in Mexico, particularly type T acute lymphoblastic lymphoid leukemia (T-ALL); which, although rare, is a complex and heterogeneous subtype at genetic level, also has fewer therapeutic alternatives.

Currently, conventional chemotherapeutics as vincristine are used, which has been associated with adverse effects such as neuronal damage and conditions in the peripheral nervous system.

Due to the clinical challenge that T-ALL represents, there is an opportunity to develop new targeted therapies such as peptides, which have been shown as promising cancer immunotherapies designed with the ability of attacking specific cells.

The objective of this work is to study the apoptotic effect of the O1P4Bad peptide (designed *in silico* to interact with Bcl-2 and promote apoptosis) on T-type acute lymphoblastic leukemia cells (Jurkat) and in healthy cells (PBMC). We used concentrations of 1, 10, 50, 100 and 200 μM . In addition, we used vincristine 10 μM as positive control to compare the cytotoxic effect. During 0, 24 and 48 hours.

The results showed a cytotoxic effect of the O1P4Bad peptide significantly greater than the control from 1 μM at 24 hours in Jurkat cells; vincristine generated a significantly greater effect than the control from 10 μM at 24 hours in Jurkat cells. The peptide effect remains at 48 hours and with all concentrations used. In contrast, the O1P4Bad peptide generated a significantly greater cytotoxic effect than the control up to 50 μM at 24 hours in PBMCs cells; while vincristine generated a significantly greater effect than the control from 10 μM at 24 hours in PBMCs cells.

These results suggested that the O1P4Bad peptide can reduce cell proliferation in cancer cells, showing a greater decrease in this than the chemotherapeutic vincristine. These data suggest the antiproliferative effect of the O1P4BAD peptide, studies to determine the apoptotic mechanisms involved are carried out at the moment.

This work was partially funded by UAQ-FOPES 2024.

IDENTIFICATION OF BIOMARKERS FOR DIAGNOSIS AND PROGNOSIS OF PEDIATRIC MEDULLOBLASTOMA

Alicia María Reveles-Espinoza¹, Andrés Rivera Chávez², Isaac Chairez³, Luis Figueroa-Yáñez⁴, Luis Gerardo Arriaga⁵, Janet Ledesma-García⁶, Jannú R. Casanova Moreno⁵, Goldie Oza⁵, José Luis Herrera Celis⁵, Ricardo A. Escalona-Villalpando⁶, Alba Adriana Vallejo-Cardona², Sergio Juárez-Méndez^{7*} and Iván Salgado Ramos[†]

¹CIDETEC-IPN, 07700, Mexico City, 2BMF-CIATEJ, 44270, Guadalajara, Jal., 3IAMSM- ITESM, 45138, Zapopan, Jal., 4BI-CIATEJ, 45019, Zapopan, Jal., 5CIDETEJ, 76703, Pedro Escobedo, Qro., 6FI-UAQ, 76010, Querétaro, Qro., 7INP, 04530, Mexico City.

E-mail: †avallejo@ciatej.mx, *ser.mend@gmail.com, †isalgador@ipn.mx

The goal of precision medicine is to direct diseases prevention, diagnosis, and treatment strategies to specific groups of people, which are classified based on genomic, environmental, lifestyle characteristics, among other; this with the purpose of providing more effective treatments, reducing side effects, and even optimizing resources¹. Precision medicine is supported by omics sciences, like genomic, transcriptomic, etc. These produce a large amount of complex data whose processing and analysis is difficult by traditional techniques. Deep learning techniques can solve these problems. Artificial neural networks (ANNs) are software tools that can be used for biomarkers identification and classification².

Medulloblastoma is the malignant solid tumor of the central nervous system more common in childhood. This type of tumor is very heterogeneous, and it has been classified in four molecular subgroups: WNT, SHH, Group 3 (G3) and Group 4 (G4). Each subgroup has different prognosis, being G3 and G4, two of the more frequent and aggressive, the least characterized³. The objective of the present work is to identify novel molecular markers for each subgroup of medulloblastoma, with emphasis on G3 and G4. Expression microarray data of medulloblastoma tumors and healthy control cerebellar tissues are downloaded from repositories, like the Gene Expression Omnibus of the National Center for Biotechnology Information, and analyzed by ANNs to determinate expression profiles for each subgroup. The identification of novel biomarkers that help in medulloblastoma diagnosis and prognosis could allow the development of better treatment strategies for this type of cancer.

References

1. Konig, I.R., et al. (2017). *Eur Respir J*, 50, 1-12.
2. Bhinder, B., et al. (2021). *Cancer Discov*, 11, 900-915.
3. Cotter, J.A., Hawkins, C. (2022). *Pediatr Dev Pathol*, 25, 23-33.

ANALYSIS OF DAPK1 METHYLATION IN APOPTOSIS RESISTANCE IN IDIOPATHIC PULMONARY FIBROSIS DERIVED FIBROBLASTS

Mariana Río de la Loza¹, David A. Lizcano¹, Remedios Ramírez¹, Uriel Camacho-Silverio¹,
Tania Valdivia-Herrera¹, Fernanda Toscano-Marquez², Moisés Selman², Annie Pardo¹, Yair Romero¹

¹Facultad de Ciencias, UNAM. Laboratorio de biopatología pulmonar. ²Instituto Nacional de Enfermedades Respiratorias (INER), Laboratorio de biopatología pulmonar.

Introduction. Idiopathic pulmonary fibrosis (IPF) is a progressive, irreversible, and fatal disease characterized by the aberrant activation of epithelial cells. This activation promotes the formation of fibroblast/myofibroblast foci that uncontrollably synthesize extracellular matrix components, reducing elasticity, altering lung architecture, and ultimately impairing gas exchange. It is known that IPF fibroblasts are resistant to apoptosis, with molecular mechanisms of evasion or resistance that have not yet been fully elucidated. This resistance leads to their accumulation and persistent activation, turning regeneration into progressive fibrosis. Specifically, these changes may result from epigenetic alterations acquired with aging, preventing proper tissue regeneration. Therefore, it is of great interest to investigate what prevents these cells from entering the apoptotic stage. The aim of this project is to analyze the relationship between the expression and methylation of genes that promote programmed cell death (apoptosis), specifically the “DAPK1” gene, in fibroblasts from patients with IPF.

Methods. For this analysis, primary cell cultures derived from patients with IPF and controls were used, following bioethical protocols. DNA and RNA were extracted and purified using the TRIZOL method. The obtained DNA was subjected to RT-qPCR with specific primers, and the amplified regions underwent bisulfite treatment, revealing methylation with MS-PCR. Subsequently, experiments with genetic and pharmacological inhibitors were conducted on the cell lines to induce DAPK loss of function, followed by apoptosis assays using TUNEL staining.

Results. The fibrotic cell lines showed significant differences in DAPK1 expression compared to controls. Additionally, methylation supported the expression results, as higher methylation was observed in IPF. Similarly, applying the genetic or pharmacological inhibitor of DAPK1 in control cells inhibited cellular apoptosis.

Conclusions. The presence of DAPK1 gene methylation is a strong inhibitor of apoptosis in fibrotic cells, promoting disease progression.

IMPACT OF PERIODONTAL DISEASE ON MENTAL HEALTH

Andrea Yamile Rivas Escobedo¹, Sergio Manuel Salas Pacheco¹, Omar Alejandro Tremillo Maldonado¹, Yareli Guadalupe Ramos Herrera¹, Oscar Almeda Ojeda¹, Javier Luis Solís Martínez¹, Marcelo Gómez Palacio Gastelum¹, Ada Agustina Carrillo² y José Manuel Salas Pacheco²

¹Facultad de Odontología, Universidad Juárez del Estado de Durango, ²Instituto de Investigación Científica, Universidad Juárez del Estado de Durango.
c.d.andrearivas@gmail.com

Keywords. Periodontal disease; Depression; Pregnant women.

Background. Periodontal disease is considered multifactorial and idiopathic; however, it is described as a chronic inflammatory condition associated with biofilm and characterized by the progressive destruction of the tooth-supporting apparatus. In Mexico, this disease has been reported to be present in up to 70% of the population, according to the American Academy of Periodontology. Multiple studies have reported a relationship between periodontal disease and some mental disorders such as depression, particularly in at-risk populations like pregnant women. However, the evidence of these findings remains contradictory. The prevalence of periodontal disease in pregnant women is 73.6%. Depression is a mental disorder characterized by the persistence of at least five symptoms of depression (sadness, loss of interest, loss of sleep, restlessness, frustration) every day for almost the entire day for at least two weeks. In Mexico, the National Psychiatric Epidemiology Survey reports an annual depression prevalence rate of 4.8% among the general population aged 18 to 65 years and 32.6% specifically among women.

Objective. To determine the CPITN (Community Periodontal Index of Treatment Needs) and the degree of depression using the Hamilton instrument, thereby evaluating the relationship between periodontal disease and depression.

Materials and methods. Evaluate the presence of periodontal disease using the Community Periodontal Index of Treatment Needs (CPITN).

Apply depression evaluation scales: Hamilton instrument to quantitatively establish the level of depression.

Compare results using statistical methods between the CPITN and Beck and Hamilton scales.

Results. A total of 14 patients were included, with an average age of 29.28 years and an average gestation period of 15.58 weeks. 72% of the population obtained a CPITN code 3, as shown in Figure 1. The degree of depression based on the Beck Inventory showed an average of 12, while the Hamilton Inventory showed an average of 8.64.

Discussion and conclusions. According to our results, 93% of pregnant women presented with periodontal disease, with 21% having a CPITN code 3. In terms of depression, 36% of the patients showed mild depression symptoms, while 7% presented severe depression, according to the Hamilton scale score. The Beck instrument showed an average score of 12 with a standard deviation of 6.07, while the Hamilton scale showed an average score of 8.64 with a standard deviation of 5.43.

References

1. Tonetti, M. S., Greenwell, H., & Kornman, K. S. (2018). Journal of Periodontology.
2. Azouni, K. G. (2014). J Clin Diagn Res.
3. Romandini et al. (2020)

CORRELATION OF GONADOTROPIN-RELEASING HORMONE RECEPTOR EXPRESSION IN BREASTCANCERCELLINESAND ISIMPLICACIONAS THERAPEUTICTARGET

Daniela Rodríguez-Cruz^{1,2}, Mauricio Flores-Fortis^{3,4}, Eunice López-Muñoz¹, Miguel Ángel López Valle⁵, Astrid Karyme Polanco López⁶, Elena Aréchaga Ocampo⁴, Arturo Aguilar-Rojas¹

¹Mexican Social Security Institute, Medical Research Unit in Reproductive Medicine, High Specialty Medical Unit in Gynecology and Obstetrics No.4, Mexico City, Mexico. ²Center for Research and Advanced Studies of the National Polytechnic Institute, Department of Molecular Biomedicine, Mexico City, Mexico. ³Metropolitan Autonomous University, Cuajimalpa Unit, Engineering and Natural Science Doctoral Program, Mexico City, Mexico. ⁴Metropolitan Autonomous University, Cuajimalpa Unit, Department of Natural Science, Mexico City, Mexico. ⁵Mexican Social Security Institute, Breast Tumor Service, High Specialty Medical Unit in Gynecology and Obstetrics No.4, Mexico City, Mexico. ⁶Mexican Social Security Institute, General Hospital of Zone No.8, Mexico City, Mexico.

*a_aguilar@unam.mx

Breast cancer (BC) is the most common neoplasm in women worldwide. There are different staging classifications according to the different types of cancer. Based on the American Joint Committee on Cancer classification, BC is classified as: stage 0 (zero), or non-invasive ductal carcinoma in situ (DCIS), and stages I to IV, representing invasive BC. In turn, the latter are subclassified into early clinical stages (IA, IB, IIA), locally advanced (IIB, IIIA, IIIB, IIIC), and metastatic (IV). Locally advanced and metastatic tumors present an aggressive course of the disease and are of particular clinical interest. The response rate of these tumors to treatment is low due to the development of resistance, and their prognosis is poor. Likewise, for stage IV patients, tumor recurrence is frequent, and most deaths occur within the first 5 years after diagnosis. All of the above underscores the importance of identifying new therapeutic targets that may be specific to these cancer stages. This is how our working group has proposed the gonadotropin-releasing hormone (GnRH) and its receptor, the gonadotropin-releasing hormone receptor (GnRHR), as a new therapeutic target. The effect shown by the GnRH analog, Buserelin, is to reduce invasion and migration in breast cancer cell line, MDA-MB-231. Considering the above information and to support the use of Buserelin as adjuvant therapy to decrease metastasis, in the present study, we have undertaken the task of evaluating the expression levels of GnRHR in different breast cancer cell lines. To achieve our objectives, we have employed the MCF10A line as non-tumoral, the MCF7 line as locally advanced clinical stage, and the MDA-MB-231 line as metastatic clinical stage. GnRHR levels were evaluated by qPCR. Based on the results obtained, we determined that in both tumor lines, there are high levels of GnRHR expression compared to the control. Like wise, the MCF7 line showed higher receptor expression levels and better response to stimulation with Buserelin. These observations correlate with the cellular invasion capacity of each line, i.e., MCF7 invades less and MDA-MB-231 invades more, which could be related, among other factors, to GnRHR expression levels. We conclude that the use of Buserelin could have a protective effect on metastasis of locally advanced tumors primarily.

This work was supported by the "Coordinación de Investigación en Salud-IMSS" Project No: R-2015-3606-18. D. R-C. is postgraduate students from the "Programa de Maestría en Biomedicina Molecular, Departamento de Biomedicina Molecular, CINVESTAV, México". M. F-F is postgraduate student from the "Posgrado en Ciencias Natural e Ingeniería, Universidad Autónoma Metropolitana, Unidad Cuajimalpa, México". They respectively received scholarships No. 1102304 and No. 472137 from "Consejo Nacional de Humanidades, Ciencia y Tecnología (CONAHCYT-México)".

EXPRESSION OF SERCA2 AND SERCA3 GENES IN A MODEL OF EPITHELIAL-MESENCHYMAL TRANSITION IN MCF10A CELLS AND BREAST CANCER CELL LINES IN RESPONSE TO RESVERATROL

Gabriela Rodríguez Rodríguez, Ángel Salmerón Hernández, Denis Yarazetl Romero Maldonado, Miguel Ángel Rodríguez Fernández, Pablo Sinuhé Gómez González, Ana Cecilia Sánchez Trujillo, Ángel Zarain Herzberg*

Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México, UNAM.
Av. Universidad 3000, 04510, Mexico City.

*Correspondence: zarain@unam.mx

Background. Cytoplasmic calcium ion (Ca^{2+}) concentration is mainly regulated by the sarcoendoplasmic reticulum Ca^{2+} -ATPases (SERCA) pumps¹. Calcium ion concentration is dysregulated in cancer and participates in different stages of carcinogenesis, such as the epithelial-mesenchymal transition (EMT)². It has been observed in breast cancer cell lines an alteration in the expression of the SERCA2b and SERCA3 pumps. SERCA expression could be affected by epigenetic mechanisms such as histone acetylation and DNA methylation³. Resveratrol (RSV) has shown activity as an inhibitor of histone deacetylases (HDACi) and DNA methyltransferases (DNMTi)⁴.

Objective. This study aims to investigate the expression of SERCA2b and SERCA3 in the non-tumor breast cell line MCF10A and the HER2+ tumor cell line SKBR3 breast cancer. We examined how the expression of these genes was influenced by treatment with RSV and in an EMT induction model (EMTi), providing insights into the role of these genes in cancer biology.

Methods. An epithelial-mesenchymal transition (EMT) model in MCF10A was induced by treatment with 5ng/ml of TGF- β and 50ng/ml of EGF. RNA was extracted from the cell cultures treated with RSV (12.5, 25, and 50 μM) and from the EMT model. We performed RT-qPCRs from this cDNA and analyzed the results using the $\Delta\Delta\text{CT}$ method to obtain the relative mRNA SERCA2b and SERCA3 isoform expression. Additionally, we conducted RNA-seq of MCF10A to analyze global mRNA expression.

Results. In MCF10A cells treated with RSV, the expression of mRNA for SERCA2b and SERCA3 was increased, while the combined treatment of the EMTi model and RSV decreased the expression of SERCA2 compared with cells treated only with RSV. In SKBR3 cells, an increase in the expression of SERCA2 and SERCA3 was observed with RSV. However, the expression of SERCA3 was decreased with RSV plus the EMTi treatment but not SERCA2b.

Conclusions. Treatment with RSV increases the expression of SERCA2 and SERCA3 mRNA in the MCF10A and SKBR3 cell lines. The EMTi induction model mitigates RSV-induced expression of SERCA2 in MCF10A and SERCA3 in SKBR3. RNA-seq data analysis of the EMTi model revealed increased SERCA3 expression and was validated by RT-qPCR.

References

1. León, et al. (2010). *Rev Med UV (11)* 1: 32-39.
2. Jones, et al. (2021). *Biomedicines (9)* 1200: 1-13.
3. Zarain, et al. (2021). *Gac. Med. Mex (175)* 4: 357-363.
4. Izquierdo, et al. (2019). *International Journal of Biochemistry and Cell Biology* 113: 37-47.

EVALUATION OF FIRST- AND SECOND-GENERATION PLATFORMS FOR MESSENGER RNA SYNTHESIS AS A POTENTIAL TECHNOLOGY IN VACCINE DEVELOPMENT

Griselda Rodríguez-Martínez^{*1}, Yolanda Patricia Alemán-García^{*1,2}, Andrea Gala Varona-Velázquez¹, Marina Ramírez-García¹, Mariana Romo-Castillo^{1,3}, Dony Karent Mayor-Villeda¹ and Víctor Manuel Luna Pineda¹

¹Laboratorio de Investigación en Patógenos Respiratorios y Producción de Biológicos, Hospital Infantil de México Federico Gómez. ²Posgrado en Ciencia en Inmunología, Escuela Nacional de Ciencias Biológicas. ³IxMx CONAHCYT.

*Estos autores contribuyeron igualmente.

Dr. Márquez 162, Doctores, Cuauhtémoc, Ciudad de México, 06720, 5538090034, griseldargzmtz123@gmail.com; luna.pineda@hotmail.com.

The first-generation (FG) RNA vaccines (mRNA-1273 and BNT162b2) based on lipid nanoparticle-encapsulated nucleoside-modified mRNA encoding the full-length SARS-CoV-2 spike protein could control COVID-19 pandemic. In February, a second-generation (SG) RNA vaccine (ARCT-154) against COVID-19 was approved in Japan. Since ARCT-154 is based on self-amplifying RNA technology, it showed non-inferiority compared to BNT162b2. In this study, the pUC-T7-RNA-VMLP (FG) and pTNT-T7-VEE-RNA-VMLP (SG) platforms were developed for mRNA synthesis as a potential technology in vaccine development. FG platform contains from 5' to 3': T7 promoter, UTR-5', a multiple cloning site (MCS), 2X UTR-3', and a *Sma*I-delimited polyA tail (n = 220). On the other hand, the SG platform contains from 5' to 3': the conserved sequence element (CSE) 5', ORFs replicase (nsp1-4) of the Venezuelan equine encephalitis virus, a subgenomic promoter, a MCS, the CSE 3' and a *Mlu*I-delimited polyA tail. *Gfp*- and *luc* sequences were cloned in both platforms, and mRNA was generated by *in vitro* transcription using nucleosides without/with modification, followed by capping with the vaccinia virus system. FG- and SG-mRNA were evaluated in HEK293T cells at different concentrations (50-1000 ng) and time-lapses (24-120 h). GFP analysis was performed by EPI-fluorescence, whereas luciferase was assayed by a luminometer. The presence of fluorescence in cells transfected with FG-mRNA-*gfp* was observed from 250 ng, with a maximum at 48h and remaining stable up to 72h, while SG-mRNA-*gfp* remained stable with lower concentration (100 ng). Cells transfected with FG-mRNA-*luc* presented bioluminescence at 24h in a concentration-dependent manner, while SG-mRNA-*luc* was observed at 100 ng. These findings indicate that both platforms can generate functional mRNA, and SG-mRNA showed non-inferiority with FG-mRNA.

PREVALENCE AND CLASSIFICATION OF C-SHAPED CANAL AND RADIX IN MANDIBULAR MOLARS USING CONE-BEAM COMPUTED TOMOGRAPHY IN MEXICAN POPULATION

Angel Gustavo Romo Mireles¹, Hugo Alejandro Bojórquez Armenta¹, Sergio Salas Pacheco¹, Omar Alejandro Tremillo Maldonado¹, Javier Solís Martínez¹, Yarely Ramos Herrera¹, Oscar Almeda Ojeda¹, José Salas Pacheco², Jesús Antonio Pescador Álvarez²

¹Facultad de Odontología, Universidad Juárez del Estado de Durango;

²Instituto de Investigación Científica, Universidad Juárez del Estado de Durango.

E-mail: angelgustavoromomireles@gmail.com

The diverse morphological configurations in dental organs present clinical challenges in root canal treatment, complicating instrumentation, and irrigation processes, which can lead to treatment failure. Understanding anatomical variations, such as C-shaped canals and radix entomolaris, enhances clinical skills and improves long-term endodontic treatment success rates. Cone-beam computed tomography (CBCT) offers superior diagnostic capabilities over conventional radiography, enabling pre-operative detection of root configurations and canal numbers, facilitating personalized endodontic treatments. Methods: A total of 2,173 dental organs of a Mexican population, including 1,057 first mandibular molars and 1,116 second mandibular molars, were studied using CBCT to identify C-shaped canals and radix configurations. Results: C-shaped canals were identified in 160 dental organs, with a prevalence of 0.2% in first mandibular molars and 14.1% in second mandibular molars. The highest frequency was in the left second mandibular molar (OD 3.7) at 14.8%. Gender differences were significant, with higher prevalence in females (27.3%) compared to males (13.3%). The most common C-shaped canal configuration was type C2 (39.3%). Radix entomolaris was found in 52 dental organs, with a prevalence of 3.4% in first mandibular molars and 1.4% in second mandibular molars. Conclusions: Early detection of anatomical variations using CBCT allows for tailored endodontic treatments, improving management of complex root canal anatomies and enhancing long-term outcomes. This study emphasizes the importance of understanding root canal morphology in mandibular molars to optimize endodontic procedures.

LOOKING FOR MOLECULAR BIOMARKER'S CHILDHOOD ACUTE LYMPHOBLASTIC LEUKEMIA

Elieel Ruiz-May¹, Silverio Soto-Alvarez², Esaú Bojórquez-Velázquez¹, Laura S. Rangel-Cova², José M. Elizalde-Contreras¹, Armando Vilchis-Ordoñez³, Miguel Ángel Palomo-Colli³, Tania Angeles Floriano³, Israel Parra Ortega³, Rosana Pelayo-Camacho⁴, Lidia F.E. Huerta-Nuñez^{2*}

¹Instituto de Ecología A.C. (INECOL), Carretera Antigua a Coatepec No. 351, El Haya, CP 91073, Xalapa, Veracruz, Mexico, ²Escuela Militar de Graduados de Sanidad, Centro de Investigación del Ejército y Fuerza Aérea Mexicana, (C.I.D.E.F.A.M), Universidad del Ejército y Fuerza Aérea Mexicana, Mexico City, C.P.11200, México, ³Hospital infantil de México Federico Gómez, Dr. Márquez 162 CP 06720, ⁴Unidad de Educación e Investigación, IMSS/Oncoinmunología y Citómica del Cáncer Infantil. E-mail: eliel.ruiz@inecol.mx

Acute lymphoblastic leukemia (ALL) is a neoplastic disease more common in children worldwide ¹. One of the major issues of this disease is the infiltration of leukemic cells into the Central Nervous System (CNS). However, leukemic cell infiltration is only detected in a minimal percentage of patients (3-5 %) or when the disease is in advanced stages of severity. It is suggested the occurrence of submicroscopic invasion during the early stage of the disease. Therefore, reference tests cannot allow the detection of leukemic cell infiltration, as demonstrated by molecular biology and flow cytometry approaches²⁻³. The limitation to determining the occurrence of infiltration of leukemic cells into the CNS jeopardizes the well-being of pediatric patients in providing an unspecific treatment that in some cases toxic nature could lead to health issues underpinned by the identification of molecular markers in the CSF. In this context proteomics approach has been an omics tool suitable for finding molecular markers associated with diseases in humans. Therefore, we conducted a comparative proteomics scrutiny-based label free quantification approach (LFQ). We compared children with the occurrence leukemic cell infiltration into the CNS to those without infiltration. Proteomics data exhibited the over-accumulation of proteins associated with acute-phase response, complemented activation and aminoglycan metabolism. Protein interaction network exhibited the clustering of three carbonic anhydrases (CA1, CA2, CA3), which were tightly associated with ROS scavenger proteins. The possible implication of CAs in regulating the redox hemostasis in leukemic cells for proper proliferation could suggest these proteins as possible targets for therapies to treat ALL in children.

References

1. Filbin, M. & Monje, M. *Nature Medicine* vol. 25 367–376 (2019).
2. Frishman-Levy, L., & Izraeli, S. *British Journal of Haematology*, 176(2), 157-167(2017).
3. Guldbrandsen, et. al., *Molecular & Cellular Proteomics*, 13(11), 3152-3163 (2014).

CA²⁺ OVERLOAD-INDUCED MITOCHONDRIAL DYSFUNCTION IS AN EARLY RISK FACTOR FOR LETHAL VENTRICULAR ARRHYTHMIAS

Felipe de Jesús Salazar-Ramírez¹, Judith Bernal-Ramírez¹, Luis Luévano-Martínez¹, Christian Silva-Platas¹, José Velasco-Bolom², Flavio F. Contreras-Torres², Guillermo Torre-Amione¹, Gerardo García-Rivas¹

¹Cátedra de Cardiología y Medicina Vascular, Escuela Nacional de Medicina, Tecnológico de Monterrey, Hospital Zambrano-Hellion, San Pedro Garza-García, N.L., México 66278, ²Tecnológico de Monterrey, Escuela de Ingeniería y Ciencias, Monterrey, NL, 64849, Mexico.

E-mail: gdejesus@tec.mx

Introduction. A significant cause of death within the cardiovascular patient population is ventricular arrhythmias, which are associated with elevated catecholamine levels. Since mitochondrial calcium (Ca²⁺) transport is necessary to elicit an adrenergic response in cardiac tissue and constant stimulation leads to mitochondrial Ca²⁺ overload and dysfunction, we evaluated its role and the effect of its modulation in arrhythmogenesis.

Objectives. Evaluate the effects of mitochondrial Ca²⁺ transport modulation in lethal arrhythmia generation.

Materials and methods. 12-15 week-old C57bl/6 male mice were administered intravenously with either Ru₃₆₀, a mitochondrial Ca²⁺ transport inhibitor, or normal saline. A baseline ECG was recorded, after which Isoproterenol (ISO, 400mg/kg) was administered subcutaneously, and ECG recording was kept for another 20 minutes. Afterward, cardiomyocytes and mitochondria were isolated for characterization studies.

Results. ISO administration caused ventricular tachycardia and fibrillation, while pretreatment with Ru₃₆₀ prevented ventricular arrhythmias. Mitochondria from ISO hearts had a higher Ca²⁺ content, indicating overload, and compromised function and membrane integrity, demonstrated by a lower respiratory control, Ca²⁺ retention capacity and mitochondrial membrane potential. ISO administration also increased peroxide production, electron leak and ROS-driven post-translational modifications as well as erratic cellular Ca²⁺ dynamics, which presented as a higher incidence of anomalous activity. Ru₃₆₀ pretreatment prevented or partially prevented these findings.

Conclusions. Mitochondrial Ca²⁺ overload promotes arrhythmias by inducing mitochondrial dysfunction and increasing oxidative stress, which compromises cellular Ca²⁺ dynamics and enables the appearance of anomalous activity. Mitochondrial Ca²⁺ transport modulation proved to be an effective tactic to prevent arrhythmias, which points towards a potential new target for the development of new anti-arrhythmic therapies.

NOVEL ANTIGIARDIAL COMPOUNDS IN LARREA TRIDENTATA AND THEIR IN VITRO EFFECTS ON GIARDIA LAMBLIA TROPHOZOITES

Angela Daniela Saldaña Alvarado¹, Xareni Zinereth Herrera Valero¹, Filiberto Gutiérrez Gutiérrez²,
Sendar Daniel Nery Flores¹, Juan Alberto Ascacio Valdés¹, Norma Paola Meléndez Rentería¹,
Raúl Rodríguez Herrera¹, Lissethe Palomo Ligas^{1*}

¹Facultad de Ciencias Químicas, Unidad Saltillo, Universidad Autónoma de Coahuila. Saltillo, Coahuila, 25280, México.

²Departamento de Farmacobiología, Centro Universitario de Ciencias Exactas e Ingenierías,

Universidad de Guadalajara, Guadalajara, Jalisco, 44430, México.

Tel: 8442118749 *email: lissethe_palomo@uadec.edu.mx

Giardia lamblia is one of the leading gastrointestinal protozoans globally, responsible for giardiasis, a disease clinically characterized by diarrhea, steatorrhea, and abdominal pain¹. The highest incidence is associated with socioeconomic and climatic factors, such as inadequate sanitation services and temperature. Additionally, this disease significantly impacts the pediatric population².

Current treatments primarily rely on nitroimidazole drugs, with metronidazole (MTZ) being the first-line medication in Mexico. However, instances of therapeutic failure have increased³. The exploration of phytochemicals as alternative anti-giardial agents is underway. *Larrea tridentata* (LAT), a perennial shrub used in traditional medicine in Mexico and the United States, has shown promise effects. Previous studies demonstrated that an organic extract of LAT affects protozoans such as *G. lamblia*, *Entamoeba histolytica*, and *Trichomonas vaginalis*, though these findings are preliminary⁴.

In this study, a hydroalcoholic extract of LAT leaves was obtained using microwave/ultrasound methods. Compounds were identified via HPLC/MS, and their effects on the growth and morphology of *G. lamblia* trophozoites were analyzed, both individually and in combination with MTZ. The analysis revealed nine compounds with previously unreported anti-giardial activity. The extract impacted the growth and morphology of the parasite both as a monotherapy and as an adjunct to MTZ. These findings demonstrate the potential of *L. tridentata* as a source of new anti-giardial compounds.

References

1. Saeedi, P. et al. (2024). "Differential gene expression analysis in individuals with a history of giardiasis." *AMB Express*, 14(1), 1.
2. Gutiérrez-Gutiérrez, F., & Palomo-Ligas, L. (2023). "Changes in the incidence of intestinal diseases caused by parasitic protozoa in the Mexican population (2015-2019) and its association with environmental and socioeconomic risk factors." *Parasitol Res*, 122(4), 903-914.
3. Argüello-García, R. et al. (2020). "Drug resistance in *Giardia*: Mechanisms and alternative treatments for Giardiasis." *Adv Parasitol*, 107, 201-282.
4. Morales-Ubaldo, et al. (2022). "Phytochemical Compounds and Pharmacological Properties of *Larrea tridentata*." *Molecules*, 27(17), 5393.

MEXICAN GANODERMA LUCIDUM MUSHROOM EXTRACTS INDUCE “BRCA-NESS” PHENOTYPE AND METABOLIC VULNERABILITIES IN TRIPLE-NEGATIVE BREAST CANCER: A COMPLEMENTARY MEDICINE STUDY

Ivan Salido-Guadarrama¹, Sandra Romero-Córdoba², Lilia Noriega-López², Maria Eugenia Meneses³,
Monica Peña-Luna⁴, Alfredo Hidalgo-Miranda⁴, Alessandra Cataldo⁵, Giulia Cosentino⁵,
Marilena Iorio⁵, D. Martínez-Carrera³

¹Instituto Nacional de Perinatología Isidro Espinosa de los Reyes, Bioinformatics and Statistical Analysis, Mexico City, Mexico. ²Instituto de Investigaciones Biomédicas- Universidad Nacional Autónoma de México UNAM, Departamento de Medicina Genómica y Toxicología, Mexico City, Mexico. ³Colegio de Postgraduados Puebla, Centro de Biotecnología de Hongos Comestibles- Funcionales y Medicinales, Puebla, Mexico. ⁴Instituto Nacional de Medicina Genómica, Laboratorio de Genómica del Cáncer, Mexico City, Mexico. ⁵Fondazione IRCCS Istituto Nazionale dei Tumori of Milan, Molecular Targeting Unit, Milan, Italy. E-mail: silvervann@gmail.com

Introduction. Triple negative breast cancer (TNBC) is the most challenging subtype to treat, lacking targeted therapies which restrict treatment options to chemotherapy. In recent years, new strategies focused on complementary medicine have proposed the use of natural products to combat tumor cells through a synergistic activity of their bioactive compounds on multiple molecular targets. However, more experimental evidence is still warranted shedding light on their action mechanism on cancer cells. Ganoderma lucidum (GI) is a hard edible mushroom with pro-health properties. In this study, we set out to evaluate the mechanisms of action of two hydroalcoholic extracts of Mexican GI.

Material and Methods. We employed a combination of cell line models of TNBC cells and in vivo mice tumorigenesis models to assess the effect of GI extracts. Further, global gene expression and functional annotation analysis were performed. Cellular metabolism was assessed with the Seahorse system.

Results and Discussions. GI extracts reduced the rate of proliferation, viability, and cell migration of TNBC cells. We observed that this cytotoxic effect is specific to neoplastic cells, suggesting reduced cytotoxicity in normal epithelium. In concordance, a profile of inactivated genes involved in cell viability, proliferation, and migration, as well as the disruption of DNA repair mechanisms, was discovered. In addition, DNA fragmentation and reduction of BRCA, ATM and PARP proteins were observed. Of relevance, co-administration of GI extracts with doxorubicin and PARP inhibitors potentiated doxorubicin antitumor effect in vitro and reduced tumor size in vivo. It is noteworthy that a disruption of the metabolism of TNBC cells was evidenced by a higher glycolytic rate and an increase in non-mitochondrial respiration.

Conclusion. Our findings show the potential use of a new biotechnological development of fungal extracts capable of sensitizing triple negative breast tumor cells through BRCA1/2 mutations to PARP inhibitors, as well as establishing an energy regulation that limits oncogenic capacities.

AUTOPHAGY INDUCTION AFTER LUNG EXPOSURE TO SACCHAROPOLYSPORA RECTIVIRGULA IN C57BL6 MICE

Andrea Montserrat Sanchez Barajas, Miguel Gaxiola, Annie Pardo Cemo, Moisés Selman, Sandra Cabrera Benítez

¹Laboratorio de Biopatología Pulmonar Ciencias-INER, Facultad de Ciencias, UNAM, Circuito interior S/N, Ciudad Universitaria, Coyoacán, 04510 CDMX.
E-mail: scb@ciencias.unam.mx; Tel. 56224800 #84076

Hypersensitivity Pneumonitis (HP) is one of the most common interstitial lung diseases. It occurs in genetically susceptible individuals whose immune system develops a hypersensitivity response to the inhalation of various antigens. It is an occupational disease and the constant exposure to antigen triggers inflammation and distortion of lung architecture. Previously, we have shown evidence of an alteration in the expression of some markers of the autophagy pathway in the lung of patients diagnosed with NH, there is the presence of the proteins LC3B, p62, ATG4B and ATG7 in bronchial and alveolar epithelial cells, as well as in both alveolar and interstitial macrophages, and neutrophils positive also for ATG5. Given the heterogeneity of the patients, we use a mouse model to reduce the multivariable challenge.

We develop the model in C57BL6 mice, which were instilled with 50µg of *Saccharopolyspora rectivirgula* (SR) in combination with 5µg of LPS three days a week for three weeks and were sacrificed three days after the last instillation.

The left lobe was embedded in paraffin and the sections were stained with hematoxylin and Masson's trichrome. Immunohistochemistry and immunofluorescence targeting autophagy markers were also performed. The right lobe was homogenized and evaluated the protein level by western blot. The lungs of the SR-exposed mice showed cellular infiltrates in the pulmonary parenchyma, mainly lymphocytes and macrophages, as well as thickening of the alveolar epithelium and collagen deposition.

In control mouse lungs, we observed low LC3B diffuse cytoplasmic staining only in some alveolar macrophages, whereas other cell types were negative. SR-exposed mouse lungs showed a strong LC3B-positive staining in neutrophils and alveolar and interstitial macrophages. Bronchial epithelium was also very reactive to LC3B in SR-exposed mouse lungs compared with control lungs that were negative. A strong p62 positive staining, quite similar to LC3B was observed in interstitial and alveolar macrophages and also in neutrophils.

By immunofluorescence we found a strong ATG4B positive signal in macrophages and bronchial and alveolar epithelium, while ATG5 was found in the plasmatic membrane of macrophages in the lungs of SR-exposed mice.

In addition, by immunoblot, we observed an increased LC3B, p62, ATG4B and ATG5 protein level in SR mouse lungs compared with control tissues, confirming the findings obtained by immunohistochemistry, suggesting autophagy could be activated in lung epithelium and inflammatory cells after *S. rectivirgula* exposure and HP pathogenesis.

This work was funded by PAPIIT IN224323 DGAPA-UNAM

A SKINOMIC APPROACH TO THE STUDY OF HEALTH, PATHOLOGIES, AGING AND ANTI-AGING OF THE SKIN

Juan Manuel Sánchez Contreras, Jorge Brito Sánchez, Maricruz Gómez Zavala, Nelly Aburto

SANGEN Biotechnologie, Universidad Autónoma de México (UNAM)
Copilco Universidad, Alcaldía coyoacán, CP: 04360, CDMX, 5590220100,
juan30msc@gmail.com

The skin is the organ with the greatest weight and surface area of the human body and serves as the first line of defense against external agents and physical-chemical damage. Additionally, it's estimated that the skin hosts around 10^{12} microbial cells. The skin microbiota plays a crucial role in the proper functioning of the skin: it contributes to its development, regulates the immune system, protects against pathogens, and assists in metabolite degradation, among other functions. However, other factors also play important roles in skin function and health.

Up to 60% of the variation in skin aging among individuals of the same age is attributed to genetic factors, while the remaining 40% can be attributed to environmental factors, mainly exposure to solar radiation and tobacco consumption. Additionally, in cases of atopic dermatitis, changes in the skin's metabolomic profile have been detected: there was an increase in levels of histamine, urate, and serotonin.

Dermagenetics investigates the interrelationship between genes, skin health, and nutrition through SNPs related to collagen degradation, skin hydration, skin pigmentation, photo-aging, elimination of reactive oxygen species, degradation of environmental pollutants, and production of pro-inflammatory molecules. However, a more comprehensive analysis of an individual's dermatological profile should be conducted through an approach that encompasses genomic, transcriptomic, proteomic, metabolomic, microbiome composition, and specialized medical examination. This approach is known as skinomics.

The objective of this study is to develop a comprehensive panel of genomic, metabolomic, and microbiome markers related to skin health, aging, and predisposition to dermatological diseases. This panel, together with specialized medical advice, will guide the diagnosis and personalized treatment (both pharmacological and nutritional) of various skin conditions, as well as improve the health, anti-aging strategies, and aesthetics of this organ. A vast number of dermagenetic, metabolomic, and microbiome studies related to skin health and disease have been reviewed. Numerous SNPs, metabolomic profiles, and microbiome compositions associated with pathological conditions and aesthetic concerns have been identified. We are currently expanding our database and will later integrate it with artificial intelligence models.

EXPLORING SARS-COV-2 GENE EXPRESSION IN MEXICAN PATIENTS: INSIGHTS FROM MOLECULAR ANALYSIS

Joana Sánchez Medina¹, Leslie Sánchez Pérez¹, Laura Itzel Quintas Granados²,
Octavio Daniel Reyes Hernández¹, Gabriela Figueroa González¹, Edgar Yebrán Villegas Vázquez¹

¹Laboratorio de Farmacogenética, Unidad Multidisciplinaria de Investigación Experimental Zaragoza, Facultad de Estudios Superiores Zaragoza, Universidad Nacional Autónoma de México, Tel:5527373237. ²Universidad Autónoma de la Ciudad de México, Colegio de Ciencias y Humanidades, Doctor García Diego 168, Colonia Doctores, Alcaldía Cuauhtémoc, 06720. E-mail: eyebran.villegas@gmail.com

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is the cause of coronavirus disease (COVID-19), which originated in Wuhan, China, in December 2019 and was subsequently declared a pandemic in March 2020. In Mexico, 7,633,355 infections were reported, with 9.57% of patients hospitalized and 334,336 deaths. The disease presents with clinical manifestations ranging from mild to moderate and, in some cases, progresses to severe. To identify genetic disparities in individual responses to COVID-19 disease severity, apart from sex, age, and pre-existing health conditions, molecular analyses have been performed, identifying several genes implicated in the severity of this disease. Given the importance of the virus, several *in silico* analyses have also been conducted, such as sequence alignment using specialized software and information banks like GISAID (<https://gisaid.org/>), where virus sequences identified worldwide are registered. This study focused on patients from Mexico, covering the period from June 2021 to July 31, 2022, with an age range of 20-60 years. For the *in silico* analysis, 70 GISAID sequences were obtained and aligned using CLUSTALW software to identify similar regions between these sequences and their mutations, which give rise to the viral variants present during this period, such as the Delta and Omicron variants. Experimentally, 30 samples from Mexican patients in the Genomic Surveillance Consortium, without comorbidities, were analyzed to evaluate the expression of the Spike, ACE-2, TNF α , β -actin, and LZTFL1 genes using semi-quantitative and quantitative PCR techniques.

Additionally, a node analysis of the potential interaction between the genes of interest was performed. The results of multiple alignments show that, for the period from the second to the fourth wave of COVID-19 in Mexico, where the predominant variants were Omicron and Delta, the highest number of mutations were identified in the region of the S protein (Spike) followed by the N (nucleocapsid) protein. Experimentally, it was found that the expression of these genes varied in the patient samples analyzed and correlated with the severity of the infection.

Funding: PAPIIT-IA208422, PAPIIT-IA206724, PAPIIT-IN222321, and PAPIIT-IN221824 to Dirección General de Asuntos del Personal Académico (DGAPA).

Reference

1. Guo, Y.R. & et, al. The origin, transmission and clinical therapies on coronavirus disease 2019 (COVID-19) outbreak—an update on the status. *Military Med Res* 7 11 (2020).

DAMAGE HETEROGENEITY DURING INDUCTION OF HEPATOCARCINOGENESIS BY CHRONIC ADMINISTRATION OF DEN AND 2-AAF THROUGH 13- AND 18- WEEKS IN WISTAR RATS

Jaime Sánchez-Meza¹, Marina Campos-Valdez¹, José A. Domínguez-Rosales¹, Juliana Marisol Godínez-Rubi², Sarai C. Rodríguez-Reyes³, Erika Martínez-López³, Adriana M. Salazar-Montes¹, Carmen M. Gurrola Díaz¹, Manuel A. Castro-García¹, Guillermo M. Zúñiga-González⁴, Laura V. Sánchez Orozco¹

¹Instituto de Enfermedades Crónicas Degenerativas, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara, 44340 Guadalajara, Jalisco, México, ²Laboratorio de Patología Diagnóstica e Inmunohistoquímica, Departamento de Microbiología y Patología, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara, Guadalajara, Jalisco, México, ³Instituto de Nutrigenética y Nutrigenómica Traslacional, Centro Universitario de Ciencias de la Salud, Guadalajara, Jalisco, México, ⁴Laboratorio de Mutagénesis, Centro de Investigación Biomédica de Occidente, Instituto Mexicano del Seguro Social, Guadalajara, Jalisco, México.
E-mail: laura.sorozco@academicos.udg.mx; Phone number: +52 33 3954 5677

Introduction. Murine animal models of hepatocellular carcinoma (HCC) induction by administration of carcinogenic agents _such as diethylnitrosamine (DEN) and N-(2-Fluorenyl) acetamide (2-AAF)_ are regularly used to study liver cancer.

Objective. To assess the changes produced by the chronic administration of DEN and 2-AAF during 13- and 18-weeks (wks) in Wistar rats.

Materials and Methods. Groups of male Wistar rats (180-200 g) were organized: 1) Control 18 wks (18-wk Ctl), 2) Damage 18 wks (18-wk Dmg), 3) Control 13 wks (13-wk Ctl), and 4) Damage 13 wks (13-wk Dmg). Each week the damage groups were treated with i.p DEN (50 mg/Kg) and i.g. 2-AAF (25 mg/Kg) on day one and the third day during 13- and 18-wks, respectively. Afterward, the animals' livers and serum were collected for histological, biochemical, and gene expression analysis. GraphPad Prism version 8 was used for statistical analysis, with Mann-Whitney U test or t-test. A *p* value < 0.05 was considered significant.

Results. Dmg Tx decreased the survival; the 18-wk Dmg group survival was 62.5% (n=8) at the tenth wk, but when the 13-wk Dmg group was included, the survival percentage until the thirteenth week was 78.5% (n= 14). Dmg Tx decrease total weight, and changes in the liver tissue were observed _such as pale coloration, differentiated nodules, and hepatomegaly (to a lesser degree in the 13-wk Dmg group). There was heterogeneity in the damage severity among the animals of both groups, which was also found at the histological level were loss of normal hepatocyte architecture (lobular structure disorder), increase in atypical cells, and collagen accumulation were observed. In the 18-wk Dmg group, possible lung metastasis was found (indicated by macroscopic damage and histological alterations). Serum levels of ALT, AST, ALKP, GGT, and total proteins were significantly increased in both Dmg groups; and the expression of *CAT*, *SOD*, *COL1A*, and *TGFBI* were also significantly altered in these groups; additionally, *IL6* was significantly increased in the 18-wk Dmg group.

Conclusion. Dmg Tx during 13-wks in Wistar rats is enough to provoke important macroscopic, histological, and gene expression changes. Nevertheless, 18-wk Tx displays probable lung metastasis. The heterogeneity in damage degree found in this model may seem like a weakness; however, this could also be seen as an equivalence or representation of the heterogeneity of liver cancer among patients in real life. **Acknowledgments.** Grant PIN 2021 and P3E 271879-2023, CUCS, U. de G.

GLI1 EXPRESSION ASSOCIATED WITH OVERALL SURVIVAL IN LOCALLY ADVANCED CERVICAL CANCER

Gabriela Carolina Morales Sandoval, Jesús Sánchez Sánchez, Erick de la Cruz Hernández, Marcela Lizano Soberón, Alejandro Avilés-Salas, Adriana Contreras Paredes

Unidad de Investigación Biomédica en Cáncer, Instituto Nacional de Cancerología–Instituto de Investigaciones Biomédicas.
San Fernando 22, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México, CDMX (5554326933)
E-mail: adrycont@yahoo.com.mx

Cervical cancer (CC) is the fourth cause of death from cancer in women worldwide¹. The treatment for patients who have been diagnosed with stages locally advanced cancer (LACC) stage IIB through IVA) consists of a combination of cisplatin-based chemotherapy and radiation. However, the 5-year survival is only 66%². The options for patients with recurrent and metastatic disease are limited, and their median overall survival is <12 months². Identifying potential new targets is urgently required to develop novel therapeutic strategies. The up-activation of the hedgehog (Hh) pathway has been correlated with developing treatment resistance in several cancers³. We examined the associations between GLI1 levels, a transcription factor of the Hh pathway, and treatment response. GLI1 levels were determined using an Immunohistochemistry assay (IHQ) in 84 biopsy tissues with LACC diagnostic obtained from the Instituto Nacional de Cancerología pathology department in México City. Clinical and pathological parameters were collected from the file. Possible correlations were examined between GLI1 levels, treatment response, overall survival (OS), and disease-free survival (DFS). We found that Gli1 expression was frequently found in LACC samples (86.9%) and was located mainly in the nucleus in samples (83.33%). Nuclear GLI1 expression showed an association with overall survival (OS) ($P= 0.004$) and disease-free survival (DFS). Our data suggest that GLI protein could have a role in the response to treatment in patients with LACC undergoing concomitant chemotherapy and radiotherapy and may be a valid therapeutic target.

The present study was partially supported by CONAHCyT (grants no. CF-2019-263979).

References

1. Globocan 2020
2. Chemoradiotherapy for Cervical Cancer Meta-analysis Collaboration (CCCMAC).. Cochrane Database Syst Rev. 2010 Jan 20;2010(1): CD008285.
3. Kotulak-Chrzęszcz A, Kmieć Z, Wierzbicki PM. (2021) Int J Mol Med. 47(6):106

β3 SUBUNIT EXPRESSION IN NORMAL BREAST EPITHELIAL CELLS AND HIGHLY METASTATIC TRIPLE-NEGATIVE BREAST CANCER CELLS

Juan Manuel Sandoval Valverde, Linley Pammely Prado Celis, Rodrigo Zamora Cárdenas, Ricardo A. Navarro Polanco, Tania Ferrer Villada*

Centro Universitario de Investigaciones Biomédicas, Universidad de Colima, Colima, Col. México.

*Av 25 de Julio 965 Col. Villas San Sebastián, C.P. 28045 Colima, Colima, México. 312-3161000 ext 47504

E-mail: tania@uacol.mx

BK channels are widely distributed throughout the human body. The pore-forming alpha subunits are coupled with β subunits, which modulate their biophysical and pharmacological properties, and exhibit tissue-specific distribution patterns. Increased expression of these channels has been reported in various types of cancers, such as invasive ductal breast cancer and glioblastoma. Furthermore, the presence of $\beta 3$ subunits, not commonly found in healthy tissues, has been associated with a poorer prognosis in glioblastoma patients, suggesting their potential as a membrane marker for these cancer cells. However, unpublished previous results from our laboratory indicate that BK current density decreases in highly metastatic triple-negative breast cancer (TNBC) cells, although the accessory β subunit that modulates the functional properties of the channel and its cellular location have not been investigated. In this study, we observed in immunocytochemistry experiments and confocal microscopy that the $\beta 3$ subunit is expressed in both normal breast epithelial cells (MCF-10A cell line) and highly metastatic TNBC cells (HCC-1428 cell line), although, unlike other types of cancer, its expression is significantly reduced in the latter. Moreover, there is a higher expression of this β subunit in the plasma membrane of MCF-10A cells (specifically in the lamellipodia) compared to that of TNBC cells, suggesting that this protein is internalized in highly metastatic TNBC cells. Additionally, we also found that the $\beta 3$ subunit exhibits a significantly higher expression within the nucleus of highly metastatic TNBC cells compared to that of MCF-10A cells. This is the first report regarding the expression of the $\beta 3$ subunit in non-cancerous breast tissue. Moreover, the decrease in $\beta 3$ subunit expression in TNBC cells and its internalization into the nucleus suggest a need to reevaluate both the role of the BK channel and the expression of other accessory β subunits in cancer cells.

DISCOVERY OF POTENTIAL INHIBITORS OF DEHYDROQUINATE DEHYDRATASE FROM METHICILLIN-RESISTANT *STAPHYLOCOCCUS AUREUS* THROUGH COMPUTER-AIDED DRUG DESIGN

Ana Karina Segovia Parra, Alfredo Téllez Valencia, Jorge Cisneros Martínez, Claudia Avitia Domínguez

Facultad de Medicina y Nutrición. Universidad Juárez del Estado de Durango. Av. Universidad y Fanny Anitúa s/n.

C.P. 34,000. Durango, Dgo., México

E-mail: claudia.avitia@ujed.mx

Bacterial resistance to antibiotics is a concerning global health problem that is constantly evolving. New resistance mechanisms are frequently presented by bacteria to evade the mechanism of action of current drugs. *Staphylococcus aureus* is a pathogen considered a high priority by the World Health Organization. It causes a wide range of infections ranging from simple skin infections to life-threatening invasive infections. The main impact of *S. aureus* lies in strains of Methicillin-Resistant *Staphylococcus Aureus* (MRSA), term established to refer to all those strains resistant to the action of β -lactam antibiotics and their derivatives, as well as other groups of antibiotics not structurally related. Although there are drugs to combat infections caused by *S. aureus*, their effect is limited due to the constant appearance of strains resistant to them. The above generates the need to design new pharmaceutical alternatives through the search for molecules that act on a different molecular target. Under this perspective, an important metabolic pathway for MRSA is the shikimate pathway, used by bacteria, plants, fungi and apicomplexan parasites for the synthesis of aromatic compounds. The enzymes of this route have been considered good targets to design novel antibiotics. In this context, the fourth step of the route, the conversion of 3-dehydroquinate to shikimate, is performed by dehydroquinate dehydratase (DHQD). In this work, a computer-aided drug design strategy, which involved a consensus scoring virtual screening protocol, was applied to a chemical library of 73000 compounds from ZINC database. Firstly, molecules were filtered according to their toxicological and druglike characteristics. Thereafter, the 4791 compounds maintained were summited at three independent virtual screening assays using Autodock Vina, Vinardo, and Dock6 programs. The data showed that after applying consensus scoring protocol, the five compounds with the highest binding score were ZINC04529370, ZINC02635395, ZINC02635396, ZINC00006486 and ZINC00156780 with affinity energies of -1.324835447, -1.275157564, -1.272133293, -1.271542629 and -1.240770812 kcal/mol, respectively. Furthermore, these compounds made interactions with residues that are important for enzyme catalysis. Finally, their ADMETox predicted properties suggested that all of them supports the structural characteristics to be considered potential drug candidates. Therefore, these compounds can serve as a starting point in the search of new antibiotics.

DETECTION OF EMT MARKERS IN THE RENAL CARCINOGENIC PROCESS OF AN *IN VIVO* EXPERIMENTAL MODEL

José Dolores Solano-Becerra¹, Francisco A. Aguilar-Alonso¹, Telma O. Pariente-Pérez¹, Ignacio Pacheco-Bernal¹, Patricia Curiel-Muñiz¹, Jorge Luis Copado-Romero¹ and María Elena Ibarra-Rubio¹

¹Laboratorio 225, Departamento de Biología, Facultad de Química, Universidad Nacional Autónoma de México, 04510 CDMX, México.
e-mail: meir@unam.mx

Renal cell carcinoma (RCC) is the most common type of kidney cancer. The diagnosis of this carcinoma generally occurs at very advanced stages leading to a high mortality rate. Therefore, the search for RCC more specific markers at early stages can contribute to a timelier diagnosis. The experimental model of RCC induction in male Wistar rats by the exposition to ferric nitrilotriacetate (FeNTA) is a valuable tool to analyze the onset and promotion of this neoplasia *in vivo*, which was implemented in our group reaching a 90% of tumor incidence. Kidney cortex from early stages harvested after 1 and 2 months of FeNTA treatment, as well as the induced tumors, show a marked oxidative stress and alterations in NF- κ B¹ and MAPKs² signaling pathways.

On the other hand, recently, attention has intensified on the participation of epithelial-mesenchymal transition (EMT) process in various carcinomas. Particularly, the presence of EMT in patients with clear cell RCC, the most common subtype, has been related to a decreased survival. Thus, the presence of EMT markers at early stages of FeNTA-induced renal carcinogenesis was assessed, as well as in tumors and in a cell line derived from them.

An increase of the EMT markers vimentin and α -SMA was observed in renal cortex extracts after 1 and 2 months of FeNTA exposition (by WB), in the induced tumors (IHC) and in the cell line obtained (ICC). These results suggest that the EMT process is active since early stages of the FeNTA induced renal carcinogenesis and support the notion that it may participate in the cell transformation and tumor maintenance.

References

1. Francisco A. Aguilar-Alonso, Et al. (2015). Mol Cell Biochem, 404, 161-170.
2. Telma O. Pariente-Pérez, Et al. (2020). Oncology Letters, 19, 3153-3164.
3. Supported by: CONAHCYT. Fondo Sectorial de Investigación para la educación (ahora programa presupuestario F003) #284155; UNAM-DGAPA-PAPIIT #IN-228716 and FQ-PAIP 5000-9109

O-GLCNACYLATION DECREASES THE EXPRESSION OF ALPHA2-6 SIALIC ACID BY ACTIVATING THE PI3-KINASE/ AKT PATHWAY IN ORAL CAVITY SQUAMOUS CELL CANCER

Jocelyne del Rosario Calvo-Ramírez¹, Luis Enrique Ambrosio-Castillo¹, Ulises González González²,
María Cristina Franco-Arellanes², Saira Karina Ramírez Thomé¹, Risk Díaz Castillejos¹,
Taurino Amilcar Sosa-Velasco², Sandra Inés Garzón-Miguel², Beatriz Xóchitl Ávila Curiel¹,
Arturo Edgar Zenteno Galindo³, Carlos Josué Solórzano Mata^{1,2*}

Facultad de Odontología¹, Facultad de Medicina y Cirugía², Universidad Autónoma Benito Juárez de Oaxaca;
Facultad de Medicina³, Av. Universidad SN, Colonia Exhacienda de Cinco Señores, CP 68130, Oaxaca de Juárez,
Oaxaca. ³Departamento de Bioquímica, Universidad Nacional Autónoma de México, Av. Universidad 3000, UNAM, CU.,
Alcaldía Coyoacán, CP. 04510, Ciudad de México.
*email: universidad99@hotmail.com

Introduction: Oral cavity squamous cell cancer (OSCC) is the most common tumor in the head and neck region. In 2020, 377,714 cases of OSCC were reported worldwide and the Global Cancer Observatory (GCO) estimates that the incidence of OSCC will increase by approximately 40% by 2040. Changes in glycosylation and the PI3-kinase/pathway Akt are a common event in cellular transformation, specifically alterations in sialylation at alpha2-6 junctions have been documented in OSCC, however, it is unknown how O-GlcNAcylation can participate in the regulation of this type of sialylation by activating the PI3-kinase pathway. **Objective:** To analyze the changes in the expression of alpha2-6 sialic acid by O-GlcNAcylation by activating the PI3-kinase/Akt pathway in oral cavity squamous cell cancer cells. **Methodology:** SCC-25 cells (HPV negative) were cultured with treatments that promote O-GlcNAcylation and the PI3-kinase/Akt pathway was activated with Epidermal Growth Factor (EGF). By immunocytochemistry, the expression of alpha2-6 sialic acid was detected with the *Sambucus nigra* lectin (SNA) and O-GlcNAc with the RL2 antibody. **Results:** SCC-25 cells with EGF expressed alpha2-6 sialic acid with a high intensity, however, cells that additionally contained treatments that favor O-GlcNAcylation decreased the expression of alpha2-6 sialic acid. **Conclusions:** O-GlcNAcylation is a post-translational modification that participates in different phenomena during cellular transformation and tumor progression, however, the functional importance of the negative regulation of sialylation in alpha2-6 by activating the PI3-kinase/Akt pathway must be studied.

Keywords: Sialylation, O-GlcNAcylation, PI3-kinase/Akt, OSCC

IBERVILLEA SONORAE (WEREKE) ENHANCES GLUCOSE SIGNALING IN THE SKELETAL MUSCLE OF DIABETIC RATS

Vanessa Wendi Suarez Barrios^{1,2}, Ana Lilia Hernández Alba^{1,2}, Juan Carlos Cabrera Angeles^{1,2},
Estefania Valle Velasquez^{1,2}, Oscar René Zambrano Vásquez², José Guillermo Gonzaga Sánchez^{1,2},
Horacio Osorio Alonso², Laura Gabriela Sánchez Lozada², Juan Gabriel Juárez Rojas²,
Abraham Said Arellano Buendia²

¹Section of Postgraduate Studies and Research, Escuela Superior de Medicina-IPN, 11340 Mexico City, CDMX,

²Lab. Renal Physiopathology, Depto. de Nefrología, Instituto Nacional de Cardiología Ignacio Chávez, 14080 Mexico City, CDMX.

E-mail: suavane41@gmail.com Tel. 52 + (55-3706-0302)

Introduction. In diabetic patients, glucose signaling is disrupted in muscle tissue, leading to insulin resistance and contributing to hyperglycemia. This study aimed to evaluate the effect of an extract of *Ibervillea sonorae* (IS) on glycemia levels and GLUT4-mediated signaling in the skeletal muscle of diabetic rats.

Methodology. Male wistar rats (280-310g) were used and three experimental groups were formed: healthy (S), diabetes (D) (streptozotocin 50 mg/kg intraperitoneal) and diabetes treated with IS (400 mg/kg/day) (DIS). After confirming diabetes (blood glucose >200 mg/dL) the three experimental groups were maintained for 1 month, administration of IS started after confirming diabetes in the group to be treated "DIS". The three experimental groups were kept with water and food on demand throughout the study. Before the sacrifice, blood and urine were collected and muscle sample was extracted. Alterations produced by glucose were evaluated by plasma glucose concentrations and protein-level expression in skeletal muscle of Glut4, insulin-1 receptor substrate (IRS-1), insulin-2 receptor substrate (IRS-2) and evelsserine/threonine kinase (AKT).

Results. At the end of the study, group D showed low body weight (294 ± 14.66 vs 457.1 ± 12.51 gr), hyperglycemia (464 ± 12 vs 108.1 ± 3.7 mg/dL), increased diuresis (79 ± 3 ml/24h vs 16 ± 2) and increased water consumption (86 ± 2 vs 32 ± 5) compared with S. These results were associated with a decrease in Glut 4 expression, IRS-1, IRS-2 and AKT in skeletal muscle respect to D. Treatment with IS significantly reduced hyperglycemia (374 ± 26.7 mg/dL), lower diuresis (55.9 ± 9.2 ml/24h), water consumption (55.4 ± 9 ml/24h) compared to group D, but these values were higher than those recorded in the group of healthy animals. In addition, IS increased the expression of Glut 4, IRS-1, IRS-2 and AKT in skeletal muscle.

Conclusion. In diabetic rats, treatment with IS enhances glucose signaling in skeletal muscle. This effect may contribute to reduced glucose plasma levels.

ANTIOXIDANT EFFECT OF THE UNSAPONIFIABLE FRACTION OF AVOCADO OIL ON OXIDATIVE STRESS AND GLUCOSE METABOLISM IN ANIMAL MODEL OF NON-ALCOHOLIC FATTY LIVER DISEASE

Olin Torres-Isidro¹, Marcela González-Montoya¹, Manuel Alejandro Vargas-Vargas¹, Elizabeth Calderón-Cortés², Mauro Manuel Martínez-Pacheco¹, Christian Cortés-Rojo¹

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolas de Hidalgo (UMSNH). Morelia, Michoacan, México., ²Facultad de Enfermería, Universidad Michoacana de San Nicolas de Hidalgo (UMSNH).

Morelia, Michoacan, México.

E-mail: 1584209e@umich.mx

Oxidative stress is involved in a variety of human pathologies that including obesity, hypertension, type 2 diabetes and non-alcoholic fatty liver disease (NAFLD). These diseases are characterized by mitochondrial dysfunction and oxidative stress. The overproduction of reactive oxygen species (ROS) alter cellular metabolism, impairing glucose utilization¹. Non-saponifiable fraction of avocado oil (UFAO) contains several bioactive molecules with antioxidant properties, including β -sitosterol, stigmasterol and campesterol. The aim of this study was test if UFAO improves glucose utilization and mitochondrial oxidative stress in rats with NAFLD. NAFLD was induced with a high-fat, high-fructose diet for 12 weeks. Males Wistar rats were divided into four groups: 1) Control (CTRL), 2) NAFLD, 3) NAFLD + UFAO and 4) UFAO. Rats were fasted for 12 h for oral glucose tolerance test (OGTT). Before sacrifice, blood was recollected and serum was obtained by centrifugation. Liver mitochondria were isolated by differential centrifugation. Mitochondrial lipid peroxidation levels were determined spectrophotometrically. Compared to NAFLD group, the NAFLD + UFAO group presented lower levels of blood glucose and lower glucose tolerance. The UFAO group did tend to have decreased levels of lipid peroxidation compared to the CTRL group. These results suggest that the UFAO improves glucose metabolism in rats with NAFLD and that decreases oxidative stress in liver mitochondria.

Acknowledgments: to CONAHCyT for the postdoctoral fellowships to M.G.M. and M.A.V.V. This work was supported by Instituto de Ciencia, Tecnología e Innovación del Estado de Michoacan with the grants PICIR23-028 (M.G.M.) and PICIR23-063 (C.C.R), and Coordinación de Investigación Científica-UMSNH (7973911 to C.C.R.).

References

1. Behl, Tepan., et al (2023). *J. Clin. Med.*, 12, 2882.

INFLUENCE OF ECM STIFFNESS AND GEOMETRIC CONSTRAINTS ON THE MORPHOLOGY AND MECHANICAL RESPONSE OF LUNG FIBROBLASTS

Fernanda Toscano-Marquez¹, Marco Espina-Ordoñez², Ángeles García-Vicente², Nancy Paola-Puga³, Genaro Vázquez-Victorio³, José Cisneros¹ and Daniel Pérez-Calixto^{3,4*}

Instituto Nacional de Enfermedades Respiratorias "Ismael Cosío Villegas" Calzada de Tlalpan 4502, Belisario Domínguez Secc 16, Alcaldía Tlalpan, CDMX, México. C.P 14080. Posgrado Ciencias Biológicas UNAM. Unidad de Posgrado, Edificio D, 1er piso, Circuito de los Posgrados, Ciudad Universitaria, Alcaldía Coyoacán, C.P. 04510, Cd.Mx., México. Departamento de Física, Facultad de Ciencias UNAM. Av. Universidad 3000, Circuito Exterior s/n Ciudad Universitaria, Alcaldía Coyoacán, CDMX, México. CP 04510. Instituto Nacional de Medicina Genómica. Periférico Sur No. 4809, Col. Arenal Tepepan, Alcaldía Tlalpan, CDMX, México. C.P. 14061.
E-mail: Daniel_Perez@ciencias.unam.mx

Introduction. In vivo, cells are embedded in complex three-dimensional (3D) environments that exhibit various physical and topographical landscapes provided by the extracellular matrix (ECM). These features are pivotal in maintaining cellular shape and function within tissues. ECM disruptions, such as stiffness or topographical architecture variations, can impact migratory patterns and nuclear reorganization, leading to aberrant cell response. Since alterations in collective behavior are associated with pathological processes such as cancer metastasis and fibrosis, it is necessary to study and characterize topological and mechanical intrinsic properties of the cellular microenvironment, as well as the influence on the collective dynamics (long-range directional order, self-organization) of cells. **Methods.** To determine the effect of ECM stiffness and geometry on cell morphology and collective dynamics, lung fibroblasts were seeded on hydrogels (HG) of 1 and 23 kPa or alveoli-shaped structures of 16 kPa. Morphological shape, nuclear deformation, topological defects in collective cell organization, and α SMA and nuclear-cytoplasmic Yap levels were detected, visualized, and quantified by optical microscopy and immunofluorescence assays. **Results.** A heterogeneous mechanical response of the cell layer associated with the stiffness and geometric constraints of the substrates was observed. In 23 kPa HG, fibroblasts exhibit greater spreading, nuclear deformation, stress fibers increase, and nuclear translocation of YAP protein. Moreover, the collective behavior of the cells is associated with the formation of topological defects in these HGs that mimic a fibrotic lung. In structures that simulate alveoli, fibroblasts adhere to the external curvature, increase their spreading, and induce stress fibers and α SMA expression. **Conclusions.** These results could provide insight into understanding cellular response to the combined influence of several external stimuli in different pulmonary pathologies.

References

1. Yu, S.-M., et Al. (2018). Substrate curvature affects the shape, orientation, and polarization of renal epithelial cells. *Acta Biomaterialia*,

PHYSIOLOGICAL RESPONSE AND ORGANIC INTERACTIONS OF BERRYCACTUS IN WISTAR RATS WITH METABOLIC SYNDROME (MS)

Victoria Ramírez¹, Claudia J. Bautista², Cesaré Ovando-Vázquez^{3,4}, Joyce Trujillo^{4,5}

¹Departamento de Cirugía Experimental, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán (INCMNSZ),

Tlalpan, CDMX 14080, México, ²Departamento de Biología de la Reproducción, INCMNSZ, Tlalpan, CDMX 14080, México,

³Centro Nacional de Supercomputo, Instituto Potosino de Investigación Científica y Tecnológica (CNS-IPICYT), San Luis Potosí,

78216, México, ⁴Consejo Nacional de Humanidades, Ciencias y Tecnologías (CONAHCYT), ⁵División de Materiales Avanzados,

IPICYT (DMA), San Luis Potosí, S.L.P. 78216, México.

E-mail: daniela.trujillo@ipicyt.edu.mx

Background. Metabolic syndrome is a cluster of abnormalities affecting visceral adipose tissue, present in ~24% of the general population. MS involves genetic and environmental factors. Abdominal obesity, with visceral fat accumulation and excess of free fatty acids. Treating MS requires a polypharmacy approach, with nutraceutical biocompounds being explored as potential adjuncts. Berrycactus juice concentrate (ByC; *Myrtillocactus geometrizans*) contains polyphenols, pectins, sterols, and betalains with hypoglycemic, hypolipidemic, anti-inflammatory, and antiproliferative properties. **Aim.** This study investigates the impact of ByC consumption on metabolism response and pathway interactions in a rat model of MS induced by a high-fat diet. **Methods.** Twenty Wistar rats were divided into four groups: control (water), ByC (200 mg/kg), high-fat diet 45% (HFD), and HFD plus ByC. After 140 days, metabolic markers were analyzed in plasma, including glucose, cholesterol, triglycerides, insulin, leptin, ALT, AST, creatinine, and BUN levels, as well as fat content in liver and adipose tissue (FOLCH method), and percentage of fat in the liver by morphometry. **Results.** HFD groups presented significant increases in body weight, glycemia, insulinemia, triglyceridemia, cholesterolemia, leptinemia, and increased fat content in hepatic and adipose tissues. ByC in fed with HFD rats induced a significant decrease in triglycerides, cholesterol, insulin, and leptin levels without modifying glucose, liver enzymes, and renal function markers. **Conclusion.** ByC with an obesogenic diet for 20 weeks showed improvement by decreasing metabolic markers of MS. These results suggest that ByC contains biomolecules that may be useful as adjunct treatment of metabolic diseases like obesity and MS. Further research is needed to understand the mechanisms and identify the metabolite responsible for these effects.

PIEZO1-RHO/PMLC/IGF1 AXIS IN SKELETAL MUSCLE UNDER MICRONUTRIENTS EFFECTS IN AN AGING MODEL

Anya A Valdez-Hernandez^{1,2}, Frida I Barcenas-Robles^{2,3}, Israel Ramirez-Sanchez², Guillermo Ceballos², Gustavo Guevara-Balcazar², Ivan Rubio-Gayosso², Palma-Flores Carlos^{2,4}

¹Faculty of Medicine. National Autonomous University of Mexico, Mexico City. Mexico. ²Postgraduate Studies and Research Section, Higher School of Medicine. National Polytechnic Institute. Mexico City. Mexico. ³University of Guadalajara. Mexico.

⁴National Council for Science and Technology. CONAHCYT, Mexico City. Mexico. C.P. 11340
E-mail: cpalma@conahcyt.mx, Cell phone number (+52) 554 04 36 29

Aging is a gradual and continuous process, which manifests itself with a decrease in the capabilities of different organs and systems. In Mexico, it is estimated that by 2050 the percentage of older adults will rise from 12% to 22%, which would result in a public health problem. One of the problems of aging is the loss of skeletal muscle, a progressive decrease in the number and size of muscle fibers, which is known as sarcopenia (pathological aging), generating a decreased ability to perform daily activities. At the cellular level, one of the factors that trigger pathological aging is the decrease of muscle regeneration potential, which is mediated by activation of muscle stem cells (MSC). One of the factors that regulates this process is the Piezo1 ion channel, which regulates the proliferation of MSCs. Among the molecular pathways that activate Piezo1 are the Rho/pMLC and PDK4/IGF1 pathway. In this context, it has been observed that molecules derived from natural products and/or micronutrients have a beneficial effect on skeletal muscle, such as: Oleanolic Acid (OA) and Resveratrol (RESV). Results of this work demonstrated in vitro and in vivo models, an increase of Piezo 1 expression under AO and RESV treatment, which correlated with the increment of muscle regeneration potential. Our preliminary data are relevant because they show that the use of functional foods modify signaling pathways that activate degeneration process.

DESIGN OF A NOVEL MULTIPLEX QPCR SYSTEM TO DETECT PATHOGENIC MUCORALEAN FUNGI FROM CLINICAL HUMAN SAMPLES

Marco Iván Valle Maldonado^{1,2}, José Alberto Patiño Medina¹, Viridiana Alejandre Castañeda¹, Isabel Sinaí Meza Abarca³, Martha Isela Ramírez Díaz¹, Victor Meza Carmen^{*}

¹Laboratorio de Diferenciación Celular, Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Morelia, Michoacán, 58030, México. ²Laboratorio Estatal de Salud Pública de Michoacán, Secretaría de Salud de Michoacán, Morelia, Michoacán, 58270, México. ³Facultad de Químico Farmacobiología, Universidad Michoacana de San Nicolás de Hidalgo, Morelia, Michoacan, 58240, Mexico.

*E-mail: victor_meza2004@yahoo.com.mx

Mycoses represent a serious public health issue worldwide. Particularly, some species of the early diverging Mucorales fungi can lead to a rare but life-threatening infection called Mucormycosis mainly in immunocompromised patients. *Rhizopus arrhizus* is the most prevalent pathogenic Mucorales species. Furthermore, some species from the genera *Lichtheimia* and *Mucor* also have clinical importance. It has been proposed that the high mortality associated with this fungal infection could be related with a delayed treatment as a result of an inopportune diagnosis or misidentification. Several technical troubles have been reported to obstruct the microscopy identification or culture isolation of clinically relevant Mucorales fungi. In this work, we present the design of a novel non-invasive molecular method based on multiplex qPCR to simultaneously detect fungal DNA from three relevant pathogenic species from Mucoralean fungi (*Lichtheimia corymbifera*, *Mucor lusitanicus*, and *R. arrhizus*). Firstly, by employing bioinformatics, we identified *tfc-1* homologues for the Mucorales species, this gene encodes a subunit of the RNA polymerase III transcription initiation factor complex. We have previously validated this housekeeping gene as a normalizer gene for qPCR analysis in *M. lusitanicus*. Afterwards, we designed hydrolysis probes with specific fluorophores and oligonucleotides for each Mucorales species, incorporating the human ribonuclease P as an internal endogenous control. A multiplex qPCR system was tested against DNA extractions from Mucorales, and the results obtained in this work provide evidence of a novel, sensitive, and specific molecular system for detecting these fungi.

MODULATORY EFFECT OF AVOCADO OIL ON MITOCHONDRIAL PERMEABILITY TRANSITION PORE IN WISTAR RATS FED A HIGH-FAT AND HIGH-FRUCTOSE DIET

Manuel Alejandro Vargas-Vargas¹, Marcela González-Montoya¹, Olin Torres-Isidro¹, Rocío del Carmen Montoya-Pérez¹, Elizabeth Calderón-Cortés², Alain Raimundo Rodríguez Orozco³ and Christian Cortés-Rojo¹

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo (UMSNH). Morelia, Michoacán, México., ²Facultad de Enfermería, Universidad Michoacana de San Nicolás de Hidalgo (UMSNH). Morelia, Michoacán, México., ³Facultad de Ciencias Médicas y Biológicas, "Dr. Ignacio Chávez", Universidad Michoacana de San Nicolás de Hidalgo (UMSNH). Morelia, Michoacán, México.
E-mail: 1371614e@umich.com.mx, christian.cortes@umich.mx

Mitochondrial dysfunction plays a critical role in the pathogenesis of metabolic disorders associated with high-fat and high-fructose (HFHFr) diets¹. The mitochondrial permeability transition pore (PTPm) is a key regulator of mitochondrial function and cell survival². This study investigates the modulatory effects of avocado oil (AO) on PTPm in a rat model with a HFHFr diet. We evaluated the effect of AO on the activity of the PTPm in Wistar rats fed a HFHFr diet. Male Wistar rats were divided into four groups: 1) CTRL 2) HFFr 3) HFFr + AO and 4) AO. The experimental period lasted 12 weeks. Liver and kidney mitochondria were isolated by differential centrifugation. Cyclosporine A (2 μ M) was added to inhibit PTPm opening and 60 μ M CaCl₂ to open it. Fatty acid composition of avocado oil was assessed by gas chromatography-mass spectrometry. Avocado oil contained 59% oleic acid, 16.5% linoleic acid and 15.7% palmitic acid. The PTPm opening in the HFFr group was decreased compared to the other groups, indicating a potential loss in pore sensitivity to Ca²⁺, suggesting an alteration in PTPm structure. However, PTPm opening in the HFFr + AO group, was similar to that of the CTRL group, indicating that AO restores PTPm opening and probably, its structure. The results suggest that avocado oil has a modulatory effect on the PTPm in liver and kidney mitochondria of Wistar rats fed a HFHFr diet. The presence of bioactive compounds, like oleic acid and antioxidants, may contribute to this protective effect. These findings suggest that avocado oil could be beneficial in mitigating mitochondrial dysfunction associated with metabolic disorders.

Acknowledgments: to the Instituto de Ciencia Tecnología e Innovación of Michoacán State for the grants ICTI-PICIR23-063 (CCR) and ICTI-PICIR23-028 (MGM); to Coordinación de Investigación Científica – UMSNH, grant 7973911.

References

1. García-Berumen CL, Vargas-Vargas MA, Ortiz-Ávila O, Piña-Zentella RM, Ramos-Gómez M, Figueroa-García MdC, Mejía-Zepeda R, Rodríguez-Orozco AR, Saavedra-Molina A and Cortés-Rojo C (2022). *Front. Pharmacol.*, 13, 1089130.
2. Endlicher R, Drahotka Z, Štefková K, Červinková Z, Kučera O. (2023). *Cells*. 12,1273.

THE UNSAPONIFIABLE FRACTION OF AVOCADO OIL IMPROVES MITOCHONDRIAL FUNCTION IN RATS WITH NON-ALCOHOLIC FATTY LIVER DISEASE

Manuel Alejandro Vargas-Vargas¹, Marcela González-Montoya¹, Olin Torres-Isidro¹, Alfredo Saavedra Molina¹, María Guadalupe Cuiniche-Mendez¹, Julio César Ontiveros-Rodríguez¹ and Christian Cortés-Rojo¹

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo (UMSNH).
Morelia, Michoacán, México. Morelia, Michoacán, México.

E-mail: marceglezmon@gmail.com, christian.cortes@umich.mx

The Western diet contains high levels of saturated fats and carbohydrates. This diet has been associated with increased prevalence of chronic degenerative diseases, including obesity, diabetes, hypertension, and non-alcoholic fatty liver disease (NAFLD). Two main features of NAFLD are inflammation and mitochondrial dysfunction. Reactive oxygen species (ROS) produced in the mitochondrial electron transport chain (ETC) promotes the progression from steatosis to steatohepatitis by stimulating the production of proinflammatory cytokines¹. Avocado oil contains oleic acid as its main fatty acid and a myriad of bioactive molecules in its unsaponifiable fraction, many of them with biological activity in mitochondria. In this study, we extracted the unsaponifiable fraction of a commercial presentation of avocado oil (UFAO) and characterized it using GC-MS/MS and ESI. 100 mg/kg body weight of UFAO was given to rats fed a high-fat and high-carbohydrate (HFHC) diet for 12 weeks to induce NAFLD. Four experimental groups were established: a control group fed a standard diet (CTRL), an HFHC diet group as the NAFLD model, a standard diet group administered with UFAO (UFAO group), and an HFHC diet group plus UFAO (NAFLD+UFAO). Mitochondria were isolated from liver and kidney, and it was determined the rate of respiration in states 3 and 4, mitochondrial membrane potential ($\Delta\psi$), and activities of the complexes I and III, the main sites of ROS production in the ETC. In both liver and kidney mitochondria, a decrease in state 3 respiration was observed in the NAFLD group compared to the CTRL group, while an increase in state 3 was seen in both the UFAO and NAFLD+UFAO groups. The activities of the complexes I and III decreased in the NAFLD group compared to the CTRL group. In contrast, the activities increased in the UFAO and NAFLD+UFAO groups. In conclusion, the UFAO improved mitochondrial function in liver and kidney mitochondria from rats with NAFLD, suggesting that UFAO may be a source of molecules for the treatment of NAFLD.

Acknowledgments. to CONAHCyT for the postdoctoral fellowships to M.G.M. and M.A. V.V. This work was supported by Instituto de Ciencia, Tecnología e Innovación del Estado de Michoacan with the grants PICIR23-028 (M.G.M.) and PICIR23-063 (C.C.R), and Coordinación de Investigación Científica-UMSNH (7973911 to C.C.R.).

FISETIN AND REGULAR AEROBIC EXERCISE ENHANCE GLUTATHIONE REDOX STATUS IN THE BRAIN OF RATS WITH DIABETES

Gabriel Vazquez Arredondo¹, Nora Camila Franco Guerra¹, Karla Susana Vera Delgado²,
Montserrat Guadalupe Vera Delgado², Luis Alberto Sánchez Briones², Sergio Márquez Gamiño²,
Elizabeth Sánchez Duarte²

¹Licenciatura en Médico Cirujano, Departamento de Medicina y Nutrición, Universidad de Guanajuato, Campus León. Blvd. Puente Milenio 1001 Fracción del Predio San Carlos, C.P. 37670 León, Guanajuato, México. ²Departamento de Ciencias Aplicadas al Trabajo, Universidad de Guanajuato Campus León, Eugenio Garza Sada #572, Lomas del Campestre Sección 2, C.P. 37150, León, Guanajuato, México.
E-mail: g.vazquezarredondo@ugto.mx

Aerobic exercise is crucial for the treatment and prevention of oxidative stress (OS) related disorders like diabetes and the resulting complications. Moreover, fisetin is a potent cytoprotective agent promoting antioxidant, anti-inflammatory effects and protects the cells against OS in *in vivo* and *in vitro* experimental models. Thus, the purpose of this study was to assess the impact of fisetin and aerobic exercise training and the combination from both on the glutathione redox status in brain of diabetic rats. Male Wistar rats (200–220 g) were randomly divided into five groups (n=6), among them: Non-Diabetic Control (ND), diabetic control (D), diabetic fisetin (DF), diabetic exercise-trained (DE), diabetic exercise-trained+Fisetin (DEF). Diabetes was induced by a single streptozotocin injection (65 mg/kg body weight), animals with fasting blood glucose levels ≥ 250 mg/dL were considered as diabetic. Training comprised 8 weeks of treadmill running (30 minutes daily, 5 days/week). Treatment with fisetin (2.5 mg/kg/day) was administered for 8 weeks. At the end of the intervention, brain tissue was collected for the determination of the concentrations of reduced glutathione (GSH), glutathione disulfide (GSSG) and the relation GSH/GSSG. Treatment with fisetin and aerobic exercise exerted remarkable effects on hyperglycemic status in diabetic rats by decreases FBG levels ($P > 0.05$), and inducing significant increases on GSH levels and an elevated GSH/GSSG ratio in the DE, DF and DEF groups with respect to the D group. The highest ratio was observed in groups DEF and the lowest in the DF group compared to D group. These findings suggest that combination of fisetin with exercise training might offer additional antioxidant effects by modulating glutathione redox state in brain of diabetic rats.

CHARACTERIZATION OF BENZIMIDAZOLE DERIVATIVES AS INHIBITORS OF SRC HOMOLOGY 2 DOMAIN-CONTAINING PROTEIN TYROSINE PHOSPHATASE 1

Héctor Abdel Vázquez Lechuga¹, Claudia Isela Avitia Domínguez¹, Jorge Arturo Cisneros Martínez¹, Jesús Oria Hernández², Adriana Castillo Villanueva², Alfredo Téllez Valencia¹

¹Facultad de Medicina y Nutrición de la Universidad Juárez del Estado de Durango, Av. Universidad y Fanny Anitúa s/n. C.P. 34,000. Durango, Dgo., México. ²Instituto Nacional de Pediatría, Insurgentes Sur 3700, Letra C, Av. Insurgentes Sur, Insurgentes Cuicuilco, Coyoacán, 04530, Ciudad de México. E-mail: atellez@ujed.mx

Cancer is a major public health and economic problem in the 21st century. The current global statistics for the year 2022 indicated that there were almost 20 million of new cases and close to 10 million deaths. Nowadays, treatments are not completely satisfactory because of adverse effects. Therefore, there is an urgent necessity of new alternatives for therapy. An approach to achieve this, is to find inhibitors of enzymes that play an important role in the development of the disease. In this context, Src homology region 2 domain-containing phosphatase 1 (SHP1) is a non-receptor tyrosine phosphatase involved in cell cycle control, cancer cell migration and invasion, and apoptosis induction, also is associated with either an increased or decreased survival rate depending on the cancer type. Therefore, it is considered an interesting target for anticancer drug design. With the aim to report SHP1 inhibitors, an in-house chemical library of benzimidazole derivatives was tested against SHP1 activity. Compounds **LTCN6**, **LTCN7**, and **LTCN8** significantly inhibited SHP1 activity at a concentration of 30 μ M, with inhibition percentages of 95%, 86%, and 92%, respectively. A blind docking protocol was applied to these compounds using AutoDock Vina, the results showed that **LTCN6** interacts with SHP1 through hydrogen bonds on Q440 and N317 with an affinity score of -8.16 kcal/mol, whilst **LTCN8** made halogen bonds with D315 and N437 with an affinity score of -8.57 kcal/mol, and **LTCN7** had an affinity score of -8.5 kcal/mol but did not exhibit specific protein-ligand interactions. The ADMETox predictions obtained using DataWarrior and SwissADME suggested that the three compounds could be considered potential drug candidates and can serve as hits for future drug development.

EVALUATION OF PLATELET FUNCTION IN WOMEN POST-CHEMOTHERAPY FOR BREAST CANCER: A PILOT STUDY

Itzel Patricia Vásquez Martínez^{1,*}, Joscelin Macías Ríos², Ismael Antonio García², Eduardo Pérez Campos³, Carlos A. Matias Cervantes⁴, Laura Pérez-Campos Mayoral¹, María Teresa Hernández Huerta^{4,*}

¹Centro de Investigación Facultad de Medicina UNAM-UABJO, C.P. 68020. ²Hospital General Aurelio Valdivieso (HGAV), 68040, Oaxaca. ³Tecnológico Nacional de México/ IT de Oaxaca. 68030, Oaxaca. ⁴CONAHCYT Facultad de Medicina y Cirugía, Universidad Autónoma "Benito Juárez" de Oaxaca, 68020, Oaxaca.

*Contacto: vami940508.fmc@uabjo.mx; mthernandez@conahcyt.mx

Currently, with the efficient response of breast cancer (BC) patients to treatments, interest in research into the quality of life of post-treatment survivors has increased. BC is characterized by dysregulation of the hemostatic system, in which platelets play a fundamental role in thrombosis and hemostasis^{1,2}. However, there are few reports on post-treatment platelet function. Our objective was to evaluate platelet function in post-chemotherapy women for BC. For this, 10 blood samples were collected from women with BC between 36 and 74 years from the HGAV under approval of the institutional ethics committee. Prothrombin (PT) and activated partial thromboplastin (APT) times, fibrinogen, D-dimer, platelet number and mean platelet volume (MPV) were determined. In addition, platelet aggregation assays were performed with platelet-rich plasma using adenosine diphosphate (ADP) (0.58, 1.17, 2.34 μM) and in platelets we used thrombin (1U). Finally, intracellular calcium concentration $[\text{Ca}^{2+}]_i$ in platelets was quantified with Fluo-3 AM using spectrofluorometry³. Results are presented below with respect to the mean and standard deviation of each parameter: 239 000 \pm 72000 platelets/ μL , PTT of 26.03 \pm 9 s, PT of 13.06 \pm 0.6 s, fibrinogen 368.6 \pm 62 mg/dL, D-dimer, 789 \pm 1091 ng/mL, and mean platelet volume of 10.56 \pm 1.46 fL. In platelet aggregation, the mean percentage with ADP 0.58, 1.17, and 2.34 μM was 33, 40, and 48%, respectively, whereas with thrombin it was 28%. And surprising, the $[\text{Ca}^{2+}]_i$ was 0 nM in all patients. In conclusion, our results suggest alterations at the post-chemotherapy platelet level for BC, even when the number of platelets is normal, morphologically, they are observed activated and smaller, resulting in loss of platelet calcium and elevation of MPV and D-dimer. Therefore, these parameters could be used to evaluate the possible residual effects of chemotherapy and susceptibilities of survivors to this or other diseases.

References

1. F, Ay C. (2022). Hemostasis and cancer: Impact of haemostatic biomarkers for the prediction of clinical outcomes in patients with cancer. *J Thromb Haemost*, **20**(12):2733-45.
2. Giannakeas V, Kotsopoulos J, Brooks JD, Cheung MC, Rosella L, Lipscombe L, et al (2022). Platelet Count and Survival after Cancer. *Cancers*, **14**(3):549.
3. Grynkiewicz G, Poenie M, Tsien RY. (1985). A new generation of Ca^{2+} indicators with greatly improved fluorescence properties. *J Biol Chem*, **25**;260(6):3440-50.

ANALYSIS OF POTENTIAL DRUGS WITH LEPTIN-BINDING CAPACITY

Brandon Jorge Romero Rodríguez¹, José Antonio Velázquez Domínguez¹
y Claudia Guadalupe Benítez Cardoza¹

¹Escuela Nacional de Medicina y Homeopatía, Av. Guillermo Massieu Helguera, No 239, Col. La Escalera,
C.P. 07320. CDMX, México.

E-mail: jvelazquezd@ipn.mx, cbenitez@ipn.mx

Introduction. The World Health Organization (WHO) defines obesity as an increased and abnormal accumulation of body fat; it is of multifactorial etiology, being a predisposing factor to the development of metabolic diseases. There is a close relationship between the increase in adipose tissue and the secretion of leptin, an adipokine responsible for energy balance and appetite inhibition. **Objective.** To analyze the effect of some drug-like compounds, previously selected by molecular docking, depicted here as AJB1, AJB2, AJB3 and AJB4, on the structure of human leptin W100E. **Materials and methods.** Human leptin W100E, was prepared as described before (Doi:10.3844/ajabssp.2014). The compounds were resuspended in DMSO, following the manufacturer's instructions. Leptin W100E was incubated in the presence of a final concentration of 25.08 mM, of each compound, at 25 °C, hided from light, while fluorescence-emission spectra were recorded every hour during 24 hours. The excitation wavelength were 280 nm and 295 nm, and emission was collected from 300 to 450 nm, using 1 cm path-length quartz cells. **Results.** Two compounds (AJB2 and AJB4) importantly modify the conformation of human leptin W100E, as judged from fluorescence emission signal. While AJB1 and AJB3, do not have any important effect on the conformation of the protein. **Conclusion.** AJB2 and AJB4 bind human leptin W100E, and can be considered as possible inhibitors of the actions of leptin, and should be further analyzed as possible new treatments of pathologies associated with hyperleptinemia.

NICOTINAMIDE DECREASES PROTEIN OXIDATION IN THE RETINA, RPE, AND LIVER IN A HYPERGLYCEMIC MODEL

J. David Villeda-González¹, Lizue B. Mancilla-Infante¹, Gustavo Sánchez-Chávez¹, Rocío Salceda-Sacanelles¹

¹Instituto de Fisiología Celular-División de Neurociencias, Universidad Nacional Autónoma de México, Ciudad Universitaria, Ciudad de México.

E-mail: jvilleda@ifc.unam.mx

Introduction. Diabetes is a multifactorial disease that is characterized by hyperglycemia. This condition contributes directly to the complications observed in various organs and tissues leading to retinopathy, nephropathy, and neuropathy. There is evidence that oxidative stress plays an important role in the development of these diseases. Coincidentally, it has been observed that increase in oxidative stress is accompanied by a decrease in the cofactors NAD⁺ and NADP⁺, which are important in the cellular redox balance, helping in the elimination of reactive oxygen species.

The objective of the study was to determine the levels of oxidized proteins in the retina, retinal pigment epithelium (RPE), and liver of diabetic rats with and without an additional intake of nicotinamide (NAM), a precursor of the cofactors NAD⁺ and NADP⁺.

Methodology. *Long Evans* rats of 200 g. Hyperglycemia was induced by streptozotocin administration (STZ, 98mg/kg i.p). Rats were distributed into four groups: 1) control (Ctrl), NAM controls (NAM), hyperglycemic (STZ), and hyperglycemic with NAM (STZ-NAM). NAM was administrated in the drinking water in a concentration of 10mM for 5 h/5 week days. Rats were considered diabetic if glucose was ≥ 250 mg/dL. Rats were sacrificed after 20 and 45 days and retina, retinal pigment epithelium (RPE) and liver extracted for the analysis of oxidized proteins, and glutathione levels.

Results. Levels of blood glucose were considerable increased by STZ, as previously reported. NAM treatment did not influence glucose levels, polydipsia or polyuria. At 45 days, weight gain was observed in the STZ-NAM group compared to the STZ, indicating an improvement in the physiological state of the rats treated with NAM.

Levels of carbonylated proteins significantly increased in the retina, RPE, and liver from hyperglycemic rats. NAM produce a decrease in the levels of oxidized proteins in retina, liver, and in a less extense in RPE. On the other hand levels of GSH were 30% reduced in the liver of diabetic rats, which were not changed by NAM. GSH levels in retina and RPE were similar in all conditions studied.

Conclusion. The results indicate increase in carbonylated proteins at early onset of hyperglycemia in all tissues studied, which might lead to tissue damage. In the liver, levels of GSH decreased in the diabetic rats, which together with the increase in carbonylated protein levels suggests the occurrence of oxidative stress, which was partially recovered by treatment with NAM.

Acknowledgments. A PAPIIT/UNAM proyect IN-204523, and CONAHCYT by the postdoctoral support through the assigned scholarship: CVU 634838.

References

1. Albert-Garay JS, Riesgo-Escovar JR, Salceda R. (2022). High glucose concentrations induce oxidative stress by inhibiting Nrf2 expression in rat Müller retinal cells in vitro. *Sci Rep.* 24;12(1):1261.
2. Villeda-González JD, Gómez-Olivares JL, Baiza-Gutman LA, Manuel-Apolinar L, Damasio-Santana L, Millán-Pacheco C, Ángeles-Mejía S, Cortés-Ginez MC, Cruz-López M, Vidal-Moreno CJ, Díaz-Flores M. (2020). Nicotinamide reduces inflammation and oxidative stress via the cholinergic system in fructose-induced metabolic syndrome in rats. *Life Sci.* 1;250:117585.

HEPATOPROTECTIVE EFFECT OF EMPAGLIFLOZIN/METFORMIN CO-TREATMENT IN METABOLIC SYNDROME

Zambrano-Vásquez Oscar R.^{1,2*}, Juan C. Cabrera-Ángeles^{1,3}, Hernández-Alba Ana L.^{1,3},
Fernando Cortes-Camacho^{1,2}, Jorge I. Castañeda-Sánchez⁴, Elena Aréchaga-Ocampo⁵, Valle-Velázquez Estefania¹,
A. Said Arellano-Buendía¹, Gonzaga-Sánchez José G.¹, Sánchez-Lozada L. Gabriela¹, Osorio-Alonso Horacio¹

¹Depto. de Fisiopatología Cardio-Renal, Instituto Nacional de Cardiología Ignacio Chávez, Mexico City, Mexico. ²Doctorado en Ciencias Biológicas y de la Salud, Universidad Autónoma Metropolitana Xochimilco, Mexico. ³Sección de Estudios de Posgrado e Investigación, Escuela Superior de Medicina-IPN, México City, México. ⁴Depto. de Sistemas Biológicos, Universidad Autónoma Metropolitana Xochimilco, Mexico City, Mexico. ⁵Depto. de Ciencias Naturales, Universidad Autónoma Metropolitana Cuajimalpa, Mexico City, Mexico.
renezambrano2513@gmail.com*

Background. Metabolic syndrome (MS) is a group of risk factors (RF) that favors fat deposits in the liver, due to non-alcoholic factors, and could cause steatohepatitis, fibrosis, and cirrhosis, alterations grouped in non-alcoholic fatty liver disease (NAFLD), a disease closely related to MS. Treatment of NAFLD is based on controlling RF through changes in lifestyle and drugs, but there is still no approved pharmacotherapy. Empagliflozin and metformin reduce obesity and insulin resistance, suggesting that could be useful as therapy in NAFLD.

Aim. To evaluate the effect of Empagliflozin/Metformin co-treatment in NAFLD.

Methodology. MS was induced in male Wistar rats (200-220g) with a type Paygen diet. Once established MS, rats were randomly assigned to MS untreated, MS + Empagliflozin/Metformin (12.5/850 mg/day gavage), Empagliflozin, and Metformin. Body weight, blood pressure (BP), fasting blood glucose, glucose tolerance test (GTT), and lipid profile were assessed as MS markers. Also, aspartate aminotransferase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP), gamma glutamiltransferase (GGT), and the histological analysis were quantified.

Results. The Paygen diet increase blood pressure, fasting glucose and impaired the GTT and the lipid profile. Also, the liver transaminases were increased. The results demonstrated establishment of MS and liver damage. The co-therapy showed improvement in BP, fasting glucose, GTT, and lipid profile compared to the MS group. The liver changes in AST, ALT, ALP, GGT, MS-induced were prevented with the co-therapy. The empagliflozin/metformin co-therapy did not restore the value of the parameters assessed respect to the control group, but were statistically different when compared with MS-untreated.

Conclusion. The Empagliflozin/Metformin co-therapy was better than each one treatment for the control of risk factors of MS, and therefore offers an hepatoprotective role, suggesting that may delay the progression to NAFLD and could be considered as a therapeutic option in NAFLD induced by MS.

THE IMPORTANCE OF NUTRITION IN ALZHEIMER'S DISEASE

Oswaldo Bivian Paredes¹, Andrés Salas Casas², Ana Hilda Figueroa Gutiérrez³

Universidad Autónoma del Estado de Hidalgo, Instituto de Ciencias de la Salud, ¹Área Académica de Nutrición, ²Área Académica de Gerontología, ³Área Académica de Medicina, Circuito Ex Hacienda la Concepción s/n Carretera, Pachuca Actopan, San Agustín Tlaxiaca, Hidalgo. C.P. 42160. Tel. 7717082324.
Correo: ana_figueroa3494@uaeh.edu.mx

Alzheimer's disease (AD) is a neurodegenerative and incurable disease, being the major cause of dementia. Its pathology is not yet fully understood, but two main features are recognized: The formation of extracellular β -Amyloid plaques and hyperphosphorylation of tau protein at the neuronal level¹. There are more than 50 million people with dementia worldwide and it is expected that there will be 65 million by 2030 and 115 million by 2050². In Mexico it is estimated that there were 800 000 cases of dementia in 2019, of which 80% were AD³.

The aim of this work is to show the importance of nutrition in AD. Among the nutrients with an impact on AD is docosahexaenoic acid with cytoprotective and neuroprotective properties. Vitamin A elevates α -secretase expression; vitamins B6, B12 and folic acid regulate DNA methylation and homocysteine metabolism; vitamin C acts as an antioxidant agent; vitamin D regulates calcium homeostasis and is characterized as anti-inflammatory and antioxidant; vitamin E prevents fatty acid peroxidation and tau phosphorylation. Polyphenols have antioxidant activity⁴.

Interesting advances are currently being made in the gut-brain axis; it connects the brain with the gastrointestinal tract, in this way the microbiota sends signals by means of substances synthesized by different microorganisms such as dopamine, serotonin and short chain fatty acids⁵.

We are using molecular docking, to study and analyze the interaction of the tau protein and the main compounds found in the diet, in this way we will determine which of them could be used to reduce cognitive impairment in people with AD.

Interestingly, curcumin, the vitamin D found in fish, showed an interaction with tau, decreasing phosphorylation and inflammation, in addition, catechins found in green tea, showed an interaction with tau. These compounds found in different foods could help slow down and aid in the treatment of AD.

In conclusion, it is important to look for new and more compounds found in the common diet that help in the treatment of AD. Curcuma, vitamin D and catechins are compounds that could be an alternative for the treatment of AD.

References

1. Monteiro, A. R., Barbosa, D. J., Remião, F., & Silva, R (2023). *Biochemical Pharmacology*, 211, 115522.
2. Prince, M., Bryce, R., Albanese, E., Wimo, A., Ribeiro, W., & Ferri, C. P (2013). *Alzheimer's & Dementia*, 9, 63-75.
3. <https://www.insp.mx/avisos/21-de-septiembre-dia-mundial-del-alzheimer#sup4>
4. Guarnieri, L., Bosco, F., Leo, A., Citraro, R., Palma, E., De Sarro, G., & Mollace, V (2024). *Ageing Research Reviews*, 95, 102210.
5. Singh, H., Chopra, C., Singh, H., Malgotra, V., Khurshid Wani, A., Singh Dhanjal, D., Sharma, I., Nepovimova, E., Alomar, S., Singh, R., Sharma, V., & Kuca, K (2024). *Journal of Functional Foods*, 112, 105915.

CHANGES IN OVARIAN MORPHOLOGY AND ULTRASTRUCTURE IN OFFSPRING OF MICE WITH POLYCYSTIC OVARY SYNDROME

Paola del Valle¹, Emilio Espinoza², David Barrera³, Rosario Ortiz¹, Rosa López-Castillero¹, Erick López-Cruz¹,
Silvia Juárez¹, Luisa Escobar¹, Olga Echeverría¹, Nayeli Torres-Ramírez¹

¹Departamento de Biología Celular, Facultad de Ciencias, UNAM. ²Departamento de Bioquímica y Biología Estructural, Instituto de Fisiología Celular, UNAM. ³Departamento de Biología de la Reproducción, INCMNSZ.
paoladelvalle@ciencias.unam.mx; nayeli.torres@ciencias.unam.mx

Mitochondria are the central organelle in energy production, but they are also the main source of reactive oxygen species that cause mitochondrial dysfunction. This dysfunction is related to various diseases, such as polycystic ovary syndrome (PCOS), characterized by hyperandrogenism, polycystic ovary morphology and oligo- or anovulation. Women with PCOS as well as animal models present mitochondrial alterations in membrane dynamics, biogenesis, and potential. Since mitochondria are maternally inherited, the aim is to evaluate reproductive and morphological mitochondrial changes in the ovaries of the offspring of mice with PCOS. For this reason, a PCOS model was created using 25-day-old Balb/c mice treated subcutaneously with dehydroepiandrosterone for 20 days. The mice were mated with healthy males and adult offspring were employed. The estrous cycle was evaluated for two weeks. During this time, the pups in the PCOS group remained in the diestrus stage, while the control group presented a normal estrous cycle. Besides, ovaries were obtained and processed for optical and transmission electron microscopy. Morphological analysis in paraffin sections stained with hematoxylin-eosin showed a decrease in primordial and primary follicles, but a greater number of secondary and antral follicles, along with higher number of atretic follicles in the offspring of PCOS group. Additionally, no healthy oocytes were observed. Ultrathin sections were stained with uranyl acetate and lead citrate. Ultrastructural characteristics of granulosa cells (GC) in the offspring of PCOS group included endoplasmic reticulum, Golgi apparatus, and mitochondrial swelling, in addition to loss of mitochondrial cristae. The oocytes of the PCOS group showed highly vacuolated mitochondria. In conclusion, maternal PCOS is associated with morphological and ultrastructural alterations in GC and oocytes in the offspring associated with loss of estrous cycle.

Reference

1. Wang F, Yu, B, Yang, W, Liu J, Lu J., Xia X. (2012). *Journal of Ovarian Research*, 5(1)

INHIBITION OF THE EARLY DEVELOPMENT OF FIBROSIS IN CHRONIC KIDNEY DISEASE

Juan Carlos Gallardo-Pérez

Departamento de Fisiopatología Cardio-Renal, Instituto Nacional de Cardiología, México.
Juan Badiano No.1 Col. Sección XVI, Tlalpan, Ciudad de México.
E-mail: carlos.gallardo@cardiologia.org.mx

In Mexico, the prevalence of kidney disease is 12.2% and has a rate of 51 deaths per 100,000 inhabitants. Chronic kidney disease (CKD) is the result of various chronic-degenerative diseases such as diabetes mellitus and high blood pressure. A particularity of CKD is the apparition, starting from the proximal tubule, of renal fibrosis (fibrogenesis), which is characterized by an excessive deposition of extracellular matrix (pathological matrix), rich in collagen, fibronectin, laminin, heparan, among others components, and is located in the interstitial space between tubules and tubular capillaries. The main mechanism by which proximal tubule damage is transferred to the interstitium, culminating in fibrosis, is mediated by epithelial-mesenchymal transition (EMT). The above implies that, due to EMT, tubular cells undergo transition towards myofibroblasts and these are responsible for the deposition of pathological matrix. There are compounds of plant origin, non-toxic, such as erythrose (rhubarb) and sulforaphane (broccoli), capable of preventing EMT in cancer; however, these have not been evaluated in the fibrotic process of CKD.

Aim: Inhibit the early development of fibrosis in kidney tubular cells using compounds of plant origin.

Therefore, LLC-PK1 cells (proximal tubule) were subjected to an EMT induction protocol modified¹ using a combination of hypoxia, hypoglycemia and lipopolysaccharides (LPS) in the culture medium, for 12 hours. Subsequently, the surviving cells were evaluated for their morphology, EMT and myofibroblast markers, oxidative stress, inflammation, migration and invasion. Likewise, the effect of the combination of erythrose and sulforaphane on these parameters was evaluated.

Results: The EMT-induced cells changed their morphology to mesenchymal, their mitochondria were elongated, the expression of alpha-SMA increased almost 6-fold, as well as a 2- to 14-fold of increase in SNAIL, Twist, vimentin, Col-1a, fibronectin, cytokeratin, desmin (markers of myofibroblasts and EMT) and a decrease in E-cadherin in comparison with the control. The processes of migration, invasion and release of extracellular cytokines such as IL-6, IL-8, HPI/AMF and TGF-beta were increased. The combination of erythrose and sulforaphane, at nanomolar concentrations before applying the EMT induction protocol, decreased all these parameters. E-cadherin, a marker of epithelia, as well as the generation of ROS, were restored to levels similar to the control indicating a protective effect of plant compounds on the onset of renal fibrogenesis.

Reference

1. *J Med Oncol.* 2023,40(3):104

ALTERATIONS IN THE FUNCTIONING OF THE HEART INDUCED BY CHANGES IN ENERGY METABOLISM DUE TO TUMORAL GROWTH

Luz Hernández-Esquivel¹, Francisco Correa², José Salud Rodríguez-Zavala¹, Álvaro Marín Hernández¹

Departamento de Bioquímica¹, Departamento de Biomedicina Cardiovascular², Instituto Nacional de Cardiología “Ignacio Chávez”. Juan Badiano #1, Col. Sección XVI Tlalpan 14080, México.
luz_hdzes@yahoo.com.mx

Significant chemotherapy side effects in the treatment of cancer include cardiotoxicity; aside from these deleterious effects, cancer *per se* may affect heart function. Cardiac muscle wasting is associated with cancer cachexia, apparently induced by inflammatory cytokines, that produces cardiac alterations (left ventricular dysfunction, fibrotic cardiac remodeling) and increases mortality in mice and rat models of cancer ^{1,2}. These alterations may be associated with changes in heart energy and intermediary metabolism. However, these metabolic alterations have not yet being studied in detail. Therefore, our goal is to conduct a metabolic, proteomic, and kinetic analysis to determine the changes in the heart's metabolic pathways induced by cancer growth.

Using a tumor-bearing rat model, we initiated the evaluation of mechanical myocardial alterations *in vivo* (rat open chest procedure) and alterations in the heart metabolism. Female rats were inoculated with AS-30D hepatoma cells or vehicle. Physiological cardiac parameters (arterial blood pressure and beat rate) were determined 4 and 7 days after hepatoma inoculation. Arterial blood pressure decreased 30% at 4 days and 50 % at 7 days compared to rats inoculated with the vehicle. Meanwhile, the beat rate decreased by 30 % at 4 days and 40 % at 7 days. When determining damage to the hearts, an infarcted area was observed in the hearts of rats with hepatoma.

In addition, some metabolites of heart energy metabolism were determined to understand why cardiac performance decreased. ATP levels decreased by around 50%, and PCr levels increased by 3- to 6-fold. The increase in PCr levels correlated with a 30-50% decrease in mitochondrial and cytosolic creatine kinase activity. Mitochondria were isolated from the hearts of control rats and rats with hepatoma (4 and 7 days). State 3 respiration with various oxidizable substrates decreased by 20 to 50%, which correlated with a 20 to 40% decrease in 2-OGDH activity, with respect to mitochondria from control hearts. On the other hand, in the case of glycolysis, a 2- to 4-fold increase in the content of some metabolites such as glucose 6-phosphate, fructose 6-phosphate, and lactate was observed.

Our results suggest that the development of AS30-D hepatoma induces a deterioration of cardiac function associated with changes in energy metabolism.

References

1. Finke, D., Heckmann, M. B., Frey, N. & Lehmann, L. H. Cancer—A Major Cardiac Comorbidity with Implications on Cardiovascular Metabolism. *Front. Physiol.* **12**, (2021).
2. Tian, M., *et al.* Cardiac alterations in cancer-induced cachexia in mice. *Int. J. Oncol.* **37**, (2010).

RAMAN SPECTRAL SIGNATURES CAN DISCRIMINATE BETWEEN MURINE AND HUMAN TUMORIGENIC TRIPLE NEGATIVE LIVE BREAST CANCER

Pérez Huerta Lexie¹, Zentella Dehesa Alejandro^{1,2,3,4}, Ventura Gallegos José Luis², Islas Sánchez Selene Rubí⁵, Hernández Vidales Karen⁵, Muñoz Castillo Juan Alejandro⁵, Saniger Blesa José Manuel⁵

¹Unidad de Bioquímica Dr. Guillermo Soberón Acevedo, Biología Celular de Cáncer, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán, México City 14080, México

²Departamento de Medicina Genómica y Toxicología Ambiental, Instituto de Investigaciones Biomédicas Universidad Nacional Autónoma de México, México City 04510, México. ³Programa Institucional de Cáncer de Mama, Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, México City 04510, México. ⁴Red de Apoyo a la Investigación, Coordinación de la Investigación Científica, Universidad Nacional Autónoma de México, México City 04510, México.

⁵Laboratorio Universitario de Caracterización Espectroscópica, Instituto de Ciencias Aplicadas y Tecnología, Universidad Nacional Autónoma de México, México City 04510, México.

Raman spectroscopy is a non-destructive technique that results from the interaction of light with chemical bounds identifying low frequency modes in chemical bounds, thus providing detailed information of the chemical structure of complex molecular mixtures.

The complex visible Raman spectrum composed of more than 1000 vibration frequencies with variable Raman Shift Scattering Intensities can provide a highly specific spectral signature and has consequently been applied to the study of whole cells and tissues^{1,2,3}. Following this principle, we have applied Raman spectroscopy and principal component analysis to discriminate the spectral signatures obtained from live tumorigenic murine (4T1) from human (MDA-MB-231) triple negative breast cancer cell lines grown on complex murine-extracellular matrix. Our results show that principal component analysis of spectral signature maps can be used to discriminate between murine (4T1) and human (MDA-MB-231) breast cancer cell lines. Our results are consistent with previous reports and support the idea of using Raman spectroscopy and principal component analysis for the identification of tumor cells and tissues to complement histopathological studies in fixed biopsies from cancer patients.

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex of atoms, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

MICROBIOLOGY

EFFECT OF EPINEPHRINE AND NOREPINEPHRINE IN THE *GALLIBACTERIUM ANATIS* BIOFILM COMPOSITION AND STRUCTURE

Alicia N. Aguilar Fuentes¹, Fernando Montes García¹, Candelario Vázquez Cruz², Martha Salcedo Álvarez¹,
Lourdes Rojas³ y Erasmo Negrete Abascal¹

¹Facultad de Estudios Superiores Iztacala, UNAM, Av. De Los Barrios 1, Los Reyes Iztacala, 54090 Tlalnepantla, Edo de México;

²Centro de Investigaciones en Ciencias Microbiológicas, ICUAP, BUAP, Puebla, México; ³Laboratorio Nacional de Servicios Experimentales, Centro de Investigación y de Estudios Avanzados del IPN, Av. IPN 2508, Zacatenco 07360. México City, Mexico.

E-mail: noafg345@gmail.com

Gallibacterium anatis is a Gram-negative bacterium pathogen opportunist. It is part of domestic and wild birds' respiratory and reproductive tract microbiomes but also causes peritonitis, air sacculitis, salpingitis, and reproductive affections, mainly when birds are stressed. Its pathogenicity has been associated with the expression of virulence factors, including biofilms. The effect of Epinephrine (E) and Norepinephrine (NE), stress hormones, on the composition and structure of *G. anatis* biofilms is evaluated here. NE diminished the amount of biofilm (50%) compared to E or the control without additions. Amounts of preformed biofilm at 24 or 48h diminished in the presence of E or NE, but there was no change at 72h. The enzymatic digestion of biofilm polymers suggests an increase in protein amount by the presence of E. Carbohydrates and DNA quantities did not present significant changes. E induces a diminishing expression of total cell extract protein patterns in the 70 to 200 kDa range but increases the expression of 110-120 kDa secreted proteins. NE induces the expression of proteolytic activities in the range of 110 to 260 kDa. A 55 kDa protease was induced at 72h by the presence of E and NE. By scanning electron microscopy, compaction, and fragmentation of biofilms with catecholamines were observed at 24h. NE diminishes the biofilm amount. At 48h, biofilm fragments are immersed in exopolymeric material, and the control biofilm shows a high quantity of filamentous cells that are not observed in the presence of hormones. *G. anatis* biofilm composition and structural changes could be important in its dispersion and pathogenesis.

Project supported by DGAPA-UNAM PAPIIT IN204122.

REVERSION OF ANTIBIOTIC-RESISTANT PHENOTYPE IN ESKAPE BACTERIA THROUGH ACTINOBACTERIA SECONDARY METABOLITES

Vanessa N. Alcántara-Garduño¹, Michelle Chávez-Hernández¹,
Lourdes Villa-Tanaca¹ and César Hernández-Rodríguez¹

¹Departament of Microbiology, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional.
Prol. de Carpio y Plan de Ayala, s/n, Col. Santo Tomás, 11340, Ciudad de México, México.
E-mail: chdez38@hotmail.com

In 2017, the World Health Organization published the document “Global Priority List of Antibiotic-resistant bacteria to Guide research, discovery, and development of new antibiotics,” which includes a list of pathogenic bacteria that present multi-resistance to most of the available antibiotics and aims to guide research and development of new antibiotics¹. The phylum Actinomycetota and the *Streptomyces* genus are of significant importance, with a notable number of metabolites exhibiting biological activity². For this reason, the collection of actinomycetes isolated from jungle soils is, therefore, an excellent source for the search for relevant biological activities. This work aimed to evaluate the ability of actinomycete supernatants to reverse the antimicrobial resistance phenotype of ESKAPE group bacteria. A series of clinical isolates from ESKAPE bacteria were subjected to testing in conjunction with a group of thirteen isolates of actinobacteria, which were tested in Müller-Hinton media containing antibiotics at sub-inhibitory concentrations. Only nine of the actinobacteria isolates demonstrated the ability to inhibit *Pseudomonas aeruginosa* in the presence of the antibiotic ceftriaxone at a concentration of 256 mg/mL. From the lyophilized supernatants, the ability to reverse the antibiotic resistance phenotype in ESKAPE group bacteria was tested in vitro. Some supernatants acted synergistically with commercial antibiotics by inhibiting resistant bacteria from the ESKAPE group. Only four supernatants decreased the MIC in bacteria with a resistant phenotype, suggesting that they have a moderate reversal effect on the resistance phenotype. A phylogenetic analysis of the isolate ENCB-J28B revealed that it represents a novel species within the *Streptomyces* genus. The identification of novel compounds that act in synergy with currently available antibiotics to reduce the MIC or inhibit various resistance mechanisms represents a highly promising avenue in the ongoing battle against infections caused by ESKAPE bacteria. This approach would permit the continued use of these antibiotics at lower concentrations than those currently required.

References

1. Chávez-Hernández M., Ortiz-Álvarez J., Morales-Jiménez J., Villa-Tanaca L., Hernández-Rodríguez C. (2023). *Microorganisms*, 11(10), 2551.
2. De Oliveira, D. M. P., Forde, B. M., Kidd, T. J., Harris, P. N. A., Schembri, M. A., Beatson, S. A., Paterson, D. L., y Walker, M. J. (2020). *Clin Microbiol Rev*, 33, 81-91.

BRIDGING NUTRITIONAL GAPS: THE ROLE OF MICROBIOMES IN HOST PRODUCTIVITY

Gerardo Mejia^{1,2} and Luis D. Alcaraz^{1*}

¹Laboratorio de Genómica Ambiental, Departamento de Biología Celular, Facultad de Ciencias, Universidad Nacional Autónoma de México, Mexico City, Mexico. ²Posgrado en Ciencias Bioquímicas, Universidad Nacional Autónoma de México, Mexico City, Mexico. *Correspondence author: Edificio A Segundo Piso Laboratorio de Biología Molecular y Genómica Facultad de Ciencias Universidad Nacional Autónoma de México 04510 Coyoacán Ciudad de México.
+52 55 5622-8222 ext. 44704; lalcaraz@ciencias.unam.mx

The symbiosis connection between the host's nutritional status and microbiome is essential for good health. We investigated some effects of the host's nutritional status and its repercussions on the composition and functioning of the microbiome. We found that under nutrient-limiting conditions, the importance of the microbiome becomes evident. We explored this using model tomato plants and their phenotypes and metagenomic and 16S rRNA gene sequencing. By inoculating tomato plants with diverse soils as a microbial source, we compared the results of the root microbiome in soils and simplified hydroponic systems. This approach allowed us to identify plant growth-promoting microbial communities. Our results indicate that it is possible to select specific microbial communities that can improve plant resilience and growth, even under nutrient-limiting conditions. We also contribute to the One Health concept by connecting microbes in production systems. Our results highlight the impact of the microbiome on host adaptation to nutritional stress, opening clues about the structuring and mechanisms of interaction with the host.

Keywords: Symbiosis, Bacteria, Metagenomics

PLANT RESPONSE TO THE PLANT GROWTH PROMOTING RHIZOBACTERIUM *ACHROMOBACTER* SP. 5B1 UNDER PHOSPHATE AND NITRATE DEPRIVATION

Joseline Suhail Alejo Guerra, Kirán Rubí Jiménez-Vázquez, José López-Bucio*

Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Edificio B3, Ciudad Universitaria, C. P. 58030, Morelia Michoacán, México.
joselinealejo@gmail.com; jbcucio@umich.mx*

Nitrogen and phosphate are the most limiting nutrients in soils, which is a major problem in modern agriculture. Biological fixation of nitrogen is a useful tool for taking up the nutrient from the air and some bacteria have this property. Although phosphate can be found in soils, it is in a form in which plants are unable to absorb it. Both situations drive plants to modify its root system architecture to enhance the absorption of the limiting nutrients by the exudation of organic acids and other nutritious compounds that attract beneficial free-living bacteria through chemotaxis. *Achromobacter* sp. 5B1 was isolated from *Prosopis* sp. (mezquite) rhizosphere that grows in a saline environment. It has the property of improving growth and productivity of *Arabidopsis* seedlings. Root inoculation with the bacterium enhanced both the auxin response and transport within the root tip and caused an agravitropic behavior¹. Here, we investigated the role of *Achromobacter* sp. 5B1 in response of *Arabidopsis thaliana* under conditions of nitrogen and phosphate scarcity. Under nitrogen limitation the plant shows a more elongated primary root compared with the plant in axenical medium. On the other hand, *Arabidopsis* seedlings under phosphate scarcity and in interaction with *Achromobacter* sp. 5B1 exhibit delay exhaustion on primary root meristem due to nutritional stress, furthermore, it was observed a modification in the expression pattern of phosphate transport gene AtPT2) and organic acid exudation through the ALMT1 and STOP1 module. Our data uncover a novel mechanism by which rhizosphere bacteria improves plant nutrition.

References

1. Jiménez-Vázquez KR, García-Cárdenas E, Barrera-Ortiz S, Ortiz-Castro R, Ruiz-Herrera LF, Ramos-Acosta BP, Coria-Arellano JL, Sáenz-Mata J, López-Bucio J. (2020). *Plant Journal* 103(5):1639-1654.

IDENTIFICATION THROUGH TRANSCRIPTOMICS, OF GENES INVOLVED IN THE SWARMING OF *PECTOBACTERIUM BRASILIENSE* BF20 OVEREXPRESSING *EXL1* GENE, AND ITS RELATION TO EXPANSIN *EXL1*

Gloria Altamirano-Cruz¹, Damien Formay², Jimenez-Arteaga Vanessa¹ and Martínez-Anaya Claudia¹

¹Instituto de Biotecnología and ²Centro de Ciencias Genómicas, UNAM. 62110 Cuernavaca, Morelos.
gloria.altamirano@ibt.unam.mx

Bacteria display different types of movement; one of them the swarming motility. Swarming is a social process for colonizing surfaces, sometimes associated to pathogenicity in species such as *Proteus mirabilis*. During swarming, bacteria differentiate and hiperproduce flagella. While swarmign occurs in laboratory conditions, swarming-like cells exist in the infection of the phytopathogen *Pectobacterium*, but its role in plant colonization and virulence remains unknown. Because swarming associates with enzyme production for soft rot, this suggests a role during infection. In *P. atrosepticum* a deletion in *exl1* gene impairs virulence and swarming, implying a function for Exl1 protein in these processes (1). This project aimed to identify other genes related to swarming in *P. brasiliense* BF20 through the overexpression of *exl1* and transcriptomic analysis in comparison to non-swarmers bacteria. Additionally, we analyzed the effect of Exl1 in the production of lipopolysaccharides (specifically antigen O) in the swarming of BF20, due to their requirement as a key surfactant for swarming (2). We obtained the transcriptome of swarmer and non-swarmers Exl1-overexpressing BF20. After quality control analyses the deduced sequences achieved acceptable values of confidence for our experiments. Then, reads were aligned to the BF20 inferred transcriptome, resulting in 751 differentially expressed genes between the two conditions. Searches in Uniprot and KEGG confirmed representation of pathways related to swarming, highlighting flagellar assembly, chemotaxis, quorum sensing and exopolysaccharide biosynthesis. Interestingly, we identified differentially regulated genes related to the type VI secretion system (T6SS) and for type IV pilus synthesis not previously reported in the swarming of *Pectobacterium*. Our results contribute to the understanding of swarming in this important plant pathogen.

Reference

1. Narváez-Barragán, et al. *Sci Rep* 10:7747. 2020
2. Bowden, et al., *Microbiol* 159 pt.11. 2013

THE SWEET COMPETITION: THE BACTERIAL LECTIN PIRB^{VP} FROM *VIBRIO PARAHAEMOLYTICUS* AND ITS IMPLICATION IN THE COLONIZATION OF ECOLOGICAL NICHES

Cassandra A. Alvarado-Verdín¹, Miriam F. Ayón-Pérez¹, Juan M. Agraz-Cibrian¹, Jorge Gutiérrez-Franco¹, Alejandro Vázquez-Reyes¹, J. Francisco Zambrano-Zaragoza¹ and Marcelo Victorio-De Los Santos¹

¹Universidad Autónoma de Nayarit. Laboratorio de Investigación en Proteínas, Biología Molecular e Inmunología, Av. De la Cultura S/N, Ciudad de la Cultura; Tepic Nayarit, México, C.P. 63000.
E-mail: mvictorio@uan.edu.mx

Vibrio parahaemolyticus (*Vp*) is a bacterium that coexists with a wide diversity of aquatic bacteria. Specific strains of *Vp* containing the plasmid pVA1, which produces the binary toxin PirAB^{vp}. These strains have been observed to cause bacterial dysbiosis in certain niches. In this work, we evaluated whether the ability of *Vp* to colonize specific niches is related to the production of the PirAB^{vp} toxin, especially the B subunit, a lectin that recognizes amino sugars through its jacalin domain. Agglutination assays were performed as well as growth inhibition assays using recombinant PirB^{vp}. Likewise, *Vp* competition tests were carried out against *V. cholerae* (*Vc*) and *Alivibrio fischeri* (*Af*), quantifying the amount of PirB^{vp} protein produced and identifying it by western blot. It was determined that PirB^{vp} recognizes glycosylated structures of *Vc* and *Af* showing a dose-dependent bacteriostatic effect, as it inhibits 66 to 70% in *Vc* and *Af*. However, when native PirAB^{vp} is present, the effect observed is bactericidal against these two strains, directly related to the increase in PirAB^{vp} production. In an *in vitro* competition test between *Vp* and *Vc*, the predominance of *Vp* and the death of *Vc* were observed as the concentration of PirAB^{vp} increases. When the competition test was carried out using a *Vp* strain without the plasmid that encode for PirAB^{vp}, it was observed that it did not cause the death of *Vc* and, on the contrary, both could coexist in the environment. *In vitro* inhibition assays showed a galactopyranoside inhibits antibacterial activity by 52 to 76% against *Vc*, suggesting that this interaction is due to blocking the recognition sites of the jacalin domain. Likewise, when using a strain of *Af* that does not express sugars in the O antigen of LPS, the bactericidal effect was not observed despite the fact that the concentration of PirAB^{vp} was increasing, which demonstrates that the colonization of the niche is related to the recognition of a sugar that acts as a ligand in bacteria.

ANALYSIS OF DIVERSITY IN THE CRISPR-CAS SYSTEM OF *CRONOBACTER SAKAZAKII* ISOLATED FROM FOOD SAMPLES

Ana Karen Álvarez Contreras^{1,2}, Elsa Irma Quiñones Ramírez¹, Carlos Vázquez Salinas²

¹Escuela Nacional de Ciencias Biológicas-IPN Santo Tomás, Manuel Carpio, Plutarco Elías Calles, Miguel Hidalgo, C.P. 11350 CDMX. ²Universidad Autónoma Metropolitana-Iztapalapa, San Rafael Atlixco 186, Leyes de Reforma 1A Sección, Iztapalapa, C.P. 09310 CDMX.
E-mail: alvacont25@gmail.com

Introduction. *Cronobacter sakazakii* is a Gram-negative opportunistic pathogen associated with foodborne diseases due to consumption of contaminated powdered infant formula, mainly affecting neonates and infants with mortality rates ranging from 40% to 80%. The presence of virulence factors contributes to its pathogenesis and the development of necrotizing enterocolitis and neonatal meningitis. It is worth mentioning that *C. sakazakii* is not considered a notifiable pathogen and its epidemiological surveillance is almost non-existent. On the other hand, Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)-associated (Cas) system is encoded by operons that exhibit a high rate of evolution due to genetic diversity and a unique spacer system. Together, they provide adaptive immunity to prokaryotes possessing them. The purpose of this study was to study the CRISPR-Cas system in *C. sakazakii* isolated from food samples.

Methodology. Sequenced genome of a *C. sakazakii* strain isolated from dehydrated coriander was used as a model to analyze the CRISPR-system. The orientation of the CRISPR spacers was determined using the CRISPRDetect program and the spacers through CRISPRCas-Finder. Subsequently, multiple alignments of the 12 DNA sequences corresponding to the *C. sakazakii* isolates were performed using MEGA software with the ClustalW algorithm.

Results. In this study, the genome of *C. sakazakii* revealed the identification of CRISPR1 and CRISPR2 loci with 22 repeated sequences and 21 variable spacer sequences contributing to genetic diversity. In the model strain analysis of *C. sakazakii*, an arrangement of seven Cas proteins (attached to the CRISPR1 locus) was found as follows: Cas2 protein (endoribonuclease), Cas1 (ribonuclease), Cas2 (endoribonuclease), CasE (CRISPR system Cascade subunit), CasC (CRISPR system Cascade subunit), and 3 hypothetical proteins. The classification of prophages was carried out using the classification established by *Escherichia coli*, where the subtype IE CRISPR-Cas was predominantly found in the conserved sites. The aligned 12 DNA sequences formed the basis for generating the phylogenetic tree, which broadly shows the incidence of horizontal gene transfer of CRISPR-Cas genes between distant species and genera. It is worth noting that 1 ATCC *C. sakazakii* and 1 ATCC *E. coli* were also used to compare homology and ancestry, which were closely related, probably due to infection by specific phages for the *Enterobacteriaceae* family.

Conclusions. The analysis of the CRISPR-Cas system offers a genetic-level perspective aiming to understand the evolution, behavior under different environmental conditions, and epidemiology of the analyzed bacterial populations.

References:

1. Simran K Kushwaha, Lakshmi P Narasimhan, Chandrananthi Chithanathan y Sandhya A Marathe. (2022). Cas system of Clustered Regularly Interspaced Short Palindromic Repeats: diversity and regulation in Enterobacteriaceae. *Future Microbiology* 17:15, pages 1249-1267.
2. Zeng, Zhang, Wu, ElW., Wu, S.MY, LiC, Lingnorte, ChenM, Wang, CaiS, LeiT, DingY, XueL. 2018. Reconstituting the evolutionary history of *Cronobacter* driven by differentiated CRISPR activity. *Appl Environ Microbiol* 84. <https://doi.org/10.1128/AEM.00267-18>
3. Zeng, H., Zhang, J., Li, C. et al. "The driving force of prophages and the CRISPR-Cas system in the evolution of *Cronobacter sakazakii*". *Scientific Reports* 7, 40206 (2017). <https://doi.org/10.1038/srep40206>

IDENTIFICATION AND CHARACTERIZATION OF CONTEMPORANEOUS CLINICAL ISOLATES OF *CANDIDA GLABRATA*

Oscar Daniel Aranda Moreno¹, Marco J. Hernández Chávez¹, Alejandro De Las Peñas¹ and Irene Castaño-Navarro¹

¹Instituto Potosino de Investigación Científica y Tecnológica, División de Biología Molecular. Camino a la Presa San José 2055, Lomas 4ª sección, C.P. 78216, San Luis Potosí, S.L.P. Tel. +52 (444) 834 2040.
E-mail: oscar.aranda@ipicyt.edu.mx

Candida glabrata is an opportunistic fungal pathogen. It is a common human commensal that is responsible for invasive candidiasis in immunosuppressed or immunocompromised patients. Nowadays, *C. glabrata* is the second most common *Candida* species responsible for invasive candidiasis just after *Candida albicans*, and it leads the high-priority group in WHO fungal priority pathogen list. This is in part because *C. glabrata* has an innate and acquired resistance to the azole class of antifungals and microevolution can occur during anti-fungal therapy leading to greater antifungal resistance. *Candida* spp have different susceptibility to azoles and echinocandins, therefore accurate and prompt detection of *Candida* spp is crucial for treating the infection and for a better patient prognosis. Moreover, the microevolution of *C. glabrata* has been studied with sequential clonal clinical isolates (longitudinal strains) obtained from patients with candidemia throughout the infection (days, weeks or months) in which genotypic and phenotypic variability can occur. In hospitals, once the pathogen is isolated, almost always only a single colony (index strain) is characterized, assuming the independent action hypothesis (IAH) or single-organism hypothesis which states that one cell of the pathogenic microorganism can be responsible for the infection, so the infecting population would be phenotypically and genotypically homogeneous. However, recent studies have documented that genotypic and phenotypic variability is already present in contemporaneous clonal strains at the time the patient is diagnosed. Therefore, when the characterization by the clinical laboratory is limited to a single colony, variability may be underestimated and traits such as antifungal resistance may not be recognized when the patient is diagnosed with candidemia. In this work, we will identify and analyze contemporaneous isolates of *C. glabrata* from different patients and different anatomical sites to determine phenotypic and/or genotypic variability and whether this variability is niche-specific.

IDENTIFICATION OF MOLECULES WITH ANTIBACTERIAL ACTIVITY FROM DIFFERENT FUNGAL PATHOGENS

Jesús Antonio Arroyo García, Guadalupe Gutiérrez-Escobedo and Alejandro De Las Peñas

IPICyT, Instituto Potosino de Investigación Científica y Tecnológica. División de Biología Molecular.

Camino a la Presa San José #2055, Lomas 4a. Sección, 78216 San Luis Potosí, S.L.P. México.

Tel: (444)-834-2040; e-mail: jesus.arroyo@ipicyt.edu.mx

Pathogenic bacteria cause infectious diseases around the world. The response against infectious diseases has been the design and development of antibiotics with different modes of action. These pathogens can survive in the presence of antimicrobials through different mechanisms: mutations in target proteins, increase in expression of efflux pumps, and uptake of genetic material from other organisms, among others. The World Health Organization (WHO) has declared a worldwide emergency in acquired antimicrobial resistance by different pathogenic bacteria. The WHO priority is the research and development for new antibiotics against multi-resistant bacteria. Yeast have shown to secrete metabolites with antibacterial activity. In our laboratory, it has been shown that the secretome of *Candida glabrata* has antibacterial activity. In this work, we found that conditioned medium from *Candida glabrata* (CgCM), which is spent media from this yeast culture grown in rich media (YPD), lysate *Escherichia coli* and *Pseudomonas aureginosa*. In addition, we found that the CgCM in combination with antibiotics, with different mode of action, presented a synergistic effect; antibacterial effect was found with sublethal concentrations of the antibiotic, meaning less concentration of the antibiotics are needed to have the antibacterial effect.

EXPLORING THE IMPACT OF GLYPHOSATE ON ORANGE TREE MICROBIOMES: A META-TAXONOMIC APPROACH

Diana Baeza^{1,‡}, Pablo Valentín Navarro-Enguilo^{1,‡}, Ernesto Tapia-Campos², Jorge Verdín^{1,*}

¹Biología Industrial, Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco. Camino Arenero 1227, El Bajío, Zapopan, JAL, México 45019. ²Biología Vegetal, Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco. Camino Arenero 1227, El Bajío, Zapopan, JAL, México 45019.

[‡]These authors contributed equally to this work; ^{*}jverdin@ciatej.mx

Glyphosate, a common herbicide in citrus cultivation, has sparked concerns about its effects, even in sublethal doses, on soil- and plant-associated microbiomes, and overall ecosystem health. It targets the shikimate pathway, crucial for synthesizing aromatic amino acids in plants and many microorganisms but absent in animals. This selective action can alter microbial communities by affecting organisms reliant on this pathway, potentially changing the composition and function of the microbiome. This raises questions about the long-term sustainability of glyphosate use in agriculture, though some studies have presented inconclusive evidence. Our study evaluated the microbiota of orange trees in conventional, transitional and organic agricultural systems in northern Veracruz using a 16S and ITS2-based metataxonomic approach; we focused on the orange tree surrounding bulk soil, rhizosphere, and phyllosphere. Results revealed significant differences in soil microbiomes between conventional and transitional practices. Conventionally managed soils had lower fungal diversity compared to soils transitioning to agroecological practices, though overall soil microbial diversity remained relatively constant across different systems.

In the rhizosphere, subtle variations were noted between conventionally and agroecologically managed orange trees. While alpha diversity differences were not significant, an enrichment of fungi was observed in the early stages of transitioning from conventional to agroecological cultivation. In the case of the phyllosphere, one of the sites sampled showed a difference in bacterial phylogenetic diversity when moving from conventional to transitional cultivation. However, upon reaching organic cultivation, the diversity was very similar to that of conventional crops. At the other location, there were no significant differences. On the other hand, fungal phylogenetic diversity in one of the plots was enriched with the change from conventional to organic practices, while in the other plot it remained constant across the different agricultural systems.

Across the three types of monitoring, perceptible differences were evident during the transition to agroecological practices compared to conventionally managed trees. Upon reaching the organic phase, the microbiome of the plots resembled that of conventionally grown orange trees, indicating resilience and succession of the microbial community. This suggests that the plant-generated microenvironments, the rhizosphere and the phyllosphere, crucially determine the composition of the microbial communities. Although agroecological practices initially alter these communities, they eventually adapt and stabilize, balancing the influence of plants and external environmental influences. In contrast, bulk soil from any agricultural practices showed a change in fungal diversity, probably due to the entry of glyphosate into the soil immediately post application.

PHENOTYPIC AND BIOINFORMATIC STUDY OF GENE *cdgE* OF *AZOSPIRILLUM BALDANIORUM* SP245

Nora Barrera¹, Sandra Reyes Carmona², María Luisa Xiqui Vázquez³, Alberto Ramírez⁴ and Beatriz E. Baca⁵

Centro de Investigaciones Microbiológicas, Benemérita Universidad Autónoma de Puebla,
Av. del Infante 2012, Cd Universitaria, Ciudad Universitaria, 72592 Heroica Puebla de Zaragoza, Pue.
E-mail: bo223470237@alm.buap.mx

The genus *Azospirillum* has been studied due to its importance in agriculture and its beneficial effects on plants. Since then it has been considered of great importance for the harvest of this grain, in addition to being a model strain for genetic and biochemical studies on the interaction of the bacteria with the plant and its use as an inoculant (dos Santos Ferreira et al. 2020)². The interaction of *Azospirillum* with the plant is due to chemotaxis and mobility. It is attracted by root exudates, allowing it to associate with the roots of the plants and adhere quickly. The formation of this biofilm is essential for the survival of the bacteria in adverse environmental conditions. This phenotype is regulated by the intracellular levels of the second messenger cyclic-di-GMP (c-di-GMP). The diguanylate cyclases (DGC) are the enzymes that catalyze the formation of c-di-GMP from two GTP molecules, being a key enzyme for the signaling cascade within the bacteria. Up to 35 genes have been described that encode proteins involved in the synthesis and degradation of di-GMP-c in *A. baldaniorum*. Of these, 20 DGC proteins are encoded with a single GGDEF domain (Ramírez-Mata et al., 2018)³. This work studies the *cdgE* gene that codes for the protein named diguanylate cyclase E (DGCE). A bioinformatic study of the gene was carried out to identify the domains that compose its structure, to identify conserved regions in the amino acid sequence, as well as predict its spatial arrangement through 3D modeling on different platforms. Additionally, the phenotype of the deletion mutant, *A. baldaniorum* $\Delta cdgE$, is evaluated. In addition, the gene complementation in the mutant is assessed too. It is inferred that the *cdgE* gene is involved in motility and biofilm formation phenotypes, among others.

References

1. dos Santos Ferreira, N., Hayashi Sant'Anna, F., Massena Reis, V., Ambrosini, A., Gazolla Volpiano, C., Rothballer, M., ... & Zilli, J. E. (2020). Genome-based reclassification of Sp245 as the type strain of *Azospirillum baldaniorum* sp. nov. *Azospirillum brasilense* International J. Systematic Evol. Microbiol, 70(12), 6203-6212.
2. Ramírez-Mata, A., Pacheco, C. M., Cruz Pérez, J. F., Sáenz, M. M., & Baca, B. E. (2018). In silico comparative analysis of GGDEF and EAL domain signaling proteins from the *Azospirillum* genomes. *BMC Microbiol*, 18, 1-14.

STUDY OF THE MECHANISM OF CONTROL OF C-5 ALGINATE EPIMERASES BY THE SECOND MESSENGER C-DI-GMP

Victor V. Barrios¹, Cinthia E. Núñez², Josefina Guzmán Aparicio, Ma. Soledad Moreno
Instituto de Biotecnología de la UNAM, Avenida Universidad 2001, Chamilpa, 62210 Cuernavaca, Mor.
Tel. +52 (777) 329 16 29. cinthia.nunez@ibt.unam.mx

A. vinelandii is a free-living Gram-negative bacterium capable of producing a type of linear exopolysaccharide called alginate, composed of mannuronic (M) and guluronic (G) acid residues. Under adverse environmental conditions, it undergoes a process of cell differentiation to form desiccation-resistant cysts. This cell consists of a central body covered by an envelope, which contains alginates with a high proportion of G residues, essential for desiccation resistance. The G residues in the polymer are derived from the activity of extracellular C-5 epimerases, AlgE1-6 (1). Previous work in our laboratory showed that the second messenger c-di-GMP exerts a positive effect on the transcription of *algE1-6* genes and thus, is essential for the formation of cysts resistant to desiccation (2). The effect of c-di-GMP on *algE1-6* gene expression was confirmed by qRT-PCR as the accumulation of *algE1-6* transcripts was elevated or reduced in genetic backgrounds with artificially high or low levels respectively of this second messenger. This result agrees with Western-Blot results, revealing that AlgE1-6 proteins were not detected at low concentrations of c-di-GMP. The genetic arrangement of the *algE* cluster was investigated by RT-PCR revealing that the *avin51240-algE6-algE4*, *algE1-algE2* cluster conforms an operon, containing a putative *s70* promoter driving its transcription. An additional AlgU-dependent promoter upstream of *algE6* was also identified. The functionality of both promoter (*Pavin51240* and *PalgE6*) was confirmed using *gusA* transcriptional fusions showing a higher activity at the stationary phase. Interestingly, both promoters were positively controlled by the sigma factor AlgU and the levels of c-di-GMP since the absence of AlgU, or reduced levels of c-di-GMP abrogated their activities. The molecular mechanism of such regulation is unknown but the effect of AlgU seems to be c-di-GMP independent as the levels of this second messenger was not altered by the absence of AlgU. Collectively, our results suggest the existence of an intermediary under the control of AlgU and c-di-GMP necessary for the transcription of *algE1-6* genes. We are currently investigating the identity of such transcriptional activator.

References

1. Segura, Daniel; Núñez, Cinthia; and Espín, Guadalupe (January 2020) Azotobacter Cysts. In: eLS. John Wiley & Sons, Ltd:Chichester.
2. Martínez-Ortiz IC, Ahumada-Manuel CL, Hsueh BY, Guzmán J, Moreno S, Cocotl-Yañez M, Waters CM, Zamorano-Sánchez D, Espín G, Núñez C. 2020. Cyclic-di-GMP-mediated regulation of extracellular mannuronan C-5 epimerases is essential for cyst formation in *Azotobacter vinelandii*. J Bacteriol.

STRUCTURAL MODELS OF THE PHOSPHATASE ENZYME AND MUTANTS IN *ESCHERICHIA COLI* INVOLVED IN THE SYNTHESIS OF TREHALOSE AND BACTERIAL DESSICATION

Bernabe-Allende Alejandra¹, Quintero-Hernández Verónica², Juárez-González Víctor Rivelino³

¹Grupo de Ecología y Supervivencia de Microorganismos (GESM), Laboratorio de Ecología Molecular Microbiana (LEMM), Centro de Investigaciones en Ciencias Microbiológicas (CICM), Instituto de Ciencias (IC), Benemérita Universidad Autónoma de Puebla (BUAP), Edificio IC11, Ciudad Universitaria, Colonia Jardines de San Manuel, Puebla, Puebla 72570, México.

²CONACYT-GESM, LEMM, CICM, IC, BUAP, Edificio IC11, Ciudad Universitaria, Colonia Jardines de San Manuel, Puebla, Puebla 72570, México. ³Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Mor., México.

Email: vquinterohe@conacyt.mx

Xerotolerant bacteria, which are resistant to intracellular desiccation, play an essential role in the rhizosphere of plants. One of their mechanisms, to success in extreme environments, is the synthesis of trehalose, a disaccharide that prevents intracellular water loss¹. *Escherichia coli* is able to produce trehalose by a path that involves the enzyme Trehalose 6 phosphate synthase that catalyzes the formation of the bond between UDP-Glucose and Glucose 6P to produce one molecule of trehalose 6P, then the enzyme Trehalose 6 phosphatase dephosphorylates trehalose 6P to produce one molecule of trehalose², therefore in this project we did a multiple sequence alignment with Clustal Omega using phosphatases sequences and the homology modeling with SWISS-MODEL, we generated a dimensional model of *Escherichia coli* phosphatase, based on the atomic coordinates, of the *Salmonella typhimurium* phosphatase structure, deposited in the PDB, in addition to proposing mutations that allowed us to learn more about the functionality of the amino acids in the active site of the enzyme. Using this knowledge will generate more active mutants than the wild enzyme, to produce more trehalose and these new mutants could be used to transform plant growth promoting bacterial strains that are not xerotolerant and will allow to produce plants of commercial interest.

References

1. Koshland D, Tapia H. Desiccation tolerance: an unusual window into stress biology. *Mol Biol Cell*. 2019 Mar 15;30(6):737-741. doi: 10.1091/mbc.E17-04-0257. PMID: 30870092; PMCID: PMC6589773.
2. Purvis JE, Yomano LP, Ingram LO. Enhanced trehalose production improves growth of *Escherichia coli* under osmotic stress. *Appl Environ Microbiol*. 2005 Jul;71(7):3761-9. doi: 10.1128/AEM.71.7.3761-3769.2005. PMID: 16000787; PMCID: PMC1168978.

DISTRIBUTION OF HUANGLONGBING IN ORANGE FRUIT PRODUCER ZONE OF SAN LUIS POTOSÍ AND NEIGHBORING STATES

Karla Grisel Calderón González, Andrés Usiel Rodríguez Rodríguez,
Luis Enrique Jiménez Vázquez, Vanessa Olivares Illana

Laboratorio de Interacciones Biomoleculares y Cáncer del Instituto de Física de la Universidad Autónoma de San Luis Potosí, UASLP. Av. Parque Chapultepec 1570, Privadas del Pedregal, 78295 San Luis Potosí, S.L.P.
vanessaolivaresillana@gmail.com

Huanglongbing (HBL) also known as “citrus greening” or “yellow dragon disease” is one of the most serious diseases prevalent in global citrus production. The disease is caused by the phloem-colonizing bacterium *Candidatus Liberibacter* (CL) that is transmitted by two psyllid vectors (*Diaphorina citri* and *Trioza erytreae*). HBL is associated with three distinct species, namely *Candidatus liberibacter asiaticus* (CLas), *Candidatus Liberibacter africanus* (CLaf), and *Candidatus Liberibacter americanus* (CLam). HBL disease is characterized by yellow shoots with pale green and yellow flushes, non-symmetrical mottled leaves, enlarged, corky mid-ribs of leaves, aborted seed, and deformed fruits. The infection with CL causes a rapid tree to decline which leads eventually to death. San Luis Potosi is the third national producer of oranges; therefore, it is very important for us to take action on this matter. The aim of this study was to determinate which variants of *C. liberibacter* are present in SLP and neighboring states. Sample collection was carried out in medium zone, center zone, Huasteca zone of San Luis Potosi, as well as in surrounding areas from Zacatecas and Tamaulipas. We developed different protocols of DNA extraction from leaves and, we standardized a nested PCR protocol for detection of different variants of *C. Liberibacter*. We concluded that the prevalent variant in SLP and surrounding areas is *C. Liberibacter asiaticus*.

EFFECT OF BACTERIAL CYCLODIPEPTIDES ON GUT MICROBIOTA COMPOSITION AND NUTRIENT ABSORPTION IN AN OBESITY MODEL

Marlene Estefania Campos Morales¹, Lorena Martínez Alcantar¹,
Asdrúbal Aguilera Méndez¹, Jesús Campos García¹

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo.
E-mail: 1435830h@umich.mx; lorena.martinez@umich.mx

Cyclodipeptides (CDPs), also known as 2,5-diketopiperazines, are the smallest cyclic peptides found in nature. In recent years, these small molecules have been a focus of research as they have been found to possess a wide range of biological activities such as antiviral, antitumor, anti-inflammatory, immunosuppressive, antifungal, and recently antibacterial capability by regulating bacterial populations^{1,2}. *Pseudomonas aeruginosa* PAO1 produces a mixture of CDPs consisting of cyclo (L-Pro-L-Tyr), cyclo (L-Pro-L-Phe), and cyclo (L-Pro-L-Tyr). This work investigated the effect of the CDPs administration in a model of obesity (a state of dysbiosis) in Wistar rats over ten weeks of treatment and determined bacterial content of the gut microbiota and its impact on nutrient absorption. Weekly fecal samples were collected and tested to determine protein, carbohydrate, lipid, and moisture content, pH quantification, and colony-forming unit (CFU) measurement. The results indicate that the administration of CDPs led to modifications in the bacterial content in the feces, correlating with causing changes in nutrient absorption. These findings suggest that the bacterial CDPs modify the intestinal microbiota and nutritional state in an obesity dysbiosis model.

References

1. Dolores Vázquez Rivera, et al., (2015). Cytotoxicity of cyclodipeptides from *Pseudomonas aeruginosa* PAO1 leads to apoptosis in human cancer cell lines. *Biomed Res Int.* 2015:197608.
2. Martínez Carranza, E., Ponce Soto, G. Y., Díaz Pérez, A. L., Cadenas, E., Souza, V., y Campos García, J. (2018). *Extremophiles*, 22(1), 73-85.

GENE OF NO ORTODOXE MULTISENSOR HISTIDINE KINASE IS INVOLVED IN MOTILITY TYPE SWARMING AND SWIMMING IN THE RHIZOBACTERIUM *AZOSPIRILLUM BALDANIORUM* SP245

Uriel Cardona-Baltazar¹, Sandra Reyes Carmona¹ and Beatriz E. Baca¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Benemérita Universidad Autónoma de Puebla. Edif. ICTI, Ciudad Universitaria, Col. San Manuel Puebla Pue, CP72570, Puebla, Mexico.

E-mail: uriel_c_b@hotmail.es

Azospirillum alphaproteobacteria, which live in the rhizosphere of many crops, are used widely as biofertilizers. To establish a successful and prolonged bacteria-plant interaction, *A. baldaniorum* can form biofilms, bacterial communities embedded in a self-made matrix formed by extracellular polymeric substances which provide favorable conditions for survival, these mechanisms can be regulated by Two-component signal systems. The Two-component signal transduction systems (TCS) are one of the primary means by which bacteria sense and adapt to fluctuating environmental conditions. Two-component signal transduction systems typically involve a membrane-bound histidine kinase that senses stimuli, autophosphorylates in the transmitter region and then transfers the phosphoryl group to the receiver domain of a cytoplasmic response regulator that mediates appropriate changes in bacterial physiology. In this study, we obtained the *A. baldaniorum* Sp245 mutant for the *hkhC* gene, which encodes the TCS of the no ortodoxe histidine kinase. Inactivation of this gene affected bacterial motility but not the formation of biofilm in NFB medium and NFB*+KNO₃. In mutant $\Delta hkhC::Km^R$ showed decreased mobility swimming type when add malate and proline but not when add lactate to medium only source carbon. The study the effects of mutant $\Delta hkhC::Km^R$ in mobility swarming type showed decreased in the proven medium. Introduction into mutant cells of the *hkhC* gene as part of an expression vector led to recovery of the phenotypes of motility. The results suggest that the HKHC under study modulates the motility of *A. baldaniorum* Sp245.

STUDY OF THE HISTIDINE-KINASE RETS AS PART OF THE MULTI-KINASE SYSTEM GACS (MKN-GACS) IN *AZOTOBACTER VINELANDII*

Miguel Castañeda Lucio¹, Araceli Rosales-Cruz¹, Jimena Reyes-Nicolau¹, Eduardo Minto-González¹, Alan Meneses-Carbajal¹, Claudia Mondragón-Albarrán¹, and Liliana López Pliego¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla. IC-11 Ciudad Universitaria Puebla, Pue. México, C. P. 72000.

E-mail: miguel.castaneda@correo.buap.mx

In bacteria, generally, signal transduction is performed by proteins belonging to signaling systems known as two-component systems (TCS). TCSs are constituted by a receptor protein (histidine kinase, HK) that, upon receiving a signal, autophosphorylates a histidine residue in the transmitter domain (H1) and activates, through transphosphorylation, the second element of the system known as the response regulator (RR). The HK-RR regulatory pair paradigm has recently been questioned by the existence of systems with two or more HKs that control the phosphorylation status of an RR but where only one HK phosphorylates the RR. The accessory HK (or HKs) is proposed to stimulate or block the kinase activity of the main HK. These signal transduction systems have been called multi-kinase networks (MKN). The TCS GacS/A in *Pseudomonas aeruginosa* has been related to other HKs. It forms a MKN in which the HK RetS negatively controls GacS activity and positively by the HK LadS. Thus, GacS, RetS, and LadS constitute the core of the GacS-MKN that controls the activation of GacA, which in turn regulates the transcription of the genes that encode the sRNAs of the Rsm post-transcriptional control system¹. Outside of the *Pseudomonas* genus, the network has only been studied in *Azotobacter vinelandii*, in this bacterium the GacS-MKN is formed with the HKs GacS, RetS and HrgS. MKN-GacS in *A. vinelandii* does not have a *ladS* homolog but possesses a distinct accessory kinase (HrgS)². In this work, we show the *A. vinelandii* RetS homolog characterization and its function in the GacS-MKN.

References

1. Francis VI, Porter SL (2019) Multikinase Networks: Two-Component Signaling Networks Integrating Multiple Stimuli. *Annu Rev Microbiol* 73:199-223. doi: 10.1146/annurev-micro-020518-115846
2. López-Pliego L, González-Acocal V, García-González et al (2022) HrgS (Avin_34990), a novel histidine-kinase related to GacS, regulates alginate synthesis in *Azotobacter vinelandii*. *FEMS Microbiology Letters* 369(1), fnac024. doi.org/10.1093/femsle/fnac024

ISOLATION AND BIOCHEMICAL IDENTIFICATION OF LACTOBACILLUS SPP. EXTRACTED FROM PRODUCTS OF ANIMAL ORIGIN

Azul Castrillo Sanchun¹, Francisco Héctor Chamorro Ramírez¹, Aida Hamdan Partida², Esmeralda Mónica Peña González¹ and Suzette Juárez Contreras¹

¹Laboratorio Veterinario de Ciencia de la Carne y Salud Pública del Departamento de Producción Agrícola y Animal, Universidad Autónoma Metropolitana Xochimilco, Ciudad de México, México. ²Laboratorio de Microbiología y Biología Molecular del Departamento de Atención a la Salud Universidad Autónoma Metropolitana Xochimilco, Ciudad de México, México. Universidad Autónoma Metropolitana Unidad Xochimilco C.P. 04960 Tel. 5611642603 E-mail: 2232801304@alumnos.xoc.uam.mx

In recent years, interest in the study of lactic acid bacteria in the food industry has increased, due to their potential in the production of bioactive compounds such as exopolysaccharides, bacteriocins, among others. Lactic acid bacteria are a heterogeneous group with morphological and physiological characteristics in common, such as the ability to acidify the environment in which they are found¹. Within this group, *Lactobacillus* is one of the most relevant genera for the food industry, currently including 261 species. Their importance initially lies in the fact that they are considered probiotics. In addition, it has been reported that these microorganisms have the potential to inhibit cognitive deficits, regulate the immune system, reduce cholesterol levels, maintain balance in the intestinal flora and reduce the risk of tumors. Therefore, its isolation and characterization are a fundamental step for the study of its influence on human health, as well as its applications in the food industry^{2,3}. The objective of the present study was to identify *Lactobacillus* isolated from 3 products of animal origin: raw milk, kefir and bacon. These products were sampled and plated on MRS agar plates and incubated at 37°C for 24–48 h. The colonies were identified by their morphology by selecting the moist, punctate colonies, whitish in color, with a smooth and shiny surface with defined edges. After isolation and reseeded of the selected colonies, microscopic identification was carried out using Gram staining where those that corresponded to Gram-positive bacteria with the shape of bacilli were selected and finally biochemical identification in which catalase, sulfide mobility of indole, triple iron sugar and Simmons citrate tests were performed. Of the 19 isolated colonies that presented the morphological characteristics, 6 were isolated that corresponded according to biochemical tests to *Lactobacillus* spp., all of them obtained from bacon, these microorganisms have been previously reported in bacon by Xinfu and collaborators in 2021⁴. As future from this perspective, we wish to evaluate the antimicrobial potential of the isolated microorganisms.

References

1. Saleem, M., Malik, S., Mehwish, H. M., Ali, M. W., Hussain, N., Khurshid, M., & Chen, Y. (2021). Isolation and functional characterization of exopolysaccharide produced by *Lactobacillus plantarum* S123 isolated from traditional Chinese cheese. *Archives of Microbiology*, 203, 3061-3070.
2. Flores-Maciél, H. A., Cordero-Soto, I. N., Martínez-Herrera, R. E., Ochoa-Martínez, L. A., & Rutiaga-Quiñones, O. M. (2024). Importancia de las bacterias ácido lácticas como productoras de exopolisacáridos. *Revista Agraria*, 21(2), 5-11.
3. Al-Tawaha, R., & Meng, C. (2018). Potential benefits of *Lactobacillus plantarum* as probiotic and its advantages in human health and industrial applications: A review. *Adv. Environ. Biol*, 12, 16-27.
4. Li, X., Xiong, Q., Xu, B., Wang, H., Zhou, H., & Sun, Y. (2021). Bacterial community dynamics during different stages of processing of smoked bacon using the 16S rRNA gene amplicon analysis. *International Journal of Food Microbiology*, 351, 109076.

PHENOTYPIC AND GENOMIC CHARACTERIZATION OF *STREPTOMYCES PAKALII* SP. NOV., A NOVEL SPECIES OF MEXICAN ACTIMONYCETE WITH ANTI-BIOFILM AND ANTI-QUORUM SENSING ACTIVITY IN ESKAPE BACTERIA

Michelle Chávez-Hernández¹, Jossue Ortiz-Álvarez², Jesús Morales-Jiménez³, Graciela Castro-Escarpulli¹, Lourdes Villa-Tanaca¹, César Hernández-Rodríguez¹

¹Departamento de Microbiología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Prol. de Carpio y Plan de Ayala s/n. Col. Sto. Tomás, 11340 Ciudad de México, México. ²Programa "Investigadoras e Investigadores por México". Consejo Nacional de Humanidades, Ciencias y Tecnologías (CONAHCYT). Av. de los Insurgentes Sur 1582, Crédito Constructor, Benito Juárez, 03940 Ciudad de México, México. ³Departamento el Hombre y su Ambiente, Universidad Autónoma Metropolitana-Xochimilco. Calzada del Hueso 1100, Villa Quietud, Coyoacán, 04960, Ciudad de México, México.

Correspondence author: César Hernández-Rodríguez.

Email: chdez38@hotmail.com

The increase in infection caused by multi-antimicrobial-resistant microorganisms has led to the search for new microorganisms capable of producing new antibiotics. This paper proposes *Streptomyces pakalii* sp. nov. as a new member of the Streptomycetaceae family. The ENCB-J15 strain was isolated from the jungle soil in Palenque National Park, Chiapas, Mexico. The strain formed beige, dry, tough, and buried colonies in the agar with no diffusible pigment in GAE (Glucose-Asparagine-yeast Extract) medium. Scanning electron micrographs showed typical mycelium with long chains of smooth and oval-shaped spores (3-10 µm). *S. pakalii* ENCB-J15 assimilated a wide diversity of organic and inorganic carbon and nitrogen sources, as well as grew at different NaCl concentrations and pH ranges. The strain also exhibited significant inhibitory activity against the synthesis of *Serratia marcescens* prodigiosin and inhibition of biofilm formation and destruction of the ESKAPE strains of *Acinetobacter baumannii* and *Klebsiella pneumoniae*. It has a genome of 7.6 Mb with a high content of G+C (71.6%), 6,833 total genes, and 6,746 genes encoding putative proteins. The phylogeny of the 16S rRNA gene, the core-proteome phylogenomic tree, and fingerprints of the virtual genome support that *S. pakalii* ENCB-J15 is a new species related to *Streptomyces badius* and *Streptomyces globisporus*. Similarly, average nucleotide identity (ANI) (96.4%), average amino acid identity (AAI) (96.06%), and virtual DNA-DNA hybridization (67.3%) provide evidence to recognize the new species. Comparative genomics revealed that *S. pakalii* and its related species maintain a well-conserved genomic synteny. In this work, *Streptomyces pakalii* sp. nov. is proposed as a novel species, exhibiting anti-biofilm and quorum-sensing activity.

EXPRESSION OF SPECIALIZED METABOLITES BY AN OSMAC APPROACH APPLIED TO *STREPTOMYCES*

Uriel Colin Camacho, Corina-Diana Ceapă

Institute of Chemistry, UNAM, Circuito Exterior s/n, C.U., 04510 Ciudad de México, CDMX.

E-mail: corina.ceapa@iquimica.unam.mx

The genus *Streptomyces*, a member of the phylum Actinobacteria, harbors an average of 39 biosynthetic gene clusters (BGCs) responsible for producing diverse specialized microbial metabolites (MEMs). However, many of these BGCs remain dormant or “silent” under laboratory conditions. The “one strain, many compounds” (OSMAC) method has been identified as a promising approach for activating these BGCs, with the potential to facilitate the discovery of novel bioactive compounds.

The primary aim of this study was to extract specialized metabolites from *Streptomyces* using the OSMAC approach. Colonies of *Streptomyces* obtained from environmental soil samples were cultivated, isolated, and purified. Subsequently, these isolates were grown in liquid SCA medium supplemented with 1% ethanol or 3% dimethyl sulfoxide (DMSO). The specialized metabolites were then extracted using ethyl acetate, followed by an assessment of their antimicrobial activity against pathogenic bacteria. A total of nine isolates were obtained, all of which exhibited positive Gram staining and filament structures.

Notably, three isolates displayed enhanced pigment production in a Petri dish assay upon the addition of 1% ethanol or 3% DMSO. From these three isolates, one strain was chosen for further specialized metabolite extraction rounds, which were subsequently subjected to analysis using high-performance liquid chromatography-mass spectrometry (HPLC-MS/MS) to identify the compounds present.

Based on phenotypic traits, it is reasonable to presume that the nine strains are species of the genus *Streptomyces*. However, further analysis, particularly genomic analysis, is imperative to confirm their taxonomic classification. Importantly, the addition of ethanol and DMSO did not exert any adverse effects on the growth of the selected strain.

Future research will encompass the evaluation of the bioactivity of the extracted compounds against pathogens, as well as the bioinformatic analysis of the genomes of the obtained strains.

BACTERIA FROM THE SKIN MICROBIOTA OF THE AXOLOTL AMBYSTOMA ALTAMIRANI SHOW GROWTH-INHIBITORY ACTIVITY AGAINST ANTIBIOTIC-RESISTANT STRAINS OF STAPHYLOCOCCUS AUREUS

Cynthia Margarita Concepción Acosta¹; Perla Jaqueline Vargas Garcia²; Eria Alaide Rebollar Caudillo³;
V́ctor Humberto Bustamante Santillán¹

¹Instituto de Biotecnología, Universidad Nacional Autónoma de México, ²Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos; ³Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México, Avenida Universidad #2001, Colonia Chamilpa, Cuernavaca, Morelos, 7773291627.

E-mail: cynthiamar921105@gmail.com y victor.bustamante@ibt.unam.mx

The World Health Organization (WHO) considers antibiotic resistance (AR) one of the main threats for humanity. In 2019, 1.27 millions of human deaths were attributed to bacteria resistant to antibiotics. Worryingly, it was estimated that by the year 2050 the deaths of humans by this cause could reach 10 million per year worldwide. In Mexico, various studies have shown a high prevalence of antibiotic-resistant bacteria, especially in hospital environments, but also in animals and nature. The WHO published a list of priority pathogens for which, due to their high level of resistance to antibiotics, the identification of new antibacterial compounds is urgent. One of these priority pathogens is *Staphylococcus aureus*, a Gram-positive bacterium that is part of the skin microbiota but is also one of the main agents causing infections in hospitalized patients; it can cause from moderate skin infections to fatal infections such as pneumonia and sepsis. Currently, different sources are being explored for the identification of new antibacterial compounds, such as the microbiota associated to animals. In this project we explored the bacteria associated to the skin of the axolotl *Ambystoma altamirani*, an amphibian endemic of Mexico. In this study, we identified several bacteria from the axolotl skin that exhibit growth-inhibitory activity against reference strains and multidrug-resistant isolates of *S. aureus*, suggesting that the compounds mediating this antibacterial activity could have novel action mechanisms. Our results show that the microbiota of the axolotl *A. altamirani* has a great potential for the identification of new antimicrobial compounds.

CLONING AND OVEREXPRESSION OF FLIC GENE FROM *AZOSPIRILLUM ARGENTINENSE* REC3 IN THE EXPRESSION VECTOR PGEX-4T1

Ana Gabriela Cordova Mejía¹, Raúl Osvaldo Pedraza², María Luisa Xiqui Vázquez¹, Claudia Mancilla Simbro¹, Sandra Reyes Carmona¹, Beatriz Eugenia Baca¹ and Alberto Ramírez-Mata¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla, Av. San Claudio S/N Apartado Postal 72000, Puebla, México. ²Facultad de Agronomía y Zootecnia, Universidad de Tucumán Argentina, Tucumán Argentina.
E-mail: alberto.ramirez@correo.buap.mx

Flagellin is the main structural protein of the flagella found in various bacterial species, including *Azospirillum argentinense* REC3. The flagellin from the polar flagellum of *A. argentinense* REC3 (AzFlap) is a glycoprotein that can activate the innate immunity of strawberry plants and protect against phytopathogens such as the fungus *Macrophomina phaseolina*, which is the causal agent of charcoal rot of roots and crown disease. AzFlap behaves as a Microbe-Associated Molecular Pattern (MAMP). Callose and lignin depositions are histological modifications induced by the bacteria and its flagellin that reinforces plant tissues helping plants to fight against pathogen infection. These traits are coordinated with other responses like stomatal closure, cell wall reinforcement, and ROS accumulation to provide plants with certain protection against diseases. Changes observed on leaves may be triggered by the biosynthesis of Salicylic Acid inducing plant immunity. To elucidate the function of AzFlap, it is essential to characterize the mechanisms by which the protein, or its individual parts, promotes the immune response. In this study, we performed a bioinformatic analysis to determine the number of polar *fliC* genes encoded in the recently sequenced *A. argentinense* REC3 genome. We cloned and overexpressed the two *fliC* genes from *A. argentinense* REC3 in the prokaryotic vector pGEX-4T1 and evaluated their expression in the heterologous system of *Escherichia coli*. We plan to use purified polar flagella protein AzFlap obtained from *A. argentinense* REC3 which contains glycosidic chains, as an elicitor in comparison with the recombinant AzFlap (produced in *E. coli* without glycosidic chains). These assays aim to understand whether the polar flagellin's glycosidic chains or the protein component alone or both are responsible for triggering the immune system of strawberry plants and protecting them against phytopathogens.

IMPACT OF THE OVER EXPRESSION OF A GENE CODIFYING FOR A HYBRID C-DI-GMP REGULATING PROTEIN OF AZOSPIRILLUM BALDANIORUM SP245

David Cortés-Sotres^{1,2}, Sandra Reyes Carmona¹, María Luisa Xiqui Vázquez¹,
Beatriz Eugenia Baca¹ and Alberto Ramírez-Mata¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Posgrado en Ciencias Microbiológicas.
Benemérita Universidad Autónoma de Puebla, Avenida San Claudio S/N. Puebla Pue. México.
E-mail: davidricardo.cortessotres@viep.com.mx

The second bacterial messenger c-di-GMP is synthesized by Diguanylate cyclase domains and degraded by Phosphodiesterase domains, which can be as an alone catalytic domain, or they can be both together domains presented in a single protein, these proteins are named as hybrid c-di-DMP regulating proteins. It has been reported that the two opposite functions diguanylate cyclase (DGC) and phosphodiesterase (PDE) proteins are switched as an on/off system by a ligand binding signalling domain present in these hybrid proteins in the specific case of the protein of our study it is a CHASE domain that would perform that task. In the sequence genome of *Azospirillum baldaniorum*Sp245 there are 35 genes that have been described to codify for c-di-GMP regulating proteins². This work aims to analyse the impact of the over expression of one of those proteins named CdgD codified by the *cdgD* gene on diverse phenotypes.

References

1. Cruz-Perez et al. (2021). Scientific reports , 11, 520.are
2. Mata et al., (2018). BMC Microbiology, 18, 1.

STUDY OF THE SMALL RNA REGULATOR (SRNA) ERS A INVOLVED IN THE SYNTHESIS OF ALGINATES IN AZOTOBACTER VINELANDII

Juan Carlos De Lima Mar¹, Miguel Castañeda-Vaquera, Miguel Castañeda Lucio¹

¹Benemérita Universidad Autónoma de Puebla, Centro de Investigaciones en Ciencias Microbiológicas, Edif. IC11.

Ciudad Universitaria Col. Jardines de San Manuel, Puebla, Puebla. C. P. 72570.

E-mail: juanc.delima.mar@gmail.com Cel: 222 333 7941

The small RNAs (sRNAs) are molecules with a regulatory role in bacteria. They modulate the expression of many genes by base pairing the mRNA promoting or avoiding their translation. ErsA is a small regulatory RNA (sRNA) reported in *Pseudomonas aeruginosa* homologous to the sRNA Spot 42 of *Escherichia coli*, which has been implicated in controlling catabolic repression. Originally ErsA, which was found bioinformatically, was known as Pseudomon-1. Subsequently, Pseudomon-1 was renamed ErsA and was reported to post-transcriptionally negatively regulate the expression of the *algC* gene. A homolog of the *ersA* gene was found in *Azotobacter vinelandii*, a nitrogen-fixing bacterium phylogenetically related to *P. aeruginosa*. In *A. vinelandii* the *algC* transcript is essential for alginate synthesis. Therefore, we originally postulated the hypothesis that in *A. vinelandii*, the *ersA* homolog negatively controlled, at the post-transcriptional level, the expression of the *algC* gene and, thus, alginate synthesis. An *ersA* mutant was generated by allelic exchange, finding, as expected, increased alginate synthesis when the bacterium was grown in a semi-gellified medium. However, we found that in a minimal liquid medium (nitrogen-free) with high aeration (200 rpm), the mutant did not grow; when aeration was decreased (< 50 rpm) or the mutant was grown in a stationary condition, growth recovered. Interestingly, adding a nitrogen source to the culture medium recovered the growth capacity of the mutant even under high aeration conditions. The above suggests that ErsA is essential for the diazotrophic growth of *A. vinelandii* under conditions of high oxygenation, probably participating in the mechanisms of nitrogenase protection.

THE LOSS OF FUNCTION OF THE METHYLTRANSFERASE KMT6 ON *TRICHODERMA ATROVIRIDE* MODIFIES THE EXPRESSION OF SECONDARY METABOLITE GENE CLUSTERS THAT COMPROMISE THE ROOT GRAVITROPISM OF *ARABIDOPSIS*

Saraí Esparza-Reynoso¹, Camilo Pérez-Salazar¹, Pablo Cruz-Morales², Alfredo Herrera Estrella¹

¹UGA-LANGEBIO Cinvestav-Irapuato, Libr. Norte carr. León km 9.6, Irapuato, Gto.

²Novo Nordisk Center for Biosustainability, [Technical University of Denmark](https://www.tu.dk).

E-mail: alfredo.herrera@cinvestav.mx

The filamentous fungus *Trichoderma atroviride* is a common inhabitant of the rhizosphere, which establishes a perdurable symbiosis with plants through the emission of volatiles, diffusible compounds, and successful root colonization. This fungus is known to produce a plethora of secondary metabolites (SMs) and its genome has revealed numerous SM gene clusters that are not expressed under conventional laboratory conditions [1]. Activation of these “silent” gene clusters involves histone modifications such as H3K4 trimethylation (H3K4me3) catalyzed by the DNA methyltransferase KMT6. Knock-down of the methyltransferase KMT6 in *T. atroviride* results in the induction of cryptic and otherwise silent secondary metabolite genes that compromise the gravitropism of *Arabidopsis* root. According to our results, the co-culture with mutant strain $\Delta kmt6$ elicits an agravitropism-like root phenotype caused by abscisic acid (ABA), which involves a wavy growth trajectory/direction and a transient perturbation of the auxin distribution. Besides, the compounds emitted by *Trichoderma* with similar activity to ABA reduce the distribution and abundance of the auxin efflux transporter PIN2 in the root tip and synergistically induce the expression of ABA-INSENSITIVE (ABI) 4 factor, which acts as a central transcription activator of ABA signalling pathway.

References

1. Khan et al. (2020). Bioactive Secondary Metabolites from *Trichoderma* spp. against Phytopathogenic Fungi. *Microorganisms* (6): 817.

THE COMPLEX REGULATION OF SIGMA FACTOR RPO_S PROTEOLYSIS IN *AZOTOBACTER VINELANDII*

Guadalupe Espín, Juliana Rojo and Soledad Moreno

†Instituto de Biotecnología, Universidad Nacional Autónoma de México. Av. Universidad s/n, Col. Chamilpa, C.P. 62219, Cuernavaca Morelos, México.
E-mail: elda.espi@ibt.unam.mx

Characterization of the regulatory mechanisms of RpoS expression and activity is of main interest, as RpoS is one of the most important alternative sigma factors in Gram-negative bacteria, due to its protecting role under stress conditions and in bacterial physiology. In *A. vinelandii* RpoS plays a pivotal function in the synthesis of biopolymers polyhydroxybutyrate and alkylresorcinols, as well in the differentiation process that undergoes this bacterium to develop desiccation resistant cysts(1, 2).

Proteolysis of RpoS in this bacterium, is carried out by the chaperon-protease complex ClpXP in exponentially growing cells, and involves proteins RssB and RssC, as mutations in either *rssB* or *rssC* genes increased the stability of RpoS protein and interaction between RssB and RssC enable RssB to recognize RpoS for its degradation(3).

In *A. vinelandii*, degradation of RpoS is also carried out by the ClpAP complex. This degradation is under the control of the PTS^{Ntr} regulatory system, as in mutant strains carrying a unphosphorylated EII^{Ntr} protein the degradation of RpoS by the ClpA complex occurs(4).

Here, we present results indicating that RpoS proteolysis is also regulated by the global Gac-Rsm regulatory system. This system widely present in Gram-negative bacteria, exerts its control on gene expression at the translational level, where the effector is RsmA, a protein that binds its target mRNAs to inhibit its translation, and GacA activates transcription of RsmZ1 and RsmZ2 that bind RsmA to counteract its repressor activity.

We found that the level of RpoS was reduced in a *gacA* mutant, and that this reduction was neither caused by a reduction in the transcription nor the translation of *rpoS*. We found that in the *gacA* mutant transcription of the *clpP* and *clpX* genes is increased, and the reduction of RpoS was caused by its proteolysis by the ClpXP complex.

References

1. Hernandez-Eligio A, Castellanos M, Moreno S, Espín G. (2011). *Microbiology*, 157, 3014-3023 1693.
2. Cocótl-Yañez M., Sampieri A., Moreno S., Núñez C., Castañeda M., Segura D., Espín G. (2011). *Microbiology*, 157, 1685-1693.
3. Rodríguez-Martínez K, Muriel-Millán LF, Ortíz-Vasco C, Moreno S, Soberón-Chávez G and Espín G. (2023). *Molecular Microbiology*.
4. Muriel-Millán L. F., Moreno S., Gallegos R., Espín, G. (2017). *Molecular Microbiology*, 104, 197-211

SYMBIOSIS INDUCTION BETWEEN *RHIZOBIUM ETLI* AND *SACCHAROMYCES CEREVISIAE*

Ilse Berenice Espinoza Hernández^{1*} and Manuel Gutiérrez Aguilar¹

¹Department of Biochemistry, Facultad de Química,
Universidad Nacional Autónoma de México, México City, 04510, México.

Microorganisms have developed strategies to coexist with other species and obtain mutual benefits. For example, it is known that bacteria of the *Rhizobium* genus can establish endosymbiotic relationships with some plants and are able to form biofilms with the baker's yeast. In the present work, we applied a published method¹ to induce endosymbiotic events between the bacterium *Rhizobium etli* and different yeast strains of *Saccharomyces cerevisiae*. Symbiotic associations were identified between a *Rhizobium etli* strain marked with the GFP protein and *S. cerevisiae* using selective mediums and confocal microscopy, in addition to PCR and Western blotting. We found that the bacterium can grow and form a reddish biofilm in selective mediums for yeasts when co-cultured. The results also show that the interaction between both microorganisms is compatible -and in some cases- events of endosymbiosis can be detected. The coexistence between the two species could be helpful for the development of biomass with low carbon and nitrogen requirements.

References

1. Uribe-Alvarez C, Chiquete-Félix N, Morales-García L, Bohórquez-Hernández A, Delgado-Buenrostro NL, Vaca L, Peña A, Uribe-Carvajal S. *Wolbachia pipientis* grows in *Saccharomyces cerevisiae* evoking early death of the host and deregulation of mitochondrial metabolism. *Microbiologyopen*. 2019 Apr;8(4): e00675.

THE ADMINISTRATION OF A RECOMBINANT TEPARY BEAN (*PHASEOLUS ACUTIFOLIUS*) LECTIN AFFECTS RAT BACTERIOME

Brandon Araujo-Mendoza¹, Roberto Esquivel-García¹, Misael Ríos-Ortiz¹, Dulce Palmerín-Carreño²,
Luis Alberto Ibarra-Sánchez¹, Roberto Álvarez-Martínez¹, Etzel Garrido-Espinoza¹, Carlos Saldaña¹, Ulisses
Moreno-Celis¹, Teresa García-Gasca¹

¹Maestría en Ciencias Biológicas. Facultad de Ciencias Naturales, Universidad Autónoma de Querétaro. ²Ingeniería Química.
Facultad de Química. Universidad Autónoma de Querétaro.
E-mail: tggasca@uaq.edu.mx

Plant lectins are proteins with the capacity for specific recognition of membrane carbohydrates, thus causing cellular effects in animal cells. We studied a concentrated lectin fraction from Tepary bean (TBLF) that induced apoptosis in colon cancer cells and inhibited early colon tumorigenesis^{1,2}. A recombinant Tepary bean lectin (rTBL-1) has been shown to maintain the biological activity, low toxicity profile and adverse effects, however, changes in the bacteriome have not been studied in depth. The objective of the present work was to evaluate the effects of subchronic administration of rTBL-1 at doses of 30 mg/kg body weight on the rat colonic bacteriome. A total of 30 five-week-old male Sprague Dawley rats were used as follows: six rats were sacrificed at the beginning of the experiment (time zero), 12 were administered daily with saline solution via intragastric cannula and 12 with rTBL-1 in saline solution. After 28 days, six animals per group were sacrificed and six animals per group continued the recovery phase of 42 days without treatment. Colonic content samples were collected stored at -80°C until DNA extraction. Bacterial communities were evaluated by massive sequencing of the 16S v3 region and analysed by bioinformatic methods to detect up to genus level of microbial communities. The rat age and the administration of rTBL-1 showed significant changes on the rat colonic bacteriome. The presence of bacterial genera associated to colonic damage and poor health were detected after the administration of rTBL-1 (*Prevotella*, *Colidextribacter*, *Oscillibacter* and *Ruminococcus*) and subsequent recovery of beneficial genera (*Alloprevotella*, *Faecalibacterium*) after six weeks without treatment was observed. The presence of bacterial genera associated to colonic damage it may also be associated to adverse effects on intestinal tissue, mainly atrophy. It is relevant to note that such colonic damage may be reversible, which must be considered to define the administration scheme in colon cancer treatment evaluation. It is important to continue with the study of metabolic inference using bioinformatic and analytical techniques to detect the presence of possible metabolites that act as indicator factors of poor intestinal health.

Keywords: Bacteriome, lectins, *Phaseolus acutifolus*, Tepary bean.

References

1. Moreno-Celis U, López-Martínez J, Blanco-Labra A, Cervantes-Jiménez R, ... García-Gasca T. 2017. *Molecules*. 22, 1670; doi:10.3390/molecules22101670.
2. Moreno-Celis U, López-Martínez FJ, Cervantes-Jiménez R, Ferríz-Martínez RA, Blanco-Labra A, García-Gasca T. 2020. *Molecules*. 25 1021; doi:10.3390/molecules25051021.
3. Dena-Beltrán, J. L., Nava-Domínguez, P., Palmerín-Carreño, D., Martínez-Alarcón, D. ,... García-Gasca, T. 2023. *Pharmaceuticals*. 16(2) <https://doi.org/10.3390/ph16020290>

ISOLATION OF PATHOGENS FROM FOOD AND ASSESSMENT OF THEIR ANTIMICROBIAL RESISTANCE

Maricela Esteban Méndez¹, Laura Silvia González Valdez¹,
Petra Laura Calzada Contreras¹, Abelardo Camacho Luis²

¹Interdisciplinary Research Center for the Integral Regional Development – Durango, National Polytechnic Institute. Sigma 119 Fracc. 20 de noviembre II. Durango, Dgo. México. C. P. 34220. ²Juarez University of the State of Durango, Faculty of Medicine and Nutrition. Av. Universidad y Fanny Anitúa s/n. C.P. 34000. Durango, Dgo. México. mesteban@ipn.mx

The Antimicrobial resistance (AMR) poses a major global threat to people, animals, plants, food and the environment (1), In 2019, it was estimated that around five million deaths were associated with RAM. The World Health Organization (WHO) published the antibiotic-resistant pathogenic microorganisms considered to be the most dangerous bacteria to health. AMR has a considerable cost for the economy and health systems as it affects the productivity of patients and their caregivers who, by prolonging hospital stays, require more expensive and intensive care. The critical priority group includes multidrug-resistant bacteria such as *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, *Enterococcus faecium*, *Staphylococcus aureus* and Enterobacteriaceae such as *Klebsiella spp*, *Escherichia coli*, *Salmonella sp* and *Proteus sp*, which can be lethal and cause infections in the bloodstream, pneumonia, gastroenteritis, some are related to poisoning food; caused by foods of animal or vegetable origin (2). The objective of this work was to analyze samples of food for human consumption to identify the pathogens present and know their AMR, the identification of pathogens was according to NOM-210-SSA1-2014 and AMR by the Kirby-Bauer method. A total of 150 food samples were analyzed and found ten strains of *E. coli*, five *Salmonella spp*, seven of *Proteus mirabilis* and four *Enterobacter aerogenes*. Of these strains evaluated for AMR two were resistant to antibiotics.

References.

1. Laxminarayan R, Sridhar D, Blaser M, Wang M, Woolhouse M. 2016. Science 353(6302):874-5. doi:10.1126/science.aaf9286.
2. <https://www.who.int/es/news/item/17-05-2024-who-updates-list-of-drug-resistant-bacteria-most-threatening-to-human-health>.

ISOLATION AND MORPHOLOGICAL CHARACTERIZATION OF SPOILAGE MOLDS OF STRAWBERRY (*FRAGARIA X ANANASSA DUCHENSE*) MARKETED IN DURANGO

Estrada-Rodriguez C.A.¹, Rojas-Contreras J.A.¹, Morales-Castro, J.¹, Rosas-Flores, W.¹, Guerra-Rosas M.I.¹

¹TecNM/Instituto Tecnológico de Durango. UPIDET. Blvd. Felipe Pescador 1830 Ote. 34080 Durango, Dgo. México.

*16040097@itdurango.edu.mx

Every year, globally, high quantities of food are lost due to spoilage caused mainly by the presence of microorganisms such as bacteria, mold, and yeast. Vegetable foods have a relatively short shelf life between 3 and 7 days. It is necessary to isolate and identify spoilage microorganisms to apply a method that retards their development. Therefore, the objective of this research was to isolate and identify spoilage molds from strawberries marketed in Durango. Deteriorated strawberries (*Fragaria x ananassa* Duchense) were collected from the local market "El Refugio" in Durango, Dgo. 1 cm³ sections were taken and placed directly in the center of Petri dishes with YPD medium with kanamycin, and incubated at 28±2 °C for 72 h. Subsequently, by morphological differentiation, each strain was reseeded by the cross-streaking method until axenic cultures were obtained. For microscopic observation of the fungal structures, the lactophenol blue technique was used, using a 40x objective. Four different axenic cultures of molds were isolated and obtained, coded as HF1, HF2, HF3 and HF4. According to the macroscopic characterization, strain HF1 presented white colonies with cotton-like appearance, slightly textured, with marked volume and defined edges, although difficult to manipulate. Strain HF2 showed white colonies with cotton-like volume and texture, with irregular edges resembling long filaments. Strain HF3 developed white colonies with cottony texture and uniform edges formed by long filaments, with a slight brownish hue. As for strain HF4, the colonies, also white and with a cotton-like texture, presented a marked volume and edges with light filaments, being also difficult to manipulate. Microscopic characterization showed that strain HF1 had elongated hyphae with visible septa. On the other hand, strain HF2 shows a combination of septate and non-septate hyphae in its fruiting structure. For strain HF3, it is characterized by fully septate hyphae, reduced length, and rectangular margins. Finally, strain HF4 has thin, septate hyphae with short sections that are spaced apart. The characterization of these strains contributes to the fundamental knowledge about the microorganisms associated with strawberry spoilage and provides information to understand their role and, therefore, to develop effective post-harvest management strategies to extend their shelf life.

EFFECTS OF CHITOSAN ON GROWTH OF FUNGAL SPECIES INVOLVED IN BUILDING BIODETERIORATION

Joshua Fernández Jasso¹, Guadalupe Guerra Sánchez², Darío Rafael Olicón-Hernández³

Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Departamento de Microbiología, Laboratorio de Bioquímica y Biotecnología de Hongos. Manuel Carpio, Plutarco Elías Calles, Miguel Hidalgo, 11350 Ciudad de México, CDMX.
E-mail: doliconh@ipn.mx

Biodeterioration is an undesirable alteration process caused by the action of biological agents. The phenomena of physical and chemical biodeterioration are caused by the growth of very different organisms, generically called "biodeteriogens"¹. Chitosan is a linear polysaccharide composed of variable amounts of (1-4) linked residues of N-acetyl-2 amino-2-deoxy D-glucose (glucosamine, GlcN) and 2-amino-2-deoxy-D-glucose residues (N-acetyl-glucosamine, GlcNAc). Chitosan is the only polycation in nature, and its charge density depends on the degree of acetylation and the pH of the medium. Chitosan exhibits antimicrobial activity against different microorganisms, including bacteria, filamentous fungi, and yeasts. Some studies point to the reduction of cell membrane permeability due to the polymer coating on the cell surface, which blocks cell access to nutrients. This process occurs due to the interaction of the NH₂ groups of the chitosan chains with the COO⁻ groups on the outer cell membranes of microorganisms, therefore, antimicrobial activity depends on the degree of acetylation². Chitosan demonstrated potential results for the inhibition of several fungi and could be effective against biodeterioration fungi. The aim of this work was to evaluate the effect of different chitosan concentrations on the radial growth of fungi isolated from biodeterioration areas. Fungi were collected from various surfaces using sponges moistened in 0.5% saline solution for cultivation on potato dextrose agar (PDA) using the imprint technique. From the sample, ten fungi were selected to be cultivated axenically and their morphological characterization was performed both macro and microscopically. Finally, an inhibition assay was conducted using low molecular weight chitosan in concentrations of 1%, 3%, and 5% to evaluate its antifungal effect using the diameter of fungal growth as the criteria. As results, in the isolated fungi, suspect genera such as *Penicillium*, *Cladosporium*, and *Aspergillus* were identified. It was found that the inhibition of the growth of these fungi begins with a chitosan concentration of 3%. However, in some isolates, a stimulation of fungal growth was observed at the same concentration. By increasing the chitosan concentration to 5%, significant inhibition of growth was achieved in all studied fungi. We concluded that chitosan inhibits the radial growth of biodeteriorating fungi depending on its concentration.

References

1. Bobu, E; Nicu, R; Obrocea, P; Ardelen, E; Dunca, S. y Balaes, T. (2016). Cellulose chemistry and Technology, 50, 689-699.
2. Aranaz, I.; Alcántara, A.R.; Civera, M.C.; Arias, C.; Elorza, B.; Heras Caballero, A.; Acosta, N. (2021). Polymers, 13, 3256.

ANALYSIS OF *bla*_{OXA-72} GENE REGULATORY REGION IN *ACINETOBACTER BAUMANNII*

Diana Flores Percino¹, Ana Karen Avendaño Gutiérrez¹, Miguel Castañeda Lucio¹ and Patricia Lozano Zarain¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla, CU, Edificio IC11, 24 sur y Av San Claudio. Colonia San Manuel. CP. 72570, Heroica Puebla de Zaragoza, Pue, México.

E-mail: diana.florespercino@viep.com.mx

Acinetobacter baumannii is a bacterium of public health interest and one of the most important causative agents of healthcare-associated infections (HAI). Indeed, in Mexico, it has been considered the fourth most common causative agent of HAI in recent years. Additionally, this bacterium belongs to the ESKAPE group as it can escape the action of antibiotics through genetic mutations or the acquisition of resistance genes using mobile genetic elements, which have allowed them to develop resistance mechanisms to multiple families of these drugs, among which we find carbapenems. Currently, there is a high incidence of *A. baumannii* strains resistant to carbapenems; these antibiotics are the last line of treatment, and of recent formulation, resistance to them has been mainly mediated by carbapenems hydrolyzing proteins, among which we find the oxacillinase-type enzymes (OXA). OXA belonging to the OXA-24/40 family are widely distributed worldwide, being reported in Asian, European and American regions. Specifically, one of the members, OXA-72, is considered one of the most critical determinants of resistance to carbapenems in Mexico in recent years. It has been reported that the upstream association of OXA genes with insertion sequences of the ISAb_a type favors the overexpression of these resistance determinants, contributing to an increase in resistance to carbapenems due to the possible participation of a strong promoter conferred by the ISAb_as. However, the regulatory regions involved in this phenomenon have not been studied, nor their exact position, either upstream of the ISAb_as sequence, within it, or in the intergenic region between this determinant and *bla*_{OXA-72}. Thus, our team analyzed by bioinformatics tools the possible promoters that would affect the transcription of *bla*_{OXA-72} in association with ISAb_a-48 in carbapenem-resistant *A. baumannii* strains isolated in a Mexican hospital. Additionally, vectors derived from the RSF1010 plasmid, reported to be stable in *A. baumannii*, were constructed. These vectors contain the *lacZ* reporter and are useful for measuring gene expression in *A. baumannii*, for which no tools have been useful to date; so they will serve to test the participation of these putative regulatory regions determined by the previous bioinformatics analysis.

EFFECT OF IRON ON THE GROWTH AND VIRULENCE OF *PSEUDOMONAS SYRINGAE PV. PHASEOLICOLA* NPS3121

Kenia Franquez Gómez¹, Jackeline Lizzeta Arvizu-Gómez², Alejandro Hernández-Morales³,
Christian González-Reyes⁴, Angélica Nallehy Rodríguez-Ocampo⁴,
Juan Campos-Guillén⁵ and Juan Ramiro Pacheco-Aguilar⁵

¹Unidad Académica de Agricultura. Universidad Autónoma de Nayarit. ²Secretaría de Investigación y Posgrado. CENITT. Universidad Autónoma de Nayarit. ³Facultad de Estudios Profesionales Zona Huasteca. Universidad Autónoma de San Luis Potosí. ⁴Unidad Académica de Ciencias Químico Biológico y Farmacéuticas. Universidad Autónoma de Nayarit. ⁵Facultad de Química. Universidad Autónoma de Querétaro.
E-mail: 20014653@uan.edu.mx; jackeline.arvizu@uan.edu.mx

P. syringae pv. phaseolicola, the causal agent of the disease “Halo blight” on bean, produces a toxin known as phaseolotoxin, which is considered as one of the major virulence factors of this bacterium and one key element for disease development¹. So far, the knowledge about the cues and/or elements belonging to signal transduction pathway of phaseolotoxin synthesis is still scarce. Previous works have suggested the iron as a element with potential influence in the physiology and virulence of *P. syringae pv. phaseolicola*². The influence of iron on diverse bacteria has showed to be specific in function of the genera and even it is differential among pathovars. Based on the above, the goal of this study was to evaluate the influence of the iron ion (FeCl₃) in the growth and phaseolotoxin production in *P. syringae pv. phaseolicola* NPS3121. The analysis of the growth kinetics performed under different iron concentrations and in function of the temperature (28 °C vs 18 °C) showed a positive effect of the iron ion in the growth rate of the bacteria. Likewise, the *in vitro* phaseolotoxin assays demonstrated greater production of phaseolotoxin in cultures growth in presence of iron. These results demonstrated that iron is part of the signal transductional pathway related with phaseolotoxin synthesis in *P. syringae pv. phaseolicola* NPS3121.

References

1. Bender, C.L.; Alarcón-Chaidez, F.; Gross, D.C. (1999). *Microbiol Mol Biol Rev*, 63, 266-92.
2. González-Villanueva, L.; Arvizu-Gómez, J.L.; Hernández-Morales, A.; Aguilera-Aguirre, S.; Álvarez-Morales, A. (2014). *Microbiol Res*, 169, 221-31

GENETIC VARIABILITY OF TUBERCULOSIS COMPLEX IN LIVESTOCK FARMERS IN OAXACA

Heidi Paola Galván Antonio¹, Mario Alfredo Urbina Mata², José Ángel Cueva Villanueva²,
Leobardo Reyes Velasco², Gabriel Sánchez Cruz², Ericel Hernández García²

¹Facultad de Medicina y Cirugía de la Universidad Autónoma Benito Juárez de Oaxaca, ²Facultad de Ciencias Químicas de la Universidad Autónoma Benito Juárez de Oaxaca, Avenida Universidad S/N Colonia Cinco Señores, C. P. 68120, Oaxaca de Juárez, Oaxaca.
E-mail: ericledimp@gmail.com

Tuberculosis is a chronic infectious disease caused by bacteria belonging to the Mycobacterium tuberculosis Complex that, in addition to affecting humans, also affects different animals. In the state of Oaxaca, bovine tuberculosis is considered a public and livestock health problem due to the high incidence of tuberculosis and the strong impact it has on the economy and zoonotic transmission to humans. The objective of this thesis was to carry out a molecular identification of the strains of the Mycobacterium tuberculosis Complex in livestock farmers in the state of Oaxaca, in order to understand the existing prevalence and genetic diversity that the bacteria present in this population and thus provide useful information to improve control and prevention strategies for this disease. Samples were collected from ranchers from different regions of Oaxaca and the Polymerase Chain Reaction (PCR) technique was used in its end-point modality to amplify and detect the Rv1818c gene, which is specific to the Mycobacterium tuberculosis Complex. After this, sequencing of the amplified DNA was carried out by the automated Sanger method to obtain the sequences that would allow the corresponding phylogenetic analysis to be carried out and thus be able to analyze the genetic diversity and determine the presence of the strains. A prevalence of 16.6% was found, since nine positive individuals were identified from a total of 54 ranchers. Three mutations were determined T524A, C939T and A1171C that are specific for Oaxacan samples, since no other sequence in the world presents them, these sequences can function as geographic molecular markers and to determine their virulence, further studies are required. By providing valuable information on the genetic diversity of the Mycobacterium tuberculosis Complex in the state's livestock farmers, the findings of this research can have a significant impact on decision-making to improve public health in Oaxaca as this can contribute to the implementation of more effective strategies for the prevention, diagnosis and treatment of tuberculosis, helping to reduce the burden of this disease in the population and improve community health.

POINT MUTATIONS IN *SALMONELLA* TYPHIMURIUM INV F TRANSCRIPTIONAL REGULATOR AFFECT ITS FUNCTION

Laura E. García-Cortés¹, Daniel Cortés-Avalos¹, Abigail Roldan-Salgado², Paul Gaytan² and J. Antonio Ibarra¹

¹Departamento de Microbiología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Prol. Carpio y Plan de Ayala S/N, Col. Santo Tomás 11340, Mexico City, Mexico. ²Instituto de Biotecnología, Universidad Nacional Autónoma de México, Av. Universidad 2001, Col. Chamilpa 62210, Cuernavaca, Mexico.
E-mail: laura.egc.03@gmail.com

Salmonella enterica is known to be a causative agent of food- and water-borne illnesses. The virulence of *S. enterica* ser. Typhimurium depends on two type III secretion systems (T3SS-1 and T3SS-2); T3SS-1 is required to initiate intestinal infection while T3SS-2 is required for systemic infection. The regulation of virulence genes ensures that these T3SSs are appropriately expressed during infection. InvF is a transcriptional regulator of the AraC/XylS family and is important for the expression of invasion genes in *Salmonella* by acting as a classical regulator. InvF interacts with SicA and with the alpha subunit of RNA polymerase, but it is unknown through which amino acid residues it does so. The purpose of this project is to generate InvF point mutants and determine their effect on protein-protein and DNA protein interactions. Mutants were obtained with three different strategies (alignment with other proteins of the family, molecular docking and by the spiked genes procedure). Therefore, the point mutants R14A, K230A, S171A-K230A, I148V, I148V-G184A-P222L, ΔI46W-I148V-E166K-G185R and I148V-I229T were generated. Most of these residues are in a conserved region at the C-terminal of the AraC/XylS, where it has been reported that other members of the same family present important DNA binding sites in this region. To evaluate the effect of these mutants, the expression of SopB-FLAG was detected. Results showed that R14A at the N-terminal region is not important for *sopB* expression, suggesting that this residue is not involved on neither protein-protein interactions or correct folding of both domains; while residues P222L, I229T and K230 seem to be important for *sopB* expression. Both of these point mutations are located in the DNA binding domain and it is possible that this is affected, although a 3D model predicted by our group suggest that this domain is also involved in protein-protein interactions. The effect of the mutants on the InvF-SicA and InvF-RpoA interactions is being evaluated, as well as in assays to determine the DNA-protein interaction to determine how these intermolecular changes affect the function of this transcriptional regulator in *Salmonella*.

INTERACTION OF *WICKERHAMOMYCES ANOMALUS* WITH FUNGI ISOLATED FROM CROPS OF ECONOMIC INTEREST

Karina García-Gutiérrez, Adriana Jazmín Legorreta-Castañeda,
Darío Rafael Olicón-Hernández, María Guadalupe Guerra Sánchez

Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Laboratorio de Bioquímica y Biotecnología de Hongos.
Prolongación de Carpio y Plan de Ayala sin número, Col. Santo Tomás, Alcaldía Miguel Hidalgo, CP 11340, CdMx.
E-mail: lupesg@hotmail.com

Most crops are treated with Captan, a fungicide that controls a wide range of soil and foliage phytopathogenic fungi on ornamentals, vegetables, and fruits. The most common diseases that affect crops are anthracnose, fusariosis, black spots, leaf spots, mildew, gray mold, scabs, and blights. It has been shown that contact with Captan can have negative effects on the eyes and skin and, if inhaled, causes an increase of tumors in the intestine after exposure to high doses, as has been shown in laboratory rats during their life. To avoid damage from using chemical substances in the field to control fungal infections, biological control alternatives are sought. It has been reported that the yeast *Wickerhamomyces anomalus* can inhibit the growth of phytopathogenic fungi without negatively affecting humans, other mammals, or the environment. In the present work, we isolated different filamentous fungi in the fields of Irapuato, Guanajuato, from different crops such as beans, chili, tomato, potato, and sunflower, and subjected them to interaction with the yeast *W. anomalus*, to evaluate its effect on the control of the growth of the phytopathogens. The fungi were grown in fungus-yeast co-culture in a liquid medium of yeast extract peptone-dextrose (YPD) at the same cell concentration for 72 hours, and aliquots were taken every 24 hours to observe the interaction between the isolated filamentous fungi and *W. anomalus*. It was evident that the yeast inhibited the growth of some fungal isolates, while others remained resistant to the action of *W. anomalus*. During the interaction of *W. anomalus* and the sensitive fungi, severe damage to the cell wall was observed in the phytopathogens producing glucanases. Additionally, different profiles of metabolites generated during the phytopathogen-yeast interaction were obtained. This study presents the yeast *W. anomalus* as an alternative to control phytopathogen fungi in crops instead of the use of synthetic chemicals that contaminate soils and fruits and damage the environment with possible harm to human health.

EFFECT OF FLAVONOID COMPOUNDS ON THE MULTIDRUG-RESISTANT YEAST *CANDIDA AURIS*

Jonathan García Hernández¹, Lourdes Villa-Tanaca¹, Dulce Andrade- Pavón¹

¹Escuela Nacional de Ciencias Biológicas del Instituto Politécnico Nacional. Plan de Ayala y Prol. Carpio,
Col. Casco de Santo Tomás, CP 11340, Ciudad de México. Cel: 2741118744.
E-mail:dandradep@ipn.mx.

Fungal infections, among which those caused by yeasts belonging to the genus *Candida* are an important cause of morbidity and mortality in hospitalized patients with an immunocompromised immune system. In recent years, the arrival of the new opportunistic pathogenic species in humans called *C. auris* has destabilized health units worldwide because it is considered a multi-resistant organism to antifungal treatment with the main classes of conventional antifungals. The antifungal treatments of choice mainly include azoles, polyenes and echinocandins, which has led to the need to propose new compounds, different from those already known. Flavonoids may be a new treatment alternative, as they are compounds present in natural sources and with potential applications in the medicinal field, among which their antifungal activity stands out. For this reason, in this work the effect of flavonoid compounds on growth, viability, synergy and toxicity in two strains of *C. auris* (clade III and IV) was evaluated. The results showed that the flavonoid compounds inhibited growth with lower MIC values than those reported for fluconazole, and also showed a decrease in viability for 24 hours when the results were compared with those obtained for some reference compounds. A synergistic effect was observed against some reference antifungal compounds as well as lower toxicity in the model a *Galleria mellonella*, suggesting that such compounds could be an alternative against infections caused by *C. auris*.

References

1. Ayala-Gaytán, Montoya, Martínez-Resendez, Guajardo Lara (2021). *Infection*, 49, 523-525.
2. Casimiro-Ramos, Bautista-Crescencio, Vidal Montiel, González (2024). *Journal of Fungi*, 10, 392.

STUDY OF THE ABCG TRANSPORTER FAMILY OF *METARHIZIUM GUIZHOUENSE* HA11-2 DURING ITS MYCORRHIZAL ASSOCIATION

Víctor Manuel García Vera, Gloria Angélica González Hernández,
Juan Carlos Torres Guzmán, Israel Enrique Padilla Guerrero

Departamento de Biología, División de Ciencias Naturales y Exactas. Universidad de Guanajuato.
Noria Alta S/N; CP. 36050; Guanajuato, Gto.
Teléfono: 461 114 75 97, E-mailpadillaguerrero@ugto.mx

Mycorrhizae form a dense filamentous network that extracts water and nutrients from deep soil and delivers them to plant roots, thereby accelerating plant growth and root development [1]. In addition to providing nutrients to plants, mycorrhizal fungi also protect plants from drought and salinity, pests and pathogens, heavy metal toxicity, and extreme environmental conditions [2]. In exchange for all these benefits, host plants provide mycorrhizal fungi with sugars and/or lipids [3]. A possible transport mechanism in the mycorrhizal association involves the use of ABCG transporters, which are in the plasma membrane [4]. These compounds are strong candidates for substrate transport in mycorrhizal interactions. In plants, these transporters are related to cuticle formation, defense mechanisms, and hormonal transport [5]. However, in mycorrhizal fungi, the function of these fungi in mycorrhization is unknown.

In this work, through bioinformatic analysis, we identified eleven transporters with characteristic domains of the ABCG family in the genome of the fungus *Metarhizium guizhouense* HA11-2, nine of the PDR type and two WBCs. Transcriptomic studies using RNA-Seq in *Metarhizium guizhouense* HA11-2 revealed high expression of some ABCG transporters during fungus–plant association. Real-time qPCR analysis under different growth conditions revealed that the two semitransporters, WBC1 and WBC2, are expressed when *Metarhizium* interacts with sorghum roots, which confirms the transcriptomics results previously obtained. In plants, this type of semitransporter WBC is related to the transport of different compounds in the rhizosphere, in addition to participating in the regulation of biotic and abiotic stress. To obtain more information on this type of semitransporter in *M. guizhouense*, a group of mutant strains was generated, which included a null mutant in the WBC2 gene, a reintegrating strain of the WBC2 gene, an overexpressing strain of the WBC2 gene, a strain for cellular localization of the WBC2 transporter and a strain that contains the GFP reporter gene under the promoter of the WBC2 gene. This group of WBC2 mutants was evaluated for root colonization and the presence of compounds from root exudates, to determine the functionality of this type of transporter in the mycorrhization process.

References

1. Shikha and Sudhakara., 2012. [2] Smith and Smith., 2015. [3] Joske R. et al., 2021. [4] Christian M. et al., 2009. [5] Lefèvre F. et al., 2018.

METAGENOMIC SEQUENCING OF RESPIRATORY SYNCYTIAL VIRUS FROM ISOLATES IN NORTHEASTERN MEXICO

Julio César Garza-Cabello¹, Alí Fernando Ruiz-Higareda¹, Kame Alberto Galán-Huerta¹, Paola Bocanegra-Ibarias², Adrián Camacho-Ortiz², Ana M. Rivas-Estilla¹

¹Departamento de Bioquímica y Medicina Molecular, Facultad de Medicina, Universidad Autónoma de Nuevo León. ²Servicio de Infectología, Facultad de Medicina, Universidad Autónoma de Nuevo León. Av. Francisco I Madero S/N col. Mitras Centro, 64460 Monterrey, N.L.
E-mail: ana.rivasst@uanl.edu.mx

Introduction. Respiratory syncytial virus (RSV) types A and B are viruses belonging to the genus Orthopneumovirus, family Pneumoviridae. The virus causes severe lower respiratory tract illnesses in infants, children, and elderly adults, including bronchiolitis and pneumonia.

Despite its impact, there is a scarcity of molecular epidemiological data on RSV in Mexico. Whole genome sequencing, offers an opportunity to enhance strain variability resolution for epidemiological surveillance and advance antiviral and vaccine development.

Objective. Characterize the genomic sequence of Respiratory Syncytial Virus (RSV) isolated from patients in northeastern Mexico using a metagenomic approach with Sequence Independent Single Primer Amplification (SISPA).

Methodology. Samples from patients with respiratory disease were collected at “Dr. José Eleuterio Gonzalez” University Hospital at UANL. Viral RNA was isolated and amplified using SISPA, followed by reverse transcription and double-strand synthesis. Sequencing was conducted on the MinION sequencer. The genome was assembled with Minimap2 using a reference genome.

Results. One complete and two partial genomic sequences were successfully obtained and analyzed, revealing it to be RSV type A, belonging to the A.D.1 clade. Phylogenetic analysis demonstrated a notable genetic relationship between the Mexican isolate and previously reported isolates from the United States. This finding suggests potential epidemiological connections and virus migration patterns between these regions.

Conclusions. This study characterized RSV isolates from northeastern Mexico using SISPA. These findings underscore the importance of continuous genomic surveillance and international collaboration to understand RSV’s epidemiology and develop effective prevention strategies.

IDENTIFICATION OF SR PROTEINS IN *USTILAGO MAYDIS*

Mayra Patricia Gaspariano Cholula, Nancy Martínez Montiel, Rebeca D. Martínez Contreras

Benemérita Universidad Autónoma de Puebla; Instituto de Ciencias Microbiológicas, Laboratorio de Biología Molecular, Edif. IC11 2do. Piso, Ciudad Universitaria. Col. San Manuel, México, Puebla, Puebla. C.P. 72570.

Tel (222)229-5500 ext 2529, rebeca.martinez@correo.buap.mx

Serine/arginine-rich proteins (SR proteins) constitute a family of RNA-binding proteins (RBPs) that are present in all metazoans, plants and some fungi. The first SR protein identified in Human was SRSF1 and to date more than 30 proteins have been identified. They are characterized by having one or two RNA binding domains (RRM) at the N-terminus and domain rich >40% of repeated arginine/serine dipeptides at the C-terminus, they have the ability to interact with ESE regions (exonic splicing enhancers) and participate in *spliceosome* assembly. Due to those characteristics, they are involved with different processes, acting as regulators of alternative splicing (AS), expanding the diversity of the proteome. It has been estimated that between 90-95% of multiexon transcripts in humans have from AS. In addition, it participates in post-splicing activities such as nuclear export of mRNA, mechanisms of mRNA degradation: NMD (nonsense-mediated mRNA decay), providing stability to the genome and also participating in the translation of the mRNA.

In general, very few SR proteins have been reported in fungi; for *Schizosaccharomyces pombe*, two SR proteins have been reported: Srp1 and Srp2, in *Candida albicans* Slr was reported and in *S. cerevisiae* Npl3. In this work we identified a total of 54 putative loci corresponding to homologs of human SR proteins in the basidiomycete *Ustilago maydis*, including both classical SR proteins, SR-like and factors related to SR proteins. The homologs for classical SR proteins were further analyzed, their expression was verified, and predictive interaction assays were performed in silico, showing a high homology with humans, as well as their ability to interact with proteins that intimately participate in processes associated with the metabolism of the RNA. The identification of these homologs in *Ustilago maydis* suggests that the regulation of alternative splicing occurs in a fine manner in this basidiomycete and constitutes a novel finding since this family of proteins is barely described in fungi.

Keywords: SR protein, alternative *splicing*, *spliceosome*, mRNA regulation, ESE.

RELEVANCE OF PROTEIN O-GLYCOSYLATION DURING THE INTERACTION OF *SPOROTHRIX SCHENCKII* WITH THE HOST

Manuela Gómez Gaviria¹, José Ascención Martínez Álvarez¹, Iván Martínez Duncker²,
Andrea Regina de Souza Baptista³, Héctor Manuel Mora Montes¹

¹Departamento de Biología, División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Guanajuato, Mexico. Noria Alta S/N, Guanajuato, Gto; C.P. 36050, ²Laboratorio de Glicobiología Humana y Diagnóstico Molecular, Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Cuernavaca 62209, Mor., Mexico; C.P.62209, ³Center for Microorganism' Research, Biomedical Institute, Fluminense Federal University, Campus Valonguinho-Alameda Barros Terra, S/N, Niterói, Rio de Janeiro, Brazil; C.P.24020-150.
E-mail: hmora@ugto.mx

Sporothrix schenckii is a globally distributed pathogenic fungus and one of the aetiological agents of sporotrichosis. It has a cell wall composed of chitin, β -glucans, and glycoproteins modified with *N*- and *O*-glycans. In pathogenic fungi, the wall is the first point of contact with the host. Therefore, its composition has been studied to obtain information on the molecular basis of pathogen-host interactions. Protein *O*-glycosylation has been studied in medically important fungi, and it is known that this pathway is mediated by two gene families, *PMT* and *MNT*. In *S. Schenckii*, the *MNT* family was characterized, and bioinformatics evidence indicates that the *PMT* gene family is also present in this organism. Therefore, we assessed the relevance of protein *O*-glycosylation during the interaction of *S. schenckii* with the host, focusing on the silencing of *MNT1* and *PMT2* genes.

The technique used to study these genes was RNAi-mediated silencing. After obtaining the silencing vectors, *Agrobacterium tumefaciens*-mediated transformation was performed, followed by transformation into *S. schenckii*. The molecular characterization of the silencing mutants for *MNT1* and *PMT2* genes was carried out using RT-qPCR, which identified mutants with different silencing levels and a single insertional event. Morphological analyses of the selected mutants revealed defects in cell morphology in yeast cells and mycelium, but these defects differed in the *MNT1* and *PMT2* mutants. When interacting with human peripheral blood mononuclear cells, mutants of both genes had a reduced ability to stimulate TNF α and IL-6; however, *PMT2* mutants stimulated higher levels of IL-10. Interaction with human monocyte-derived macrophages was also altered, increased phagocytosis was observed in mutants of both genes, but *PMT2* mutants were more phagocytosed. Also, survival assays using the *Galleria mellonella* model indicated that silencing of both genes affects the ability of *S. schenckii* to kill the host. For *PMT2* mutants, 60-70% of individuals survived when mutants with medium-high silencing levels were injected. For *MNT1* mutants 60% of individuals survived. Our data demonstrate that *MNT1* and *PMT2* silencing affects different aspects of the *S. schenckii* interaction with the host.

This work was supported by Consejo Nacional de Ciencia y Tecnología (Ciencia de Frontera 2019-6380), and Red Temática Glicociencia en Salud (CONACYT-México).

STRUCTURAL STUDY OF TLA-1 β -LACTAMASE IN COMPLEX WITH TAZOBACTAM

César Alberto González Guzmán¹, Enrique Rudiño Piñera¹

¹Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología / UNAM, Av. Universidad # 2001, Col. Chamilpa, Cuernavaca, Morelos, C. P., 62210. Tel +52 1 777 191 9575..
E-mail: enrique.rudino@ibt.unam.mx

The great survival capacity of microorganisms has led humanity to face a significant challenge: antimicrobial resistance. In Mexico, a multidrug-resistant strain of *Escherichia coli* was isolated¹, which contains the β -lactamase TLA-1, an enzyme capable of degrading a wide range of antibiotics². The structure of TLA-1 in complex with clavulanic acid was determined, revealing two inhibitor intermediates bound to Ser70 and Ser237, a phenomenon not previously reported³. Currently, there are no other TLA-1 structures in complex with other frequently used inhibitors such as tazobactam. Therefore, this project focused on obtaining the crystallographic model of TLA-1 in complex with tazobactam and observing if this complex exhibits the same behavior as with clavulanic acid. TLA-1 was expressed in a recombinant *E. coli* strain and purified using ion exchange chromatography. TLA-1 crystals were obtained via the sitting-drop method, and the crystals were soaked with tazobactam. Subsequently, the crystals soaked were diffracted at the BL13-Xaloc beamline (Alba), and a dataset was processed using various programs (Suite XDS, Phaser, Refmac5, Phenix, Coot) to obtain the crystallographic model. The crystallographic structure of TLA-1 in complex with tazobactam was determined (2.22 Å, $R_{\text{work}}=0.19$, $R_{\text{free}}=0.23$), with the *trans*-enamine intermediate acylated at Ser70. An important finding is the apparent preference of the residues Trp105, Arg132, and His170 for the sulfate group in the *apo* structure (PDB: 6NVT) or sulfonamide in the structure of TLA-1 in complex with tazobactam. However, these residues are closer to the sulfate group (*apo*) than the sulfonamide group (TLA-1 + TZB), suggesting that interactions are crucial only for ligand-receptor recognition. Regarding Ser237, the electron density was fitted to a sulfate molecule instead of another molecule of tazobactam. Probably, tazobactam has no affinity for Ser237, or the sulfate that is present during crystallization conditions may have greater affinity for Ser237 than tazobactam (but less than clavulanic acid). Nevertheless, the results do not rule out the possibility of tazobactam binding to the second catalytic serine, although further crystallization conditions need to be explored. The results of this project provide the crystallographic model of the TLA-1 in complex with tazobactam, where the *trans*-enamine intermediate of tazobactam is observed only in the vicinity of Ser70, not Ser237.

References

1. Silva, J., Aguilar, C., Becerra, Z., López-Antuñano F., & García, R. (1999) *Microb Drug Resist* 5(3), 189–193.
2. Silva, J., Aguilar, C., Ayala, G., Estrada, M. A., Garza-Ramos, U., Lara-Lemus, R., & Ledezma, L. (2000). *Antimicrob Agents Chemother* 44(4), 997–1003.
3. Cifuentes-Castro, V., Rodríguez-Almazán, C., Silva-Sánchez, J., & Rudiño-Piñera, E. (2020). *BBRC*, 522(2), 545–551.

THE MITOCHONDRIAL FUSION PROTEIN FZO1 IS ESSENTIAL IN THE FUNGUS *PODOSPORA ANSERINA*

Mariana Aline González Manríquez, Sara K. Schroder, Leonardo Peraza Reyes

Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, UNAM.

Ciudad Universitaria, 04510, Cd. México, México.

Tel. 5556225628. lperaza@ifc.unam.mx

Mitochondria are essential organelles that undergo dynamic fission and fusion events, which define their arrangement and connectivity, and that are essential to regulate their function, removal and quality control. The fission process is mediated by dynamin-related protein 1 (Drp1/Dnm1), which is recruited to mitochondria by membrane receptor proteins, like Fis1. These proteins also conduct the division of peroxisomes. The mitochondrial fusion process is divided into two sequential steps, the fusion of the mitochondrial outer membrane, regulated by Mitofusins (Mfn1/2/Fzo1), and the fusion of the inner membrane, regulated by Optic Atrophy 1 (OPA1/Mgm1). In the filamentous fungus *Podospora anserina*, we have shown that Dnm1 and Fis1 are required for mitochondria and peroxisome fission throughout the life cycle. Moreover, we have discovered that peroxisome segregation at different developmental stages relies on these proteins, and that sexual development is compromised when either of these proteins is missing. Our results have shown that sexual development depends on a dynamic regulation of mitochondria, which involves the activity of their fission machinery. However, the contribution of the fusion process is unknown. Here we analyzed the function of the *P. anserina* mitofusin Fzo1. We deleted *FZO1* gene and showed that *P. anserina* bearing the *Dfzo1* allele can only grow in the presence of wild-type (*FZO1*⁺) nuclei. Analysis of the meiotic progeny of sexual crosses of these strains to the wild type corroborated that strains possessing *fzo1* deletion could only be recovered in a heterokaryotic context (i.e., *Dfzo1* / *FZO1*⁺). Interestingly, we found that homokaryotic double mutant *Dfzo1Ddnm1* strains could be recovered from the meiotic progeny of crosses of *Dfzo1*/*FZO1*⁺ strains to a strain deleted for *DNM1*, showing that *Dfzo1* lethality is suppressed by *Ddnm1*. Our results show that a precise balance between the mitochondrial fusion and fission processes is critical for cell viability in *P. anserina*. This research was supported by grant IN227823 from PAPIIT-DGAPA. Thanks to Beatriz Aguirre López for the moral and technical support during the completion of this work.

UNCOVERING THE FUNCTIONAL ROLE OF THE RND PUMPS IN *R. ETLI* CFN42

Adrián González, María M. Banda, Ma. de la Paz Salas, Marisa Rodríguez, Mishael Sánchez-Pérez, Georgina Hernández and Lourdes Girard Cuesy

Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México.
Av. Universidad 1001, col. Chamilpa, Cuernavaca, Mor., México.
adriangm@ccg.unam.mx

Multidrug-resistant efflux pumps (MDR) represent the primary mechanism for resistance and physiological adaptation in bacteria. MDR pumps play an essential role in bacteria resistance to broad toxic compounds. They are transport proteins involved in extruding a wide range of substrates (antibiotics, antimicrobials, and metabolites) from the cellular interior to the external environment. Currently, five families of bacterial drug efflux pumps have been described, including the ATP-binding cassette (ABC) family, the major facilitator superfamily (MFS), the resistance-nodulation-cell division (RND) family, the small multidrug resistance (SMR) family, and the multidrug and toxic compound extrusion (MATE) family. Efflux pumps belonging to the RND superfamily are exclusive of Gram-negative bacteria and consist of an active pump protein in the outer membrane (OMP), an inner membrane protein (RND), and a periplasmic adapter protein (PAP) that connects the OMP to the RND and facilitates passage of a wide of substrates into the external medium.

Despite the importance of MDR efflux pumps in the active extrusion of toxic compounds from cells, their role is relatively unknown in plant symbiotic bacteria such as rhizobia. It is well known that plants produce diverse antimicrobial secondary metabolites to protect themselves against pathogen infection and it has been shown its participation in phytopathogenic bacteria to overcome the chemical barriers formed by host plants. *Rhizobium etli* CFN42 interacts with common bean roots to induce the formation of nodules, within which this bacterium converts nitrogen to ammonia that becomes available to the plant. Its genome encodes 44 putative drug efflux pumps, of which eight belong to the RND family.

We use a combination of molecular and genetic approaches to elucidate the functional role of the RND pumps in preventing the accumulation of stressful compounds both in free-living conditions and due to the presence of those produced during the interaction of *R. etli* CFN42 with bean plants. To that end, we constructed a set of *R. etli* CFN42 mutants obtained by vector insertion. Our preliminary results show the negative effect on the bacterial growth of diverse phenolic compounds, antibiotics, osmotic stressors, and oxidative agents in some mutant derivatives. By quantitative RT-qPCR experiments, we determined that the presence of bean root exudates induces expression levels of genes that encode the RND pumps' components in *R. etli* CFN42. We will present genetic evidence supporting that RND pumps enable *R. etli* CFN42 to circumvent the effects of toxic compounds naturally present in its habits.

Acknowledgments. This work is partially supported by PAPIIT-DGAPA (grant IN208823).

IDENTIFICATION OF VIRULENCE FACTORS (EFAA, ASA1, ACEL AND ESP) IN BIOFILM-FORMING ENTEROCOCCUS FAECIUM AND E. FAECALIS

Christian González-Reyes*, Erick Nereu Chávez Fonseca, Jackeline Lizzeta Arvizu-Gómez,
Abril Bernardette Martínez Rizo, Angélica Nallelhy Rodríguez-Ocampo

Unidad Académica de Ciencias Químico Biológicas y Farmacéuticas, Universidad Autónoma de Nayarit,
Ciudad de la Cultura, S/N, col. Centro, Tepic, Nayarit, México. C.P. 63000.

*E-mail: christian.gonzalez@uan.edu.mx

The genus *Enterococcus* are Gram-positive bacteria, considered part of the normal microbiota, although some can cause serious infections in humans, especially in hospital environments. *Enterococcus faecalis* and *E. faecium* are the two important species within this genus, which have been associated with the formation of biofilms and isolated from patients and contaminated water sources. The major threat is the presence of virulence and antibiotic resistance related genes among enterococci bacteria that cause health problems in humans. Biological and chemical contamination of surface water bodies has important consequences, it renders them unusable for human consumption. The most common sources of contamination are sewage discharge and litter trash. In this research, the strains used were obtained from wastewater from three hospitals in the city of Tepic, Nayarit. In this area there is no strict monitoring of the quality of the wastewater discharged by these hospitals into the municipal sewage network, it is possible to obtain strains with virulence characteristics from these discharges. Therefore, in this study, 50 *Enterococcus* strains were included. Biofilm formation capacity was evaluated by crystal violet method, specie identification and the presence of virulence genes *efaA*, *asa1*, *acel* and *esp* were carried out by PCR technique, to determine the relationship between the presence of virulence factors and biofilm formation. The strains analyzed, 70% were identified as *E. faecium* and 30% as *E. faecalis*. Evaluation of biofilm formation showed that 28% of the strains were non-biofilm producers. Meanwhile, the prevalence of biofilm formation was 72%. Among the biofilm-forming strains, 26% had a strong biofilm phenotype, 18% a moderate biofilm phenotype and 28% had a weak biofilm phenotype. The prevalence of genes *efaA*, *asa1*, *acel*, and *esp* in biofilm-forming strains was 77.8%, 66.7%, 52.8% and 5.6%, respectively. In this work, it was found that *efaA* and *asa1* genes are the most prevalent in biofilm-forming strains.

Research supported by Patronato-UAN, SIP-PU22-10.

PHENOTYPIC, GENOTYPIC AND METABOLOMIC CHARACTERIZATION OF *STREPTOMYCES ALBIDOFLOAVUS* J29 ORI2 WITH ANTIFUNGAL ACTIVITY ON *CANDIDA* SPP.

Adilene Gonzalez Silva¹, J. Alfredo Hernández García¹, Humberto L. Mendoza Figueroa², Luis Gerardo Zepeda Vallejo¹, Lourdes Villa-Tanaca¹, César Hernández-Rodríguez^{1*}

¹Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Prol. Carpio y Plan de Ayala, CP 11340, Casco de Santo Tomás, Ciudad de México, México. Teléfono 5729-6000 Ext. 62554. ²Escuela Superior de Medicina, Instituto Politécnico Nacional, Salvador Díaz Mirón esq. Plan de San Luis, CP 11340, Casco de Santo Tomás, Ciudad de México, México.

*E-mail: chdez38@hotmail.com

Streptomyces is a genus of Gram-positive bacteria with high GC content. It is soil saprophytes with linear genomes. *Streptomyces*' importance lies in their ability to produce secondary metabolites with different biological activities, including antifungals, antibiotics, siderophores, and, antiparasitics, among others.

The aim of this work is to analyse the antifungal activity of *Streptomyces albidoflavus* J29 ori2 on *Candida* strains, sensitive and resistant to fluconazole and to analyse at genome and metabolome level, the possible metabolites responsible for the activity.

As a first step, the inhibitory activity of *Streptomyces albidoflavus* J29 ori2 on *Candida* spp. was tested; once inhibition was observed, *Streptomyces* DNA was extracted for genome sequencing. The sequenced genome was assembled and annotated for secondary metabolite biosynthetic clusters using a bacterial antiSMASH version. The results suggest that the possible metabolite responsible for the antifungal activity is candididin, other biosynthetic clusters with various biological activities, such as siderophores, antibiotics, among others, were also identified.

On the other hand, the strain was grown in a liquid medium, and the supernatant was concentrated by freeze-drying for a dose-response assay. The freeze-dried supernatant was analyzed by HPLC-DAD and NMR to determine the type of metabolites present in the sample. In addition, the culture medium was optimized by substituting the carbon (FC) and nitrogen (FN) sources to select the one in which the highest inhibitory activity was detected. Mono, di, and polysaccharides were tested as FC, while amino acids, both acidic, neutral, and basic, as well as aromatics, were tested as FN. The results suggest that mannitol and tyrosine are a good combination to increase the production of the secondary metabolites of interest.

The work concludes is that *Streptomyces albidoflavus* J29 ori2 is a strain with potential antifungal activity, and it has metabolites of different chemical nature with this activity.

Keywords: *Streptomyces*, *Candida*, antifungal, HPLC, NMR.

ROLE OF THE *S. CEREVISIAE* ORTHOLOGOUS PROTEINS *KLNRG1* AND *KLRTG3* FROM THE AEROBIC YEAST *K. LACTIS* IN RESPIRATORY METABOLISM

Yael González Tinoco¹, Carlos Campero-Basaldúa¹, Edgar Ramírez¹, Francisco Padilla-Garfias¹, Norma Silvia Sánchez¹, Alicia Gonzalez-Manjarrez¹

Instituto de Fisiología Celular, UNAM¹.
Cto. Exterior s/n, C.U., Coyoacán, 04510 Ciudad de México, CDMX
E-mail: ytinoco@ifc.unam.mx

In the yeast *Saccharomyces cerevisiae*, Nrg1 and Rtg3 regulatory proteins form a novel and recently described hybrid complex (Nrg1-Rtg3), which plays an essential role in the maintenance of mitochondrial DNA integrity. If either the Nrg1 or Rtg3 encoding genes are deleted, the respiratory metabolism is nullified, and the mutant strains are unable to grow on ethanol as the sole carbon source and growth is achieved exclusively through fermentative metabolism. These results indicated that Nrg1 and Rtg3 could form a dimeric hybrid whose physiological role contributed to the maintenance of respiratory metabolism. Accordingly, a more detailed study showed that mtDNA integrity is disrupted in either *Scnrg1*Δ or *Scrtg3*Δ mutants which are unable to integrate the ScNrg1-ScRtg3 complex.

Considering that the petite-negative yeast *Kluyveromyces lactis*, which presents a predominantly respiratory metabolism, possesses the *KINRG1* and *KIRTG3* orthologous genes, we have analyzed whether as well as in *S. cerevisiae*, their encoded proteins play a role in respiratory metabolism.

Through the use of the directed gene mutation technique, we obtained *Klnrg1*Δ and *Klrtg3*Δ single mutant strains. Growth curve assays in ethanol as the sole carbon source, showed that the two mutant strains display similar growth rates to those found in the wild-type strain, indicating that in this yeast, lack of either *KINrg1* or *KIRtg3* does not affect respiratory metabolism. Accordingly, measurement of oxygen consumption in the *K. lactis* mutants showed that as opposed to that found in *S. cerevisiae* *Scnrg1*Δ or *Scrtg3*Δ mutants, those of *K. lactis* show wild-type oxygen consumption when grown on ethanol. These results suggest that *K. lactis* *KINrg1* and *KIRtg3*, do not constitute a *KINrg1-KIRtg3* hybrid heterodimer which in *S. cerevisiae* plays a crucial role in the maintenance of mitochondrial DNA integrity. Further work will be presented addressing the physiological role of *KINrg1* and *KIRtg3*.

OBTAINING BIOETHANOL FROM BREWER'S MALT BAGASSE

Venus Melissa Gómez Lagunas, Teresita Paz Maldonado, Martha Imelda Maldonado Cervantes,
Jesús Ramon Castillo Hernández, Minerva García Rangel, María Guadalupe Martel Gallegos,
Enrique Maldonado Cervantes

Unidad Académica Multidisciplinaria Zona Media, Rioverde-San Ciro Highway Km. 4,
Col. Puente del Carmen, Rioverde, San Luis Potosí.
E-mail: venusgola02@gmail.com

By-products from the agri-food industry can represent a valuable source of nutrients that can be reused to obtain high added value products¹. In this research project, light and dark must, a by-product of the brewing industry, were used to obtain ethanol by fermentation with *Saccharomyces cerevisiae* yeast.

For the characterization of the raw material, a quantification of total sugars was carried out, obtaining 36.87 mg/l for the light must and 80.27 mg/l for the dark must; Reducing sugars were also quantified, obtaining 320.24 mg/ml in the light must and 387.82 mg/ml in the dark must. Fermentation of light and dark musts was carried out in a bioreactor, inoculating *S. cerevisiae* in a mixture of water and must (10% w/v) for 24 hours and sampling was performed to quantify reducing and total sugars, cell growth and ethanol concentration. The results of these analyses show a yeast growth of 511%, a decrease in total and reducing sugars of up to 69%, and an increasing production of ethanol, reaching a concentration of 16.9% in 24 hours for both types of must. Subsequently, the ethanol produced will be distilled for purification and identification.

References

1. Espitia C., Gutierrez I., Espitia H. (2009). Producción de etanol a partir de cebada no malteada hidrolizada con α y β amilasas comerciales. *Universitas Scientiarum*, vol. 14, pp. 164-172.

VARIABILITY OF LONGITUDINAL CLINICAL ISOLATES OF *CANDIDA GLABRATA*

Marco Josué Hernández Chávez and Irene Castaño-Navarro

Instituto Potosino de Investigación Científica y Tecnológica, División de Biología Molecular.
Camino a la Presa San José 2055, Lomas 4ª sección, C.P. 78216, San Luis Potosí, S.L.P., México. Tel. +52 (444) 834 2040.
marco.hernandez@ipicyt.edu.mx

Candida glabrata is a fungal opportunistic pathogen responsible for invasive candidiasis in immunocompromised and immunosuppressed patients. Nowadays, it is the second most common species responsible for systemic candidiasis worldwide after *C. albicans*. This pathogen adapts to diverse and changing conditions within the host where genotypic and phenotypic variability can take place. This variability can confer the pathogen a set of advantages regarding antifungal susceptibility, oxidative stress tolerance, and virulence. Phenotypic and genotypic variability has been previously studied in sequential clonal clinical isolates obtained from patients with systemic candidiasis throughout the infection (longitudinal isolates).

In this study, we characterized four longitudinal isolates obtained from one patient (P12) with systemic candidiasis and found genotypic and phenotypic changes compared to our laboratory strains. We looked for chromosomal rearrangements and determined the susceptibility to two classes of antifungals, metals, and oxidative stress.

We also determined the cell wall composition and its porosity to gain a better understanding of the pathogen-host interaction and found differences compared to our laboratory strains and a set of additional longitudinal strains from a different patient, suggesting that cell wall composition and porosity are commonly variable among clinical isolates of *C. glabrata*.

Next, we determined the susceptibility of P12 isolates to cell wall perturbing agents. Even though no difference in susceptibility to these agents was found among P12 isolates, there is a high tolerance of *C. glabrata* isolates and laboratory strains compared to *Candida albicans*, a trait that may be crucial in virulence in this pathogen and is currently being investigated.

Finally, we tested the ability of P12 isolates to survive the attack of human neutrophils *in vitro*, resulting in relatively high survival rates compared to another set of longitudinal isolates. This characterization suggests that P12 longitudinal isolates behave similarly among them but possess certain traits that confer the pathogen an advantage during host interaction.

IDENTIFICATION OF PROTEINS ASSOCIATED WITH ISOFORMS OF THE EUKARYOTIC TRANSLATION INITIATION FACTOR 5A (EIF5A) IN *FUSARIUM GRAMINEARUM*

Jorge Eduardo Hernández-Espinosa¹, Estela Ruiz-Baca², J. Félix Gutiérrez-Corona¹, Ana Lilia Martínez-Rocha^{1*}

¹University of Guanajuato, Division of Natural and Exact Sciences, Department of Biology. *Noria Alta s/n, Col. Noria Alta, Guanajuato, Gto. 36050, Mexico. Tel: +52 473 7320006 ext.8296. ²University Juárez of Durango State, Faculty of Chemistry Sciences.

*E-mail: anamartinez@ugto.mx.

Fusarium graminearum an ascomycete fungus, is the causal agent of Fusarium Head Blight (FHB) in wheat and cob rot (CR) in maize, causing losses by contaminating crops and grains with mycotoxins such as Deoxynivalenol (DON) and Zearalenone (Zea) [1]. *F. graminearum* is difficult to control with conventional fungicides, therefore, finding new targets to control FHB and CB is imperative. The eukaryotic translation initiation factor 5A (eIF5A), which plays a crucial role in translation and elongation, is a small protein undergoing a post-translational modification forming the amino acid hypusine for its activation [2]. Hypusine biosynthesis involves two steps, first, the enzyme deoxyhypusine synthase (DHS) transfers a 4-aminobutyl moiety from spermidine to produce the intermediary isoform eIF5A-deoxyhypusine (eIF5A-Dhp) and second, deoxyhypusine (DOHH) hydroxylates the eIF5A-Dhp to form the eIF5A-Hypusine [3]. Hypusination of *F. graminearum* eIF5A was studied by overexpressing DHS and DOHH enzymes resulting in contrary phenotypes, as well as the production of different isoforms of eIF5A [4]. This project aims to identify the proteins associated with isoforms of eIF5A containing a FLAG tag in the mutant DOHHoex and the wild type strain by immunoprecipitation and mass spectrometry (LC-MS). For this, we determine if the mutants carrying eIF5A with the FLAG tag have the same genotype and phenotype as the parental strains. The correct integration of DOHH under the strong promoter P_{gpd1} and the FLAG tag in eIF5A from WT and DOHHoex were verified by PCR. Phenotypic analyses were conducted on complete medium plates (CM), with and without different stress conditions such as oxidative, osmotic, and cell wall stress, showing similar phenotypes between the parental and the FLAG tag strains. The identification of differential proteins associated to eIF5A isoforms could reveal new functions and targets associated to eIF5A to control this devastating phytopathogen.

References

1. Buttar, Z. A., Cheng, M., Wei, P., Zhang, Z., Lv, C., Zhu, C., Ali, N. F., Kang, G., Wang, D., & Zhang, K. (2024). *Plants*, 13(8), 1159.
2. Legrand, F., Picot, A., Cobo-Díaz, J. F., Chen, W., & Floch, G. L. (2017b). *Biological Control*, 113, 26-38.
3. Park, M. H. (2006). *Journal Of Biochemistry/The Journal Of Biochemistry*, 139(2), 161-169.
4. Martinez-Rocha, A. L., Woriedh, M., Chemnitz, J., Willingmann, P., Kröger, C., Hadelers, B., Hauber, J., & Schäfer, W. (2016). *Scientific Reports*, 6(1).

MICROBIOLOGICAL AND PHYSICOCHEMICAL CHARACTERIZATION OF KOMBUCHA

Liliana Alejandra Hernández Espinosa¹, Ana Karen Álvarez Contreras¹, Iván Natividad-Bonifacio¹, Carlos Vázquez Salinas² and Elsa Irma Quiñones Ramírez¹

¹Escuela Nacional de Ciencias Biológicas IPN, Santo Tomás, Manuel Carpio, Plutarco Elías Calles, Miguel Hidalgo, C.P. 11350 CDMX, ²Universidad Autónoma Metropolitana, Iztapalapa, San Rafael Atlixco 186, Leyes de Reforma 1A Sección, Iztapalapa, C.P. 09310 CDMX.
E-mail: helaqbp@gmail.com

Introduction. Kombucha, a fermented beverage derived from sugared green tea, undergoes fermentation facilitated by a symbiotic culture of bacteria and yeast (SCOBY) along with a starter liquid of previously fermented kombucha. This process, primarily driven by acetic acid bacteria (AAB) supplemented by yeasts and sometimes lactic acid bacteria (LAB), provides the beverage with its distinctive flavor profile¹. Throughout fermentation, both the SCOBY and kombucha experience dynamic shifts in microbial composition and metabolic activity, yielding various beneficial effects upon consumption². While numerous studies have elucidated kombucha's microbial makeup, few have investigated its temporal changes and antimicrobial efficacy across fermentation stages. The aim of this study is to understand the correlation between microbial proportions in kombucha, its physicochemical attributes, and antimicrobial potential. **Methodology.** Physicochemical parameters were assessed on days 0, 7, 14, and 21 of fermentation, including saccharose content via Brix densimeter, protein concentration via Bradford method, pH via potentiometer, and titratable acidity. Isolates were cultured from kombucha and SCOBY serially diluted samples on the same fermentation days, subject to macroscopic and microscopic characterization, Gram staining, oxidase, catalase, and calcium carbonate solubilization tests for classification into AAB, LAB, and yeast groups. Antimicrobial activity was tested against *Listeria monocytogenes*, *Escherichia coli*, *Salmonella enterica* sbsp. *enterica* serovar *typhi*, *S. enterica* sbsp. *enterica* serovar *abony*, *Cronobacter sakazakii*, *Staphylococcus aureus* and *Bacillus cereus* using agar well diffusion method. **Results.** Titratable acidity, pH, saccharose content, and protein concentration declined over time, contrasting with an increase in titratable acidity. Day 0 exhibited diverse morphotypes, yet day 7 boasted the highest microbial counts. AAB population rose throughout fermentation, while yeast decreased and LAB remained stable. Antimicrobial activity intensified over time, transitioning from minimal to significant inhibition by day 14. **Conclusion.** These findings underscore the intricate interplay between microbial communities and metabolic processes during kombucha fermentation. Such dynamics likely contribute to the beverage's health-promoting attributes, including its antimicrobial potency.

References

1. Aghogho Ohwofasa, Manpreet Dhama, Christopher Winefield, Stephen L W (2024). Current Research in Food Science, 18.
2. Nurten Abaci, Fatma Sezer Senol Deniz, Ilkay Erdogan Orhan (2022). Food chemistry, 14.

CHARACTERIZATION OF THE MYCELIAL GROWTH OF FUNGI FROM AN ARID ENVIRONMENT

Miguel Emiliano Hernández-Fernández¹, Karina García-Gutiérrez,
Darío Rafael Olicón-Hernández¹, Guadalupe Guerra Sánchez¹

¹Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Microbiology Department. Mexico City, Mexico.
E-mail: mguerras@ipn.mx

Fungi are worldwide distribution organisms that inhabit extreme environments despite not having optimal growth, known as extremotolerant. An arid environment presents conditions such as low water activity, high salinity, temperature changes throughout the day, and UV radiation. Fungi have adapted to osmotic stress and dehydration². This work aimed to characterize the mycelial growth of isolates from arid areas of Baja California, Mexico, analyzing the effect of nutrients, temperature, and salinity on their growth. Three growth media were tested for fungal growth: YPD, PDA, and MM (Minimum Media). To determine the effect of temperature, fungi were incubated in a YPD medium at temperatures of 4, 28, 37, 45, and 50°C. To observe the effect of salt stress, different concentrations of NaCl (2.5, 3.8, and 5.2 M) were added to the best growth medium. The results showed differences in mycelial size and morphology, conidiation, and pigmentation of the colonies. The media with the higher concentration of protein and inorganic nitrogen promoted a larger mycelial growth and was YPD. Most of the isolates grew better at 37°C, the mycelial morphology changed with the temperature. Thirteen isolates grew from 4 to 45°C and 60% of the isolates tolerated 2.5 M NaCl in the growth media showing a reduction of the size and mycelial morphology. The fungi showed the capacity to grow at extreme conditions, as a response to their adaptations to the arid environment where they establish their habitat

References

1. Zhang, X., Li, S. J., Li, J. J., Liang, Z. Z., & Zhao, C. Q. (2018). Novel natural products from extremophilic fungi. *En Marine Drugs* (Vol. 16, Número 6). MDPI AG. <https://doi.org/10.3390/md16060194>
2. Coleine, C., Stajich, J. E., & Selbmann, L. (2022). Fungi are key players in extreme ecosystems. *En Trends in Ecology and Evolution* (Vol. 37, Número 6, pp. 517–528). Elsevier Ltd. <https://doi.org/10.1016/j.tree.2022.02.002>

ANTIMICROBIAL RESISTANCE IN MEAT SAMPLES

Hugo Antonio Hernández Pérez¹; Orlando Vizuet Uscanga¹

¹Departamento de Biología, Facultad de Química, UNAM, México.
Ciudad Universitaria, 04510, Ciudad de México, México. Tel. 56223696.
E-mail: hhernand@quimica.unam.mx.

According to Mexican Official Norm 194-SSA1-2004, meat is defined as the skeletal striated muscular structure, accompanied or not by connective tissue, bone, and fat, as well as nerve fibers, lymphatic and blood vessels, which originates from animals intended for consumption. The high water and protein content of meat, along with other water-soluble constituents, make meat and its products a suitable medium for the growth of microorganisms. Nowadays, within the field of microbiology, the resistance of microorganisms to antimicrobial agents has caused significant concern worldwide due to its serious implications for public health. These antimicrobial-resistant microorganisms can be found in humans, animals, food, and the environment (water, soil, and air). The use of antimicrobial agents in animals intended for food production represents an important potential risk factor for the selection and spread of resistant microorganisms to humans through food consumption.

The main objective of this study is to isolate and characterize antimicrobial-resistant bacteria from meat products. The specific objectives include developing an experimental protocol to determine the presence of microorganisms resistant to antimicrobial substances in meat samples obtained from slaughterhouses, and finally, using MALDI-TOF technology for identification purposes.

The development of the experimental protocol took place at the Faculty of Chemistry, UNAM, since August 2022. Different meat samples were analyzed using an experimental protocol designed to isolate microorganisms resistant to five different antibiotics.

The isolation of bacteria resistant to antimicrobial substances followed a four-stage experimental protocol based on international norms and standards. From the isolates obtained, a significant proportion were identified as *E. coli*. The beef viscera sample exhibited a high number of microorganisms, likely due to the intestine being a site with a greater diversity and quantity of microorganisms. The use of MALDI-TOF biotype equipment proved to be a useful tool for the rapid microbial identification of the isolates obtained.

References

1. CDC. (2019). Antibiotic Resistance Threats in the United States . Atlanta:U.S. Department of Health and Human Services.
2. Clinical and Laboratory Standards Institute . (2022). M100 Performance Standards for Antimicrobial Susceptibility Testing . Estados Unidos : CLSI.
3. Kraft, A. (1986). Meat Microbiology.Orlando: Academic Press.
4. Secretaría de Salud. (18 de Septiembre de 2004). NOM-194-SSA1-2004.Productos y servicios. Especificaciones sanitarias en los establecimientos dedicados al sacrificio y faenado de animales para abasto, almacenamiento, transporte y expendio.
5. OPS. (2019). Tratamiento de las enfermedades infecciosas 2020-2022(Octava ed.). Washington, D.C.:Organización Panamericana de la Salud.Obtenido de Organización Panamericana de la Salud Web site.

PARTICIPATION OF THE ORF PA2305/AMB B IN THE VIRULENCE OF *PSEUDOMONAS AERUGINOSA* PAO1

Ximena Hernandez Ramos¹, Carla Isabel Tena Fuentes¹, Jesús Campos García¹

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo.
Gral Francisco J. Mújica S/N, Ciudad Universitaria, 58030, Morelia Mich. 4431460411.
E-mail: 1590734d@gmail.com

Pseudomonas aeruginosa is a Gram-negative bacterium considered an opportunistic pathogen, affecting individuals with compromised immune systems and causing severe respiratory and soft tissue infections, often associated with hospital environments. This bacterium produces several virulence factors (biofilms, pyocyanin, siderophores, lipopolysaccharides, etc.) essential for host infection development. The biosynthesis of these virulence factors is regulated by quorum sensing (QS), a cell-to-cell communication mechanism where various molecules, including cyclic dipeptides (CDPs), modulate QS systems, thereby regulating virulence factor production [1]. This study investigated the role of genes encoding non-ribosomal peptide synthetases (NRPS), whose products, like CDPs and other metabolites, could modulate QS and, thus, bacterial pathogenicity. The study focused on the ORF PA2305, called *ambB*, due to their unknown role in CDPs synthesis and *P. aeruginosa* virulence. A Mutant for the ORF PA2305 was constructed by gene disruption through inserting a gentamicin resistance cassette, and their production of virulence factors, CDPs production, and pathogenicity were evaluated using a *Caenorhabditis elegans* model. Quantifying virulence factors showed that the mutant's general protease and pyocyanin production did not significantly differ from the PAO1 reference strain. Pyoverdine production decreased, while pyocyanin production increased in the *ambB::Gm* mutant. The virulence factors biofilm, rhamnolipid, and lipopolysaccharide increased the biosynthesis. The CDPs cyclic(L-Pro-L-Ile), cyclic(L-Pro-L-Tyr), cyclic(L-Pro-L-Val), and cyclic(L-Pro-L-Phe) were identified in the supernatants showing a significant increase in cyclic(L-Pro-L-Ile) compared to the *P. aeruginosa* PAO1 strain. Assays in the *C. elegans* model with bacterial cultures showed that the *ambB::Gm* mutation in the *P. aeruginosa* PAO1 strain increased lethality, correlating with the increased virulence factors and pyocyanin. These results indicate that the NRPS encoded in the PA2305 ORF in *P. aeruginosa* PAO1 is involved in virulence factor production, influencing bacterial pathogenicity.

Reference

1. Dela Ahator, S., & Zhang, L. (2019). Small Is Mighty-Chemical Communication Systems in *Pseudomonas aeruginosa*. *Annu. Rev. Microbiol.* 8;73:559-578.

MICROBIOLOGICAL CHARACTERIZATION OF TENATE CHEESE THROUGH A MICROBIOLOGICAL-GENOMIC ANALYSIS

Mariana Suárez¹*, Carmen Julia Figueredo-Urbina² and Silvia Armenta^{1*}

¹Universidad Autónoma del Estado de Hidalgo (UAEH), Instituto de Ciencias Agropecuarias (ICAp), Rancho Universitario, Av. Universidad Km. 1 Ex-Hda. de Aquetzalpa AP 32, CP 43600, Tulancingo. Hidalgo, ²Investigadora por México CONAHCYT-Instituto de Ciencias Agropecuarias, Tulancingo de Bravo, Hidalgo.

*E-mail: he379568@uaeh.edu.mx, silvia_armenta@uaeh.edu.mx

Tenate cheese is an artisanal cheese native to Mexico, specifically produced in Hidalgo. This dairy product is made from raw milk, and its main peculiarity is the handcrafted tenate basket in which it is molded. The interest in tenate cheese lies in its microbiota and the functional properties they impart to the cheese. Despite its cultural and economic importance, there is not enough data about its microbiological diversity. Researchers have primarily focused on understanding the organoleptic characteristics and identifying specific microorganisms. Tenate cheese contains a high proportion of lactic acid bacteria, with *Lactobacillus paracasei* being an important strain. This particular strain was isolated using MRS agar and then identified through molecular methods like 16S rRNA sequencing [2]. *L. paracasei* has demonstrated probiotic properties, making it suitable for further research as a representative model for studying the *Lactobacillus* genus. Interestingly, previous research on artisanal cheeses has demonstrated the significant potential of these dairy products. For instance, an analysis of Cotija cheese using metagenomics discovered a novel *Escherichia coli* strain with non-pathogenic genes and uncovered the metabolic capabilities of specific bacteria in producing various flavor compounds. Furthermore, the study identified genes linked to bacteriocin production and immunity [1].

Therefore, this research aims to be approached from a microbiological-genomic analysis viewpoint. By using specific media for bacterial and fungal growth and carrying out DNA extraction for partial or complete sequencing with 16S rRNA, we can identify the bacteria that play a role in the processes of elaboration, maturation, and final characteristics of tenate cheese, allowing us to investigate the most important microorganisms present. Furthermore, a metagenomic analysis will provide a comprehensive understanding of the microbial diversity and metabolic functions in tenate cheese. Finally, it provides information about how this microbiota can affect the cheese's organoleptic and functional characteristics, contributing to the enhancement of current knowledge and cultural and social perspectives.

References

1. Escobar Zepeda, Sanchez Flores, Quirasco Baruch (2016). Food Microbiology.
2. N. Falfán-Cortés, N. Mora-Peñaflor, C. Gómez-Aldapa, E. Rangel-Vargas, A. Acevedo-Sandoval, J. Franco-Fernández, J. Castro Rosas (Year). Journal of Food Protection, vol 85, 112-121.

THE *pqsA* MUTATION IN *PSEUDOMONAS AERUGINOSA* PAO1 MODIFIES THE PRODUCTION OF VIRULENCE FACTORS DEPENDING ON THE GROWTH STAGES

Jaime Adrian Hurtado Solorzano¹, Lorena Martínez Alcantar¹, Jesús Campos García¹

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo.
Francisco J. Múgica S/N, Ciudad Universitaria, 58030 Morelia, Mich.
+52 443 367 7256, adriansolorzano190@gmail.com

Pseudomonas aeruginosa is an opportunistic pathogenic bacterium causing plant, animal, and human diseases. It is part of the ESKAPE group of bacteria, which are of primary biomedical importance due to their pathogenesis and antibiotic resistance. The pathogenicity of *P. aeruginosa* is attributed to virulence factors production, such as elastase, protease, pyocyanin, siderophores, and biofilm, which enable its adaptation and survival in the host. The production of these virulence factors is regulated by quorum sensing (QS). *P. aeruginosa* has three main QS systems (Las, Rhl, and Pqs). The Pqs system is crucial in bacterial production of virulence factors, stress resistance, and immune response evasion phenomenon [1]. Consequently, it has been reported that null mutants in the Pqs system reduce the production of virulence factors and exhibit an avirulent phenotype in mouse burn, plant, and nematode infection models [2,3]. This study evaluated the production of virulence factors in a *pqsA* mutant strain obtained from the *P. aeruginosa* PAO1 strain. For this purpose, the *pqsA* gene was mutated by interruption with a resistance cassette encoding 3-N-acetyltransferase. The obtained mutant was evaluated for producing acyl homoserine lactones (AHLs), proteases, elastases, pyocyanin, pyoverdine, and rhamnolipids. The findings showed that besides the fact that *pqsA* is essential for the biosynthesis of the autoinducer PQS, its phenotype contrasts with what was previously reported [2,3]. The *pqsA* mutant showed an increase in elastase and pyocyanin amounts since the exponential growth stage, but the AHLs were severely affected in the stationary growth stage, which suggests a novel QS mechanism that controls the production of virulence factors in *P. aeruginosa* in a PQS-dependent fashion.

References

1. Bala, A., Kumar, L., Chhibber, S., & Harjai, K. (2015). Augmentation of virulence related traits of *pqs* mutants by *Pseudomonas* quinolone signal through membrane vesicles. *Journal of Basic Microbiology*, 55(5), 566-578.
2. Chadha, J., Harjai, K., & Chhibber, S. (2022). Revisiting the virulence hallmarks of *Pseudomonas aeruginosa*: a chronicle through the perspective of quorum sensing. *Environmental microbiology*, 24(6), 2630-2656.
3. Lin, J., Cheng, J., Wang, Y., & Shen, X. (2018). The *Pseudomonas* quinolone signal (PQS): not just for quorum sensing anymore. *Frontiers in cellular and infection microbiology*, 8, 230.

TRANSCRIPTION FACTORS INVOLVED IN ROOT CAP DEVELOPMENT DRIVE ROOT GROWTH DIRECTION UPON INOCULATION WITH THE BENEFICIAL RHIZOBACTERIUM *ACHROMOBACTER SP. 5B1*

Kirán Rubí Jiménez-Vázquez¹ and José López-Bucio^{1*}

¹Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Edificio B3, Ciudad Universitaria, Morelia, Michoacán, C. P. 58030, México.

kiran.jimenez@umich.mx, jlbucio@umich.mx*

The root cap is a multilayered organ that assists the root in exploring the soil. Its location at the distal end of roots protects the meristem and directs root growth towards positive stimuli such as gravity and nutrients. Although the root cap maintains a close interaction with the rhizosphere, how it perceives soil microorganisms is unknown. Here, through direct root cocultivation with the plant growth promoting rhizobacterium *Achromobacter sp. 5B1*, we show that specific elements in the development of the root cap control root growth direction. In *Arabidopsis* seedlings with mutations in the transcription factor FEZ, the rhizobacterium caused susceptibility of roots to form supercoils, whereas in seedlings harboring mutations in *SOMBRERO* the change in root growth direction did not occur. These effects coincided with expression analysis of *FEZ::FEZ-GFP* and *SMB::SMB-GFP* in the root cap. Interaction with *Achromobacter sp. 5B1* modifies the structure of the root cap, while the loss-of-function of *SOMBRERO* and FEZ could be associated to the repression of auxin transporters PIN1, PIN2, and PIN3 at this region and exhibits a synergistic effect upon bacterial inoculation, resulting in the inhibition of asymmetric auxin distribution in the root tip in *smb-3* mutant. Our data indicate that the root cap senses the rhizobacterium *Achromobacter sp. 5B1* through root cap transcription factors and contributes to the deviation of root growth forming turns, coils and branches that help plants to more efficiently explore the soil.

References:

1. Jiménez-Vázquez KR, García-Cárdenas E *et al.*, 2020. The plant beneficial rhizobacterium *Achromobacter sp. 5B1* influences root development through auxin signaling and redistribution. *Plant J.* 103(5):1639-1654.
2. Jiménez-Vázquez KR, López-Hernández J *et al.*, 2024. The plant growth promoting rhizobacterium *Achromobacter sp. 5B1*, rescues *Arabidopsis* seedlings from alkaline stress by enhancing root organogenesis and hormonal responses. *Microbiol Res.* 281:127594.

THE QSCR MUTATION IN *PSEUDOMONAS AERUGINOSA* PAO1 MODIFIES THE BIOSYNTHESIS OF ACYL HOMOSERINE LACTONES DEPENDENT ON THE GROWTH PHASE

Kevin Ledesma Muñiz¹, Lorena Martínez Alcantar¹, Jesús Campos García¹

¹Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo.
Francisco J. Múgica S/N, Ciudad Universitaria, 58030 Morelia, Mich. +52 443 471 6978.
1630208D@umich.mx

Pseudomonas aeruginosa is an opportunistic pathogen responsible for various nosocomial infections worldwide, causing chronic and acute diseases in immunocompromised patients. The pathogenicity of *P. aeruginosa* is related to its ability to produce a wide variety of virulence factors, which are regulated by the Quorum Sensing system (QS) [1]. The regulator QscR (quorum-sensing-controlled-repressor) is an essential element that has not been fully described, which influences the synchronization and timely activation of QS to prevent an overproduction of virulence factors [2]. We are interested in evaluating the participation of QscR in the regulation of the QS and its impact on the production of virulence factors. In this study, assays were conducted in the *qscR* mutant from *P. aeruginosa* PAO1 to quantify virulence factors and signaling molecules of the QS at different growth stages. The results indicate that QscR impacts the production of virulence factors in *P. aeruginosa* PAO1 by modulating the activation of QS systems through intervention in producing acyl homoserine lactones. Additionally, QscR showed a variable degree of regulation on the production of virulence factors highly dependent on the growth phase.

References

1. Ledgham, F., Ventre, I., Soscia, C., Foglino, M., Sturgis, J. N., & Lazdunski, A. (2003). Interactions of the quorum sensing regulator QscR: interaction with itself and the other regulators of *Pseudomonas aeruginosa* LasR and RhIR. *Molecular microbiology*, 48(1), 199–210.
2. Ding, F., Oinuma, K. I., Smalley, N. E., Schaefer, A. L., Hamwy, O., Greenberg, E. P., & Dandekar, A. A. (2018). The *Pseudomonas aeruginosa* Orphan Quorum Sensing Signal Receptor QscR Regulates Global Quorum Sensing Gene Expression by Activating a Single Linked Operon. *mBio*, 9(4), e01274-18.

STUDY OF *BACILLUS CEREUS* FROM FOOD SAMPLES: PREVALENCE, TOXIGENIC PROFILE AND ANTIBIOTIC RESISTANCE RESPONSE

Brenda Sofía Loaeza Cruz¹, Ana Karen Álvarez Contreras¹, Carlos Ramón Vázquez Quiñones¹,
Elsa Irma Quiñones Ramírez¹ y Carlos Vázquez Salinas²

¹Escuela Nacional de Ciencias Biológicas-IPN Santo Tomás, Manuel Carpio, Plutarco Elías Calles, Miguel Hidalgo, C.P. 11350 CDMX. ²Universidad Autónoma Metropolitana-Iztapalapa, San Rafael Atlixco 186, Leyes de Reforma 1A Sección, Iztapalapa, C.P. 09310 CDMX.
E-mail: sofia.loaeza99@gmail.com¹

Introduction. *Bacillus cereus* is a Gram-positive, sporulating, opportunistic, facultative anaerobic, cosmopolitan pathogen that causes two syndromes: diarrhoeal syndrome and emetic syndrome, associated with the consumption of contaminated foods such as meat products, dairy products, vegetables, and starchy foods such as rice, potato, flour and some cereals. The outbreaks that occur are usually self-limiting, however, they can be life-threatening due to virulence factors and the resistance profile that *B. cereus* can show. Since reporting of this pathogen is not mandatory in Mexico, there are few reports on the prevalence of this microorganism, so this study aims to detect the toxigenic profile of *B. cereus* isolates from samples of raw rice and pasteurised milk marketed in different parts of CDMX, as well as to determine the antimicrobial resistance profile. **Methodology.** From 50 samples of raw rice and 50 samples of pasteurised milk, the isolation and identification of *B. cereus* was carried out using the methodology described in chapter 14 of the FDA Bacteriological Analytical Manual. For the detection of the toxigenic profile of each isolate, the genes *hblACD* (haemolysin), *nheABC* (non-haemolytic), *cytK* (cytotoxin K), *ces* (cerulide), *entFM* (FM enterotoxin) and the *tE* (toxin E) were amplified by PCR. Antimicrobial susceptibility profiling was performed using the Kirby Bauer method, described in the Clinical and Laboratory Standards Institute (CLSI) manual, testing 12 antibiotics: trimethoprim-sulfamethoxazole (SXT), tetracycline (TET), ciprofloxacin (CIP), penicillin (PEN), gentamicin (GEN), cephalothin (CF), furazolidone (FUR), vancomycin (VA), fosfomicin (FO), oxacillin (OX), clindamycin (CC), erythromycin (EM). **Results.** Of the total number of samples analysed, 56% (28/50) from raw rice and 82% (41/50) from pasteurised milk were positive, with a total of 68 isolates identified. Regarding the toxigenic profile, of the isolates obtained, the *hblA* gene was detected in 75% (51/68), *hblC* in 67.6% (46/68), *hblD* in 64.7% (44/68), *nheA* in 45.5% (31/68), *nheB* by 67.6% (46/68), *nheC* by 94.1% (64/68), *cytK* by 83.8% (57/68), *ces* by 10.2% (7/68), *entFM* by 33.8% (23/68) and *tE* by 27.9% (23/68). In relation to the resistance profile, antibiotics were grouped according to antimicrobial activity, with FUR (100%), OX (91.7%), CF (85.2%), PEN (85.2%), GEN (73.5%) and VA (54.4%); antibiotics with intermediate sensitivity are TET (57.3%) and SXT (48.5%) and finally, sensitive antibiotics are FO (83.8%), CIP (75%), EM (54.4%) and CC (50%). **Conclusions.** The consumption of this type of food represents a risk for the consumer, due to the presence of *B. cereus* isolates with more than one gene coding for emetic and diarrhoeal syndrome toxins, in addition, they show a resistance profile to antibiotics that can be used for the treatment of infections.

References:

1. Dietrich, R., Jessberger, N., Ehling-Schulz, M., Märtlbauer, E., & Granum, P. E. (2021). The Food Poisoning Toxins of *Bacillus cereus*. *Toxins*, 13(2), 98. <https://doi.org/10.3390/toxins13020098>
2. Rodrigo, D., Rosell, C. M., & Martínez, A. (2021). Risk of *Bacillus cereus* in Relation to Rice and Derivatives. *Foods (Basel, Switzerland)*, 10(2), 302. <https://doi.org/10.3390/foods10020302>

ROLE OF CELL-END PROTEIN TEA-5 IN THE GROWTH AND DEVELOPMENT OF *NEUROSPORA CRASSA*

Pedro A. Lopez-Garcia¹, Olga A. Callejas-Negrete¹, Astrid N. Espino-Vázquez¹, Rosa R. Mouriño-Pérez¹

¹Centro de Investigación Científica y de Educación Superior de Ensenada. Carretera Ensenada-Tijuana No. 3918, Zona Playitas, C.P. 22860, Ensenada, B.C. México.
lgpedro@cicese.edu.mx

TEA (Tip Elongation Aberrant protein) complex has been associated with the interaction between microtubules and the actin cytoskeleton in growing sites of *Schizosaccharomyces pombe*. This complex comprises three proteins: Tea1, localized at the microtubules plus end and cell tip; Tea4, an SH3 domain-containing protein; and Mod5, a Tea1 receptor in the apex. The complex seems to regulate formin function, promoting actin nucleation and polymerization in the cell poles. To decipher the role of the TEA complex during the polarized growth of *Neurospora crassa*, we identified and deleted the three components of the complex: TEA-1, TEA-4, and TEA-5 (homologs for Tea1, Tea4, and Mod5, respectively). Notably, the absence of TEA-1 or TEA-4 does not significantly impact the development of the fungus, but *tea-5* gene deletion was ascospore lethal. Thus, the phenotype of a heterokaryon was evaluated. The $\Delta tea-5^{Het}$ mutant strain showed a decrease of 43% in the elongation rate, 57% in biomass production, and 68% in conidia production ($p < 0.05$). The branching rate was two-fold higher in the $\Delta tea-5^{Het}$ mutant ($p < 0.05$) than in the WT. Mature hyphae of $\Delta tea-5^{Het}$ strain showed a small and unstable Spitzenkörper and slightly disorganized actin and microtubular cytoskeleton. In conclusion, TEA-5 is essential in *N. crassa*, and the partial silencing of the gene strongly affects the symmetry during hyphal development and the organization of actin filaments in the apical region of *N. crassa*.

THE PLANT GROWTH PROMOTING RHIZOBACTERIUM ACHROMOBACTER SP. 5B1, RESCUES ARABIDOPSIS SEEDLINGS FROM ALKALINE STRESS BY ENHANCING ROOT ORGANOGENESIS AND HORMONAL RESPONSES

José López Hernández, Kirán Rubí Jiménez-Vázquez, Jesús Salvador López-Bucio,
Enrique Ibarra-Laclette, José López-Bucio

Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Edificio B3,
Ciudad Universitaria, C. P. 58030, Morelia, Michoacán, México.
Tel. 4432452898; jlbucio@umich.mx

Soil alkalinity is a critical environmental factor for plant growth and distribution in ecosystems. An alkaline condition (pH > 7) is imposed by the rising concentration of hydroxides and cations, and prevails in semiarid and arid environments, which represent more than 25% of the total arable land of the world. Despite the great pressure exerted by alkalinity for root viability and plant survival, scarce information is available to understand how root microbes contribute to alkaline pH adaptation. Here, we assessed the effects of alkalinity on shoot and root biomass production, chlorophyll content, root growth and branching, lateral root primordia formation, and the expression of CYCB1, TOR kinase, and auxin and cytokinin-inducible transgenes in shoots and roots of *Arabidopsis* seedlings grown in Petri plates with agar-nutrient medium at pH values of 7.0, 7.5, 8.0, 8.5, and 9.0. The results showed an inverse correlation between the rise of pH and most growth, hormonal and genetic traits analyzed. Noteworthy, root inoculation with *Achromobacter* sp. 5B1, a beneficial rhizospheric bacterium, with plant growth promoting and salt tolerance features, increased biomass production, restored root growth and branching and enhanced auxin responses in WT seedlings and auxin-related mutants *aux1-7* and *eir1*, indicating that stress adaptation operates independently of canonical auxin transporter proteins. Sequencing of the *Achromobacter* sp. 5B1 genome unveiled 5244 protein-coding genes, including genes possibly involved in auxin biosynthesis, quorum-sensing regulation and stress adaptation, which may account for its plant growth promotion attributes. These data highlight the critical role of rhizobacteria to increase plant resilience under high soil pH conditions potentially through genes for adaptation to an extreme environment and bacteria-plant communication.

ISOLATION AND GENOME SEQUENCING OF *VIBRIO PARAHAEMOLYTICUS* STRAINS IN *LITOPENAEUS VANNAMEI* SPECIMENS FROM AN AQUACULTURE FARM IN SONORA

Jesús Daniel López Loera¹, Héctor Manuel Acuña Ramírez², Sergio Abrahán Gutiérrez Payan¹, Edén Noé Cervantes Leal¹, José Antonio Magaña Lizárraga¹, Hilda Martina Gracia Valenzuela², Rodolfo Bernal Reynaga¹, José Guadalupe Rendón Maldonado¹, María Elena Báez Flores¹

¹Unidad de Investigaciones en Salud Pública "Dra. Kaethe Willms", Facultad de Ciencias Químico-Biológicas, Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México; ²Instituto Tecnológico del Valle del Yaqui, Bacum, Sonora, México.
E-mail: jesus.daniel.lopez.loera.fcqb2022@uas.edu.mx

White shrimp (*Litopenaeus vannamei*) has become the most valuable marine product in the world, representing a source of quality protein for the population. Considering the volume of national shrimp aquaculture production, Sonora is the second shrimp-growing state. The regular consumption of shrimp represents a risk to public health, since it can harbor bacteria of the *Vibrio* genus that can cause gastrointestinal conditions and even septicemia in immunocompromised patients, with *V. parahaemolyticus* being the most prominent; a bacteria linked to large losses in aquaculture production. Regarding the global problem of antimicrobial resistance, *V. parahaemolyticus* has developed resistance to some antibiotics. This threatens both public health and aquaculture, since it poses challenges in the treatment of foodborne infections, as well as in the management of diseases in shrimp farms, potentially compromising the profitability of this activity. Therefore, it's important to know the antimicrobial resistance and virulence profiles of *V. parahaemolyticus* through NGS technologies. The resistance profiles of four *V. parahaemolyticus* strains (AHPND-VP7, AHPND-VP8, AHPND-VP12, and AHPND-VP14) were studied using the VITEK®2 system (v9.02). The genomes of these strains were then sequenced on the Illumina MiSeq platform using a paired-end read strategy (2x150 bp). The strains studied were resistant to ampicillin and intermediately resistant to cefuroxime. In addition, they demonstrated susceptibility to a total of nine antibiotics; these included β -lactamase blocking penicillin (ampicillin/sulbactam), cephalosporins (cefotaxime, ceftazidime, and cefepime), carbapenem (meropenem), aminoglycosides (amikacin, gentamicin), fluoroquinolones (ciprofloxacin), and sulfonamides (trimethoprim/sulfamethoxazole). The genomic sequences obtained were deposited in the National Center for Biotechnology Information (NCBI) under Bioproject No: PRJNA1071127 and the Sequence Read Archive (SRA): SRR27848365 (AHPND-VP7), SRR27848364 (AHPND-VP8), SRR27848363 (AHPND-VP12), SRR27848362 (AHPND-VP14). Bioinformatics analysis is underway to determine the resistome and virulome that these genomes harbor.

SEARCH FOR PATHOGENIC BACTERIA AND ANTIBIOTIC RESISTANCE IN THE WATER OF XOCHIMILCO CANALS

Daniel López Pastrana¹, Jaime Bustos Martínez²,
María Guadalupe Figueroa Torres², Víctor Humberto Bustamante Santillán¹

¹Instituto de Biotecnología de la UNAM, Av. Universidad 2001, Colonia Chamilpa, Cuernavaca, Morelos. C.P. 62210. ²Universidad Autónoma Metropolitana-Xochimilco. Ciudad de México.
E-mail:daniel.lopez@ibt.unam.mx;victor.bustamante@ibt.unam.mx

Antibiotic resistance has become a priority issue in public health. In 2019, 4.95 and 1.27million deaths were associated with and attributable to antibiotic resistance, respectively. It was estimated that if adequate measures are not taken, by 2050, antibiotic-resistant bacteria will be the leading cause of death worldwide. Therefore, studies monitoring antibiotic resistance are required, not only in hospital settings but also in the environment, for the establishment of sanitary measures to avoid health risks for humans and animals.

The Xochimilco canals are an iconic tourist destination in Mexico City, comprising approximately 116 km of waterways. They have great historical, cultural, and natural value. In 1987, the canals were declared a world heritage site; however, their deterioration due to pollution has become very evident, mainly due to the expansion of urban communities. The main source of water for the canals comes from water treatment plants. However, a great number of black or grey water discharges to the canals have been reported. There are some studies indicating contamination in these canals, including the presence of bacteria, viruses, and other infectious agents.

In this study we aimed to investigate whether the water of the Xochimilco canals contains pathogenic bacteria like *Klebsiella*, *Shigella*, *Salmonella*, *Enterobacter*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*, as well as whether antibiotic resistance can be found in this water. For this, 16S rDNA amplicon metagenomics and microbiological analyses were performed in samples collected from five different sites of the canals expecting to have different anthropogenic impact. Notably, the metagenomic analysis revealed very different bacterial communities in the site with the greatest anthropogenic impact, compared with the other four sites that showed bacterial communities very similar among them. For the sites showing different bacterial communities, the most abundant bacteria were related to the sulfur biogeochemical cycle, organic matter degradation, and some were human and animal gut commensal. For the other four sites, the most abundant bacteria were related to organic matter degradation, as well as to the biogeochemical cycle of Oxygen, Nitrogen, Carbon and Hydrogen. Microbiological analysis revealed the presence in the five sampling sites of bacteria from genera like *Pseudomonas*, *Aeromonas*, *Citrobacter*, *Enterobacter*, *Klebsiella*, *Enterococcus*, *Staphylococcus*, *Bacillus*, *Priestia* and *Rosellomorea*, most being environmental bacteria. Analysis of antibiotic resistance revealed low percentages of resistance for ampicillin, nalidixic acid, kanamycin, streptomycin, and tetracycline. Our study indicates low presence of pathogenic bacteria and of antibiotic resistance in the samples analyzed of the water from Xochimilco canals.

DETERMINATION OF ISONIAZID TOLERANCE OF BCG PASTEUR ATCC 35734 AND ITS ISOGENIC MUTANT DERIVATIVE BCG Δ BCG1419C AS PLANKTONIC AND BIOFILM CULTURES

Lorenzo Rodriguez Luis Manuel¹, Flores Valdez Mario Alberto², Jeannette Barba León³, Bravo Madrigal Jorge²

¹Maestría en Microbiología Médica en Centro Universitario de Ciencias de la Salud (UdG), 2. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco (CIATEJ). 3. Centro Universitario de Ciencias Biológicas y Agropecuarias de la Universidad de Guadalajara. Av. Normalistas #800, Lomas de la Normal, CP. 44270, Guadalajara, Jal. Tel. 7474988016. E.mail: luislorenzo9810@gmail.com

Introduction. *Mycobacterium tuberculosis* (MTB) is the etiological agent of tuberculosis in humans, which has acquired great importance as a public health problem at present, due to the large number of cases and deaths it causes (1). The bacterium has developed natural survival mechanisms within the host, through its adaptation to stress changes in its environment. In vitro biofilm formation has been documented as one of these mechanisms, which contains free mycolic acids (2), cellulose and extracellular polysaccharides (3,4), and has been shown to increase tolerance to first-line antimycobacterial agents such as isoniazid, commonly used for the treatment of tuberculosis, as compared with planktonic cells. Similarly, the BCG Δ BCG1419c mutant has been reported to have increased biofilm formation in vitro. No tolerance assays have been documented in BCG thus far to support the similarity of the results already reported in MTB. Here, by using the BCG Δ BCG1419c mutant strain, we aim to determine whether its increased biofilm production in vitro correlates or not with increased drug tolerance compared with BCG. **Objective.** The purpose of this work is to analyze the tolerance of biofilm and planktonic cells of wild type BCG and BCG Δ BCG1419c to the use of isoniazid (INH) *in vitro*. **Methods.** Viability was determined by colony forming units per milliliter (CFU/mL), before and after challenge with INH, in planktonic bacteria, under static and shaking conditions, as well as in biofilm cultures. The biofilm formed by these strains was quantified by crystal violet staining read at OD_{595nm}. The results were evaluated by the Shapiro-Wilk test for data distribution, then the hypothesis was evaluated by Student's t-test or Mann-Whitney test, and ANOVA or Kruskal-Wallis. **Results and discussion.** We found that planktonic cells, either in static environment or in shaken cultures, had a similar decrease in CFU/mL, whereas a higher drug tolerance was observed for biofilm cultures of both BCG strains, with a greater amount of CFU/mL observed in the mutant strain, coinciding with previous reports of the relationship of the biofilm with tolerance to INH. **Conclusion.** Our results are in agreement with those reported in MTB, where the biofilm is implicated in greater tolerance to INH, in contrast to planktonic growth, which did not present an important difference under static or shaken conditions. Of note, in biofilm growth, higher CFU/mL were obtained for BCG Δ BCG1419c compared with wild type BCG after INH treatment, therefore showing its increased tolerance to this antibiotic under this condition. **Keywords.** *Mycobacterium tuberculosis*, tolerance, BCG, BCG Δ BCG1419c

References

1. Organization WH. Global Tuberculosis Report 2022 [Internet]. 2022. Disponible en: <http://apps.who.int/bookorders>.
2. Ojha AK, Baughn AD, Sambandan D, Hsu T, Trivelli X, Guerardel Y, et al. Growth of *Mycobacterium tuberculosis* biofilms containing free mycolic acids and harbouring drug-tolerant bacteria. *Molecular Microbiology*. julio de 2008;69(1):164.
3. Trivedi A, Mavi PS, Bhatt D, Kumar A. Thiol reductive stress induces cellulose-anchored biofilm formation in *Mycobacterium tuberculosis*. *Nat Commun*. el 25 de abril de 2016;7(1):11392.
4. Chakraborty P, Bajeli S, Kaushal D, Radotra BD, Kumar A. Biofilm formation in the lung contributes to virulence and drug tolerance of *Mycobacterium tuberculosis*. *Nat Commun*. el 11 de marzo de 2021;12:1606.

ANTIMICROBIAL ACTIVITY FROM MAGNOLOL AND HONOKIOL FROM *MAGNOLIA SPP* ON PERIIMPLANTITIS CAUSING PATHOGEN

Jesús Agustín Loyola Alonso¹, Ximena Valdés Sánchez¹, Antonio Ramses Vargas Lara¹, Alma Rosa González Pérez², Suria Gisela Vásquez Morales², Ismael Secundino Velazquez³, Cruz Eugenia Martínez Palacios², Juana Elizabeth Reyes Martínez², Ruth Reyes Cortes²

¹Licenciatura en Químico Farmacéutico Biólogo, Universidad de Guanajuato,²División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Noria Alta S/N; C.P. 36050; Guanajuato, Gto. ³Universidad Lasalle Bajío, Facultad de Odontología, Av. Universidad 602, Col. Lomas del Campestre, León, Gto. México.
Tel: (+52) 473-7320006 Ext: 1451; E-mail: reyes.ruth@ugto.mx

Nowadays, implantology has become widely used and a treatment of choice for teeth loss. However, recently an increased number of cases of periimplantitis and mucositis have been observed, the exacerbated inflammation causes the loss of periodontal tissue. Among the most common etiological agents causing periimplantitis have been identified anaerobic facultative microorganisms as *Streptococcus spp.* and *Enterococcus spp.* or *Staphylococcus aureus*, also anaerobic organisms as *Porphyromonas gingivalis*, *Prevotella intermedia* causing polymicrobial infections on a complex biofilm. Due the unregulated use of antibiotics, has been emerged a drug resistant pathogens clindamycin, doxycycline, metronidazole, amoxicillin and some combinations are frequently found resulting in chlorhexidine topical use as alternative treatment.

In this research was characterized a bacterial culture isolated from an 82-year-old female with periimplantitis to further investigate the antimicrobial activity of bioactive compounds magnolol and honokiol identified in Magnolia botanical extracts which previously shown to affect the bacterial growth of some reference strains. However, the activity on drug resistant clinical isolates causing periimplantitis has not been tested before. The growth conditions were standardized resulting Brain Heart Infusion media (BHI) and Cation Adjusted Muller Hilton (CAMH) at 37° C and aerobiosis condition as optimal for growth. Gram-positive cocci were observed forming short chains. Biochemical and molecular test were performed resulting in the identification of an *Enterococcus spp* pathogenic strain. To test the antimicrobial activity of mentioned bioactive compounds, diffusion disc test was performed resulting in antibacterial activity over *Enterococcus faecalis* ATCC 29212 strain but not on the clinical isolate.

References

1. Sánchez-Gárce, M. A., & Gay-Escoda, C. (2004). Periimplantitis. *Medicina oral, patología oral y cirugía bucal*, 9 Suppl, 69. PMID: 15580138
2. Smeets, R., Henningsen, A., Jung, O., Heiland, M., Hammächer, C., & Stein, J. M. (2014). Definition, etiology, prevention and treatment of peri-implantitis—a review. *Head & face medicine*, 10, 34. <https://doi.org/10.1186/1746-160X-10-34>

ANTI-QUORUM SENSING EFFECT OF THE CYCLODIPEPTIDE CYCLO (PRO-TYR), CYCLO (PRO-VAL), AND CYCLO (PRO-PHE) ON THE RHL SYSTEM IN *PSEUDOMONAS AERUGINOSA* PAO1

Valeria Madrigal Pineda¹, Lorena Martínez Alcantar¹, Jesús Campos García¹

¹Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo
F. J. Mújica s/n, Edificio U3, Ciudad Universitaria, Morelia, Michoacán, México, C.P. 58030.
E-mail: lorena.martinez@umich.mx

Pseudomonas aeruginosa regulates the production of a wide variety of virulence factors through quorum sensing systems. Quorum sensing (QS) within bacterial populations involves producing, secretion, and exchanging diffusible small molecules or peptides called autoinducers. Once the autoinducer reaches a minimal concentration, it binds to its cognate transcriptional regulator, modulating gene expression [1]. The major quorum sensing network in *P. aeruginosa* consists of three interconnected systems: Las, Rhl, and PQS. The Rhl system consists of the RhlI synthase and the transcriptional regulator RhlR. RhlR binds to the autoinducer *N*-butanoyl-L-homoserine lactone (C4-HSL), the product of the RhlI synthase. RhlR:C4-HSL directs the expression of many genes, including those encoding virulence factors such as pyocyanin, elastases, and rhamnolipids [2]. Cyclodipeptides (CDPs), also known as 2,5-diketopiperazines, are the minor cyclic peptides commonly found in nature and are synthesized mainly by microorganisms [3]. Data points to CDPs as a novel group of biomolecules influencing QS signaling. To understand the physiological involvement of the CDPs produced by *P. aeruginosa* on the Rhl-dependent quorum system was evaluated the effect of a mixture of the CDPs cyclo (Pro-Val), cyclo (Pro-Tyr), and cyclo (Pro-Phe). Virulence factors such as proteases, elastases, pyocyanin, pyoverdine, rhamnolipids, and biofilm were quantified in cultures of the *rhlI*Δ and *rhlR*Δ mutant strains supplemented with different concentrations of the CDPs mixture. The results showed that adding the CDPs did not modify the growth kinetic of the strains; however, these differentially modify the production of the virulence factors evaluated. Findings suggest that the CDPs may be considered a novel QS system interfering with the Rhl system and modulation of the *P. aeruginosa* pathogenesis.

References

1. García-Diéguez, L., et al., (2023). Periodically disturbing biofilms reduce the expression of quorum sensing-regulated virulence factors in *Pseudomonas aeruginosa*. *iScience*, 26(6):106843.
2. Mukherjee, S., et al., (2017). The RhlR quorum-sensing receptor controls *Pseudomonas aeruginosa* pathogenesis and biofilm development independently of its canonical homoserine lactone autoinducer. *PLoS Pathogens*, 13(7), e1006504.
3. Mishra, A. K., et al., (2017). Cyclodipeptides: An overview of their biosynthesis and biological activity. *Molecules*, 22(10), 1796.
4. Canu, N., et al., (2020). Cyclodipeptide synthases: a promising biotechnological tool for the synthesis of diverse 2, 5-diketopiperazines. *Natural Product Reports*, 37(3), 312-321.

DESIGN OF A MULTIPLEX PCR FOR THE DETECTION OF MYCOPLASMA AND UREAPLASMA SPECIES ASSOCIATED WITH REPRODUCTIVE DISORDERS IN CATTLE

Anabelle Manzo-Sandoval¹, Ana Victoria Zermeño-Granados^{1,2}, Ana Silvia González-Méndez¹,
Laura Jaramillo-Meza¹, Fernando Díaz-Otero¹, Laura Hernández-Andrade¹

¹Centro Nacional de Investigación Disciplinaria en Salud Animal e Inocuidad (CENID-SAI), Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP), México City, México, ²Facultad de Medicina Veterinaria y Zootecnia, Universidad Nacional Autónoma de México, México City, México.

E-mail: manzo.anabelle@inifap.gob.mx; teléfono: 5538718700 Ext: 80390

Some species of the genera *Mycoplasma* and *Ureaplasma* are pathogenic for humans and animals. In dairy cattle, *Mycoplasma bovis*, *Mycoplasma bovis* and *Ureaplasma diversum* can cause pneumonia, conjunctivitis, polyarthritis, otitis, mastitis, agalactia, abortion, granular vulvovaginitis and infertility. Traditional detection and identification methods rely on culture and serological techniques, necessitating a lengthy incubation period of 7 to 10 days, leading to several limitations in detection. Due to the above, both endpoint PCR and real-time PCR are used for the rapid detection of these microorganisms; however, most of the proposed protocols are based on the identification of a single species (simple PCR). Therefore, the objective of this study is to design and standardize a multiplex PCR for use in studies to understand the potential involvement of *M. bovis*, *M. bovis* and *U. diversum* in reproductive disorders on dairy farms in Mexico. Primers for the PCR assay were designed and evaluated using different software tools including Primer 3 Plus, the Primer Designing Tool from NCBI, the PrimerQuest Tool from Integrated DNA Technologies (IDT) and Geneious. These primers were designed based on specific regions of the *uvrC* gene (encodes deoxyribodipyrimidine photolyase, an enzyme of the excision DNA-repair system) for *M. bovis* and the 16S-23S rRNA internal transcribed spacer regions for *M. bovis* and *U. diversum*, sourced from the GenBank database of the National Center for Biotechnology Information (NCBI). On the other hand, with Geneious software, synthetic genes (gBlocks) were constructed and synthesized to be used as templates to standardize the PCR. Under the established conditions, the expected products of 429 bp (*M. bovis*), 294 bp (*U. diversum*) and 141 bp (*M. bovis*) were amplified. The specificity of this PCR was determined with the bacterial genera *Salmonella*, *Streptococcus pyogenes*, *Escherichia coli*, *Staphylococcus aureus*, *Proteus*, *Klebsiella* y *Pseudomonas*, no amplification of nonspecific products was observed. Assays are currently underway to determine the analytical sensitivity of this molecular test. The proposed multiplex PCR can be a good diagnostic alternative to differentiate between these three species of interest, it does not require the viability of the pathogens as with traditional microbiological isolation, rapid results are obtained compared to culture and it is more economical.

ANTIFUNGAL ACTIVITY OF IMMOBILIZED CYSTATIN MICROPARTICLES OF ALBUMIN

María Karina Manzo Valencia¹; Víctor Olalde Portugal¹; Silvia Edith Valdés Rodríguez*

¹Departamento de Biotecnología y Bioquímica, Centro de Investigación y de Estudios Avanzados del IPN-Unidad Irapuato, Irapuato, Guanajuato, México. *Corresponding author: silvia.valdes@cinvestav.mx Tel. 4626239600 Ext. 9638

Fungi are a major cause of disease in economically relevant crops, significantly reducing the quality and quantity of agricultural products, and making them difficult to control. The need to reduce the use of chemically synthesized pesticides is leading to the implementation of various methods, including biological control. Nanotechnology plays an important role in sustainable agriculture, the application of protein nanoparticles derived from plants for disease control has been reported. Phytocystatins are cysteine protease inhibitors involved in the regulation of several endogenous processes, defense against pests and pathogens, and response to abiotic stress. Phytocystatins have been used as effective molecules against a variety of phytopathogenic fungi, including *Aspergillus*, *Fusarium*, *Botrytis*, and *Phytophthora*¹. The aim of the present study was to evaluate the antifungal activity of immobilized cystatin microparticles of albumin and purified cystatin (AhCPI) on *Fusarium oxysporum* and *Alternaria* spp. *in vitro*. The fungi were isolated from diseased tomato plants. The microparticles were obtained based on the protocol described by Khramtsov et al., 2022. To determine the inhibitory activity, two tests were carried out: a) conidial germination: different concentrations of cystatin microparticles of albumin and purified cystatin were tested; spore suspensions of 1×10^4 conidia/well were inoculated into microtiter plates (24 well) containing Potato Dextrose Broth medium and incubated for 8-24 h, microscopic observations of samples were done, and germinated and ungerminated spores were counted; b) Mycelial inhibition: Potato Dextrose Agar plates containing different concentrations of the cystatin microparticles of albumin and purified cystatin were inoculated at the center with mycelium of each strain and incubated at 28 °C in the dark, fungal growth (colony diameter) was measured in two directions, during eight days each 24 h. Five replicates for each concentration were performed. The assays was repeated three times. It was found that cystatin microparticles of albumin and purified cystatin inhibited radial mycelial growth and reduced the germination and viability of spores of *Alternaria* spp. and *Fusarium oxysporum*. This strategy could provide an innovative way to control plant diseases caused by fungi.

This research work is supported by CONAHCYT project CF-2023-I-1432

References

1. Madeirus Lima, A., Pinho Dos Reis, S., and Batista de Souza, C. R. (2015). Phytocystatins and their potential to control plant diseases caused by fungi. *Protein and peptide letters*, 22, 104-111
2. Khramtsov, P., Kropaneva, M., Bochkova, M., Timganova, V., Kiselkov, D., Zamorina, S., and Rayev, M. (2022). Synthesis and application of albumin nanoparticles loaded with Prussian blue nanozymes. *Colloids and Interfaces*, 6(2), 29.

EFFECT OF EPINEPHRINE AND NOREPINEPHRINE ON THE EXPRESSION OF VIRULENCE FACTORS IN PASTEURELLA MULTOCIDA

Mayra Martínez Alfonzo¹, Juan Fernando Montes García¹, Candelario Vázquez Cruz²,
Tomas Villamar Duque¹ and Erasmo Negrete Abascal¹

¹Carrera de Biología, Facultad de Estudios Superiores Iztacala, UNAM. ²Centro de Investigaciones en Ciencias Microbiológicas, ICUAP, BUAP, Puebla, Pue., México. Av. De los Barrios #1, Los Reyes Iztacala Tlalnepantla Edo. De México CP 54090
Tel 56231254; negretee@yahoo.com

Pasteurella multocida is a Gram-negative bacterium that is part of the oropharyngeal microbiota of various domestic and wild animals, a member of the Pasteurellaceae family. It is considered of medical and economic importance because it is an opportunistic pathogen that affects various animals and humans. This work aimed to analyze the effect of epinephrine (EP) and norepinephrine (NE); hormones associated with the stress response in the host. The effect of Ep and NE (10-50 μ M) was evaluated on bacterial growth, biofilm formation and dispersion, Congo red (RC) binding, protein patterns, and proteolytic activity. Ep and NE favor growth by up to 30%, being more evident with NE after 3 hours of cultivation. The formation of biofilms increases in the presence of NE and EP in a dose-dependent manner; with 30 μ M, it increases by up to 25% compared to the control. The dispersion of biofilms is induced by NE and EP, and the amount of biofilm decreases up to 23% from 20 μ M. Both hormones increase (10%) the binding of Congo red with 20 and 30 μ M. In the protein pattern of the total extract (ExT), the decrease of a 270 kDa protein in the presence of NE and the expression of 3 bands in the 80-130 kDa range with EP is observed. In the pattern of secreted proteins (PS), a band of 95 kDa is no longer expressed with NE and another 200 kDa with EP. Proteolytic activity in 52 kDa bands increases in the absence of Ep but decreases with 20-30 μ M of NE in ExT samples. This activity disappears in proteins secreted from cultures with Ep and NE. These findings suggest that catecholamines may regulate the expression of virulence factors in *P. multocida*.

Project supported by DGAPA-UNAM PAPIIT IN204122.

ACTINOMYCETE METABOLITES ISOLATED FROM MEXICAN JUNGLE SOILS WITH ANTIMICROBIAL AND ANTI-VIRULENCE ACTIVITY AGAINST ANTIMICROBIAL-RESISTANT PATHOGENIC BACTERIA

Araceli Martínez-Larios¹, Michelle Chávez-Hernández¹, Itzel Plascencia-Ortiz¹, Graciela Castro-Escarpulli¹, Lourdes Villa-Tanaca¹, César Hernández-Rodríguez¹

¹Departamento de Microbiología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Prol. de Carpio y Plan de Ayala s/n. Col. Sto. Tomás, 11340 Ciudad de México, México. Correspondence author: César Hernández-Rodríguez. Email: chdez38@hotmail.com

Microorganisms resistant to various antimicrobial agents, usually Gram-negative bacteria, have become one of the most important public health problems, particularly within the hospital environment and healthcare facilities. Infections caused by microorganisms, resistant to antibiotics that are not properly treated, lead to the natural development of the disease and consequently increase the severity of the case and put the lives of patients at risk. On the other hand, due to the great diversity in secondary metabolism, actinomycetes represent the microbial group from which most of the bioactive compounds and antibiotics discovered and in use are derived. Currently, more than 80% of antibiotics of bacterial origin come from actinomycetes. In the past, most companies have conducted extensive programs for the discovery of metabolites with antimicrobial activity, but most of these programs have been abandoned due to the lack of commercial interest in these types of molecules. In this work, the search was carried out for actinomycete supernatants with activity on new targets in pathogenic bacteria causing diseases in humans. These lyophilized supernatants exhibited a wide variety of effects, such as, for example, antimicrobial activity, production of volatile compounds capable of inhibiting growth or modifying the growth of the colony, microscopic morphology, and inhibiting the acquisition of iron on bacterial strains of *Pseudomonas aeruginosa*, *Acinetobacter baumannii*, *Klebsiella pneumoniae*, *Salmonella Typhi*, *Staphylococcus aureus*, and *Aeromonas hydrophila*. The molecular identification was made by sequencing the 16S rRNA gene, which identified them within the genus *Streptomyces* spp.

ASSESSING ANTIBIOTIC RESISTANCE IN BACTERIA ISOLATED FROM STREET FOOD STALLS

Meneses Sánchez A. A.¹, Lira Reyes A. I.¹, Vega-Marín, C.¹, Martínez-Rojas R.¹,
Aguilar Melo C. M.*¹, González Ramírez D. F.²

¹UPIBI-IPN, Av. Acueducto, La Laguna Ticomán, Gustavo A. Madero, 07340, CDMX, ²UPIITA-IPN, Av. I.P.N. 2580, Col. Barrio La Laguna Ticomán, Gustavo A. Madero, C.P. 07340, CDMX.
E-mail*: cmaguilarm@ipn.mx

Antibiotic resistance is a public health concern because of the low response to medical treatments increase pathogens propagation and health complications. Low-fiber diets promote facultative anaerobes proliferation enhancing antibiotic resistance¹. This research analysed the antibiotic sensibility of Gram-negative bacteria isolated from food stalls located in a polygon of 1920 m in GAM borough of CDMX. The food samples were analysed by the NOM-210-SSA1-2014 and evaluated with the antibiogram disk. Each multidisc has standardized solution of 12 antibiotics (amikacin, ampicillin, carbenicillin, cephalothin, cefotaxime, ciprofloxacin, chloramphenicol, gentamicin, netilmicin, nitrofurantoin, norfloxacin, and sulfamethoxazole/trimethoprim). The results from the antibiotic sensibility test of 18 bacteria isolated from 10 food stalls indicated that all the bacteria have resistance to ampicillin. The bacteria are mostly confirmed *Escherichia coli* (*E. coli*) by the INViC test. It was observed 44% of bacteria resistance to carbenicillin. While 11% of the isolated bacteria presented resistance to Cephalothin. Nearly all the isolated bacteria have an indeterminate sensibility to the tested antibiotics. The only antibiotics that proved still effective against bacteria were chloramphenicol, ciprofloxacin, and cefotaxime. In Latino America, *E. coli* is associated with most diarrhoea cases and antibiotic resistance due to the acquisition of mobile genetic elements². The results confirm the resistance of *E. coli* isolated to antibiotics of the β -lactams class that have clinical widespread use like ampicillin case³.

References

1. Babines-Orozco, I., Balbuena-Alonso, M. M., Barrios-Villa, E., Lozano-Zarain, P., Martínez-Laguna, Y., Del Carmen Rocha-Gracia, R., & Cortés-Cortés, G. (2024), *Food and Health*, 43, 4-12.
2. Varela M.F., Stephen J., Lekshmi M., Ojha M., Wenzel N., Sanford L.M., Hernandez A.J., Parvathi, A., Kumar S.H., (2021), *Antibiotics*, 10, 593.
3. Oliver, A., Xue, Z., Tang, Y., Durbin-Johnson, B., Alkan, Z., Taft, D., Liu, J., Korf, I., Laugero, K., Stephensen, C., Mills, D., Kable, M., & Lemay, D. (2022). *Current Developments in Nutrition*, 6, 1023.

MICROBIOLOGICAL ANALYSIS OF INDOOR AIR QUALITY IN THE UNIVERSIDAD IBEROAMERICANA TORREÓN

José Antonio Martínez Villalba, Giuliana Zanella Braña, Regina Reyes Montelongo and Myriam Michell García Luján

Universidad Iberoamericana Torreón, Calzada. Iberoamericana No. 2255 Ejido la Unión. CP 27420 Torreón, Coahuila. México. Tel (871)7051019; e-mail: jose.martinez@iberotorreon.edu.mx

Some microorganisms are related to indoor contamination of building causing certain health problems to its occupants. The objective of this investigation was to evaluate the spatial variability and microbiological quality of indoor air in the most frequented sites of the university. The obtainment of the biological samples took place in 66 sites (33 indoor and 33 outdoor) by the sedimentation method. Petri dishes were placed open for 60 min and evenly distributed at the standard height of 1.20 m from the floor. Nutrient and selective agars were used to obtain bacteria and fungi. After incubation, their metabolic characteristics were determined by biochemical tests. Colony forming units (CFU) were counted per m³ of air according to the equation described by Bogomolova and Kirtsideli¹ (2009): $N=5a \cdot 10^4 (bt)^{-1}$. In addition, using the climatological data from the METPAK II station of the University, an exploratory analysis of the biological material of the sampled sites was carried out for IDW and Kriging interpolations using the WRPLOT and Qgis software. The biological material collected at the sampling sites was mostly identified as: 1) pathogenic bacteria of the genera *Salmonella*, *Klebsiella*, *Escherichia*; and 2) fungi of the genera; *Aspergillus*, *Trichophyton*, *Candida*. In indoor spaces was an average of 5,680 CFU/m³ of bacteria and 5,810 CFU/m³ of fungi in the period from february to april 2024. The places with the highest detectable concentrations in internal areas were the cafeteria, auditorium and library. Factors such as poor ventilation, particle resuspension, the dust storms of the region and humidity favor the reproduction of microorganisms indoors. Exposure to these pathogens could lead to mucosal sensitization, allergies, asthma, development of respiratory symptoms, chronic cough, laryngitis, sinusitis and urticaria that have been experienced by users. It is important to mention that in Mexico there is no normative parameter for indoor air microbiological quality.

References

1. Bogomolova, E. & Kirtsideli, I. (2009). Airborne fungi in four stations of the St. Petersburg Underground railway system. *International Biodeterioration & Biodegradation*, 63(2), 156-160. <https://doi.org/10.1016/j.ibiod.2008.05.008>

ANALYSIS OF NAD⁺ SELF-SUFFICIENCY IN *AVIBACTERIUM PARAGALLINARUM*

Sergio Alberto Matías Vera¹, María Patricia Sánchez Alonso¹, Ma. Elena Cobos Justo¹, Vladimir Morales Erasto², Erasmo Negrete Abascal³, Candelario Vázquez Cruz¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla, CU, Edificio IC11, 24 Sur y Av. San Claudio. San Manuel, CP. 72570, Puebla. ²Facultad de Medicina Veterinaria y Zootecnia, UNAM, CU, Av. Universidad No. 3000, Coyoacán, CP. 04510, CDMX. ³Facultad de Estudios Superiores Iztacala UNAM, Av. de los Barrios no. 1. Los Reyes Iztacala, 54090 Edo. de México.
E-mail: sergio.matiasvera.icuap@viep.com.mx; ecobacilos@yahoo.mx

Avibacterium paragallinarum is a Gram-negative pathogenic bacterium of poultry, a member of the *Pasteurellaceae* family, which causes losses in egg and meat production in the poultry sector. The most well-known and studied *A. paragallinarum* strains are the NAD-dependent variety¹. However, *A. paragallinarum* NAD-independent strains have also been reported; some authors have suggested that the presence of plasmids in these strains is responsible for the independence of NAD, although this has not yet been proven in Mexican strains². For this reason, we studied 4 NAD-independent strains of *A. paragallinarum*, we have identified through growth curves that strains VLA23 and VLA24 have better growth in synthetic medium than NAD-dependent strains. To know the possible genes that could confer NAD independence to these strains, the genome of strain VLA23 was sequenced by Illumina technology, to later be assembled with SPAdes and annotated on the RAST platform, obtaining an assembly with 47 contigs and 40.78% GC. Finally, bioinformatics analysis was performed for comparison of NAD-independent and NAD-dependent strains available in GenBank to study the phylogenetic relationship and conservation between genomes. Additionally, the presence of deduced genes and proteins “Nicotinate phosphoribosyl transferase” encoded by *nadV_{avpg}-nampt* gene of 1095 bp and “NAD synthetase” encoded by *nadE* gene of 738 bp was observed. The NadV protein is conserved in members of the *Pasteurellaceae* family, but distantly related to the *Enterobacteriaceae*, whereas NadE is more conserved in both groups of bacteria. In conclusion, in this work, we report for the first time the genes that could be responsible for the independence of NAD in Mexican strains of *A. paragallinarum*.

References

1. Horner, R. F., Bishop, G. C., & Haw, C. 1992. Avian Pathology, 21(3), 421–427.
2. Soriano-Vargas, E., Sánchez-Morales, J., Morales-Erasto, V. & García-Sánchez, A. 2013. Veterinaria México, 44(1), 57-62

IDENTIFICATION AND IMPLICATIONS OF THE N501Y MUTATION IN SARS-COV-2 ISOLATES FROM A MEXICAN POPULATION

Angel Medina Reyes¹, Francisco Josué Carrillo-Ballesteros², Diana Emilia Martínez-Fernández², Dulce Yamileth Medina Valenzuela¹, Sara Landeras-Bueno³, Cesar Arturo Nava-Valdivia¹, Jose Angel Regla-Nava¹

¹Department of Microbiology and Pathology, University Center for Health Science (CUCS), University of Guadalajara, Sierra Mojada 950, Col. Independencia, C.P. 44340, Guadalajara, Jalisco, México, ²Transdisciplinary Institute of Research and Services (ITRANS), University of Guadalajara, Av. José Parres Arias 5, Rinconada de la Azalea, Industrial Belenes, C.P. 45150, Zapopan, Jalisco, México, ³Universidad Cardenal Herrera-CEU, CEU Universities, Carrer Lluís Vives, 1, 46115 Alfara del Patriarca, Valencia, España.

E-mail: jose.regla@academicos.udg.mx, Phone: +52 33 10585200 Ext 34192

Introduction. Coronavirus disease 2019 (COVID-19) is a highly contagious respiratory disease caused by the recent and novel coronavirus SARS-CoV-2. It is an RNA virus that is known to accumulate many mutations in the genome. Specifically, the spike protein tends to have a higher mutation rate, particularly in the Receptor Binding Domain (RBD). These mutations in genome can result in aminoacidic change, which could then impact the structure or special conformation of the proteins, thus affecting several of its functions. In the case of SARS-CoV-2, the RBD region is a critical component for union receptor and a major target of neutralizing antibodies, increasing transmissibility, disease severity and vaccine development. **Objective.** In this study, we analyzed mutations in the RBD region derived from SARS-CoV-2 patient samples in the city of Guadalajara between December 2020, and March 2023. **Methodology.** To accomplish the identification of the mutations, we performed polymerase chain reaction (PCR) to amplify the RBD, which was then sequenced using sanger methodology to detect the mutations. To analyze its impact on the conformational change, 3D modelling of the RBD domain with the mutations were made and compared with reported structures of wildtype SARS-CoV-2 RBD. Finally, we performed a thermodynamic analysis of the possible impacts of structural and conformational change via the analysis and comparisons of binding free energy, hydrogen bonds, salt bridges and dissociation constant of the mutation against the WT variant. **Results.** We found that the N501Y mutation was present in our population, and its presence affected the free binding energy of the interaction between the RBD and the ACE2 receptor. We also hypothesized that this N501Y mutation was implicated in the escape of antibody neutralization, and was consistent with others experimental results, where they observed that this change has necessary for the higher rate of transmission of the Omicron variant of SARS-CoV-2. Further monitoring of the evolution of mutations in other proteins may also contribute to the severity, immune escape, and vaccine development.

References

1. Uzoeto HO, Ajima JN, Arazu AV, Ibiang GO, Cosmas S, Durojaye OA. Immunity evasion: consequence of the N501Y mutation of the SARS-CoV-2 spike glycoprotein. *J Genet Eng Biotechnol.* 2022 Jan 14;20(1):10.
2. Laffebber C, de Koning K, Kanaar R, Lebbink JHG. Experimental Evidence for Enhanced Receptor Binding by Rapidly Spreading SARS-CoV-2 Variants. *J Mol Biol.* 2021 Jul 23;433(15):167058.
3. Harvey WT, Carabelli AM, Jackson B, Gupta RK, Thomson EC, Harrison EM, Ludden C, Reeve R, Rambaut A; COVID-19 Genomics UK (COG-UK) Consortium; Peacock SJ, Robertson DL. SARS-CoV-2 variants, spike mutations and immune escape. *Nat Rev Microbiol.* 2021 Jul;19(7):409-424.

BACTERIAL ADHESINS IMPLICATED IN ROOT AND GUT ATTACHMENT

Gerardo Mejia^{1,2}, Andrés Moya^{3,4,5} and Luis D. Alcaraz^{1*}

¹Laboratorio de Genómica Ambiental, Departamento de Biología Celular, Facultad de Ciencias, Universidad Nacional Autónoma de México, Mexico City, Mexico. ²Posgrado en Ciencias Bioquímicas, Universidad Nacional Autónoma de México, Mexico City, Mexico. ³Instituto de Biología Integrativa de Sistemas (I2Sysbio), UV-CSIC, Valencia. ⁴Fundación para el Fomento de la Investigación Sanitaria y Biomédica, FISABIO, Valencia. ⁵CIBER en Epidemiología y Salud Pública (CIBEResp), Madrid.

*Correspondence author:

Edificio A Segundo Piso Laboratorio de Biología Molecular y Genómica Facultad de Ciencias, Universidad Nacional Autónoma de México 04510 Coyoacán Ciudad de México. +52 55 5622-8222 ext. 44704
lalcaraz@ciencias.unam.mx

Adhesins are proteins that allow bacteria to adhere to biotic and abiotic surfaces. In bacteria, adhesion to host cells is crucial for colonization and interaction. In addition to mediating interactions with host cells, adhesins may be involved in biofilm formation and other bacterial interactions. Many bacteria, including pathogens and mutualists, express adhesins on their surface to start host colonization. Bacteria attachment to roots and the human gut marks the beginning of interactions that can negatively or positively influence the host's health. Furthermore, the study of adhesins has significant implications for developing vaccines against pathogens and understanding the importance of beneficial bacteria.

Despite the advances in the search for adhesins using bioinformatic methods, much remains to be understood about whether these proteins are shared between the bacteria inhabiting plant roots and those in the human gut. In this study, we aim to investigate adhesins shared between rhizosphere-associated bacteria and human gut-associated bacteria, as well as those unique to each system. Adhesins found in both systems may be key to attaching to both environments. On the other hand, specific adhesins are exclusive to each environment and could represent the adhesion proteins that are especially important in each. Studying how bacteria attach to biotic surfaces could help develop techniques to prevent pathogen infections or improve host health by modifying or controlling bacterial adhesion in contexts such as human health or agriculture.

Keywords. Root, gut, microbiome, adhesion, adhesins.

PRODUCTION AND CHARACTERIZATION OF CHITINASE ACTIVITY BY BACILLUS SPP. STRAINS WITH ANTAGONISTIC ACTIVITY AGAINST FUSARIUM VERTICILLIOIDES

Ma. Fernanda Meneses-Hernández¹, Stefhany Segoviano Correa¹,
A. Fabiola Camacho-Gamiño¹, Javier Plasencia de la Parra^{1*}

¹Depto. de Bioquímica, Facultad de Química, UNAM., 04510. Cd. Mx. México.
315249536@quimica.unam.mx

Globally, maize production losses range between 30% and 60% due to abiotic and biotic factors. Among microbial pathogens in maize, *Fusarium verticillioides* is the predominant fungus, causing diseases in seedlings, stems, and ears. *F. verticillioides* produces several mycotoxins that have harmful health effects on animals and humans. Disease control through fungicide application is often not feasible due to costs and the environmental contamination caused by chemical residues. A sustainable alternative to chemical control is the use of microorganisms with antagonistic activity against plant pathogens. Following this approach, we isolated two *Bacillus spp.* strains (CEL1 and AC1) that exhibit strong antagonistic activity against *F. verticillioides* in culture. One potential mechanism of action for this antagonism is the production of chitinases and β -1,3-glucanases, which target the fungal cell wall. We tested the ability of both strains to produce these enzymes by culturing them in potato dextrose broth supplemented with various substrates as inducers. We found that adding 0.02% wheat bran to the liquid media resulted in higher chitinase and glucanase activities. The proteins were precipitated with ammonium sulfate (60% saturation) and dialyzed against 50 mM acetate buffer. Chitinase activity was tested at various pH levels (4, 6, 8, and 10), but no differences in activity were observed. Similarly, the enzymatic activity remained constant at 28°C, 37°C, and 50°C. The protein preparation was sterilized through filtration and tested for antifungal activity in a microtiter plate, showing a 25% reduction in fungal growth. Our results indicate that hydrolytic enzyme production contributes to the antagonistic activity of *Bacillus spp.* against *F. verticillioides*. Experiments are underway to further characterize and compare the activities between the two strains. Funding: PAIP 5000-9121 FQ-UNAM.

MICROBIAL EXAMINATION OF FOOD: COMPARATIVE STUDY OF FAECAL COLIFORMS PRESENCE IN COOKED VS. UNCOOKED FOODS

Lira Reyes A. I.¹, Meneses Sánchez A. A.¹, González Ramírez D. F.*², Aguilar Melo C. M.¹, Salgado Ramos I. de J.³

¹UPIBI-IPN, Av. Acueducto, La Laguna Ticomán, Gustavo A. Madero, 07340, CDMX, ²UPIITA-IPN, Av. I.P.N. 2580, Col. Barrio La Laguna Ticomán, Gustavo A. Madero, C.P. 07340, CDMX., ³CIDETEC, Av. Juan de Dios Bátiz S/N, Nueva Industrial Vallejo, 07700 Gustavo A. Madero, CDMX.
E-mail*: denisse_amr@hotmail.com

In Mexico City, street food is a common occurrence. In 2023 according to the Mexican Ministry of Health, Mexico City registered 110,783 cases of intestinal infections¹. The most vulnerable to complications due to gastrointestinal infections are children below 4 years old whose deaths reached 495 in 2022². This study comprehended a polygon of 1920 m at Gustavo A. Madero borough in Mexico City, where 10 food stalls were chosen to determine the presence of faecal coliforms by the NOM-210-SSA1-2014. The food samples cooked and uncooked were blended and inoculated on lauryl tryptase broth at 45.5 ± 0.2 °C for 48 hours. The positive samples were grown in EMB agar at 36 °C for 24 h to confirm that they were *Escherichia coli* (*E. coli*) observing the characteristic metallic green colonies and negative testing for Gram stain. The samples of uncooked food tested positive for *E. coli* in nearly all the sites except for Site #1, #3, and #4 where the samples consisted of salads of lettuce and carrots. Regarding the cooked food, the food samples (all of them with sauce) in sites #5, #7 and #8 tested positive against *E. coli*. From the results, we conclude that most of the stalls have *E. coli* in their food despite being cooked or uncooked. The handling of food under unsanitary conditions might be the cause of *E. coli* presence. Since the SARS-Cov-2 pandemic, there has been more awareness of handwashing and surface disinfection. However, evidence indicates that restaurants are the main cause of foodborne disease even after the pandemic³. On the other hand, Mexico is a country that presents marked income disparities which limits the obligation to adapt and assess sanitary conditions in food stalls⁴.

References

1. Secretaria de Salud. (2024). Boletín epidemiológico.
2. INEGI. (2023). Estadísticas de defunciones registradas (EDR) 2022.
3. Stewart, C. K., Freid, R. D., Brewster, R. K., Sutherland, E. Q., Best, E. A., & Cappello, M. A., (2023) J Public Health Emerg, 7, 12-12.
4. Housni, F.E.; Lares-Michel, M. (2024) Sustainability, 16, 2811.

MICROBIOLOGICAL CHARACTERIZATION OF MOTHER OF VINEGAR AND ITS INHIBITORY EFFECT

Gloria Beatriz Molina Valdés¹, José Carlos Parada-Fabián¹, Elsa Irma Quiñones Ramírez¹
Iván Natividad-Bonifacio¹, Marcos Francisco Hernández-Robles¹, Carlos Ramón Vázquez Quiñones¹,
Carlos Vázquez Salinas², Ana Karen Álvarez Contreras¹

¹Escuela Nacional de Ciencias Biológicas-IPN Santo Tomás, Manuel Carpio, Plutarco Elías Calles, Miguel Hidalgo, C.P. 11350 CDMX. ²Universidad Autónoma Metropolitana-Iztapalapa, San Rafael Atlixco 186, Leyes de Reforma 1A Sección, Iztapalapa, C.P. 09310 CDMX.
E-mail: gloria.02.10.2000@gmail.com

Introduction. Mother of vinegar is a cellulose matrix formed by a microbial consortium between bacteria and yeasts, in which ethanol and acetic acid are present as final products. There are reports about the presence of microorganisms with probiotic potential in commercial vinegar and fruit vinegar, which have the capacity to inhibit foodborne pathogens growth.

Methodology. The pH, titratable acidity and isolation were performed on different fermentation days (0 and 7 days). Specific culture media were used for isolation: Man, Rogosa and Sharpe, yeast extract glucose agar with calcium carbonate, mannitol egg yolk agar, and acidified potato dextrose agar. To select those isolates with probiotic potential, they were pooled and evaluated for antimicrobial activity against pathogenic bacteria such as: *Salmonella* Typhi, *Salmonella* Abony, *Escherichia coli*, *Bacillus cereus*, *Staphylococcus aureus*, and *Cronobacter sakazakii*; using the modified Kirby Bauer method. In the evaluation of the antimicrobial activity of the vinegar mother and artisanal vinegar, inhibition tests were carried out at 0, 3 and 7 days of fermentation, using the method and pathogens mentioned above.

Results. At day 0, vinegar pH was 3 with a titratable acidity of 4 meq; at day 7, pH was 4, acidity remained constant. 100 isolates were obtained (65 cellulose matrix, 35 vinegar) with 60% Gram negative bacteria, 20% Gram positive bacteria, 10% yeasts, and 10% yeast-Gram negative bacteria consortium. 27.7% (5/18) of the pools showed antimicrobial activity against all pathogens. Mother of vinegar and artisanal vinegar exhibited inhibition against all pathogens at 0 and 3 days except for *E. coli* on day 7.

Conclusion. During mother of vinegar fermentation at different periods of time a variation of pH was observed, while the titratable acidity remained constant. The microbial population with the highest predominance in the sample was Gram negative bacteria. The pool isolates showed antimicrobial activity against all the pathogens used in this study, as well as the mother of Vinegar and the artisanal vinegar.

References

1. Maciuga, O. M., Sánchez-Juanes, F., & Menéndez, E. (2022). Isolation and identification of microorganisms present in probiotic beverages: the case of kombucha. *FarmaJournal*, 7(2), 29-39.
2. Dávalos Cerrón, P. Y. (2022). Isolation and characterization of native strains of *Komagataeibacter xylinus* and comparison of their growth on different substrates. *Universidad Técnica de Ambato*

KILLER YEAST AND GLUCOSE MEDIATED SYNTHESIS OF NANOPARTICLES WITH BIOLOGICAL EFFECT

Carlos Alberto Molina-Vera¹, Verónica Morales-Tlalpan^{1,2}, Adriana Gallardo-González², Jorge Chávez-Servín¹, Juan Campos-Guillén³, Luis Gabriel Brieba-de-Castro⁴ and Carlos Saldaña^{1,2*}

¹Laboratorio de Biofísica de Membranas y Nanotecnología. ²Laboratorio Nacional de Visualización Científica Avanzada Facultad de Ciencias Naturales (LAVIS-FCN-UAQ). ³Facultad de Química. Universidad Autónoma de Querétaro. ⁴Structural Biochemistry Group, Unidad de Genómica Avanzada (LANGE BIO), Centro de Investigación y de Estudios Avanzados del IPN, Guanajuato, Mexico.

*carlos.saldana@uaq.mx Tel. 442 192 1200, ext 65332

Recently, novel nanobiotechnological approaches have been proposed to reduce the impact of pollutants during the synthesis of nanoparticles (Nps). To do so, green synthesis methods are now in trend, using biomolecules to produce high-quality and stable nanoparticles. Some strains of *Saccharomyces cerevisiae* can produce toxins, for the case of strain 42300 (ATCC), *K1* is produced. The *killer* effect of this toxin is related to the recognition of the cell wall receptor 1-6- β -D-Glucan, the channel Tok1p, and the receptor Kre1p². The inhibition effect is evident in sensitive strains of *S. cerevisiae*, showing inhibition zones due to the activity of *K1*. This study aims to use the *K1* toxin produced by *S. cerevisiae*, and D-(+)-Glucose as both, a reducing agent, and a capping agent to produce Magnesium nanoparticles using the green synthesis method. The synthesis of MgK1 Nps was prepared using a cell-free overnight culture medium of *S. cerevisiae* 42300 containing the secreted *K1* toxin. In parallel, D-(+)-Glucose was used to obtain MgGlu-capped Nps, and the combination MgGluK1. Different temperatures, pH, and concentrations were tested to get the highest yield in the synthesis of Nps. The obtained nanoparticles were detected using UV-Vis spectroscopy. The toxic effect was determined against *S. cerevisiae*. Additionally, the activity of Nps on mitochondria was tested using MitoTracker Deep Red. The optical property of Mg depends highly on the concentration of the precursor. The absorption spectra of Mg nanoparticles were recorded in the range of 200-600 nm, showing a single absorption peak obtained between 288-300 nm for the distinct preparations, corresponding to the SPR of Mg nanoparticles reported previously³. The effect of synthesized Nps shown to be effective against *S. cerevisiae*, affecting the viability of mitochondria, and decreasing their activity completely.

We extend our sincere gratitude to the funding institutions: FONDEC-UAQ, FONFIVE 2024 and CONAHCyT A1-S-26966 for their generous support and financial backing provided to Carlos Saldaña. Carlos Molina-Vera CVU: 1003111.

References

1. Gier, S., Schmitt, M., & Breinig, F. (2020). *mSphere*, 5(1) e00979-19.
2. Ying, S., Guan, Z., Ofoegbu, P. C., Clubb, P., Rico, C., He, F., & Hong, J. (2022). *Environmental Technology & Innovation*, 26. 102336.

THE POST-TRANSCRIPTIONAL REGULATOR RSM A MEDIATES THE PROTECTIVE RESPONSE AGAINST PYOCYANIN OVERPRODUCTION IN *PSEUDOMONAS AERUGINOSA* ID4365

Luis Fernando Montelongo-Martínez¹, Abigail González Valdez², Gloria Soberón-Chávez², Miguel Cocotl Yañez¹

¹Departamento de Microbiología y Parasitología, Facultad de Medicina, UNAM, México. ²Departamento de Biología Molecular y Biotecnología, Instituto de Investigaciones Biomédicas, UNAM, México.

E-mail: miguel.cocotlyanez@iibiomedicas.unam.mx

Pseudomonas aeruginosa (PA) is an opportunistic human pathogen, which causes infections that are difficult to treat, this results in high mortality rates worldwide. The virulence of this bacterium is multifactorial; however, the production of secondary metabolites with redox activity such as phenazines stands out. Clinical and environmental isolates can produce five different phenazines, among them, pyocyanin (PYO) is the most important phenazine for the pathogenesis and competence due to its stability and chemical properties. Currently, PYO production represents interest in diverse areas including biotechnology and medicine. Although PYO production provides advantages during infection and colonization, its synthesis could be toxic to the bacterial producers because of its pro-oxidant activity and ROS production. Due to these PYO-toxic effects, its synthesis is tightly regulated at different levels and involves different regulators, one of them is the posttranscriptional Rsm system where RsmA is the protein effector which prevents ribosome binding in RNA targets and blocks its translation. It has been documented that the *rsmA* deletion increases PYO production in the reference strains PAO1 and PA14, nevertheless, the environmental ID4365 and its *rsmA* mutant strain surpass PYO production compared with these reference strains in the same growth conditions. This suggests an optimized protection response against PYO overproduction, which has not been previously characterized.

To delve into this, we performed a proteomic analysis growing the ID4365 strain and its *rsmA* mutant in a low phosphate medium (PPGAS, pH 7.5) at 37 °C and 24 h, where PYO levels are increased, in order to identify targets modulated by RsmA and deepen into proteins with a protective role in these conditions. After ID_{rsmA} vs ID4365 proteomic comparison, we identified 437 differentially abundant proteins (DAPs) that were affected by *rsmA* deletion, this means that levels of 164 proteins were increased whereas 237 were reduced. This corresponds to 7 % of the total proteins codified in the genome of this marine strain (6,318 CDS). Although the vast majority of the identified proteins have not yet been classified, we identified proteins that could protect PA against PYO overproduction like efflux pumps, antioxidant enzymes, chaperones, etc. We constructed transcriptional and translational *lacZ* fusions with the most interesting targets like *groESL*, *dhpCF*, *katA*, *mexGHI-opmD*, *dhpB*, *rpoS*, and *clpP*. We showed that RsmA indirectly modulates oxidative stress response via the principal redox regulators OxyR and SoxR. Also, we reported that RsmA controls GroESL, RpoS, and ClpP at the post-transcriptional level.

THE CHORIONIC GONADOTROPIN HORMONE DECREASES THE EXPRESSION OF POSSIBLE VIRULENCE FACTORS OF *KLEBSIELLA PNEUMONIAE* IN VITRO

Juan Fernando Montes García¹, Graciela Villeda Gabriel², Erasmo Negrete Abascal¹

¹Carrera de Biología, Facultad de Estudios Superiores Iztacala, UNAM. ²Departamento de Inmunología e Infectología, INPerIER. fernando.montesg@iztacala.unam.mx

K. pneumoniae (*Kp*) is an opportunistic pathogen that causes frequent respiratory and urinary tract infections and is associated with nosocomial infections, lung infections (2%), and urinary tract infections (UTI, 4%). However, this percentage is increasing; women are more susceptible than men. In pregnant women, the risk increases due to physiological changes. Pregnancy-associated hormones excreted in the urine, such as estrogen, progesterone, and human chorionic gonadotropin (hCG), generate a very particular microenvironment in the urine, altering the microbiota and possibly the virulence of the pathogens associated with UTI. This work explored the effect of hCG (2.5, 25, 50, 250, 2500 mIU/ml) on the growth, motility, and expression of *Kp* virulence factors *in vitro*. hCG reduces the growth rate of *Kp* in a dose-dependent manner. The amount of biofilm formed decreased by 45% compared to the control with 25 mIU/ml; that same concentration induces biofilm dispersion (50%), but the dispersion is lower at a higher concentration of hCG. *In vitro*, motility increased at 25 and 50 mIU/ml. Secreted proteins of 10, 12, 18, 20, and 38 kDa were expressed in the presence of hCG; total cell extract proteins of 27, 35, and 130 kDa were induced with 50 mIU/ml. The proteolytic activity of the total cell extract in bands of 50 kDa increases but disappears in bands of 200 kDa with 25 mIU/ml of hCG or higher. The opposite is observed with secreted proteolytic activities. *K. pneumoniae* is capable of binding Congo red. hCG modifies the expression of *Kp* virulence factors.

Project supported by DGAPA-UNAM PAPIIT IN204122

IDENTIFICATION OF A PESTICIDAL CRYSTAL PROTEIN (CRY8BA) IN THE GENOME OF THE *METARHIZIUM BRUNNEUM*

Aida Gabriela Mora Acebedo, Israel Enrique Padilla Guerrero, Gloria Angélica González Hernández, Ma. Azul del Rocío Martínez Vázquez, Juan Carlos Torres Guzmán¹

¹Departamento de Biología, División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Col. Noria Alta, C.P. 36050, Guanajuato, Gto, México.
E-mail: torguz@ugto.mx

Metarhizium is a genus of entomopathogenic fungi that can colonize plant roots and promote their growth¹. Due to these properties, it is a biological control agent against various pests. It is believed that the ancestor of *Metarhizium* was endophytic, and its entomopathogenic ability arose through the acquisition of horizontal transfer genes (HGT)². We identified an HGT in the genome of *M. brunneum* using the bioinformatics tool ALIENNESS. Based on sequence homology, this gene was classified as *Cry8Ba*, which encodes a pesticidal crystal protein like Cry proteins from *B. thuringiensis*. We determined its distribution in the different genomes of the various *Metarhizium* species available on the NCBI Platform. We did not find this gene in other species, suggesting a unique and intriguing aspect of *M. brunneum*. This finding hints at the possibility that the acquisition of this gene occurred after speciation. Cry-type proteins are insecticidal proteins with three structural domains. The N-terminal domain of the protein forms pores in the membranes of intestinal cells that cause insect death³. In this work, we predict the 3D structure of *Cry8Ba* from *M. brunneum* and compare it with Cry proteins from *B. thuringiensis*. The N-terminal domain of *Cry8Ba* from *M. brunneum* forms 7 alpha-helices that structurally match the N-terminal domain of *Cry8Ba* from *B. thuringiensis*.

To investigate whether this gene, like the genes of *B. thuringiensis*, has an insecticidal effect, we deleted it by homologous recombination in *M. brunneum*. The deletion plasmid was constructed using the “One Step Construction of Agrobacterium-Recombination-ready-plasmids”⁴.

This project is supported by Conahcyt 2024, Ciencia Básica y de Frontera: CBF2023-2024-1745.

References

1. St. Leger, R., & Wang, J. (2020). *Open Biology*, 10(12).
2. Zhang, et al.,. (2019). *PNAS*, 116(16), 7982-7989.
3. Pardo-López, L., Soberón, M., & Bravo, A. (2013). *FEMS Microbiol Rev*, 37(1), 3-22.
4. Paz, Z., García-Pedrajas, M., Andrews, D., Klosterman, S., Baeza-Montañez, L., & Gold, S. (2011). *Fungal Genet Biology*, 48, 677-684.

ROLE OF *SARGASSUM FLUITANS* AS A BIOLOGICAL REDUCER OF GOLD IN SYNTHESIZING NANOPARTICLES AND ITS BACTERICIDAL IMPACT

Leonardo Naredo-Rojas¹, Mou Pal², Alvin Noé Suarez Castillo³,
Lucía Soto-Urzúa¹ and Luis Javier Martínez-Morales^{1*}

¹Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias. ²Instituto de Física. ³Centro Regional de Investigación Acuícola y Pesquera en Puerto Morelos, Instituto Mexicano de Investigación en Pesca y Acuicultura Sustentable. Benemérita Universidad Autónoma de Puebla. C.P. 72570. Av San Claudio y 24 sur Col. San Manuel. Puebla, Pue. México.

Brown algae of the genus *Sargassum* have gained prominence in recent years due to the large quantities that arrive annually on the Atlantic Ocean coasts. The overpopulation of holopelagic species of this genus in the ocean causes several tons of biomass to wash ashore yearly, with records indicating that concentrations have increased annually since 2000. In 2011, the University of Florida discovered, through satellite images, a large amount of sargassum in an unrecorded area, which they named the "sargassum belt." This belt has continued to grow, affecting countries such as Mexico, the United States, Colombia, Venezuela, and Brazil, and even reaching Ghana and the Ivory Coast (Hernández-Zanuy A. C. 2018, Optical Oceanography Lab, 2018, García-Sánchez M. et al., 2020).

In 2018, there was a significant atypical increase in these species. Satellite images show that the largest sargassum bloom in history occurred in March, April, and May of that year. In Mexico, particularly in the state of Quintana Roo, it was reported that 522,226 tons of sargassum were collected from beaches and coastal areas (Martínez-González G, 2019, Espinosa-L. A., 2020).

This situation is problematic because it affects economic activities in tourist areas. When the algae reach the beach, they begin to decompose, affecting the sand quality and completely altering the ecosystem of endemic species. It has been reported that the arrival of sargassum to reefs directly impacts endemic algae and coral species by blocking sunlight (Hernández-Zanuy A. C. 2018).

This project aims to repurpose this biomass by converting it into a reducing agent to generate metallic nanoparticles. These nanoparticles have been reported to have various applications in different fields, one of which is Microbiology. Therefore, we will verify their bactericidal activity to provide alternative methods for combating bacterial infections.

The experiments conducted so far indicate that *Sargassum fluitans*' extracts function as a reducing agent. Various extracts at different temperatures have been tested, and the optimal temperature for extracting the reducing substance has been determined. UV-visible spectroscopy has confirmed the reduction of Au ions and the production of the gold nanoparticles' plasmon. Micrographs show the nanoparticles produced in these syntheses. Additionally, we have tested the bactericidal activity at different concentrations on previously identified strains of *E. coli* and *S. aureus*. HPLC is being performed on the extract to identify the reducing metabolite extracted at its optimal temperature.

Referencias:

- Hernández-Zanuy A, C. (2018). Adaptación basada en Ecosistemas: alternativa para la gestión sostenible de los recursos marinos y costeros del Caribe. Editorial Instituto de Ciencias del mar, ICIMAR. ISBN: 978-959-298-043-3.
- Optical Oceanography Lab. (2018a). Satellite-based Sargassum Watch System (SaWS). Recuperado el 30 de diciembre de 2019, a partir de <https://optics.marine.usf.edu/projects/SaWS.html>
- Espinosa, L. A. (2020). *El riesgo del sargazo para la economía y turismo de Quintana Roo y México*. Bbvaresearch.com. Recuperado el 30 de mayo de 2024, de https://www.bbvaresearch.com/wp-content/uploads/2020/02/Riesgo_Sargazo_Big_Data.pdf
- Martínez-González, G. (2019). Sargassum: The atypical irruption of an ancient ecosystem. *Salud pública de México*, 61(5), 698-700. <https://doi.org/10.21149/10838>
- García-Sánchez, M., Graham, C., Vera, E., Escalante-Mancera, E., Álvarez-Filip, L., & van Tussenbroek, B. I. (2020). Temporal changes in the composition and biomass of beached pelagic Sargassum species in the Mexican Caribbean. *Aquatic Botany*, 167, 103275. <https://doi.org/10.1016/j.aquabot.2020.103275>

STUDY OF THE ROLE OF THE HISTIDINE-KINASE RPF2 IN ALGINATE BIOSYNTHESIS IN *AZOTOBACTER VINELANDII*

Mariana Ochoa López¹, Miguel Castañeda Lucio¹

¹Centro de investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla, CU, Edificio IC11, 24 Sur y Av. San Claudio. Colonia San Manuel, CP. 72570, Puebla.
mariana.ochoalopez.icuap@viep.com.mx

Azotobacter vinelandii is a Gram-negative, strictly aerobic and widely distributed free-living soil bacterium with many interesting features, including the ability to grow on a wide variety of carbohydrates, N₂ fixation, and alginate production. Alginate is an exo-polysaccharide composed of β-1,4-linked mannuronic acids and its epimer, guluronic acid, that has material properties appropriate for plenty of applications in industry as well as in medicine. Currently, this polymer is manufactured from brown algae, but given the intrinsic variation in their composition, bacterial culture has been proposed as a source for a tailor-made production.

Cyclic bis-(3', 5')-guanosine monophosphate, c-di-GMP, is a second messenger that regulates many cellular processes, including exo-polysaccharide production. The levels of c-di-GMP regulate at a post-translational level the biosynthesis of alginate in *A. vinelandii*, affecting the physical properties of the polymer and favoring the production of high-molecular-mass alginates.

Some two-component systems can contribute to modulating the c-di-GMP levels in response to environmental factors. A novel kinase, named RpfC2, was found in the *A. vinelandii* genome and down-stream of rpfC2 a gene that encodes a response regulator containing the conserved GGDEF domain that could contribute to the c-di-GMP pool. In this work, mutants were constructed by insertion and insertion-deletion, in which the role of this kinase in the synthesis of alginates was characterized.

DEVELOPMENT OF A BIOSENSOR FOR THE DETECTION OF *STREPTOCOCCUS PNEUMONIAE*

José de Jesús Olivares-Trejo^{1*}, María Elizabeth Álvarez-Sánchez¹,
Diego Jeovani Huerta-Flores¹, Jorge Alfredo Campoy-Ramírez¹

Posgrado en Ciencias Genómicas, Universidad Autónoma de la Ciudad de México (UACM), San Lorenzo 290,
Col. Del Valle, 03100, Mexico City, Mexico. Proyecto: UACM CCYT-CON-08.

*Corresponding author: jose.olivares@uacm.edu.mx, olivarestrejo@yahoo.com

Streptococcus pneumoniae is a bacterium considered a public health problem, it is the causative agent of infections such as otitis and sinusitis or more serious infections such as pneumonia or meningitis. This pathogen has a high rate of morbidity and mortality, attributed principally to a failure in diagnosis, making the time for diagnosis the most important factor to choose the best treatment. Therefore, it is imperative to develop new identification methods, which in a short time allow the identification of the pathogen to generate an adequate treatment scheme. A good alternative for this purpose is the methods based on biosensors, they can give an accurate diagnosis and fast in a very low cost. The objective of this research is to create a device that allows the identification of *S. pneumoniae* in a short time, therefore, a colorimetric biosensor based on an aptamer and gold nanoparticles (AuNP) was developed for the specific detection of this pathogen. The aptamer chosen was specific and it was used as a recognition element; upon binding with the AuNP, the complex interacts as a colorimetric indicator. The binding of the aptamer to the AuNP allowed them to be stabilized, maintaining the solution red in the absence of *S. pneumoniae*. When *S. pneumoniae* is present in the solution, the aptamer dissociates from the AuNP, resulting in a visual color change in the solution from red to blue. The color change was quantified by spectrophotometry, which allowed obtaining a relationship between the number of CFU and the absorbance of the solution. The results of this new method are compared with the results obtained from the CFU count. The quantification of CFU by the two methods gave us the same results, however, the biosensor has the great advantage of obtaining results in a time of 25 min, without having to wait the required 48 h for the traditional plate counting method.

Keywords. Biosensor, gold nanoparticles, aptamer, *S. pneumoniae*, bacterial detection.

BIOTECHNOLOGICAL POTENTIAL OF THERMOTOLERANT WASTEWATER YEASTS FOR HIGH-TEMPERATURE BIOFUEL PRODUCTION

Miguel Angel Orozco-González¹, Dana Vázquez-de los Reyes¹, Viviana Escobar-Sánchez,
Miguel Rosas-Paz¹, Francisco Padilla-Garfias², Jessica K. Suastes-Rivas³, María Teresa Orta Ledesma³,
Claudia Segal-Kischinevzky¹ and James González¹

¹Departamento de Biología Celular, Facultad de Ciencias. ²Departamento de Genética y Biología Molecular, Instituto de Fisiología Celular. ³Instituto de Ingeniería, UNAM, Ciudad Universitaria, CDMX, México.
Main institution: Facultad de Ciencias, UNAM. Address: Av. Universidad 3000, Circuito Exterior s/n, Alcaldía Coyoacán, C. P. 04510, Ciudad Universitaria, Ciudad de México.
Phone number:5525377704 E-mail: miguel.orozaez@gmail.com

The pressing need for eco-friendly fuel sources drives the search for microorganisms with valuable biotechnological properties. Yeasts isolated from wastewater represent a viable alternative due to their adaptation and survival in hostile environments. This research investigates these yeasts, which offer several advantages such as rapid growth, controllable growth conditions, and scalability, making them suitable for industrial production. This study examines yeast isolated from a wastewater treatment plant, co-cultured with microalgae, collected monthly from 2023. Using Sanger sequencing of the ITS region, we identified reddish yeasts of the genus *Rhodotorula*, as well as yeasts from the genera *Pichia*, *Candida*, *Naganishia*, *Cystobasidium*, and *Nakaseomyces*. Our analysis included cell viability at different temperatures, biomass production, ethanol production and lipid biosynthesis in a lipogenic medium. *Nakaseomyces glabratus* was found to be a thermotolerant yeast capable of surviving at 37-45 °C, producing 3.81 g/L of bioethanol, and accumulating 4.5 g/L of biomass at ambient temperature, highlighting its potential for ethanol synthesis. *Pichia sp.* also showed promising results, with biomass production of 3.49 g/L and bioethanol production of 3.61 g/L. The high biomass and bioethanol production by *N. glabratus* and *Pichia sp.* make them strong candidates for large-scale biofuel production through simultaneous saccharification and fermentation at high temperatures.

MODULATION OF THE VIRULENCE OF MULTIDRUG-RESISTANT *E. COLI* O104:H4 BY SUBINHIBITORY CONCENTRATIONS OF AMPICILLIN

Yaraymi Ortiz Reyes, Carolina Ortiz Murillo, Vianey Ramírez Lechuga, Eduardo Palomino Reyna, Santos García and Norma Heredia

Universidad Autónoma de Nuevo León, Facultad de Ciencias Biológicas, Laboratorio de Bioquímica y Genética de Microorganismos, Avenida Manuel Barragán S/N, Ciudad Universitaria, San Nicolás de los Garza CP 66455, Nuevo León, México.
E-mail: yortizr@uanl.edu.mx

The serotype *E. coli* O104:H4 has been recognized for carrying virulence factors such as Shiga toxin production and aggregative adhesion that increase the risk of developing haemolytic uremic syndrome. Also, is a multidrug-resistant bacteria that produces the TEM-1 β -lactamase responsible of ampicillin (AMP, subgroup penicillin) hydrolyses (Chowdhury et al. 2015). An interesting question is, whether the virulence of this bacteria is altered by exposure to subinhibitory concentrations of antibiotics to which bacteria are normally resistant. To clarify, was determined the effect of AMP concentrations (inducer of TEM-1 β -lactamase) on virulence factors of *E. coli* O104:H4. Sensitized inoculum was prepared by growing bacteria in Luria Bertani media with 0.1, 0.3, or 0.5 mg/ml of AMP (that not affected viability of bacteria) for 4 h at 37°C. This inoculum was used for assays of transformation capacity with exogenous DNA (plasmids and PCR-amplified DNA sequences), swarming motility, biofilm formation, and curli production. The expression of related virulence and resistance genes was analysed by real time PCR (control normalized to 1; >1 upregulated, <1 downregulated). Results showed that AMP at the concentrations used, increased the transformation ability, detecting the highest number of transformants (>10⁴ CFU/ng DNA; p \leq 0.05) after exposure to DNA sequences of resistance to spectinomycin, altering the expression of genes related to homolog recombination (*recA* and *lexA*). In addition, bacteria sensitized with 0.5 mg/ml of AMP and then exposed to 0.1 and 0.3 mg/ml of antibiotic during the swarming and biofilm assays, exhibited higher swarming mobility (up to 7.6 cm, vs 6.0 cm of control; p \leq 0.05) and biofilm production (up to 1.9-fold; p \leq 0.05 compared to control). Also, significant overexpression of the genes *flhC* (\leq 16.1-fold), *fliA* (\leq 22.1-fold), *csgA* (\leq 3.6-fold), *csgD* (\leq 9.1-fold), *stx2a* (\leq 32.2-fold) and *blaTEM-1* (\leq 5.5-fold, p \leq 0.05) was observed, showing positive correlation with *blaTEM-1* in most of cases. Qualitative curli formation increased when bacteria was exposed to 0.5 mg/ml during assay, however the expression of genes *csgA* and *csgD* was upregulated and downregulated with a non-defined trend. In conclusion, AMP-resistant *E. coli* O104:H4 alters its virulence factors when was exposed to some subinhibitory concentrations of this antibiotic. This information should be considered for therapeutic measures against this foodborne pathogen, and recommends studying the potential risks of antibiotic residues in food or environmental samples on the pathogen.

Reference

1. Chowdhury, PR., Charles, IG., & Djordjevic, SP. (2015). PLOS ONE 10(4):e0126197.

VISUALIZATION OF THE NEUROSPORA CRASSA DEVELOPMENTAL CYCLE IN MUTANT STRAINS LACKING HYDROPHOBINS

Omar Páez, Sammy Gutiérrez-Terrazas, Wilhelm Hansberg
Dpto de Biología Celular y del Desarrollo, IFC-UNAM

The cell wall is an essential structure for normal physiology and survival of fungi. The conidiation process involves remodeling of the cell wall and deposition of hydrophobins. Our hypothesis is that these hydrophobic proteins together with other cell wall components are mechanisms for cell insulation from environmental dioxygen. Here, we study several *N. crassa* strains lacking the hydrophobins. Absence of hydrophobins results in wall instability and precocious germination of spores. Visualizing aerial structures in filamentous fungi has been elusive given the high fragility of these hyphae and easy dispersal of conidia. Hence, obtaining confocal images of these living structures is very unlikely to succeed. We take advantage of an innovative technique for visualizing wall status in aerial hyphae and conidiophores by confocal microscopy. The imaging of walls in these hydrophobin deficient strains presented here serves as a demonstration that aerial structures can, indeed, be subject to evaluations in cell wall changes by fluorescent molecules opening exciting avenues for the studies of aerial sporogenic development. We thank PAPIIT grant No. IN226723 and CONACYT Fronteras de la Ciencia grant No. CBF2023-2024-1748

MITOCHONDRIA PROTEOME OF MOSQUITO CELLS INFECTED WITH DENGUE VIRUS

Santos Ramírez Carreto¹, Lourdes Gutierrez Xicotencatl¹, Lidia Huerta-Nuñez²,
Exsal Albores-Mendez², Cesar V. F. Batista², Victoria Pando-Robles¹

¹Centro de Investigación Sobre Enfermedades Infecciosas, Instituto Nacional de Salud Pública. Cuernavaca, Morelos 62100. México. ²Laboratory of Pharmacology, Escuela Militar de Graduados de Sanidad, Universidad del Ejército y Fuerza Aérea Mexicana, Mexico City 11200, Mexico.
E-mail: victoria.pando@insp.mx

Aedes aegypti is a principal mosquito vector that transmits the dengue virus (DENV) in tropical and subtropical regions worldwide, putting an estimated 3 billion people at risk of dengue disease. DENV-infected individuals exhibit symptoms ranging from subclinical or mild febrile diseases to severe haemorrhagic fever. Infected mosquitoes, however, do not show detectable signs of disease, despite the virus maintaining a lifelong persistent infection.

Mitochondria are dynamic organelles that control the cell's energy metabolism, immune response, and lifespan. These functions are primarily performed and regulated by proteins localized to the mitochondria. Although the mitochondrial genome only encoding 13 proteins, it is currently estimated that over 1000 proteins reside within mitochondria or are transiently associated with them. These mitochondrial proteins represent a functional subcellular protein network encoded by mitochondrial and nuclear genomes and significantly vary during viral infection.

In vertebrates, viruses frequently manipulate mitochondrial processes by remodelling mitochondrial ultrastructure and inter-organellar communication to enhance viral replication and pathogenicity. However, the role of mitochondria in arthropod vectors remains poorly understood. Recently, our group published findings indicating that DENV infection in mosquito cells alters redox metabolism and mitochondrial membrane potential without significantly affecting the cellular ATP pool or viability. To understand how the dengue virus modulates mitochondrial function in the vector, we present the first comprehensive analysis of the mitochondrial proteome during DENV infection in mosquito cells.

INTERACTION OF THE MIBR PROTEIN WITH THE REGULATORY REGION OF THE *IPDC* GENE OF *AZOSPIRILLUM BRASILENSE* SP7

José Cristian Pérez Gómez¹, María Luisa Xiqui Vázquez¹, Alberto Ramírez-Mata¹,
Beatriz Eugenia Baca¹, Sandra Reyes Carmona¹

Laboratorio de la Interacción Bacteria-Planta, Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla, Puebla 72570, México.

Correspondence: sandra.reyes@correo.buap.mx

Azospirillum brasilense is a plant growth-promoting rhizobacterium. The promotion of plant growth and development occurs by several mechanisms, one of them being the production of phytohormones such as auxins¹. Indole-3-acetic acid (IAA) is the most important and predominant auxin in plants; however, its effects go beyond this, as it is involved in the regulation of bacterial physiology, adaptation to stressful conditions and plays a key role as a signaling molecule. IAA biosynthesis in *Azospirillum brasilense* is mainly carried out through IPyA pathway, with the indole pyruvate decarboxylase (PPDC) enzyme encoded by the *ipdC* gene. Mutant strain generated in this gene, reduced drastically IAA production². Therefore, to study the transcriptional regulation of the *ipdC* gene, a protein that binds to regulatory region was identified and classified as a member of the MarR family. This protein was named MibR (MarR-like indole-3-acetic acid biosynthesis regulator). In analyses with mutants of this protein, a decrease in the expression of the *ipdC* gene, as well as in the biosynthesis of IAA was demonstrated³. The members of the MarR family act mostly as negative regulators, requiring binding to a ligand the interface between the dimerization and DNA binding domains, causing a conformational change that reduces the DNA binding affinity⁴. In this work, expression and purification of MibR protein in *E. coli* BL21 strains was carried out using the expression vector pGEX-4T1. Electrophoretic mobility shift assays (EMSA) of the MibR protein interacting with the regulatory region of the *ipdC* gene were performed to test whether it binds specifically to it and explore its functions as a negative transcriptional regulator in the expression of *ipdC* gene.

References

1. Duca, D., & Glick, B. (2020). Indole-3-acetic acid biosynthesis and its regulation in plant-associated bacteria. *Applied microbiology and biotechnology*, 104(20), 8607–8619.
2. Tang, J., Li, Y., Zhang, L., Mu, J., Jiang, Y., Fu, H., & Zhang, Y. (2023). Biosynthetic Pathways and Functions of Indole-3-Acetic Acid in Microorganisms. *Microorganisms*, 11(8).
3. Jijón-Moreno, S. (2017). Estudio de la regulación transcripcional del gen *ipdC* que participa en la biosíntesis de ácido indol-3- acético en cepas de *Azospirillum brasilense*. Tesis doctoral. BUAP.
4. Baksh, K. A., & Zamble, D. B. (2020). Allosteric control of metal-responsive transcriptional regulators in bacteria. *The Journal of biological chemistry*, 295(6), 1673–1684.

CHARACTERIZATION OF THE ANTI-QUORUM SENSING AND ANTI-BIOFILM ACTIVITY OF DIVERSE ACTINOMYCETES ISOLATED FROM SOILS OF THE MEXICAN JUNGLE AGAINST PATHOGENIC BACTERIA RESISTANT TO ANTIBIOTICS

Itzel Plascencia-Ortiz¹, Michelle Chávez-Hernández¹, Araceli Martínez-Larios¹, Graciela Castro-Escarpulli¹, Lourdes Villa-Tanaca¹, César Hernández-Rodríguez¹

¹Departamento de Microbiología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Prol. de Carpio y Plan de Ayala s/n. Col. Sto. Tomás, 11340 Ciudad de México, México.

Correspondence author: César Hernández-Rodríguez.

Email: chdez38@hotmail.com

Multidrug-resistant pathogenic bacteria have become one of the most important public health problems, especially in the hospital setting. Failure of antibiotic therapy causes the natural development of the disease, increases the severity of the case and the length of hospital stay, and compromises the lives of patients. In 2017, the WHO launched a global alert to draw attention to the problem of spreading the phenomenon of multi-antibiotic resistance and called on scientists to restart the search for new antimicrobial molecules. Actinomycetes are bacteria with a very broad secondary metabolism that maintains a potential interest in the discovery of new anti-virulence activities. This work aims to search for metabolites that inhibit quorum sensing, a cell-to-cell communication process that regulates various biological phenomena such as biofilm formation, mobility, and production of bacterial pigments, among others, in lyophilized supernatants produced from a collection of actinomycetes isolated from Mexican jungle soils. Several lyophilized supernatants exhibited inhibition and destruction activity of biofilms formed by multidrug-resistant strains of the species *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Acinetobacter baumannii*, and *Staphylococcus aureus*. Lyophilized supernatants capable of inhibiting various biological processes regulated by quorum sensing such as the production of pigments, motility and extracellular enzymes were also recognized in strains of *Serratia marcescens*, *P. aeruginosa* and *Chromobacterium violaceum*. All actinomycetes with biological activity were identified as species of the genus *Streptomyces* and some of these could be new species of this genus.

EVALUATION OF THE IMMUNOGENIC PROPERTIES OF SURFACE PROTEINS FROM *GALLIBACTERIUM ANATIS* 12656-12

Ana Karen Ponce Quiroz¹, María Elena Cobos Justo¹, Ma. Patricia Sanchez Alonso¹, Miguel Angel Ávalos Rangel¹, Candelario Vázquez Cruz¹, Erasmo Negrete Abascal²

¹Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias Benemérita Universidad Autónoma de Puebla, CU, Edificio IC11, 24 Sur y Av. San Claudio. Colonia San Manuel, CP. 72570, Puebla Pue, ²Carrera de Biología, Facultad de Estudios Superiores de Iztacala, UNAM. Av. de los Barrios no. 1. Los Reyes Iztacala, 54090 Edo. de México.
E-mail: pq223470252@alm.buap.mx y ecobacilos@yahoo.com

Gallibacterium anatis is a bacterium belonging to the *Pasteurellaceae* family, and two biovars have been reported: a hemolytic and a non-hemolytic biovar. It is a Gram-negative, non-motile, capsulated coccobacillus, which on blood agar forms semitransparent grayish, circular colonies 0.5 to 1 mm in diameter. It is catalase and oxidase negative but positive for phosphatase, porphyrin, and alanine aminopeptidase [1]. It is common to find *G. anatis* in the respiratory tract of apparently healthy chickens, indicating that it may be part of the bird's autochthonous microbiota; however, epidemiological studies have shown that this bacterium is an opportunistic pathogen since it mainly affects the host when it is immunosuppressed or under stress conditions [2]. One of the most common pathologies it causes is salpingitis with or without peritonitis, causing lesions in the reproductive tract of hens and consequently a significant decrease in egg production. This pathogen has different virulence factors that allow colonization, invasion, and evasion of the host immune response. Therefore, the secretion of proteases that degrade IgG, the ability to agglutinate red blood cells, the production of a cytosolic toxin Gtx, and the presence of adhesins are known. Nowadays, there are vaccines against *G. anatis*. However, these have not been enough to control the gallibacteriosis disease. We consider it important to innovate vaccines against this disease, which can induce an efficient immune response in birds. To fulfill this purpose, we considered it necessary to know the surface antigens of *G. anatis*, so we made a bioinformatic exploration in 90 proteomes of this microorganism. We found that there are 28 outer membrane proteins, of which 26 have unique functions and 2 have duplicate functions. Of these, the ones with the highest immunogenic potential are hemagglutinins, Gtx toxin, and OmpA porin. Some of the coding genes have already been cloned, which is expected to corroborate their expression and the immunogenicity of the recombinant proteins. In conclusion, knowing and testing the antigenic potential of these outer membrane proteins in an animal model is essential to propose new useful and efficient components as vaccine agents.

References

1. Christensen, H., et al. (2003). International Journal of Systematic and Evolutionary Microbiology. 53: 275-287.
2. Chantes-Guerra A, et al. (2022). Front Microbiol. Aug 16;13:951173.

THE UNEXPLORED DIVERSITY OF YEASTS FROM OPEN FERMENTATIONS IN MEXICO

Nancy Judit Quintana Rodríguez^{1,2,3}, Eugenio Mancera² and Alexander De Luna¹

¹Unidad de Genómica Avanzada, ²Departamento de Ingeniería Genética, Unidad Irapuato and ³Posgrado en Biología Integrativa, Centro de Investigación y de Estudios Avanzados, 36824 Irapuato, Mexico.
nancy.quintana@cinvestav.mx; eugenio.mancera@cinvestav.mx; alexander.deluna@cinvestav.mx

Artisanal production of traditional beverages in Mexico usually involves open fermentation processes, where environmental bacteria and yeasts transform sugars and other compounds in substrates such as agave, cacao, corn, and prickly pears. In agave fermentation, microbial diversity may vary with biogeographical factors, underscoring the importance of understanding these microbial communities to preserve the production processes of these beverages of significant cultural and industrial value. Despite a growing interest of agave spirits in global markets, there is still limited knowledge about the microbial consortia involved in their production. In a recent collaborative effort, we reported the YMX-1.0 culture collection, consisting of 4524 yeast strains isolated from traditional distilleries across Mexico, encompassing diverse climatic, geographical, biological, and cultural contexts ¹. While this report included species usually associated with agave fermentation, such as *Saccharomyces cerevisiae*, *Pichia kudriavzevii*, and *Kluyveromyces marxianus*, it also revealed at least two possible novel species of the *Pichia* clade, based on sequences of their ITS and D1/D2 regions. In this project, using whole-genome sequencing and phenotyping profiling, we will formally characterize these two possible new-to-science yeast species. In addition, we will conduct a thorough analysis of the YMX-1.0 culture collection, which may harbor additional new fungal species associated with this relatively understudied fermentative environment.

References

1. Gallegos-Casillas, et al. (2024). Yeast diversity in open agave fermentations across Mexico. *Yeast*, 41, 35–51.

GENOME ANALYSIS OF *GLUCONACETOBACTER* SP. UAPS01-405, ISOLATED FROM THE NON-PHOTOSYNTHETIC PARASITIC PLANT *CONOPHOLIS ALPINA*

Rigel Quintero Luis¹, Miguel Castañeda Lucio¹, Yagul Pedraza Pérez¹, José Antonio Munive Hernández¹,
María del Rocío Bustillos Cristales¹, Gilberto Juárez Munguía¹,
Liliana López Pliego¹ and Luis Ernesto Fuentes Ramírez¹

¹ Centro de Investigación en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla, Ciudad Universitaria, Avenida San Claudio S/N, Puebla Pue., México C.P. 72570.
E-mail: liliana.lopez@correo.buap.mx; luis.fuentes@correo.buap.mx

The Acetic Acid Bacteria (AAB) are members of the *Acetobacteraceae* family. Since their ability to metabolize ethanol, glucose and acetate, they play a key role on the production of vinegar, kombucha and other products of commercial relevance. Some species have been isolated from plants, flowers, herbs, fruits and roots, participating on plant-microbe interactions, promoting vegetal growth, nitrogen fixation, phosphate solubilization and/or release of antimicrobial substances¹.

Conopholis alpina is a parasitic plant, that colonizes the roots of the oak trees, covered from the light by the trees canopy, and feeding from its host². To our knowledge, there are no previous work related to this plant microbiology. This work aims to study one *Acetobacterium* isolate obtained from the flowers of this plant.

The isolate UAPS01-405 grows on LGI medium and shows characteristic phenotypic traits of the genus *Gluconacetobacter*. The assembly, annotation and analysis of its genome showed that this microbe is closely related to other members of this genus such as *G. liquefaciens* and *G. dulcium*, nonetheless its species identification remains to be determined.

Acknowledgments: This work was partially funded by Vicerrectoría de Investigación y estudios de Posgrado, BUAP.

References

1. Rodrigo José Gomes, Maria de Fatima Borges, Morsyleide de Freitas Rosa, Raúl Jorge Hernan
2. Castro-Gómez and Wilma Aparecida Spinosa (2018). Food Technology and biotechnology, 56, 139-page.
3. Anuar Rodrigues, Shana Shaya, Timothy A. Dickinson, and Saša Stefanović (2013). American Society of Plant Taxonomists, 38, 795-804.

ALDEHYDE DEHYDROGENASE DIVERSITY AND 6-OL-ALDH OVEREXPRESSION IN *AZOSPIRILLUM* GENOMES

Alberto Ramírez-Mata¹, Ricardo Cuatlayotl Olarte¹, Claudia Mancilla Simbro¹, María Luisa Xiqui Vázquez¹, Sandra Reyes Carmona¹ and Beatriz Eugenia Baca¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Benemérita Universidad Autónoma de Puebla,
Avenida San Claudio S/N, Puebla Pue, México. C.P. 72570
E-mail: alberto.ramirez@correo.buap.mx Tel: 222 867 0044

This study presents the first comprehensive analysis of the aldehyde dehydrogenase (ALDH) superfamily within fully sequenced *Azospirillum* genomes. We identified a remarkable diversity of ALDH, encompassing 17 distinct families further classified into 31 subfamilies. This classification scheme provides a valuable tool to understand the extent of ALDH variation and potential redundancy across bacterial genomes. It paves the way for future investigations into the unique properties and functions of each ALDH family. Interestingly, the study proposes the ALDH19 family as a powerful phylogenetic marker for *Azospirillum* species. This suitability stems from its high degree of sequence conservation and lack of redundancy across various *Azospirillum* strains. The observed diversity of ALDH among different *Azospirillum* strains may contribute to their adaptability in diverse environmental conditions. Future research elucidation the specific functions of these enzymes could hold of *Azospirillum*, scientists may develop strategies to enhance crop productivity. To begin the analysis in removing toxic aldehydes we overexpressed a 6-OL-ALDH to evaluate its ability to degrade hydrocarbons.

References

1. Kuykendall, J.R.; Kuykendall, N.S. 15.19—Aldehydes. 2018. In *Comprehensive Toxicology*, 3rd ed.; McQueen, C.A., Ed.; Elsevier: Oxford, UK, 2018; pp. 352–388.
2. Deza-Ponzio, R.; Herrera, M.L.; Bellini, M.J.; Virgolini, M.B.; Hereñú, C.B. Aldehyde Dehydrogenase 2 in the Spotlight: The Link between Mitochondria and Neurodegeneration. *Neurotoxicology* 2018, 68, 19–24.
3. Muzio, G.; Maggiora, M.; Paiuzzi, E.; Oraldi, M.; Canuto, R.A. Aldehyde Dehydrogenases and Cell Proliferation. *Free. Radic. Biol. Med.* 2012, 52, 735–746.
4. Shortall, K.; Djeghader, A.; Magner, E.; Soulimane, T. Insights into Aldehyde Dehydrogenase Enzymes: A Structural Perspective. *Front. Mol. Biosci.* 2021, 8, 659550.

STUDY OF THE ENDOPHYTIC CAPACITY OF ENTOMOPATHOGENIC FUNGI

Jesús Ernesto Ramírez Nieto¹, Areli Duron Castellanos¹, Angélica González Hernández¹, Juan Carlos Torres Guzmán¹ and Israel Enrique Padilla Guerrero^{1*}

¹Departamento de Biología, División de Ciencias Naturales y Exactas. Universidad de Guanajuato, Noria Alta S/N; CP. 36050; Guanajuato, Gto.

*E-mail: ie.padillaguerrero@ugto.mx

Entomopathogenic fungi are organisms that infect and kill insects, can also establish symbioses with plants through the colonization of their roots¹. However, in the colonization process, it is not completely clear whether entomopathogenic fungi can be endophytes.

This work focused on understanding the endophytic capacity of “sleeper”² or “creeper”² strains of the fungi *Metarhizium robertsii* and *Beauveria bassiana*. Sleeper strains have a phenotype in which more conidia are produced under dark conditions², while creeper strains develop as mycelia in darkness².

To determine the endophytic capacity, strains of *M. robertsii* and *B. bassiana* expressing fluorescent proteins were used, and the study strains were transformed by *Agrobacterium* with the T-DNAs of the plasmids pPK2-BAR-mCherry and pPK2-BAR-GFP. Subsequently, the fluorescent strains were inoculated into the roots of sterile rice (*Oryza sativa*) and bean (*Phaseolus vulgaris*) plants. The seedlings were sown, and the interaction was monitored at 10, 20 and 60 days through fluorescence microscopy. The levels of colonization and endophytism were analyzed through histological sections of the roots, as well as the determination of colony-forming units.

References

1. Behie, S. W.; Jones, S. J.; Bidochka, M. J. (2015). Plant tissue localization of the endophytic insect pathogenic fungi *Metarhizium* and *Beauveria*. *Fungal Ecology*, 13(), 112-119.
2. Angelone, S., Piña-Torres, I. H., Padilla-Guerrero, I. E., & Bidochka, M. J. (2018). “Sleepers” and “Creeper”: A Theoretical Study of Colony Polymorphisms in the Fungus *Metarhizium* Related to Insect Pathogenicity and Plant Rhizosphere Colonization. *Insects*, 9(3), 104.

PHENOTYPIC CHARACTERIZATION OF THE *LIBR* GENE OF *AZOSPIRILLUM BRASILENSE* SP7

Sandra Reyes Carmona¹, Antonio Pérez Carillo¹, María Luisa Xiqui Vázquez¹,
Alberto Ramírez-Mata¹ and Beatriz E. Baca¹

¹Laboratorio de la Interacción Bacteria-Planta, Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla. Av. San Claudio S/N Edif. IC11, Ciudad Universitaria. Puebla, Pue. México.
E-mail: sandra.reyes@correo.buap.mx

Azospirillum brasilense Sp7 is a plant growth-promoting rhizobacteria that produces the phytohormone indole-3-acetic acid (IAA), a signaling molecule involved in bacteria-plant interaction¹. IAA biosynthesis in *Azospirillum* occur mainly through the indole-3-pyruvic acid (IPyA) pathway, in which the phenylpyruvate decarboxylase (PPDC) encoded by the indole pyruvate decarboxylase gene (*ipdC*), is responsible of the mayor biosynthesis of the phytohormone². Is well known that several environmental factors control the biosynthesis of IAA at the physiological level³, however there are few studies about the transcriptional regulation of the *ipdC* gene. Previously, we identified a putative transcriptional regulator with REC and HTH domains named LibR (LuxR-family indole-3-acetic acid biosynthesis regulator). The mutant in this protein decreases the IAA biosynthesis, while the complemented strain restores this effect. In this work, we presented the evaluation in several phenotypes like growth curve, biofilm formation and colony morphology in *A. brasilense* Sp7 (wt), *libR* mutant and *libR* complemented strains, to explore the effect on this protein in the *Azospirillum* biology.

Acknowledgments. We thank to Vicerrectoria de investigación y estudios de posgrado (VIEP) for financial support.

References

1. Duca DR, Glick BR. (2020). *Applied Microbiology and Biotechnology*. 104:8607–8619
2. Jijón-Moreno S, Marcos-Jiménez C, Pedraza RO, Ramírez-Mata A, de Salamone IG, Fernández-Scavino, Claudia A. Vázquez-Hernández, Lucía Soto-Urzáa, Beatriz E. Baca. (2015). *Antonie van Leeuwenhoek, International Journal of General and Molecular Microbiology*. 107(6):1501–17.
3. Carreño-Lopez R, Campos-Reales N, Elmerich C, Baca BE. (2000). *Molecular and General Genetics*. 264(4):521–30.

BIOCHEMICAL AND MICROBIOLOGICAL CHARACTERIZATION OF THE INTESTINAL MICROBIOME OF THE *TENEBRIO MOLITOR* LARVA

Axel Reyes Coatl¹, María Rosete Enríquez¹

Facultad de Ciencias Biológicas, Benemérita Universidad Autónoma de Puebla. Blvd. Valsequillo y Av. San Claudio Edificio 112-A, Ciudad Universitaria, Col. Jardines de San Manuel, Puebla, Pue. México, C.P. 72570.
E-mail: axel.reyesco@alumno.buap.mx Tel: 22-21-27-23-35

The study of the intestinal microbiome of the larva of the beetle *Tenebrio molitor* (mealworm) has been of great relevance due to its potential applications in plastic degradation and environmental bioremediation¹. The objective of the present study is the biochemical characterization of culturable bacteria isolated from the intestine of *Tenebrio molitor* larvae. For this purpose, six larvae were sacrificed by cooling at 4°C for 10 minutes, subsequently, all body surfaces were disinfected with absolute ethanol and a saline solution rinse. The intestines of each larva were obtained by microdissection under sterile conditions. Each intestine was shaken in 50% peptone water to make a set of serial dilutions to culture an aliquot in different culture media (LB, MacConkey, EMB, Sal and Mannitol), which were incubated for 24 hours at 30°C. As a control, hemolymph samples cultured in LB medium were used. From each type of culture medium, representative bacterial colonies were selected due to their abundance to isolate them in their corresponding medium to carry out the characterization of the bacterial and colonial morphology considering size, shape, color, etc.² Finally, five bacterial colonies were selected to carry out the biochemical study by analyzing miniaturized galleries of API20E tests². In general, the highest colonial diversity was observed in the EMB medium and the lowest in the MacConkey medium. 22 types of representative culturable bacterial colonies were identified in the four culture media. In addition, of the set of representative colonies, 18 are Gram (-) cocci and 4 are Gram (-) bacilli. The colonial morphologies fundamentally presented circular shapes with entire edges, flat elevation, with soft consistency, smooth texture, medium size, and white, yellow, and pink colors. Biochemical tests mainly revealed positive reactions to the presence of enzymes ONPG (β -galactosidase), TDA (tryptophan deaminase), URE (urease); production of CIT (citrate), VP (acetoin) and NO₂ (nitrogen dioxide); degradation of GLU (glucose), ARA (arabinose) and INO (inositol). These results indicate the presence of an intestinal microbial diversity with differential metabolic activities that could help in the digestive activities that favor the growth and survival of the *Tenebrio molitor* larva³. Likewise, it opens the possibility to study the probable biotechnological applications of culturable bacteria from insect intestinal microbiomes.

References

1. Rodríguez-Carreón, Anlui, Ortiz-Rivera, Yuridia, Hernández-Peña, Claudia Carolina, & Figueroa, Coyolxauhqui. (2021). Biodegradación de espumas plásticas por larvas de insectos: ¿una estrategia sustentable? TIP. Revista especializada en ciencias químico-biológicas, 24, e311. Epub 15 de noviembre de 2021. <https://doi.org/10.22201/fesz.23958723e.2021.311>
2. Galvis Salazar, J. D., Quiñones, R., & Jiménez, P. (2016). Aislamiento de Microorganismos del Tracto Digestivo de Larvas de Coleópteros y Lepidópteros Detritívoros y Evaluación, In Vitro, de su Efecto Antagónico en una Cepa de *Fusarium oxysporum*. Revista Facultad De Ciencias Básicas, 5(1-2), 106–113. <https://doi.org/10.18359/rfcb.2124>
3. Machona, O., Chidzondo, F., & Mangoyi, R. (2022). *Tenebrio molitor*: possible source of polystyrene-degrading bacteria. BMC biotechnology, 22(1), 2. <https://doi.org/10.1186/s12896-021-00733-3>

PHYSIOLOGICAL AND SYMBIOTIC DIFFERENCES OF PYRUVATE CARBOXYLASE AND PHOSPHOENOLPYRUVATE CARBOXYLASE IN *RHIZOBIUM PHASEOLI*

Alma Ruth Reyes González, Noé Arroyo Mozo, Carmen Vargas Lagunas,
Lourdes Girard Cuesy, Humberto Peralta, Jaime Mora†

Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México. Av. Universidad s/n Col. Chamilpa 62210
Cuernavaca, Morelos. argonzal@ccg.unam.mx

Pyruvate carboxylase (PYC) and phosphoenolpyruvate carboxylase (PEPC) enzymes produce oxaloacetate, an essential metabolite of the tricarboxylic acids cycle (Koendjibharie *et al.*, 2021). In *Rhizobium phaseoli* CIAT652, both genes are present; a *pyc*- mutant had optimal growth in succinate medium but it did not grow in medium with pyruvate, glucose or aspartate as a carbon source. Thus, PYC enzyme is necessary to supply oxaloacetate using different carbon sources. PYC activity is commonly present; however, PEPC activity has not been found in many organisms in free life. *Corynebacterium glutamicum* has both enzymes and has a great plasticity in its metabolism as well as *R. phaseoli* CIAT652. Curiously, the carbon flux partition in gluconate/pyruvate was 10 per cent PEPC/ 90 per cent PYC, however lysine production increased in PEPC overexpression but not in PYC overexpression. That means that PEPC is playing an important role in the replenishment of the TCA intermediaries. In *R. phaseoli* CIAT652, PEPC apparently has not a function in free life, when grown on different carbon sources. A significant difference was that *pyc* and *pyc-pepc* mutants presented no growth with acetate in *R. phaseoli*, while *C. glutamicum pyc* mutant grew well on such source (Peters-Wendisch *et al.*, 1998). This suggested that *R. phaseoli* CIAT652 uses the glycolytic pathway, while *C. glutamicum* uses the glyoxylate pathway. To confirm which pathway is used, we will measure the expression of genes in different carbon sources by qRT-PCR. Apparently, PEPC has a special function during symbiosis with common bean, because PEPC activity in nodule was high. These data point out that both carboxylases in CIAT652 are necessary for free life and symbiotic nitrogen fixation.

†In memory of Dr. Jaime Mora Celis. We thank for his contribution to the development of scientific research in the area and for his support received by each of us.

References

1. Koendjibharie Jeroen G., van Kranenburg Richard., and Kengen Servé W. M. (2021). FEMS Microbiology Reviews, 45:1-19.
2. Peters-Wendisch Petra G., Kreutzer Caroline., Kalinowski Jörn., Pátek Miroslav., Sahm Hermann., and Eikmanns Bernhard J. (1998). Microbiology, 144: 915-927.

THE BARA/SIRA AND CSR SYSTEMS REGULATES THE EXPRESSION OF A PUTATIVE CARBONIC ANHYDRASE ENZYME ENCODED IN THE PSLT VIRULENCE PLASMID OF *SALMONELLA ENTERICA* SEROVAR TYPHIMURIUM

Yoselin Xiomara Rodríguez Muñoz; Víctor Humberto Bustamante Santillán

Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Uiniversidad 2001, Colonia Chamilpa, Cuernavaca, Morelos 62210, México. Teléfono: 7773291627
E-mail: xiomara.rodriguez@ibt.unam.mx y victor.bustamante@ibt.unam.mx

The BarA/SirA and Csr systems are found in many bacteria where they control the expression of genes encoding a wide diversity of cellular functions, like carbon metabolism, motility, biofilm production, quorum sensing, stress response, and virulence. BarA and SirA form a two-component system; BarA is the sensor kinase that autophosphorylates in response of short-chain fatty acids, such as formate and acetate, and subsequently transfers the phosphate group to the response regulator SirA. Phosphorylated SirA activates the expression of the *csrB* and *csrC* genes that code for the small RNAs CsrB and CsrC, respectively. BarA/SirA and CsrB/CsrC controls gene expression through CsrA, a protein that binds near to the Shine-Dalgarno motif and thus mainly blocks translational initiation of target mRNAs; however, CsrA can also favor translation and can regulate positively or negatively RNA stabilization or transcriptional termination. When present, CsrB and CsrC bind to CsrA and thus counteracts its action. The BarA/SirA and Csr systems control the expression of multiple genes in *Salmonella*, including virulence genes. For instance, BarA/SirA and CsrB/C induce expression of the SPI-1 genes, required for the invasion of *Salmonella* to host cells, by counteracting CsrA-mediated translational repression on the *hilD* gene. Furthermore, BarA/SirA and CsrB/C decrease expression of the *pdu* and *eut* genes, required for the replication of *Salmonella* in the intestinal lumen, by counteracting CsrA-mediated translational induction on these genes. A recent proteomic analysis from our group revealed new target genes for SirA and CsrB/CsrC. One of these genes is *SLP1_0061*, which is present only in *Salmonella*, located in the pSLT virulence plasmid, and codes for a putative carbonic anhydrase enzyme. Carbonic anhydrase enzymes catalyze a fundamental chemical reaction, the interconversion between CO₂ and bicarbonate, and are involved in different biological process such as metabolic biosynthetic pathways, pH regulation, virulence, and survival in different niches. In this study, we confirm that SirA and CsrB/C negatively control translation of the *SLP1_0061* gene. Currently, we are investigating whether SirA and CsrB/CsrC regulate *SLP1_0061* through CsrA, as well as the probable biological function of *SLP1_0061*. To our knowledge, *SLP1_0061* is the first gene located in plasmid shown to be regulated by SirA and CsrB/CsrC.

THE POTENTIAL OF *PERICONIA MACROSPINOSA* HAGJ2 ISOLATED FROM *AGAVE TEQUILANA* TO CONTROL PLANT PATHOGENS

Fernando Uriel Rojas Rojas¹, Ingrid Melissa Gómez-Vázquez¹,
Harumi Shimada-Beltrán¹, Julio C. Vega-Arreguín¹

¹Laboratorio de Ciencias Agrogenómicas, Escuela Nacional de Estudios Superiores Unidad León, Universidad Nacional Autónoma de México (UNAM). Boulevard UNAM 2022, Col. Predio El Saucillo y El Potrero, León, Guanajuato, Mexico.
frojas@enes.unam.mx, jvega@enes.unam.mx

Periconia macrospinoso is a fungi belonging to the dark septate endophytes (DSE). This species has limited information regarding the colonization of roots of nonmycorrhizal plants and its anti-phytopathogenic characteristics. The strain HAGJ2 was isolated from the root tissue of *Agave tequilana*, a crop located in Atotonilco El Alto, Jalisco. The strain was identified by microscopical observations and the sequencing and analysis of the ITS region. To date, there is no record of the presence of this genus or species in the *A. tequila* microbiome or in the geographical location. The aim of this study was to identify the potential of *P. macrospinoso* HAGJ7 as a biocontrol agent, considering its possible impact in reducing the use of chemical fertilizers and pesticides, which alter soil composition and contaminate water.

We determined the antifungal and antioomycete activity of *P. macrospinoso* HAGJ2 in different culture media. The antimicrobial properties depended on the culture media and the inoculation method of HAGJ2 and the pathogens. *P. macrospinoso* HAGJ2 was capable of completely inhibiting or stressing fungi and oomycetes such as *Phytophthora capsici*, *Sclerotium rolfsii*, *Fusarium proliferatum*, *Fusarium oxysporum*, *Alternaria alternata*, *Botrytis cinerea*, *Rhizoctonia solani*, and *Colletotrichum gloeosporioides* on PDA plates; these microorganisms are economically important phytopathogens. We have also identified that PDA is the best culture media to produce antimicrobials and the specific time of growing of *P. macrospinoso* HAGJ2 to produce enough antimicrobial metabolites on PDA plates to show antagonistic activity. We are currently analyzing the genome sequence of HAGJ2 to identify putative genes related with the antimicrobial activity of this fungi.

THE ROLE OF EPINEPHRINE AND NOREPINEPHRINE IN PROMOTING THE GROWTH AND VIRULENCE FACTOR EXPRESSION OF *MANNHEIMIA HAEMOLYTICA* IS SIGNIFICANT

Verónica Rosales Islas¹, Tomas Villamar Duque¹, Candelario Vázquez Cruz², Erasmo Negrete Abascal¹

¹Facultad de Estudios Superiores Iztacala, UNAM. Av. De Los Barrios 1, Los Reyes Iztacala, 54090 Tlalnepantla, Edo de México, México. ²Centro de Investigaciones en Ciencias Microbiológicas, ICUAP, BUAP, Puebla, México.
E-mail: verito0622@gmail.com

Mannheimia haemolytica is an opportunistic pathogen bacterium in healthy bovines' amygdaline crypts. However, in stress conditions, this bacterium reproduces rapidly and descends into the lungs, causing acute fibrinous pneumonia, also known as shipping fever. This work aimed to know the effect of Epinephrine (Epi) or Norepinephrine (Nepi), stress hormones, on *M. haemolytica* growth and expression of different virulence factors. The growth of *M. haemolytica* was improved when cultured in the presence of physiological concentrations of Epi or NEpi (1 to 10 ng/ml). A higher effect was observed with 3 ng/ml of both hormones. Total, outer membrane (OMP) and secreted protein patterns of *M. haemolytica* grown in the presence of catecholamines presented differences with respect to control cultures without hormones. There was observed an increase and diminish, respectively, of 40, 50, 175 kDa, and 35 kDa bands in the total cell extracts pattern, of 50 and 55 kDa bands in OMPs and 64 and 100 bands in secreted proteins when cultures were in the presence of Epi. Similarly, in the presence of Nepi, increase and diminish of bands of 60 and 175 and in a band of 180 kDa in total cell extracts; 80 kDa band, respectively, in OMPs, and increase of 55 and 127 secreted protein bands. Proteolytic activity of a 60 kDa band was increased in the presence of Epi but not by NEpi. Catecholamines induce changes in protein patterns and favor the growth of *M. haemolytica*.

Project supported by DGAPA-UNAM PAPIIT IN204122, VRI is a CONAHCYT (1326191) scholarship and a Posgrado en Ciencias Biológicas- UNAM student.

STUDY OF SPLICING AND NONSENSE MECHANISM DECAY (NMD) FACTORS IN *USTILAGO MAYDIS*

Kate Ariadna Rossano Gutiérrez¹, Nancy Martínez Montiel¹, Rebeca D. Martínez Contreras¹

¹Benemérita Universidad Autónoma de Puebla, Av San Claudio s/n, Cd. Universitaria, La Hacienda, 72592.
Heroica Puebla de Zaragoza, Pue, México.
E-mail: kate.rossanog@alumno.buap.mx

Nonsense Mechanism Decay is a quality control mechanism for mRNAs, which recognizes premature termination codons (PTC's) and leads to their degradation, thus preventing the translation of aberrant transcripts and controlling gene expression. There are central factors in "trans" that are responsible for this, such as the proteins UPF1, UPF2 and UPF3. Other types of factors associated with NMD are RNPS1, U1-70K and SRm160, which participate in splicing; however, the relationship between these two mechanisms is unknown. For years, *S. cerevisiae* has been the most outstanding model, because most of its genes are destined for cellular functions. Despite this, this model still has certain difficulties, since it lacks cellular processes that are present in the animal cell, which is why the search for new fungal models has been proposed; A clear example of this is the fungus *U. maydis*. 40% of *U. maydis* genes suffer from alternative splicing events, which are mostly intron retention, which could lead to PTC formation and activation of the NMD pathway. The study of NMD factors, as well as their association with splicing, will allow us to understand their regulation and function in the degradation of aberrant transcripts under normal and inhibitory cellular conditions. Using bioinformatic tools, at least 63 proteins that participate in the NMD pathway have been identified. Each of them is expressed differentially in the different families of fungi. In *Ustilago maydis*, the participation of 47 factors that detect and arrest anomalous transcripts has been described. Furthermore, the expression of the genes that encode the UPF1, UPF2 and UPF3 proteins in *E.coli* DH5α has been verified by PCR. Finally, the data obtained show that *U. maydis* has the necessary factors to activate the NMD pathway, which allows us to perceive this fungus as an excellent study model in the regulation of transcripts and, eventually, extrapolate it in humans to understand abnormal processes. such as cancer, as well as proposing new detection methods and or alternative therapies.

ELUCIDATING THE FUNCTION OF *AZOTOBACTER VINELANDII* PHASINS PROTEINS: PHBP2 AND PHBP3 ARE REQUIRED FOR THE DEGRADATION OF THE BIODEGRADABLE PLASTIC POLYHYDROXYBUTYRATE

Jessica Ruiz Escobedo, Josefina Guzmán Aparicio, Soledad Moreno León, Guadalupe Espín Ocampo, Carlos Peña Malacara, Daniel Segura González

Instituto de Biotecnología / UNAM., AV. Universidad # 2001, Col. Chamilpa, Cuernavaca, Morelos, C.P. 62210.
Tel. 52 (777) 329 1629; daniel@ibt.unam.mx

Polyhydroxybutyrate (PHB) is a natural polyester synthesized by *Azotobacter vinelandii*. This polymer can be used industrially as biodegradable plastic. PHB accumulates intracellularly in the bacterium in the form of granules when the carbon source is abundant, serving as a carbon and energy reserve for the bacterium, and is degraded when the carbon source is depleted. The PHB granules are surrounded by several proteins: those involved in PHB metabolism, both in its synthesis or degradation (1), and other proteins called phasins, that constitute the major protein content on the PHB granule but do not have enzymatic activity. However, phasins can modulate the PHB synthesis or degradation enzymes in some PHB producing bacteria (2). This work focuses on determining the role of PhbP2 and PhbP3 phasins in the PHB metabolism of *A. vinelandii*. For this purpose, the PHB accumulation of strains with disrupted phasin genes (*phbP2* and *phbP3*) and the wild strain OP was compared. In addition, PHB synthase and PHB depolymerase activity assays were performed in the OP strain and mutants. The results show that the phasins PhbP2 and PhbP3 are involved in PHB metabolism, because the absence of these gene caused a decrease in PHB degradation, which could be related to a low PHB depolymerase activity or to a regulation in the genes of the PHB depolymerase enzymes. Furthermore, the expression of *phbP2* and *phbP3* in a heterologous system (*E. coli*), together with PHB biosynthetic enzymes, considerably increased PHB accumulation, showing a stimulatory role on PHB synthesis rather than a control of degradation. It was also shown that PhbP2 and PhbP3 do not intervene in the structuring of the PHB granule, which was expected, since they are minor phasins. However, they could be playing a role as anchors of enzymes involved in the metabolism of PHB to the granule or indirectly regulating its gene expression, since the interruption of *phbP2* and *phbP3* modified the protein banding in a discontinuous SDS-PAGE-Tricine. With the results obtained in this work we show that by manipulating the expression of the phasins PhbP2 and PhbP3 it is possible to increase the production of biodegradable bioplastics in *A. vinelandii*.

References

1. Galán, B., Dinjaski, N., Maestro, B., de Eugenio, L. I., Escapa, I. F., Sanz, J. M., García, J. L., & Prieto, M. A. (2011). *Mol. Microbiol.* 79(2), 402–418.
2. Wieczorek, R., Pries, A., Steinbüchel, A., & Mayer, F. (1995). *J. Bacteriol.* 177(9), 2425–2435.

WHOLE GENOME SEQUENCING OF CANDIDA AURIS STRAIN ISOLATED IN MONTERREY, MEXICO

Alí Fernando Ruiz-Higareda¹, Kame Alberto Galán-Huerta¹, Julio César Garza-Cabello¹, Paola Bocanegra-Ibarias², Adrián Camacho-Ortiz², Ana M. Rivas-Estilla¹

¹Departamento de Bioquímica y Medicina Molecular, Facultad de Medicina, Universidad Autónoma de Nuevo León.

²Servicio de Infectología, Facultad de Medicina, Universidad Autónoma de Nuevo León. Av. Francisco I Madero S/N Col. Mitras Centro, 64460 Monterrey, N.L.
E-mail: amrivas1@yahoo.ca

Introduction. *Candida auris* is a multidrug-resistant yeast causing invasive infections with high mortality rates. Unlike other *Candida* species, *C. auris* is highly contagious and persists in healthcare environments, leading to outbreaks. *C. auris* infections have since spread across six continents. Genomic studies have identified four major clades linked to specific regions: East Asia, South Asia, South Africa, and South America. Understanding its phylogeographic structure is crucial for tracking its spread and developing control measures.

Objective. To characterize the genomic sequence of *C. auris* isolated from a patient at the Hospital Universitario “José Eleuterio González” in 2021 and infer its phylogeny.

Methodology. A clinical isolate of *C. auris* was obtained from a patient at the Hospital Universitario “José Eleuterio González” in 2021. Genomic DNA was extracted and quantified, with 700 ng used to prepare the library using the Ligation Kit (SQK-LSK109) from Oxford Nanopore Technologies. The library was loaded onto a FLO-MIN106 R9.4.1 flow cell on a MinION device. Sequencing was conducted for 72 hours using super high accuracy basecalling (Dorado v.0.6). The genome was assembled de novo using Canu v2.2, resulting in seven chromosomal sequences and a mitochondrial genome with a total length of 12,375,136 base pairs.

Results. The genomic analysis revealed that the Mexican isolate of *C. auris* is similar to strain B12342 from Colombia. Phylogeographic analysis calibrated with BEAST 1 indicated that the isolate belongs to Clade IV, suggesting that *C. auris* was introduced to Mexico in 2014 from Colombia. Mutations identified in the *ERG11* gene (K143R, T220L, and F105L) confer resistance to fluconazole. No mutations associated with resistance to echinocandins were found.

Conclusions. This study characterized the genomic sequence of *C. auris* isolated in northeastern Mexico, highlighting its close relationship with Colombian strains and its resistance to fluconazole. Continuous genomic surveillance and international collaboration are essential to understand the epidemiology of *C. auris* and to develop effective prevention and control strategies.

CHARACTERIZATION OF GUT MICROBIOTA IN CAFETERIA DIET-FED MICE MODEL OF OBESITY

Jessica Ruvalcaba Galindo¹, Luis Marcelino Ramírez Maldonado¹, Diana Laura Cortés Figueroa¹,
Kristy Elizabeth Jiménez Meráz¹, José Luis Rodríguez Mejía¹, Julio Guerrero Castro¹,
Yair Cárdenas-Conejo², Andrómeda Liñán Rico¹

¹Centro Universitario de Investigaciones Biomédicas-Universidad de Colima. Colima. Col., México.28040.

²Laboratorio de Agrobiotecnología-Universidad de Colima, Col., México.

Email: andromeda_linan@ucol.mx

Obesity is a multifactorial disease characterized by a caloric imbalance and the accumulation of adipose tissue. The gut microbiota consists of trillions of microorganisms, such as bacteria, fungi, viruses, and archaea. Unhealthy eating habits induce gut dysbiosis, which promotes obesity and intestinal low-grade inflammation. Metagenomic tools have allowed us to study gut microbial diversity in obesity models. However, bacterial cultures are crucial to analyze host-microbiota interactions. Our goal was to characterize the gut microbiota through metagenomic sequencing and to isolate a collection of culturable bacteria from mice fed two different diets: control diet (CTRL) and cafeteria diet (CAF).

C57BL mice were fed with CAF or a standard diet for 8 weeks. Body weight and glucose levels were recorded. Fecal DNA was extracted, and changes in gut microbiota were monitored by sequencing the V3-V4 region of the 16S rRNA gene. Additionally, the isolation and identification of culturable bacteria were performed through morphological characterization and taxonomic assignment via 16S rRNA gene sequencing. The CAF diet promoted a gradual increase in body weight, disturbed glucose metabolism and intestinal inflammation. Sequence analysis revealed a dysbiotic profile in CAF-fed mice. The CAF group displayed a remarkable increase of *Proteobacteria* and a decrease in *Campilobacterota* phyla. Bacterial hallmarks of health status were reduced in CAF-fed mice, including f_*Prevotellaceae* and g_*Lactobacillus*. On the contrary, the relative abundance of g_*Bacteroidetes* increased. We isolated 40 aerobic and 50 anaerobic bacteria using culturing techniques, classified as either Gram-positive or Gram-negative. Sequencing the 16S gene of these bacterial isolates will allow its taxonomic assignment and subsequent description. The microbiological collection will provide reference material for subsequent studies focused on understanding the role of dysbiotic microbiota in the pathophysiology of obesity, as well as their use to obtain metabolites or products of therapeutic interest.

THE WILD PLANT *CONOPHOLIS ALPINA* AND THEIR VISITING HONEY BEES (*APIS MELLIFERA*) CARRY IDENTICAL BACTERIAL STRAINS UNDER NATURAL CONDITIONS

Salas-Limón, M.J.¹, Bustillos-Cristales, M.R.¹, García-de los Santos, A.², Miguel Castañeda Lucio¹, Quintero-Hernández Veronica¹, R. Carreño-López¹, Munive-Hernández, J.A.¹, Lara-Ochoa, C.³, A. Báez-Rogelio¹ and Fuentes-Ramírez, L.E.¹

¹Centro de Inv. Microbiológicas, IC-11, Ciudad Universitaria, Benemérita universidad Autónoma de Puebla, Puebla, Pue.;

²Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México, Cuernavaca, Mor.; ³Centro de Detección Biomolecular, Ciudad Universitaria, Benemérita universidad Autónoma de Puebla, Puebla, Pue.

E-mail: antonino.baez@correo.buap.mx; luis.fuentes@correo.buap.mx

Nowadays pollination is considered a dynamic process. In addition to its important ecological role, growing evidence proposes pollination as a driver of microbial exchange between pollinators and flowers. Culturable methylotrophic microbial bacteria were isolated from flowers and stems of the non-photosynthetic parasitic plant *Conopholis alpina* and from the proboscis and the guts of honey bee specimens visiting blossoms of the same plant. By 16S rDNA sequence analysis, the following OTUs were isolated, both from the plant and the pollinator: *Paenibacillus glycanilyticus*, *Bacillus subtilis*, *Methylobacterium radiotolerans*, *Serratia quinovorans*, *Pseudomonas libanensis*, and *Herbaspirillum huttiense*. The isolates of those OTUs were analyzed with BOX-PCR. Notably, the parasitic plant and its pollinator share the same strain of both, *Paenibacillus glycanilyticus*, and *Methylobacterium radiotolerans*, but the other OTUs strains are not shared in those environments. Our results provide evidence of microbial exchange between a non-cultivated plant and its pollinator under natural conditions.

Acknowledgments. This work was partially funded by Vicerrectoría de Investigación y estudios de Posgrado, BUAP.

IDENTIFICATION OF ANTIBIOTIC RESISTANCE IN ORAL BACTERIA FROM DOGS WITH PERIODONTAL DISEASE

Yosahandy Palacios Castañón¹, Nailea Zambrano Pérez¹, Sergio Andrés Martínez Hidalgo²
Eduardo Mandujano García² e Ismael Secundino Velazquez¹

¹Facultad de Veterinaria, Universidad La Salle Bajío. ²Hospital Veterinario para Pequeñas Especies. Facultad de Medicina Veterinaria y Zootecnia, Universidad La Salle Bajío. Avenida Universidad 602, Col. Lomas del Campestre. C.P. 37150, León Guanajuato. Tel: 477 8500 ext. 2900.
E-mail: isecundino@lasallebjio.edu.mx

Periodontal disease is a frequent oral pathology found in dogs which consists of inflammation of the periodontium (gingiva, periodontal ligament, and alveolar bone) caused by accumulation of bacterial plaque. While periodontal disease is often considered a localized disease affecting only the dog's mouth, it impacts multiple systems, including the cardiovascular, immune, digestive, and musculoskeletal systems. The reported incidence rate of periodontal disease is 80-90% of dogs older than 3 years. Addressing periodontal disease involves eliminating dental biofilm accumulation by dental cleaning under anesthesia followed by administration of antibiotics, such as metronidazole, clindamycin, enrofloxacin and ciprofloxacin, which might contribute to antimicrobial resistance when they are not administered as indicated. In humans, antibiotic resistant bacteria were found in 83% of isolates responsible of periodontitis. However, the emergence of antibiotic resistance in oral bacteria from dogs has not been addressed. Of note, bacteria inside biofilm are more resistant to antibiotics. This study is focused on determining the antibiotic resistance and the biofilm-forming capacity of oral bacteria. Seventeen dogs with periodontal disease were analyzed (ten males, seven females). Two healthy dogs were included as controls (one male, one female). The mean age of patients with periodontal disease was 3.23 ± 2.74 years (mean \pm SD). The probing depth was 3.08 ± 1.41 mm (mean \pm SD). Healthy dogs aged 2.04 ± 1.46 years (mean \pm SD), with a probing depth of 1.5 ± 0.5 mm (mean \pm SD). Given that bacteria responsible of periodontal disease are found on subgingival space, where a low concentration of oxygen is prevalent, favoring the proliferation of anaerobic microorganism. Thus, bacterial plaque were collected from subgingival plaque using Gracey's cures, then microorganisms were grown under anaerobic conditions. To evaluate antibiotic resistance, bacteria were cultured in the presence of metronidazole, clindamycin, enrofloxacin and ciprofloxacin. To determine biofilm formation, bacteria were cultured in polystyrene wells for 24 h. Subsequently, the biofilm was stained with 0.2% crystal violet and its optical density was measured at 630 nm. Preliminary results suggest that strain P01, which corresponds to one-year-old Doberman breed had a strong capacity of biofilm formation. We are currently evaluating the antibiotic resistance in periodontal pathogens. This study will provide an insight into the antimicrobial resistance in oral bacteria from dogs with periodontal disease.

SEARCH OF MAIZE SEED ENDOPHYTES WITH ANTAGONISTIC ACTIVITY AGAINST *FUSARIUM VERTICILLIOIDES*

Stefhany Segoviano Correa¹, Ma. Fernanda Meneses-Hernández¹,
A. Fabiola Camacho-Gamiño¹, Javier Plasencia de la Parra^{1*}

¹Depto. de Bioquímica, Facultad de Química, UNAM., 04510. Cd. Mx. México.
311112553@quimica.unam.mx

Mexico maize production as well as worldwide, is hindered by both abiotic and biotic factors. *Fusarium verticillioides* is the main fungal pathogen in this crop. This ascomycete can survive as an endophyte in maize tissue or as saprophyte in stubble. It causes seedling blight, stalk rot, ear rot, and synthesizes several mycotoxins harmful to humans and animal. Fumonisin B1 (FB1) is the main toxin produced by this phytopathogen, acting as a virulence factor that promotes fungal colonization and as a contaminant in moldy corn. Because chemical control of this plant pathogen is not economically feasible, biological control agents offer an alternative. We isolated several fungal and bacterial endophytes from maize seeds and identified them through microbiological and molecular techniques. Monosporic or single-colony cultures were obtained, and genomic DNA was purified for PCR amplification. Fungal isolates were analyzed by DNA sequencing of the ribosomal RNAs Internal Transcribed Sequence (ITS) identifying: *Fusarium* sp., *Acremonium* sp., *Phialemoniopsis* sp., *Talaromyces* sp. For bacterial isolates, the 16S ribosomal RNA gene was amplified and sequenced, founding CEL1 and AC1 identified as *Bacillus* spp. inhibits the growth of several *F. verticillioides* strains in a plate assay with different patterns. While CEL1 strain showed a rapid growth that hindered *Fusarium* growth, AC1 strain inhibition pattern suggested the production of diffusible compounds. When CEL1 strain was cocultured with *F. verticillioides* in potato dextrose broth, FB1 production was diminished. In both bacterial strains, glucanase and quitinase activities were recorded. Both enzymes hydrolyze fungal cell wall components. To test their ability as potential biocontrol agents, we developed a formulation based on kaolin and vermiculite, used for coating maize seeds challenged with *F. verticillioides*. In our preliminary tests, plants derived from seed coated with the bacterial formulation showed a more robust system and the aerial growth was like those plants which were not inoculated with *F. verticillioides*. Assays are under way to improve the bacterial formulation and to determine the mode(s) of action of this potential biocontrol agents.

Funding. PAIP 5000-9121 FQ-UNAM.

STUDY AND CHARACTERIZATION OF NEW COMPOUNDS WITH ANTIVIRULENCE AND ANTIFUNGAL ACTIVITY PRODUCED BY *STREPTOMYCES ALBIDOFLOAVUS* J25 ISOLATED FROM MEXICAN JUNGLE SOIL

Daniela Elysa Solorio-Escamilla, Michelle Chávez-Hernández¹, Adilene Gonzalez Silva¹, Juan Alfredo Hernández-García¹, César Hernández-Rodríguez¹, Lourdes Villa-Tanaca¹

¹Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Prol. Carpio y Plan de Ayala, CP 11340, Casco de Santo Tomás, Ciudad de México, México. Teléfono 5729-6000 Ext. 62569.

Correspondence author: Lourdes Villa-Tanaca.

Email: mvillat@ipn.mx

The increase in antimicrobial resistance represents a public health problem, since it has been increasing in recent years, the development of new compounds with inhibitory activity has been compromised because pathogens develop resistance mechanisms faster and faster. The WHO has issued multiple alerts for the search and development of new compounds that are not only focused on inhibiting growth but are also capable of inhibiting different virulence factors. Within the search for new compounds is the genus *Streptomyces*, which is the main producer of secondary metabolites with a wide variety of chemical structures, and therefore, of biological activities on various microbial groups. First, the *Streptomyces albidoflavus* J25 strain was evaluated to inhibit the growth of different *Candida* strains, both sensitive and resistant to fluconazole, using punch tests, the MIC of the supernatant and the possible molecular target was also performed. Anti-virulence activity was demonstrated by inhibiting the formation and destruction of biofilms of antibiotic-resistant strains of *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Staphylococcus aureus*. It also showed anti-quorum sensing activity in *Serratia marcescens*. With the results mentioned above, it was interesting to sequence the genome of this strain to know at the genome level, the biosynthetic clusters (BGC) of sequential metabolites responsible for the different activities. The genome was sequenced using the Illumina HiSeq 2000 platform, once the sequences were obtained, it was assembled, and annotated, phylogenomic identification was carried out and the analysis of the BGCs was carried out with the help of the antiSMASH server. The data obtained from this analysis were as follows: the strain was identified as *Streptomyces albidoflavus* J25, the possible metabolites responsible for the antifungal activity are polyenes, as well as some surugamides, on the other hand, the possible metabolites of the antivirulence activity are class III lantipeptides.

DETECTION OF *BACILLUS CEREUS* AS A STRATEGY TO ASSESS FOOD SAFETY IN MEXICO CITY

María Denisse Tejeda Ramírez, José Carlos Parada-Fabián, Ana Karen Álvarez Contreras, Marcos Francisco Hernández-Robles, Iván Natividad-Bonifacio, Elsa Irma Quiñones Ramírez, Carlos Ramón Vázquez Quiñones, Carlos Vázquez Salinas

National Polytechnic Institute, National School of Biological Sciences, Microbiology Department, Mexico City, Mexico.
Manuel Carpio, Plutarco Elías Calles, Miguel Hidalgo, C.P. 11350 CDMX.
E-mail: jparadaf@ipn.mx, 5525632598

Background and Objective. *Bacillus cereus* is a Gram-positive, spore-forming, facultative anaerobic bacterium widely distributed in the environment and commonly found in a variety of foods. It produces two main types of enterotoxins: the emetic or heat-stable toxin and the diarrheal or heat-labile toxin. The emetic toxin has been identified as a major cause of food poisoning and foodborne illnesses associated with *B. cereus*. In Mexico, the detection of *B. cereus* is not mandatory, thus data on its prevalence are unknown. Obtaining more information about this microorganism allows for a better understanding to implement preventive measures. **Methodology.** Sixteen samples of red rice were obtained from markets in the 16 districts of Mexico City. Isolation and phenotypic characterization followed the methodology proposed by the Food and Drug Administration (FDA) Analytical Bacteriology Manual (BAM). From positive cultures of *B. cereus*, toxin presence was detected using molecular biology by PCR, employing a commercial DNA extraction kit and a standardized PCR to amplify the genes of *B. cereus* emetic toxin. **Results.** *B. cereus* was isolated from red rice samples using MYP plates, exhibiting characteristic morphology after 24 hours of incubation. Biochemical tests allowed for identification consistent with the metabolism described in the BAM literature. Cereulide toxin detection was conducted via molecular biology, categorizing the different districts of Mexico City by biotypes. **Conclusion.** The detection of *B. cereus* and its toxin underscores the importance of rigorous monitoring and control of red rice samples obtained from markets in Mexico City. Although the isolation of *B. cereus* from food is not mandatory, there is a need to gather more information about this microorganism to implement appropriate preventive strategies and measures to ensure food safety in the community.

References

1. Yang, S., Wang, Y., Liu, Y., Jia, K., Zhang, Z., & Dong, Q. (2023). Cereulide and Emetic *Bacillus cereus*: Characterizations, Impacts and Public Precautions. *Foods (Basel, Switzerland)*, 12(4), 833. <https://doi.org/10.3390/foods12040833>
2. M. Tallent, S., Knolhoff, A., Jeffery R., E., M. Harmon, S., & W. Bennett, R. (2021, 29 junio). BAM Chapter 14: *Bacillus cereus*. U.S. Food And Drug Administration. <https://www.fda.gov/food/laboratory-methods-food/bam-chapter-14-bacillus-cereus>

NERNST EQUATION VERSUS KILLER EFFECT IN *SACCHAROMYCES CEREVISIAE*

Brenda Tellez de la Garza¹, Verónica Morales-Tlalpan^{1,2}, Carlos Saldaña^{1,2*}

Laboratorio de Biofísica de Membranas y Nanotecnología. Laboratorio Nacional de Visualización Científica avanzada (UNAM-UAQ). Facultad de Ciencias Naturales, Universidad Autónoma de Querétaro. Juriquilla, Qro., México. CP 76230. Phone + 52 4421921200 x65332. *carlos.saldana@uaq.mx

Saccharomyces cerevisiae needs high extracellular $[K^+]$ to grow (low μM up to 2.5M), while in the intracellular medium $[K^+]$ is low (200-200 mM); it's fundamental maintaining the importation and exportation K^+ systems finely adjusted. Although, yeast can modify its affinity to K^+ and this depends on its availability. Besides, *K1* toxin can inhibit H^+ pumping on sensitive cells and activates K^+ transport to counteract the effect of H^+ loss; this makes yeast develop an internal pH of 6.5 and an external pH of 4.6. So, *K1* acts on sensitive yeast cells to perturb K^+ homeostasis and it's activity depends on temperature, pH, biomolecules, etc. *K1* is shown to activate the plasma membrane K^+ channel (*TOK1*; two-pore outwardly rectifying K^+ channel) and depleting the intracellular medium with K^+ , producing sensitive cells death by unbalancing the electrochemical gradient. The aim of this report is to determine how the extracellular controlled increase of K^+ modifies *K1* response. Experiments were made on YPD culture medium (in %: Yeast Extract 1.0. Peptone 2.0, Dextrose 2.0, Agar 2.0 and 100mM of KH_2PO_4 , pH was adjusted to 4.7 with citric acid); the medium had different $[K^+]$ (0-1000 mM). On the culture medium was made a lawn with 5x47 strain (sensible; sensible; 5x47 MATa/MATalpha his1/+ trp1/+ +/ura3 [KIL-o]), and cell spots with 42300 strain (producer: MAT α Ade2/+Thr1/+sKi2-1/+ [KIL-K1]). Inhibition halo were measured on ImageJ and graphics were made on Origin2019b. Our results suggest that *Killer* effect is affected by 500 mM KCl, indicated by the decrease on the inhibition halo size. These results suggest that *TRK* transport systems, during the *K1* response, are working to compensate K^+ loss when *TOK1* is activated.

Acknowledgments. We extend our sincere gratitude to the funding institutions: FONDEC-UAQ, FONFIVE 2024 and CONAHCyT A1-S-26966 for their generous support and financial backing provided to Carlos Saldaña.

ANTIMICROBIAL POTENTIAL OF HELIOTROPIMUM ANGIOSPERMUM AGAINST SALMONELLA SP., ISOLATED FROM CHICKEN

David Tirado Torres¹, Claudia Cecilia Ramírez Ramírez¹, María del Rosario Abraham Juárez², Jesús Rubén Rodríguez Núñez³, Felicitas Calderón Vega¹ and Eduardo Jahir Gutiérrez Alcántara⁴

¹Departamento de Ingeniería Civil y Ambiental, División de Ingenierías, Campus Guanajuato, Universidad de Guanajuato, Av. Juárez No 77, Col. Centro, Guanajuato, Gto., México. ²Departamento de Ingeniería en Alimentos, División de Ciencias de la Vida, Universidad de Guanajuato, Hacienda El copal Km 9, carretera Irapuato-Silao CP: 36500, ³Departamento de Ingeniería Agroindustrial, División de Ciencias de la Salud e Ingenierías, Campus Celaya-Salvatierra, Universidad de Guanajuato, Privada de Arteaga S7N, zona centro, Salvatierra, Guanajuato, ⁴Facultad de Ciencias Químico-Biológicas, Universidad Autónoma de Campeche, San Francisco de Campeche CP: 24085, Campeche, México. E-mail: d.tirado@ugto.mx

The objective of this study was to evaluate the antimicrobial action of *Heliotropium angiospermum* Murray against antibiotic-resistant strains of *Salmonella* isolated from chicken. Antibiotic resistance is currently recognized by the World Health Organization as an emerging public health problem, which has driven the search for natural antimicrobials, such as *Heliotropium angiospermum* Murray, capable of combating multidrug-resistant microorganisms. Twenty chicken samples were collected and processed using traditional methods. The Kirby-Bauer method was used for dendrogram construction. Extracts of *Heliotropium angiospermum* Murray were obtained following the methodology of Tirado-Torres et al., 2019. The antimicrobial activity of these extracts was analyzed using the disk diffusion method. A 70% prevalence of *Salmonella* was found. Ampicillin was the least effective antibiotic, with 100% resistance, while amikacin had a 70% resistance rate. The ethanolic extract showed the greatest inhibitory effect, with zones of inhibition measuring 23 mm. Multidrug-resistant strains were identified. The results indicate that *Heliotropium angiospermum* Murray could be an effective alternative for controlling infections caused by *Salmonella*.

Acknowledgments. A la Universidad de Guanajuato a través del apoyo Fortalecimiento de la Capacidad Académica (249/2024).

References

1. Domham, C., Uhl, P., Meinhardt, A., Zimmermann, S., Kleist, C., Linder, T., Leotta, K., & Wink, M. (2018). International Journal of Antimicrobial Agents, 52, 52-62.
2. Maka, L., Maćkiw, E., Sciezynska, H., Pawlowska, K., & Popowska, M. (2020). Food Control, 36, 199-204.

EVALUATION OF THE ANTIMICROBIAL ACTIVITY OF *Buddleja scordioides* AGAINST *SALMONELLA* SP., ISOLATED FROM CHICKEN

Guadalupe Vázquez Rodríguez¹, Jaime Juan Huerta Delgado¹, David Tirado Torres¹, Juan Carlos Baltazar Vera¹, Saúl Villalobos Pérez¹ and Juan Esteban Bello Lara²

¹División de Ingenierías, Campus Guanajuato, Universidad de Guanajuato, Av. Juárez No 77, Col. Centro, Guanajuato, Gto., México. ²Universidad Autónoma de Nayarit, Xalisco, Nayarit, C.P: 63780, México.
E-mail: d.tirado@ugto.mx

The purpose of this study was to investigate the antimicrobial action of *Buddleja scordioides* against antibiotic-resistant strains of *Salmonella* isolated from chicken. Antibiotic resistance, currently recognized by the World Health Organization as a growing public health problem, has prompted the search for natural antimicrobials like *Buddleja scordioides* capable of combating multidrug-resistant microorganisms. Twenty chicken samples were collected and processed using traditional methods. The Kirby-Bauer method was used for dendrogram construction. Extracts of *Buddleja scordioides* were obtained following the methodology of Tirado-Torres et al., 2019. The antimicrobial activity of these extracts was evaluated using the disk diffusion method. A 70% prevalence of *Salmonella* was observed. Ampicillin was the least effective antibiotic, with 100% resistance, while amikacin showed a 70% resistance rate. Acetonic and ethanolic extracts resuspended in aqueous phase were analyzed, with the ethanolic extract showing the greatest inhibitory effect, with zones of inhibition measuring 13 mm. Multidrug-resistant strains were identified. The results suggest that *Buddleja scordioides* could be an effective alternative for controlling infections caused by *Salmonella*.

Acknowledgments. A la Universidad de Guanajuato a través del apoyo Fortalecimiento de la Capacidad Académica (249/2024).

References:

1. Jamali, H., Paydar, M., Radmerhr, B., Ismail, S., Dadrasnia, A. (2019). *Food Cont*, 54, 383-388.
2. Maka, L., Maćkiw, E., Scieczynska, H., Pawlowska, K., & Popowska, M. (2020). *Food Control*, 36, 199-204.

BIOINFORMATIC ANALYSIS OF GLYCINE DERIVATIVE OF 22-OXOCOLESTENYL AS POTENTIAL ANTI-CANCER AGENT

Ana Karen Villa Merlán^{1,2}, Beatriz Eugenia Baca^{1,2}, Alan Carrasco Carballo²

¹Centro de Investigaciones Microbiológicas, Av. San Claudio, S/N, Ciudad Universitaria Edif. Edif. IC111, Puebla Pue.

²Laboratorio de Elucidación y Síntesis en Química Orgánica, Instituto de Ciencias de la Universidad Autónoma de Puebla (ICUAP), Benemérita Universidad Autónoma de Puebla, Puebla 72570, México.

E-mail: p22mcm0032@viep.com.mx

Cancer is a multifactorial disease that is distributed worldwide and is considered as one of the main causes of death (1). Steroidal derivatives, such as 22-oxocolestano, have been reported as molecules with antitumoral potential, however, aminoester derivatives have not yet been evaluated (2). In the present study, the bioinformatic analysis of a glycine aminoester derivative of 22-oxocolesteryl was carried out with the objective of evaluating its biological potential as an anti-cancer compound. Then, the analysis on the PASSOnline platform, potential biological activities were identified, including antineoplastic, immunosuppressive activity and as a treatment of proliferative disease. These activities were preserved as well as the derivative and the original compound, suggesting the preservation or increase of activities. Through SWISS Target Prediction, it was found that cancer-associated activities can be due to potential molecular targets, highlighting the 5 α -reductase enzyme, associated to prostate cancer and benign prostatic hyperplasia (3). To determine if the derivative can exert an inhibitory effect in the enzyme, a molecular docking analysis was performed, where a better coupling energy of the derivative in contrast to its original compound and the endogenous substrate were observed (testosterone). The amino acids involved in the interaction were analysed, finding Asn193, Tyr98 and Asp164 as the main residues. Despite not being the residues of the catalytic site of the enzyme, the interactions occur near to Glu57 and Tyr91 residues, which suggests a steric inhibition. These results propose Glycine aminoester derivative of 22-oxocolesteryl as a molecule with anticancer potential.

References

1. Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A. (2021). *CA Cancer J Clin.* 71: 209–49.
2. Aguilar-Barrientos JP, Moo-Puc RE, Villanueva-Toledo JR, Murillo F, Cáceres-Castillo D, Mirón-López G. (2022). *Steroids.* 183: 109030.
3. Korassa YB, Saptarini NM, Mustarichie R, Hendriani R, Tjitda PJP. (2022). *Int J App Pharm.* 49–57.

MITOGEN-ACTIVATED PROTEIN KINASE HOG1: KEY REGULATOR FOR ENHANCING RIBOFLAVIN OVERPRODUCTION IN THE YEAST *DEBARYOMYCES HANSENI*

Diana Villarreal-Huerta¹, Lucero Romero-Aguilar², Nayeli Torres-Ramírez¹, Miguel Ángel Rosas-Paz¹, Norma Silvia Sánchez³, Roberto Arreguín-Espinosa⁴, Mohammed El-Hafidi⁵, Viviana Escobar-Sánchez¹, Claudia Segal-Kischinevzky¹, James González¹

¹Departamento de Biología Celular, Facultad de Ciencias, UNAM. ²Departamento de Bioquímica, Facultad de Medicina, UNAM. ³Departamento de Genética Molecular, Instituto de Fisiología Celular, UNAM. ⁴Departamento de Química de Biomacromoléculas, Instituto de Química, UNAM. ⁵Departamento de Biomedicina Cardiovascular, Instituto Nacional de Cardiología.
Main institution: Facultad de Ciencias, UNAM. Address: Av. Universidad 3000, Circuito Exterior s/n, Alcaldía Coyoacán, C. P. 04510, Ciudad Universitaria, Ciudad de México.
Phone number: 56225433. villadianahuerta@ciencias.unam.mx

The industrial production of vitamin B2 (riboflavin) using flavinogenic microorganisms has largely replaced chemical synthesis due to its superior cost-effectiveness for large-scale production. Although *Ashbya gossypii*, *Bacillus subtilis* and some *Candida* species exhibit favorable riboflavin yields, they have both advantages and limitations. *Debaryomyces hansenii* (synonym *Candida famata*) is a well-known flavinogenic yeast previously used in riboflavin production, but discontinued due to genetic instability in overproducing-strains. Besides, *D. hansenii* is a valuable model for studying stress response pathways and lipogenesis, being an halotolerant and oleaginous yeast. Its halotolerance is primarily attributed to an enhanced High Osmolarity Glycerol (HOG) pathway, which is more robust than in non-halotolerant yeasts like *Saccharomyces cerevisiae*. Additionally, the lipogenic pathways in *D. hansenii* are not fully elucidated. This study aims to characterize lipid accumulation and riboflavin production in a *Dhhog1Δ* mutant under lipogenic conditions with NaCl. The results show that the *Dhhog1Δ* mutant increases lipid production, and a yellow pigment appears in the medium when NaCl is added. Fluorescence measurements (440 nm/535 nm) revealed significantly increased riboflavin content in the supernatant. We will discuss the connection between the HOG pathway, stress response, and riboflavin biosynthesis.

ANALYSIS OF GENES *CDGE* AND *HKHB* IN *AZOSPIRILLUM BALDANORIUM* SP245 INVOLVED IN BIOFILM FORMATION AND MOTILITY

María Luisa Xiqui Vázquez¹, Carlos Daniel Cordero-Rivera¹, Miguel A. Ávalos Rangel¹,
Sandra Reyes Carmona¹, Alberto Ramírez-Mata¹ and Beatriz E. Baca¹

¹Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla,
Ciudad Universitaria, Av. San Claudio S/N Edif. IC11, Puebla, Pue. México.
beatriz.baca@correo.buap.mx

The plant growth-promoting bacterium *Azospirillum baldanorium* Sp245 contains several genes encoding proteins involved in the biosynthesis and degradation of the second messenger cyclic-di-GMP¹, which may control key bacterial functions, such as biofilm formation and motility. Here we described the function of two genes that likely encoding by a diguanylate cyclase (*CdgE*), and a histidine kinases (*HkhB*). To study these genes, it was constructed the following mutants: an insertional *cdgE::gusA-Km^R* mutant, the deletion Δ *cdgE*, Δ *hkhB* and Δ *cdgE*- Δ *hkhB* mutants. It was determined the swimming motility of mutants. The results showed a decrease in mobility compared to the WT. This is an unusual phenotype because the mutants in the genes encoding for diguanylate cyclases has an increase of motility phenotype². Preliminary result of RT-PCR indicates that genes *cdgE* and *hkhB* form a transcriptional unit. The data obtained suggest that a putative operon is implicated, and may be associated in a complex signaling pathway. Moreover, we constructed a corresponding complemented mutant strain harboring the wild-type *cdgE* gene to determine the effect of the mutation on biofilm formation and motility, to compare to the WT.

References

1. Alberto Ramírez Mata, César Millán Pacheco, José F. Cruz Pérez, Martha Minjárez Sáenz, and Beatriz E. Baca. *In silico* comparative analysis of GGDEF and EAL domain signaling proteins from the *Azospirillum* genomes. BMC Microbiology doi.org/10.1186/s12866-018-1157-0.
2. U. Jenal., A. Reinders, C. Loiri. Cyclic di-GMP: second messenger extraordinaire. Nat Rev. Microbiol. . 2017 May;15(5):271-284. doi: 10.1038/nrmicro.2016.190.
3. Acknowledgments: We thank to Vicerrectoria de investigación y estudios de posgrado (VIEP) for financial support

CHARACTERIZATION OF THE ANTIBIOTIC EFFECT AND METAL RESISTANCE IN THE C4 STRAIN OF *PROVIDENCIA STUARTII*

Gokabeth Zarco Hernández², Aldo Agustín Pérez Fonseca², Jorge Luis Álvarez Zapata², Rocío Alejandra Silva Contreras², Aurelio Álvarez Vargas¹, Carmen Cano Canchola¹, Juan Francisco Sánchez López²

¹Universidad de Guanajuato. Departamento de Biología. División de Ciencias Naturales y Exactas. Guanajuato, Gto., México. C.P. 36000. ²Instituto Politécnico Nacional-Unidad Profesional Interdisciplinaria de Ingeniería Campus Guanajuato. Silao, Gto., México. C.P. 36275. Tel. (55)5729-6000 ext. 81380. jusanchezl@ipn.mx

The C4 bacterial strain is part of our collection of bacteria isolated from mining tailings. In this work, an axenic culture of this strain was carried out, followed by growth kinetics in different culture media in order to analyze the antibiotic effect in the supernatants obtained on Gram positive or Gram negative bacteria. On the other hand, resistance or sensitivity tests to heavy metals were carried out as well as antibiogram tests to different antibiotics. Likewise, the identification of this C4 strain was carried out through biochemical tests and PCR amplification of the 16S ribosomal gene. Finally, in order to detect the presence of extra chromosomal material, plasmids were extracted and purified in strain C4 and subsequently cut with restriction enzymes. As part of the results, an antibiotic effect was detected in the supernatants of strain C4 on *E. coli* from the second day onwards, evidenced by the formation of a halo of inhibition of growth of *E. coli* in solid medium. The results on sensitivity to different antibiotics showed that strain C4 presented resistance to Pb^{+2} , Zn^{+2} and Cu^{+2} at the concentrations analyzed (0.1, 0.2, 0.4 and 0.6 mM); in the case of Mn^{2+} , concentrations of 10 and 15 mM affected its growth. Regarding sensitivity to different antibiotics, the C4 strain presented 5 resistances (PE, AM, DC, CF, and STX), 4 intermediate tolerances (NF, CFX, CTX and CL) and only 3 sensitivities (NET, AK and GE). Biochemical and molecular tests confirmed that strain C4 is an isolate of *Providencia stuartii*. Finally, the presence of a plasmid was detected in strain C4, with a molecular weight of 3,000 bp, revealed by cutting with the *EcoRI* enzyme, which is being characterized to determine its possible relationship with resistance to metals.

Keywords. *Providencia stuartii*, heavy metals, antibiotic resistance, plasmids.

GENETIC STUDY OF MELANIN SYNTHESIS IN *AZOTOBACTER VINELANDII*

Biol. María Fernanda Chávez Jacobo¹, Biol. Diego García González¹, D.C Dalia Molina Romero¹,
M.C Nohely Aimeé Pizano Sosa¹, D.C Liliana López Pliego¹, D.C Miguel Castañeda Lucio¹

Centro de Investigación en Ciencias Microbiológicas, Benemérita Universidad Autónoma de Puebla, Avenida San Claudio S/N,
Puebla Pue, Mexico. C.P. 72570¹

E-mail: maria.chavezj@alumno.buap.mx; Cel: 2214303571

Melanins are polymeric pigments produced by a wide range of organisms. These pigments arise through the oxidation of phenolic or indolic substrates and subsequent random polymerization that results in melanins: eumelanin, pheomelanin, neuromelanin, pyromelanin, or allomelanins². These play an essential role in the survival mechanisms against environmental stresses like resistance to UV radiation, thermoregulation, antioxidation, and chelation of metals. Their physicochemical features give them a vast potential for multiple applications in industry³. Species of *Bacillus*, *Rhizobium*, *Azospirillum*, and *Azotobacter*, were reported to produce melanins. Particularly in *Azotobacter*, the studies are mostly limited to *A.nigricans*, *A. salinestris*, and *A.chroococcum*⁶. The latter is the most studied in which the structure of these pigments was characterized¹, and several physiological studies reveal its ability to produce them from different substrates such as tyrosine or malonil-CoA; however, there are no genetic or biochemical studies to support it. Besides, the synthesis of these biopolymers has not been reported in the model organism *A.vinelandii*. A comparative genome study between *A.vinelandii* and *A.chroococcum* reported the presence of putative genes involved in melanins synthesis⁴. The significance of our research lies in the potential applications of our findings. We isolated an *A. vinelandii* strain that produces a brown-black melanin on the differential Ashby medium, with benzoate as the carbon source. Since benzoate, a widely used food preservative, could serve as an economical precursor for melanin synthesis in the biotechnology industry. Our findings could potentially revolutionize melanin production, making it more sustainable and cost-effective. We carried out transposon random mutagenesis to characterize possible genes involved in melanin production. To select mutants with alterations in pigment synthesis, we formulated a selective culture media supplemented with benzoate. Four mutants were selected, and we have started their characterization.

References

1. Banerjee, A., Supakar, S., & Banerjee, R. (2014). PloS one, 9(1)
2. Pavan, M. E., López, N. I., & Pettinari, M. J. (2020). Applied microbiology and biotechnology, 104(4), 1357–1370.
3. Rizvi, A., & Khan, M. S. (2018). Ecotoxicology and Environmental Safety, 157, 9–20.
4. Robson, R. L., Jones, R., Robson, R. M., Schwartz, A., & Richardson, T. H. (2015). PloS one, 10(6).
5. Singh, S., Nimse, S. B., Mathew, D. E., Dhimmar, A., Sahastrabudhe, H., Gajjar, A., Shinde, P. B. (2021). Biotechnology Advances, 107773. doi:10.1016/j.biotechadv.2021.107773
6. Shivprasad, S., & Page, W. J. (1989). Applied and environmental microbiology, 55(7), 1811–1817.

RISK OF THE PREVALENCE OF *STAPHYLOCOCCUS AUREUS* IN ARTISANAL CHEESES SOLD IN MEXICO CITY

Marcos Francisco Hernández-Robles^a, Carlos Ramón Vázquez Quiñones^a, Iván Natividad-Bonifacio^a,
Ana Karen Álvarez Contreras^a, José Carlos Parada-Fabián^a,
Elsa Irma Quiñones Ramírez^a, Carlos Vázquez Salinas^b

^aNational School of Biological Sciences, National Polytechnic Institute. Calle Carpio y Plan de Ayala s/n, Col. Santo Tomás, Del. Miguel Hidalgo, C.P.11340. Mexico, Mexico City. ^bAutonomous Metropolitan University-Iztapalapa, San Rafael Atlixco #186, Col. Vicentina, Del. Iztapalapa, C.P. 09340 Mexico, Mexico City.

*E-mail: marcos_ibq@hotmail.com

Staphylococcus aureus is considered a pathogenic bacterium of importance in food safety, causing food poisoning due to the production of heat-stable staphylococcal toxin. Main foods contaminated by this microorganism are dairy products so the objective of this study was to demonstrate the presence of *S. aureus* in artisanal cheeses sold in public streets of Mexico City. The methodology used was established in NOM-210-SSA1-2015. One hundred samples of artisanal cheeses were analyzed (25 Oaxaca type, 25 panela, 25 grated, and 25 double cream). To confirm the identity of the *S. aureus* strains, the *nuc* gene, which encodes the TNAsa enzyme, was amplified by PCR, and the ability to produce enterotoxins was evaluated using the Visual Immunoassay of Staphylococcal Enterotoxins (VIA of SET). Additionally, the activity of TNAsa was evaluated by modifying factors such as temperature, pH, and sodium chloride concentration; the *S. aureus* ATCC 29213 strain was used as a positive control. In 50% of the samples analyzed (11 in Oaxaca cheese, 14 in fresh cheese, 17 in grated cheese, and 8 in double cream cheeses), colonies characteristic of *S. aureus* were observed on Baird Parker agar. A total of 50 presumptive colonies of *S. aureus* were obtained, and all amplified the *nuc* gene. Regarding the quantification of TNAsa activity, it was not affected under the evaluated conditions of NaCl and pH, while the temperature at which the enzyme had the highest activity was 37°C. These results show the risk of consuming these products due to the likelihood that consumers may experience food poisoning with children, the elderly, and immunocompromised individuals being at higher risk, given that these products are not properly preserved and the production conditions are unknown.

AVIN 41190: A NOVEL REGULATORY ELEMENT IN THE MULTIKINASE NETWORK GACS (MKN-GACS) IN AZOTOBACTER VINELANDII

M.C Itzel Rodríguez Guerra¹, D.C Liliana López Pliego¹, D.C Miguel Castañeda Lucio¹

Centro de Investigación en Ciencias Microbiológicas, Benemérita Universidad Autónoma de Puebla, Avenida San Claudio S/N,
Puebla Pue, Mexico. C.P. 72570¹

E-mail: Itzelrodriguez94@hotmail.com; Cel: 2228196245

Two-component systems act as a basic stimulus-response coupling mechanism to allow organisms to sense and respond to changes in many different environmental conditions¹. This mechanism involves a Histidine-Kinase (HK) that activates a Response Regulator (RR) through phosphorylation, which activates a downstream effector domain that triggers a specific response². However, a recently emerging study field highlights multikinase networks (MKNs), where multiple HKs collaborate to detect and integrate diverse signals into different responses, explaining the widespread diversity and adaptive capabilities in bacteria. GacS-MKN from *P. aeruginosa* is the best characterized, where besides of GacS, other accessory kinases (LadS and RetS) form a part of this MKN⁴. In *Azotobacter vinelandii*, we have started the characterization of MKN-GacS; in this bacterium, the accessory kinases are RetS and HrgS³. In both bacteria, GacS-MKN controls the activity of the response regulator GacA. The RR GacA positively regulates the transcription of genes that encode small RNA regulators (sRNAs) belonging to the Rsm (Csr) family, which, in turn, counteracts the activity of the post-transcriptional repressor RsmA³. *A. vinelandii* is an aerobic gamma proteobacterium that can fix nitrogen and produce alginate⁵. This polysaccharide is used as a thickening agent, stabilizer, and emulsifier in the food, medical, and pharmaceutical industries. The alginate is one of the targets of regulation of MKN-GacS in *A. vinelandii*. Avin_41190, a novel Histidine-Kinase, shares a structural similarity with HrgS. Both kinases feature a 7TMRDISMED periplasmatic sensor domain, a highly conserved protein architecture found only in select kinases from *Azotobacter* and *Pseudomonas*. The aim of this study is to shed light on the potential role of Avin_41190 in the MKN-GacS network in *A. vinelandii*. Understanding how Avin_41190 interacts with the kinases in this network and its relationship with the Rsm system could significantly contribute to our knowledge of alginate production regulation.

References

1. Stock, A. M. Robinson V. L., Goudreau P. N. (2000) *Annu. Rev. Biochem.* 69:183–215
2. Francis, V. I., & Porter, S. L. (2019). *Annual review of microbiology*, 73, 199-223.
3. López-Pliego, L., González-Acocal, V., García-González, D. L., Reyes-Nicolau, J. I., Sánchez-Cuapio, Z., Meneses-Carbajal, A. S., Castañeda, M. (2022) *FEMS Microbiology Letters*, 369(1).
4. Chambonnier, G., Roux, L., Redelberger, D., Fadel, F., Filloux, A., Sivaneson, M., de Bentzmann, S., & Bordi, C. (2016). *PLoS genetics*, 12(5).
5. Castañeda, M., Guzmán, J., Moreno, S., and Espín, G. (2000) *J. Bacteriol.* 182:2624-2628

STUDY OF BACTERIOPHAGES TO CONTROL OF MULTI-DRUG RESISTANT BACTERIA

Omar Alejandro Sepúlveda Robles¹, Iasha Jocelyn Ferreyra-Luna², Sayuri Salgado-López³, Mario Rafael Reyes-Torres³, Maribel Ibarra-Sarlat⁴, Roberto Joaquín Robles-Ramírez⁵, Juan Manuel Ruíz-Ruiz⁶, Francisco Javier Torres-López⁷, María Guadalupe Miranda-Novales⁸, Haydeé Rosas Vargas¹

¹Unidad de Investigación Médica en Genética Humana, UMAE Hospital de Pediatría, CMN Siglo XXI, IMSS. ²Facultad de Estudios Superiores Cuautitlán, UNAM. ³Facultad de Ciencias de la Salud, Universidad Autónoma de Tlaxcala. ⁴Unidad de Terapia Intensiva Pediátrica, UMAE Hospital de Pediatría, CMN Siglo XXI, IMSS. ⁵División de Auxiliares de Diagnóstico y Tratamiento, UMAE Hospital de Pediatría, CMN Siglo XXI, IMSS. ⁶Laboratorio Clínico de Microbiología, UMAE Hospital de Pediatría, CMN Siglo XXI, IMSS. ⁷Unidad de Investigación en Enfermedades Infecciosas y Parasitarias, UMAE Hospital de Pediatría, CMN Siglo XXI, IMSS. ⁸Unidad de Investigación en Análisis y Síntesis de la Evidencia, UMAE Hospital de Pediatría, CMN Siglo XXI, IMSS.
*E-mail: sero_82@icloud.com

Background. The World Health Organization has recognized antimicrobial resistance (AMR) as one of the most serious public health threats to the treatment of infectious diseases worldwide. Indeed, it was estimated that AMR mortality could exceed deaths caused by cancer in 2050. Thus, it is important to develop some alternate treatments for the antibiotic usage. In this sense, “phage therapy” –the use of bacterial viruses (bacteriophages or phages) as a treatment for infections– is a promising option, since phages are, by nature, the main predator of bacteria. The aim of the present study was to create a collection of phages capable of infecting multidrug-resistant bacteria. **Methods.** It was isolated and characterized by antibiotic resistance and pulsed field gel electrophoresis, bacteria strains causing infections in hospitalized patients. Bacteria were used to phage isolation from wastewater samples, which were characterized by host range, DNA restriction and electronic microscopy. **Results.** A collection of 597 bacteria strains causing infections was formed, they had a variable phenotypic antimicrobial susceptibility pattern. The leading pathogens isolated were *P. aeruginosa*, followed by *S. aureus*, *S. epidermidis*, *K. pneumoniae*, *E. coli*, *A. baumannii*, *S. haemolyticus*, *E. cloacae*, *S. hominis*, *S. marcescens*, and *S. maltophilia*. A total of 115 phages specific to *P. aeruginosa*, *S. aureus*, *K. pneumoniae*, *E. coli*, *A. baumannii*, *S. maltophilia*, *P. mirabilis* and *Salmonella spp.* were isolated; 28 of which showed an infection capacity against 40.7% of the bacteria strains. **Conclusions.** A collection of phages with potential therapeutic use against multi-drug resistant bacteria and prevalent at the Pediatric Hospital of XXI Century National Medical Center of IMSS was formed. Completing the characterization of these phages is essential to offer phage therapy as an alternative treatment against bacterial infections when antibiotics are not effective.

GENOMIC INSIGHTS INTO THREE CLINICAL *SERRATIA MARCESCENS* STRAINS OF ENVIRONMENTAL ORIGIN

Faviola Tavares-Carreón¹, Mishael Sánchez-Pérez², Angel Andrade³,
Karla de Anda-Mora³, Rodolfo García Contreras⁴

¹Facultad de Ciencias Biológicas, Universidad Autónoma de Nuevo León. ²Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México. ³Departamento de Microbiología, Facultad de Medicina, Universidad Autónoma de Nuevo León. ⁴Departamento de Microbiología y Parasitología, Facultad de Medicina, Universidad Nacional Autónoma de México. ¹Faviola Tavares-Carreón. Facultad de Ciencias Biológicas, Universidad Autónoma de Nuevo León. Av. Universidad s/n, Ciudad Universitaria, San Nicolás de los Garza, Nuevo León, C.P. 66450. México.
Email: ftavaresc@uanl.edu.mx

Serratia marcescens (SM) is most commonly known as an opportunistic pathogen causing nosocomial infections. It, however, was shown to infect a wide range of hosts apart from vertebrates such as insects or plants as well, being either pathogenic or growth-promoting for the latter. The recent genomic analysis have been highlight important differences in diversity, plasticity and niche adaptation of SM. The latest report suggests five deeply demarcated SM clusters based on comparative whole genomic analyses. Cluster 1 is associated with clinical settings, while Cluster 3 shows enrichment in environmental sources such as soil, plants, and water. Cluster 5 is enriched in environmental and animal sources, and Clusters 2 and 4 are not associated with any specific isolation sources. Thus, it appears that there are genomic differences between clinical isolates and those inhabiting natural environments. To date, thousands of clinical SM strains have been deeply scrutinized to know the antimicrobial resistance and virulence factors but genomic analyses properties and distinct trends related to isolation sources is poorly studied.

In this study, we present the whole-genome sequencing data of three prodigiosin-producing strains isolated from different patients, namely HU1848, HU2225, and HU2228. Analysis of Mash distances, ANI, and phylogenetic tree construction reveals that these three pigmented strains are closely associated with the environmental cluster (Cluster 5) and distantly related to the clinical cluster (Cluster 1). The identification of antimicrobial resistance genes using the RGI database indicates that these strains harbor several genes encoding RND and MFS efflux pumps, responsible for extruding a broad range of antimicrobial compounds. Additionally, genes encoding broad-spectrum β -lactamases such as vancomycin, fosfomicin, and tetracycline were detected in HU1848, HU2225, and HU2228. Notably, despite this resistance profile, these strains remain susceptible to aminoglycosides, similar to other environmental strains like Db11. Furthermore, HU1848, HU2225, and HU2228 encode a diverse array of virulence factors, including fimbriae and exoenzymes (particularly phospholipase, hemolysin, serralyisin, proteases, and chitinase). Interestingly, these strains also possess genes involved in the degradation of environmental xenobiotics such as atrazine and haloalkanes. In summary, the HU1848, HU2225, and HU2228 strains exhibit a stronger association with the environmental cluster rather than the clinical cluster, demonstrating an ability to adapt to various niches, whether human or environmental, shedding light on their genomic plasticity and potential to cause illness in humans.

RISK OF THE PRESENCE OF *ESCHERICHIA COLI* IN VEGETABLES MARKETED IN MEXICO CITY

Carlos Ramón Vázquez Quiñones^a, Iván Natividad-Bonifacio^a, Marcos Francisco Hernández-Robles^a, Ana Karen Álvarez Contreras^a, José Carlos Parada-Fabián^a, Carlos Vázquez Salinas^b, Elsa Irma Quiñones Ramírez^a

^aNational School of Biological Sciences, National Polytechnic Institute. Calle Carpio y Plan de Ayala s/n, Col. Santo Tomás, Del. Miguel Hidalgo, C.P.11340. Mexico, Mexico City. ^bAutonomous Metropolitan University-Iztapalapa, San Rafael Atlixco #186, Col. Vicentina, Del. Iztapalapa, C.P. 09340 Mexico, Mexico City.

*E-mail: vazquezqc1973@gmail.com

Escherichia coli is found colonizing the lumen of intestines in mammals acting as a commensal bacterium; However, there are some strains that have genes encoding virulence factors. According to the characteristics of their pathogenicity mechanisms they are classified as enteropathogenic *E. coli* (EPEC), enteroaggregative (EAEC), enterotoxigenic (ETEC), enteroinvasive (EIEC), diffusely adherent (DAEC) and Shiga-type toxin-producing (EHEC). In order to determine the presence of these pathotypes, 90 vegetable samples (30 lettuce, 30 cabbage and 30 coriander) from different markets of the Tlahuac municipality in Mexico City were analyzed according to the methodology described in the Bacteriological Analytical Manual of the FDA. *E. coli* was isolated in 33.3% of the lettuce samples, 43.3% on cabbage and 60% on coriander. Subsequently, the search for genetic markers for each pathotype was carried out, obtaining the following distributions: in the analyzed lettuce samples, the ETEC pathotype was identified in 16%, and in 3% EPEC and ETEC in the same sample, in cabbage the ETEC pathotype in 20%, EPEC in 13% and in 10% of the samples EPEC and ETEC were identified in the same sample. In relation to the analyzed cilantro samples, the ETEC pathotype was identified in 23%, EPEC in 10%, and EAEC in 1%. These results showed that the vegetables that are marketed and consumed by the population of these municipalities have the possibility of containing pathotypes of *E. coli*, with the risk of causing foodborne diseases among residents, especially if these products do not have a hygiene process. and disinfection before consumption.

The background of the entire page is a composite image. On the left, there is a close-up of a plant stem with several green leaves. On the right, there is a glowing, golden-yellow molecular structure, possibly representing a protein or a complex of atoms, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

NEUROSCIENCES & NEUROBIOLOGY

COGNITIVE STIMULATION INCREASES MICROGLIAL PHAGOCYTOSIS IN AN ALZHEIMER'S DISEASE MODEL

David F. Aguilar-Ávila¹, Gustavo J. Fabian- Rodríguez¹, Héctor E. López-Valdés¹,
Isabel Arrieta-Cruz², Mónica Torres-Ramos³ and Hilda Martínez-Coria^{1*}

¹Departamento de Fisiología, Facultad de Medicina, UNAM, CDMX. ²Departamento de Investigación Básica, INGER, CDMX.

³Ciencia Traslacional. Centro de Investigación sobre el Envejecimiento del CIVESTAV -Instituto Nacional de Neurología y Neurocirugía Manuel Velasco Suárez.

*hildamcoria@gmail.com, Telephone 5534360647

Microglia are crucial for modulating cognition, memory, behavior, oxidative stress, and inflammation and they are considered the main phagocytic cells in the central nervous system. The process of microglial phagocytosis is crucial for maintaining adult brain homeostasis and eliminating potential harmful factors. Dysfunction of microglia has been linked to neurodegenerative and neuroinflammatory disorders.

We previously demonstrated that cognitive stimulation improves cognitive impairment and decreases microglia-associated neuroinflammation in 3xTg-AD mice. We sought to investigate the effects of cognitive stimulation by Enrichment Environmental (EE) on microglial phagocytosis.

Males and females 12-month-old 3xTg-AD were housed in EE that was changed every 7 days and trained for 5 days every month for 3 months. At the end of the stimulation program, they were tested in a Barnes Maze paradigm and compared with 3xTg-AD of the same age and sex without stimulation. After the memory test, we obtained the brain, and forty-micrometer frozen sections were prepared as described previously and used Iba-1, TNF α , and TREM2 antibodies to identify the immunophenotype of microglia.

Our results showed that cognitive stimulation increased phagocytic activity of microglia, decreased inflammation, and improved memory decline in 3xTg-AD female mice.

AGE-RELATED INCREASES IN SKELETAL MUSCLE ALPHA-SYNUCLEIN MONOMERIC AND OLIGOMERIC FORMS

Ana Leticia Arriaga-Guerrero^a, Vianney Ramírez-Adoney^b, Ibrahim Ramirez-Soto^c and Alicia Ortega-Aguilar^{a*}

^aDepartamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México. ^bDepartamento de Genética y Bioestadística, Facultad de Medicina Veterinaria y Zootecnia, UNAM. ^cDepartamento de Kinesiólogía, Universidad de Waterloo, Canadá. *Corresponding: Postal address: Dto. de Bioquímica, Facultad de Medicina, Av. Universidad 3000, Copilco, CDMX, 04510, email: aortega@unam.mx, telephone: 5556232253

Nowadays Parkinson's disease (PD) is a major disability and a public health issue due to motor and cognitive symptoms, affecting predominantly older adults. The main protein with PD has been linked is alpha Synuclein (α Syn), this is a soluble 140 amino acids protein with a non-native form. However, in contact with membrane lipids leads to oligomers and aggregates formation. Recently our research group has been identifying equal and even higher levels of α -Syn in skeletal muscle plasma membrane than brain. Skeletal muscle (SM) represents approximately 40% of a mammal's body mass and during aging, changes occur in all organs especially in SM due to this tissue not being divided after birth. Proteomics studies in muscle during aging have identified a synthesis decrease of structural and contractile proteins. Muscle fiber is constantly exposed to mechanical and metabolic damage that worsen during aging resulting in proteolysis related toxic effects. These conditions could trigger muscle degenerative diseases which are similar to neurodegenerative diseases in protein aggregate formation associated with aging.

On this work, we identified the presence of α Syn monomeric and oligomeric forms in young healthy rats sarcolemma and brains, further α Syn was found phosphorylated at serine 129 and when α Syn expression was compared between old and young rats, we identified an overexpression of this protein in old rats sarcolemma and brains. These findings are interesting given that the presence of oligomers and phosphorylation have been proposed as pathogenic. On the other hand, we identified that α Syn was localized in non-lipids rafts, differing from the proposed that α Syn it was localized in lipid rafts. In the same way we identified the α Syn ability to enter and exit the muscle in vivo, therefore, muscle could be a source of this protein in the brain. Although α Syn oligomers have been related to PD, it has not yet been elucidated how the formation of oligomers occurs in contact with membrane lipids. Due to this we decided to study the physicochemical and thermodynamic behavior of the recombinant human α Syn reconstituted in lipids isolated from old rats sarcolemma. For this purpose, a recombinant human α Syn (r- α Syn) has been purified using our system, this protein has been characterized through SDS-PAGE and Western blot proving the integrity and purity. In the same manner, we identified in the r- α Syn the serine 129 phosphorylation. We are currently working on characterization of r- α Syn interactions with isolated lipids from sarcolemma from old rat SM.

LONG AND VERY-LONG CHAIN CERAMIDES PROMOTES AN ANXIETY-LIKE BEHAVIOR IN FEMALE MICE THROUGH MICROGLIA ACTIVATION IN CORTEX

Sofía Bernal-Vega^{1,2}, Alberto Camacho-Morales^{1,2}

¹College of Medicine, Biochemistry and Molecular Medicine Department, Universidad Autónoma de Nuevo León, Monterrey, Mexico, ²Neurometabolism Unit, Center for Research and Development in Health Sciences, Universidad Autónoma de Nuevo León, Monterrey, Mexico.
E-mail: acm590@hotmail.com

Anxiety disorders are characterized by excessive and enduring fear, commonly associated with an exacerbated excitability state in the nervous system, affecting 4% of the global population. Risk factors encompass stress, genetics, substance use and fetal exposure to adverse factors. Neuroinflammation and sphingolipid metabolism alterations contribute to anxiety pathogenesis. Clinical and animal studies link plasma ceramides to anxiety symptoms, yet their exact molecular mechanisms remain unclear. Maternal exposure to high-energy-dense nutrients induces anxiety-like behavior and increases plasma levels of long-chain ceramides in offspring. Furthermore, ceramides can modulate proinflammatory pathways in microglia, suggesting a role in anxiety. This project aims to identify the effect of ceramides on the induction of anxiety-like behavior in mice through microglia activation. C57/BL6J 3-month-old male and female mice received intravenous ceramides (C16:0, C18:0, C22:0, C24:0, C24:1), and behavioral tests were conducted. Microglia cells were obtained from hippocampus, striatum, and cerebral cortex and phenotyped by flow cytometry. Phagocytic activity was determined in microglia immortalized cultures following ceramide stimulation by using fluorescent latex beads, cells were analyzed using a flow cytometer. For the elevated plus maze we found a significant increase in the time spent on the closed arms in female subjects administered with ceramides. In male mice, we did not observe a significant difference in any of the analyzed parameters. In the open field test, there were no significant differences in any of the assessed parameters for both female and male mice. For the dark/light box test we found a significant increased latency to first enter the light compartment in female mice administered with ceramide. In male subjects no statistically significant differences were detected. In the novelty suppressed feeding test, we found that female mice administered with ceramides experienced longer latency to reach the food pellet in the center of the arena. We did not observe statistically significant differences in the analyzed parameters for males. We analyzed if ceramides promote changes in microglia phenotype. Ceramides increased proinflammatory CD86⁺ cells in the cortex and decreased anti-inflammatory CD206⁺ cells across striatum, cortex and hippocampus. In vitro, ceramides enhanced microglial phagocytic activity. These findings suggest long and very long-chain ceramides induce anxiety-like behavior in female mice and promotes cortical microglial activation, potentially mediating anxiety.

IMPAIRED HIPPOCAMPAL NEUROGENESIS AND SPATIAL LEARNING DEFICIT IN ADULT WISTAR RATS WITH ACQUIRED HYPOTHYROIDISM

Pablo Edson Bustamante Nieves^{1,2}, Paola Gisell Guillén Herrera², Norma Osnaya Brizuela², Armando Valenzuela Peraza², Jorge Pacheco Rosado¹, Karla Berenice Sánchez Huerta²

¹Laboratorio de Neurofisiología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, Av. Wilfrido Massieu 399, Nueva Industrial Vallejo, Gustavo A. Madero, 07738 CDMX, ²Laboratorio de Neurociencias, Subdirección de Medicina Experimental, Instituto Nacional de Pediatría, Av. Insurgentes Sur 3700-Letra C, Insurgentes Cuicuilco, Coyoacán, 04530 CDMX
E-mail: ksanchezh@pediatria.gob.mx

Background. Acquired hypothyroidism (AH) is a deficiency of thyroid hormones that occurs during childhood and adulthood. Adult patients with AH exhibit memory deficits, lower intellectual quotient, and a higher prevalence of anxiety and depression¹. The cause of cognitive and mental alterations in patients with AH is unknown. However, studying the alterations in adult hippocampal neurogenesis in murine models could contribute to the understanding of these causes, as this biological process participates in the consolidation of cognitive functions and mood control in rodents. **Objective.** To evaluate the effect of AH on neurogenesis in the dentate gyrus (DG) of the hippocampus and its association with changes in learning and spatial memory in adult Wistar rats. **Methodology.** Male Wistar rats (aged 60 days) were divided into three experimental groups: control, hypothyroid (Hyp, treated with methimazole), and hypothyroid with hormonal replacement (Hyp + T₄, treated with methimazole and levothyroxine). Neurogenesis was assessed by quantifying the number of immature granular neurons (IGNs) positive for the doublecortin marker (DCX⁺). Learning and spatial memory were determined with the Morris water maze test. The association between neurogenesis and cognition was determined by quantifying the number of neuronal cells (c-Fos⁺) and IGNs (DCX⁺/c-Fos⁺) that were activated after learning-memory experience. **Results.** The Hyp group exhibited a lower number of IGNs and a delay in the acquisition of spatial learning, without alterations in memory. The learning-memory experience caused an increase in the activation of neuronal cells compared to the control group, but the IGNs did not participate in the activation response. The Hyp + T₄ group showed no changes in neurogenesis or neuronal activation post learning-memory, but presented a delay in the acquisition of spatial learning. **Conclusion.** AH leads to a reduction in hippocampal neurogenesis and hyperactivation of the DG, which was associated with a delay in the acquisition of spatial learning.

References

1. Chaker, L., Razvi, S., Bensenor, I.M. et al. Hypothyroidism. *Nat Rev Dis Primers* 8, 30 (2022).

EFFECTS OF ACETYL-L-CARNITINE AND L-CARNITINE ON THE DIFFERENTIATION EFFICIENCY AND THE MORPHOLOGY OF MOTOR NEURONS DERIVED FROM MOUSE EMBRYONIC STEM CELLS

Maricela Cabrera-Castro¹, Fernando Becerra-Velez¹, Iván Velasco-Velázquez^{1,2}

¹Instituto de Fisiología Celular–Neurociencias, Universidad Nacional Autónoma de México. Circuito Exterior S/N, Ciudad Universitaria, CDMX 04510, México. ²Laboratorio de Reprogramación Celular, Instituto Nacional de Neurología y Neurocirugía. Insurgentes sur 3877, CDMX 14269, México.
E-mail: maricela@ifc.unam.mx. Tel: +52 (55) 5556225649

Currently, there are various neuromuscular diseases that hinder proper control of the limbs, and result in a low quality of life for affected patients. One of the reasons behind these pathologies is the death of spinal motor neurons (MNs), that innervate skeletal muscle, sometimes caused by oxidative stress. In the absence of *in vivo* studies, *in vitro* models offer the opportunity to study the effect of molecules on reducing oxidative stress and promoting neuronal survival. While carnitines are primarily involved in lipid metabolism, by facilitating the transport of fatty acids into the mitochondria, there is also evidence suggesting that carnitines act as neuroprotective agents under conditions of mitochondrial dysfunction and oxidative stress, by functioning as antioxidants and mitochondrial stabilizers. Additionally, there are studies supporting their role as inhibitors of apoptosis, neuronal damage, neuroinflammation, and astrogliosis in the adult brain. Specifically, in primary MNs cultures, L-Carnitine (LCAR) and Acetyl-L-Carnitine (ALCAR) have been characterized as promoters of survival. In this work, we aim to adapt previously reported methodologies and assess the effects of administering these molecules at a concentration of 100 μ M for 7 days in cultures of MNs differentiated from mouse embryonic stem cells (mESCs), to determine changes in morphology and cell survival using markers that indicate MN fate, and imaging studies that allow observation of neuronal morphology, as well as the expression of transcripts related to carnitine metabolism, oxidative stress, and cell survival via RT-PCR. The preliminary results are intended to improve spinal MN differentiation protocols that include the use of ALCAR or LCAR *in vitro* for neuromuscular modelling, and in the future test possible therapeutic interventions, accelerating the development of effective treatments that might translate to the clinical setting.

Keywords: Neuromuscular diseases; Oxidative stress; Neuronal survival; Motor neurons.

Acknowledgements: This study was supported by PAPIIT IN219122. We thank the technical support of Dr. Itzel Escobedo-Avila.

EFFECT OF HIGH-CARBOHYDRATE DIET ON MITOPHAGY AND MITOCHONDRIAL DYNAMICS OF THE FRONTAL CORTEX OF MALE WISTAR RATS

Karen Carreto Meneses¹, Diana Moroni González¹, Samuel Treviño Mora^{1*}

¹Laboratorio de Investigaciones Químico Clínicas, Facultad de Ciencias Químicas, Benemérita Universidad Autónoma de Puebla. Av. San Claudio, Col. San Manuel, Puebla, Puebla, México. C.P. 72570.
E-mail: samuel_trevino@hotmail.com

Introduction. The current lifestyle promotes physical inactivity and consumption of high-calorie diets, resulting in the development of metabolic disorders such as metabolic syndrome (MS) and type 2 diabetes (DT2), characterized by the presence of insulin resistance (IR), increasingly linked to the progression of neurodegenerative diseases through mitochondrial dysfunction. To maintain healthy mitochondrial function, a balance between mitochondrial fission and fusion is necessary, and biogenesis and mitophagy are also key mechanisms for regulating mitochondrial mass and quality in response to cellular energy needs. **Methodology.** Two groups of male Wistar rats (n=6, 1-month-old, and weighing 100 g) were fed for 120 days with a high-carbohydrate diet (Patent: MX/E/2013/047377) or regular chow (NCD; LabDiet 5001; diet for laboratory rodents). At the end of feeding, metabolic characterization was performed. Glucose, triglycerides, insulin, cholesterol, and their fractions were quantified using commercial kits. HOMA-IR and Matsuda-DeFronzo insulin sensitivity indexes were calculated. From a frontal cortex sample, mitophagy and mitochondrial dynamics were analyzed by western blot. Protein samples were separated by SDS-PAGE and transferred to polyvinylidene difluoride (PVDF) membranes, which were blocked in 5% non-fat dry milk for 2 hours and incubated with primary antibodies at 4 °C overnight. The primary antibodies were as follows: Mfn1 (1:1000), Opa-1 (1:1000), Drp-1 (1:1000), Fis1 (1:1000), Pink1 (1:1000), Parkin (1:1000), β -actin (1:1000). Densitometry analysis of each band was quantified using ImageJ and normalized against those of β -actin protein in each sample. **Results.** Wistar rats fed high carbohydrate content developed MS characterized by hyperglycemia, hyperinsulinemia, dyslipidemia, and IR. Pink and Parkin expression are significantly decreased and the expression of proteins related to mitochondrial dynamics shows changes associated with the development of MS in rats. **Conclusion.** MS modified the mitophagy and mitochondrial dynamic in the frontal cortex of male Wistar rats.

LC NORADRENERGIC NEURONS ARE INVOLVED IN THE LACK HYPOTHALAMUS-PITUITARY-THYROID AXIS RESPONSE TO COLD IN STRESSED ANIMALS

Andrea Castillo-Campos¹, Patricia Joseph-Bravo¹, Fidelia Romero¹, Jean Louis Charli¹, Rosa María Uribe¹

¹Instituto de Biotecnología, UNAM, Av. Universidad 2001, col. Chamilpa 62210. Cuernavaca, Mor. México.

E-mail: rosa.uribe@ibt.unam.mx

Hypothalamus-Pituitary-Thyroid (HPT) axis plays an important role in maintaining energy homeostasis, its activity is regulated by energy demands such as cold exposure, exercise, and stress. Chronic psychological stress inhibits HPT axis activity, which impacts on efficient metabolic activity and promotes alterations such as depression and anxiety. In rats, we have shown that an acute increase in peripheral corticosterone level or chronic stress inhibits the appropriate response of the HPT axis to energy demands, as acute cold exposure^{1,2}. Noradrenergic (NA) activity controls the increase of Median Eminence (ME) TRH release and Paraventricular (PVN) TRH mRNA synthesis in response to cold. It has been proposed that chronic stress diminished PVN NA levels, so we will attempt to define the role of Locus Coeruleus (LC) noradrenergic neurons, in the lack of response of the HPT axis to cold exposure in chronically stressed male rats. Using a chronic restraint paradigm in adult male Wistar rats for 14 days compared to naïve animals, we found that chronic restraint caused hyperthermia as reported, and deficient body temperature control in response to cold exposure. Naïve and chronically stressed animals showed a strong response to cold exposure on serum corticosterone levels. As expected, cold exposure did not increase serum thyrotropin in chronic stress compared to naïve animals. In adenohypophysis, we found a significant increase in TRHR1 mRNA levels in the stressed group compared to the naïve group, supporting a decrease of basal ME TRH release by chronic stress. Also, we observed low levels of prolactin (PRL) mRNA in stressed compared to naïve, however, an increased PRL mRNA level in cold response only on stressed rats. As well, we detected an increase in TRHDE mRNA levels in response to cold exposure in naïve and stressed rats. In order to establish the role of LC-NA neurons on HPT cold stress response in chronically stressed rats, we propose quantifying the co-expression of cFos and DBH on LC. Preliminary results show that LC-NA neurons co-express cFos mRNA in naïve 1h cold exposure animals.

References

1. Sotelo-Rivera et al., (2014) J. Neuroendocrinol. Dec; 26(12):861-9
2. Castillo-Campos et al., (2021) J. Endocrinol Invest Apr,44(4):713-723

CO-INFECTION MODEL OF CYTOMEGALOVIRUS AND ZIKA VIRUS IN HUMAN NEURAL PROGENITOR CELLS

Edson I. Rubio-Hernández, Ana Sonia Silva-Ramírez, Ana Sofía García-García, Verónica García-Betancourt, Itzel Abigail Loredó-Alonso & Claudia Castillo

CIACYT-Facultad de Medicina, Universidad Autónoma de San Luis Potosí, San Luis Potosí, México.

Zika virus (ZIKV) is a single-stranded positive-sense RNA virus (ssRNA (+)) belonging to the genus *Flavivirus*. Bites from infected mosquitoes primarily transmit it. Between 2015 and 2016, there was a significant outbreak of ZIKV in Latin America and the Caribbean. During this outbreak, an unusual increase in cases of microcephaly in newborns was observed, whose mothers had been infected with ZIKV during pregnancy. On the other hand, Cytomegalovirus (CMV) is a double-stranded DNA virus (dsDNA) belonging to the *Herpesvirus* family. It is a common virus among people of all ages; however, the immune system of a healthy person usually prevents the virus from causing disease. Among the factors that might be associated with the generation of microcephaly are co-infections.

One possible explanation for why ZIKV and CMV co-infection could be more detrimental relates to the host's immune response. Therefore, the co-infection of ZIKV and CMV was studied in the hNS-1 cell line, which was used as a study model in the laboratory. These cells have been extensively studied in our lab, and the characteristics of the cells in the proliferation state and the populations resulting from differentiation are well known. When determining the metabolic activity of ZIKV and CMV mono-infections and ZIKV-CMV co-infections, using the Resazurin assay, it was found that at five days post-infection (dpi), the cells in proliferation conditions significantly decreased their metabolic activity compared to the control. However, at nine dpi, the ZIKV mono-infection and ZIKV-CMV co-infection showed decreased metabolic activity under both proliferation and differentiation conditions.

In contrast, the CMV mono-infection did not show changes in activity compared to the control. This suggests that the cells may have transitioned from a lytic infection to a latent phase. Furthermore, the total number of cells in ZIKV and CMV mono-infections and co-infections at 5 and 9 dpi under proliferation conditions and nine dpi under differentiation conditions was evaluated. In both cases, there was a lower number of cells than the control group, indicating a more significant reduction in the co-infection group. The obtained data are descriptive, as the experiment was conducted in duplicate. The results suggest that ZIKV and CMV co-infection could be related to the incidence of newborns with microcephaly and other alterations, as it was demonstrated that there is an impact on the proliferation of neural progenitor cells and, to a lesser extent, on cells in the differentiation state.

STUDY OF TIBOLONE ADMINISTRATION ON NADPH OXIDASE EXPRESSION IN A MURINE MODEL OF TRAUMATIC SPINAL CORD INJURY

Nadia Tzayaka Castillo Mendieta¹, Stephanie M Sánchez-Torres¹, Carlos Orozco-Barrios², Hermelinda Salgado Ceballos¹, Christian Guerra-Araiza³, Angélica Coyoy-Salgado²

¹Unidad de Investigación Médica en Enfermedades Neurológicas, Hospital de Especialidades, Centro Médico Nacional Siglo XXI, Instituto Mexicano del Seguro Social. ²CONAHCYT- Instituto Mexicano del Seguro Social, Unidad de Investigación Médica en Enfermedades Neurológicas, Hospital de Especialidades, Centro Médico Nacional Siglo XXI. ³Unidad de Investigación Médica en Farmacología, Hospital de Especialidades, Centro Médico Nacional Siglo XXI, Instituto Mexicano del Seguro Social. Zip code: 06720. E-mail: acoyoy@email.ifc.unam.mx. Phone: 55 5627 6900 Ext. 21367.

Traumatic spinal cord injury (LTME) is a disabling condition, it can have severe consequences for patients, including paraplegia or quadriplegia, and even lead to death. The incidence is estimated to be approximately 15 to 40 cases per million population worldwide. The pathophysiology of LTME can be categorized into primary and secondary injury. During secondary injury, cytokines are released and promote acute and chronic inflammation as a result of microglia activation, macrophage invasion and activation of the enzyme NADPH oxidase (NOX). The NOX enzyme is a primary source of reactive oxygen species (ROS), and there are several NOX homologs, including NOX2 and NOX4, which increase their expression in LTME. To date, there is no treatment that allows the complete recovery of lost functions after LTME, which makes necessary the development of new therapeutic strategies. In the present work we evaluated the effect of tibolone (TB), a synthetic hormone with neuroprotective properties, on the number of astrocytes, microglia and neurons; we also evaluated the effect of TB on the expression of NOX2 and NOX4 in astrocytes, microglia and neurons, in the spinal cord after 3 days of LTME. Methodology. Adult male rats of the Sprague Dawley strain, which underwent LTME at the level of the ninth thoracic vertebra (T9) by moderate-intensity contusion, were randomly divided into 3 groups (n=3): LTME and two groups with LTME plus TB administration (doses, 1 mg/kg and 2.5 mg/kg TB, respectively), treated daily. Three days after LTME the animals were perfused intracardially, the spinal cord was removed, histological sections were made, in which the presence of astrocytes was evaluated with GFAP, microglia with Iba1, neurons with tubulin-beta3 or NeuN, and the expression of NOX2 and NOX4, using the immunohistochemistry technique. Results. TB did not modify the number of astrocytes or microglia, compared to the LTME group; interestingly, the number of neurons is higher with TB treatment, with respect to the LTME group. Regarding the number of NOX2- and NOX4-positive cells, TB treatment decreased the number of NOX2-positive neurons and NOX4-positive astrocytes compared to the LTME group. Conclusion. TB has a neuroprotective effect, probably regulating NOX2 and NOX4, homologues of NADPH oxidase.

Funding: This research was funded by FORDECYT-PRONACES (grant number 845110). Acknowledgments: Tzayaka Castillo-Mendieta would like to thank the Post-Doctoral Fellowship Program of CONAHCYT.

INDUCTION, PHOSPHORYLATION AND PROTEIN INTERACTIONS OF PEBP1 UNDER EARLY FOCAL CEREBRAL ISCHEMIA/REPERFUSION IN THE RAT HIPPOCAMPUS

Febe Elena Cazares Raga¹, Daniel Corzo-Toledo¹, J. Ángel Rubio-Miranda¹, Erick R. Corzo-Toledo¹, Leticia Cortés Martínez¹, Hugo Sánchez-Hernández², Alma Ortiz-Plata³, Fidel de la Cruz Hernández-Hernández¹

¹D. Infectómica y Patogénesis Molecular, Cinvestav; ²Universidad La Salle Cancún; ³Neuropatología Experimental, Instituto Nacional de Neurología y Neurocirugía. fcazares@cinvestav.mx

Cerebral ischemia (CI) affects all ages, mainly the elderly population, being a leading cause of mortality and morbidity. Focal cerebral ischemia (FCI) is the sudden loss of blood flow in a specific area of the brain and, reperfusion (R) is a secondary brain process in which the blood supply is restored. Phosphatidylethanolamine-binding-protein-1 (PEBP1), a scaffold protein inhibitor of protein kinase regulating several cell signaling pathways through phosphorylation at Ser-153 and multiple protein partners. PEBP1 participates in several neurodegenerative process including CI. Under oxidative stress generated during CI, PEBP1 participates in the regulation of ferroptosis, interacting with GPX4 and 15 lipoxygenase forming 15LOX/PEBP1 complex, whereby it is considered that PEBP1 has neuroprotective functions, which has not been enough characterized. Using the Middle Cerebral Artery Occlusion/Reperfusion (MCAO/R) model in adult Wistar male rats during 30, 60 and 90 min of ischemia/reperfusion (I/R). PEBP1 was analyzed in hippocampus under I/R by 2DE and SDS-PAGE and WB and, IH using antibodies α -PEBP1 and α -pPEBP1-S153. To identify PEBP1 interaction with other proteins, at 60 min FCI co-immunoprecipitation (co-IPP) and Mass Spectrometry (MS) were used. In addition, PEBP1 interaction with antioxidant proteins were searched using STRING interactome database and molecular docking with candidates was made. The results showed that PEBP1 expression and phosphorylation increased at 60 min I/R and diminished after 90 min; PEBP1 was detected as a couple (37 and 25 kDa) by WB. In hippocampus tissue an increase of PEBP1 was also observed by IH. Using Co-IPP and MS, interaction of PEBP1 with dynein and desmin was detected. PEBP1 interaction with catalase, GPX4 and SOD3 was detected and supported by interactomes and molecular docking. During FCI, PEBP1 increased and form complex with Catalase, SOD3, GPX4 and possibly modulating antioxidant response. Interaction of PEBP1 with dynein and desmin suggests its participation in the intraneuronal protein mobilization.

EFFECTS OF 2-APB ON DOPAMINERGIC PATHWAYS

Rebeca Chávez¹, Carlos Ángel Guzmán¹, Elizabeth Estevez¹, Alberto Alatorre¹,
Marvin Antonio Soriano¹, Eunice Dalet Farfan¹, Enrique Querejeta¹

¹Av. Plan de San Luis y Salvador Díaz Mirón, Col. Casco de Santo Tomas, 11340 Miguel Hidalgo, Mexico
E-mail: chavez.campos.rebeca@gmail.com

2-Aminodiphenyl boronic acid (2-APB) is a compound known to regulate calcium channels in various cells, including neurons, with effects that vary depending on the dosage and can lead to cytosolic toxicity. This study focuses on investigating the effects of 2-APB on dopaminergic pathways related to motor control, observing motor and neurological alterations in animals treated with the compound. The results reveal that 2-APB induces neurotoxicity, especially at doses of 100 mM, leading to changes in behaviour and an increase in neuronal death. Additionally, it is highlighted that the co-administration of 2-APB and 6-OH (6-Hydroxydopamine) exacerbates the lesion and its manifestations.

Studying the neurotoxic effects of 2-APB is essential for understanding and addressing neurological diseases such as Parkinson's, which potentially can reduce its incidence and severity. These investigations can also improve the quality of life of affected individuals by enabling more precise therapeutic interventions. Furthermore, the dissemination of these findings can increase public awareness of the risks associated with certain chemicals, promoting measures to avoid unnecessary exposures, and advocating for stricter regulations.

In terms of its importance in research, 2-APB is crucial in neuroscience for studying intracellular signalling and the function of calcium channels, as well as in disease models for investigating neurological disorders such as Parkinson's disease. Additionally, its use in pharmacology allows for evaluating the efficacy of compounds as well as in models of neurological disorders.

TIBOLONE IMPROVES MOTOR RECOVERY, REGULATES NEUROINFLAMMATION AND GLIOSIS IN A MODEL OF TRAUMATIC SPINAL CORD INJURY

Angélica Coyoy-Salgado*, Stephanie M Sánchez-Torres, Carlos Orozco-Barrios, Luis Ángel León-Cholula, Hermelinda Salgado Ceballos, Isabel Arrieta-Cruz, Christian Guerra-Araiza, Julio Morán*
CONAHCYT-Unidad de Investigación Médica en Enfermedades Neurológicas, Hospital de Especialidades, Centro Médico Nacional Siglo XXI, Instituto Mexicano del Seguro Social. Av. Cuauhtémoc 330, Doctores, Cuauhtémoc, 06720 Ciudad de México, CDMX.
E-mail: acoyoys@gmail.com

Spinal cord injury (SCI) is a destructive neurological and pathological state that causes major motor, sensory and autonomic dysfunctions. The pathophysiology of SCI can be divided in a primary and secondary phase; inflammation contributes to the secondary injury through the non-specific activation of the innate immune response, which activates immediately after the stimulation by the SCI. Glial scar formation (gliosis) is a reactive cellular mechanism that is facilitated by astrocytes and occurs during the chronic secondary phase of SCI.

It is important to develop a treatment for SCI, the use of synthetic steroids, such as tibolone, has been proposed, since it exerts the neuroprotective effects of sexual hormones, but does not promote the development of cancer. Some of the neuroprotective effects of steroids occur through the regulation of pro inflammatory and anti-inflammatory cytokines, and gliosis regulation. Thus, in this work we studied the effect of tibolone in the regulation of neuroinflammation, gliosis and locomotor functional recovery after SCI in a biological model.

Male Sprague Dawley rats were used, on which SCI was performed at thoracic vertebra 9, TIB was orally administered daily at doses of 1 and 2.5 mg per kg of body weight. The concentration of pro- and anti-inflammatory cytokines in the injured site of the spinal cord was quantified using MILLIPLEX kits, to study the effect of TIB on gliosis, immunochemistry assay was performed, and motor recovery was determined using the Basso, Beattie, and Bresnahan (BBB) scale. The results obtained show that tibolone improves locomotor function by modulating neuroinflammation and gliosis in a rat model of SCI. In conclusion, TIB treatment immediately after SCI reduced structural damage to nerve tissue and effectively preserved tissue around the injury site modulating neuroinflammation and gliosis in this model of SCI and importantly TIB could be a therapeutic alternative for the recovery of motor function after SCI.

This research was funded by FORDECYT-PRONACES (Project 845110).

CEPHALIC ALTERATIONS PRODUCED BY CONDITIONAL INACTIVATION OF *PLPP3* IN THE *WNT1* EXPRESSION DOMAINS

Sheyla Cruz-Cruz, Angelica Piedra-Ramírez, Erick Antonio Rojas-Pacheco, Alejandro Elliot Flores-Oliva, Valeria Martínez-Silva, Monserrat Llaguno-Munive, Diana Escalante-Alcalde

¹Instituto de Fisiología Celular, UNAM
E-mail: descalan@ifc.unam.mx

Phospholipid phosphatases (PLPPs) are a group of integral membrane enzymes that, depending on the cell type, are located in plasma membrane, Golgi apparatus, endoplasmic reticulum and endosomes. They catalyze the dephosphorylation of phosphate esters of a wide variety of phospholipids. Among their substrates are bioactive lipids such as sphingosine-1-phosphate (S1P) and lysophosphatidate (LPA)⁴. These lipids activate multiple intracellular signaling pathways through the activation of specific G-protein couple receptors⁸.

Our laboratory studies *Plpp3*, which is essential during embryonic development, and its absence causes early embryonic lethality around E9.5, with a diversity of phenotypes such as delayed development, abnormalities in the vascularization of the yolk sac, defects in vascular and cardiac development, among others².

Our research group has obtained data linking *Plpp3* to neural crest cells (NCC) development. The experiments conducted suggest that *Plpp3* deficiency could alter NCC migration^{7,9}. Furthermore, other group, using single-cell RNA analyses, detected the expression of *Plpp3* and some bioactive lipid receptors in NCC during embryonic development (E8.25-E8.5)⁶ and in many of its derivatives³. This suggested that PLPP3 could be relevant for NCC development. Using the *Wnt1::Cre2* mouse line¹ to inactivate *Plpp3* in the expression domains of *Wnt1* (i.e. premigratory neural crest cells, telencephalon and in the region of the midbrain organizing center), dysmorphology of the head has been observed in embryos and postnatal individuals represented by: reduction in the size of the telencephalon; reduction in the distance between the eye and the frontonasal process; reduction in the size of the frontal bone and the central anterior region of the interparietal bone, which are structures derived from the neural crest^{5,9}. These observations suggested defects in the migration, survival and/or differentiation of NCC and/or their derivatives caused by alterations in the signaling of *Plpp3* substrates.

We hypothesize that the conditional inactivation of *Plpp3* in cranial NCC causes alterations in their migration and in the ossification dynamics of the group of cells that give rise to the frontal bone. To establish the cellular and/or molecular mechanisms responsible of this phenotype, lineage tracing experiments, analysis of the ossification domains during development, the dynamic of ossification measured by the expression of molecular markers and bone deposition by Micro-CT were performed and preliminary findings will be discussed.

References

- Dinsmore, C., Ke, C. & Soriano, P. (2022). *Genesis (New York, N.Y.: 2000)*, 60(3). <https://doi.org/10.1002/dvg.23468>
- Escalante-Alcalde, D., Hernandez, L., Le Stunff, H., Maeda, R., Lee, H., Jr-Gang-Cheng, Sciorra, V., Daar, I., Spiegel, S., Morris, A. & Stewart, C. (2003). *Development (Cambridge, England)*, 130(19): 4623–4637. <https://doi.org/10.1242/dev.00635>
- Escalante-Alcalde, D., Morales, S. & Stewart, C. (2009). *Int J Dev Biol*, 53(1):139-47. Doi: 10.1387/ijdb.082745de.
- Kok, B., Venkatraman, G., Capatos, D. & Brindley, D. (2012). *Chemical reviews*, 112(10): 5121–5146. <https://doi.org/10.1021/cr200433m>
- Piedra, A. (datos no publicados) Caracterización del fenotipo originado por la deficiencia de *Plpp3* en la cresta neural cefálica del ratón. Tesis de Maestría
- Pijuan-Sala, B., Griffiths, J., Guibentif, C., Hiscock, T., Jawaid, W., Calero-Nieto, F., Mulas, C., Ibarra-Soria, X., Tyser, R., Lian Ho, D. Wolf Reik, W., Srinivas, S., Simons, B., Nichols, J., Marioni, J. & Berthold Göttgens, B. (2019). *Nature* 566, 490–495. <https://doi.org/10.1038/s41586-019-0933-9>
- Ramírez-Corona, A. (2018). El papel de la fosfatasa de fosfolípidos 3 en el desarrollo del cerebro medio y posterior. Tesis de Licenciatura, Universidad Nacional Autónoma de México. <http://132.248.9.195/ptd2018/junio/0775641/Index.html>
- Tang, X. & Brindley, D. (2020). *Biomolecules*, 10(9): 1263. <https://doi.org/10.3390/biom10091263>
- Vázquez-Díaz, J (datos no publicados) Análisis de la participación de la fosfatasa de fosfolípidos-3 (*Plpp3*) en los dominios de expresión de *Wnt1* durante el desarrollo embrionario. Tesis de Maestría.

MATERNAL DIABETES MODIFIES THE TRANSCRIPTION OF GENES IMPORTANT FOR THE DIVISION OF CORTICAL NEURAL STEM CELLS: ROLE OF FOXP2

Diana Sarahi De la Merced-García^{1,2}, Rocío Valle-Bautista¹,
Néstor Fabián Díaz-Martínez¹ and Anayansi Molina-Hernández¹

¹Instituto Nacional de Perinatología "Isidro Espinoza de los Reyes", Lomas de Chapultepec IV Secc, Miguel Hidalgo, Ciudad de México, México. ²Doctorado en Ciencias Biomédicas, UNAM, Ciudad Universitaria, Coyoacán, Ciudad de México, México
E-mail: anayansimolina@gmail.com

Maternal diabetes has been related to neurodevelopmental alterations in children, mainly affecting cognitive, motor, and verbal abilities, functions that depend on the structural integrity of the cerebral cortex¹. The development of this structure depends on changes in the pattern of neural stem cell (NSC) proliferation to generate specialized cell types at specific periods of embryo development. Within the complex molecular network that guides gene expression during cortical development, the transcription factor FOXP2, through the activation and repression of its transcriptional targets, participates in the transition from NSC to intermediate progenitors² for the acquisition of neuronal commitment to deep-layer cortical phenotype and the formation of neuronal circuits important for movement and speech. However, the mechanisms by which FOXP2 exerts regulatory control over its transcriptional targets during corticogenesis have not been elucidated. On the other hand, in murine models of maternal diabetes, increased early neurogenesis, decreased NSC proliferation, and increased FOXP2 nuclear translocation are observed in the cortical neuroepithelium, suggesting an important role of FOXP2 in these processes^{3,4}. Hence, in an attempt to identify the early mechanism involved in the altered cortical neurogenesis, cytoarchitecture, and function in embryos and 21-day-old (P21) rats previously reported⁵, the aim of this work is to determine changes in FoxP2 transcriptional targets in the cortical neuroepithelium of 12-day-old embryos (E12) exposed to high glucose compared to control embryos. From an RNA-seq analysis of the cortical neuroepithelium of E12 embryos from diabetic rats, we identified 23 genes differentially expressed in the diabetic group that have been reported as transcriptional targets of FOXP2. Here, through RT-qPCR we were able to validate 12 FOXP2 targets differentially expressed in the cortical neuroepithelium. Interestingly, these targets are involved in cell cycle processes such as mitotic spindle elongation, microtubule organization, and depolymerization. The results obtained so far contribute to understanding the mechanisms that underline intellectual, motor, and language deficits related to FOXP2 and that have been reported in the children of diabetic mothers. The above is supported by changes in ultrasonic vocalization in P21 male and female offspring from diabetic rats (unpublish data). This work was supported by INPer 2019-1-11.

References

1. Rodolaki, K., Pergialiotis, V., Iakovidou, et al (2023). *Frontiers in endocrinology*, 14, 1125628.
2. Tsui, D., Vessey, J., Tomita, H. et al (2013). *The Journal of Neuroscience*, 33, 244-258.
3. De la Merced-García, D. S., Sánchez-Barrera, Á., Hernández-Yonca, J. et al (2023). *Cells*, 12, 510.
4. Solís, K. H., Méndez, L.I., García-López, G. et al (2017). *Frontiers in Neuroscience*. 11,1-14.
5. Valle-Bautista, R., Márquez-Valadez, B., Fragoso-Cabrera, A. D. et al (2020). *Frontiers in cell and developmental biology*, 8, 564561.

NEW ANTIEPILEPTIC TARGETS MODIFIED BY LEVETIRACETAM TREATMENT IN DENTATE GYRUS OF RATS WITH TEMPORAL LOBE EPILEPSY

Veronica Díaz-Villegas^{1,4,3}, Luz Adriana Pichardo-Macías¹, Sergio Juárez-Méndez², Iván Ignacio-Mejía³, Noemí Cárdenas-Rodríguez⁴, Marco Antonio Vargas-Hernández⁵, Mercedes Edna García Cruz⁴, Julieta Griselda Mendoza-Torreblanca^{4,*} and Sergio R. Zamudio^{1,*}

¹Departamento de Fisiología, Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Mexico City 07738, Mexico; vdiazv1000@alumno.ipn.mx; lpichardom@ipn.mx; ²Laboratorio de Oncología experimental, Instituto Nacional de Pediatría, Secretaría de Salud, Mexico City 04530, Mexico; ser.mend@gmail.com; ³Laboratorio de Medicina Traslacional, Escuela Militar de Graduados de Sanidad, UDEFA, Mexico City 11200, Mexico; ivanignacio402@gmail.com.mx; ⁴Laboratorio de Neurociencias, Subdirección de Medicina Experimental, Instituto Nacional de Pediatría, Mexico City 04530, Mexico; noemicr2001@yahoo.com.mx; ⁵Subdirección de Investigación, Escuela Militar de Graduados de Sanidad, UDEFA, Mexico City 11200, Mexico; mavh78@yahoo.com.mx

Temporal lobe epilepsy (TLE) is one of the most frequent types of focal epilepsy. Levetiracetam (LEV) is an antiepileptic medication whose mechanism of action at the genetic level has not been entirely elucidated during chronic epilepsy. The lack of change expression of the gene encoding the SV2A protein in TLE rats indicates that new molecular mechanisms could be involved in the effectiveness of LEV and suggests novel targets. Therefore, the aim of the present study was to evaluate the outstanding gene expression changes in the dentate gyrus (DG) of LEV-treated rats with pilocarpine-induced TLE. Whole-transcriptome microarrays were used to find the differential genetic profiles of control (CTRL), epileptic (EPI), and EPI rats treated for one week with LEV (EPI + LEV). Quantitative RT-qPCR was utilized to evaluate the RNA levels of the genes of interest. During treatment (week eight), SRSs were absent in the EPI + LEV group; in contrast, the rats in the EPI group continued to present SRSs. Subsequent using a heatmap, the gene expression profiles that changed according to epileptic condition were showed several opposite green and red areas respect to those in the EPI + LEV conditions. The EPI vs. CTRL analysis showed that, 685 genes were differentially expressed. In turn, in the analysis of the EPI + LEV vs. EPI groups, 675 genes were differentially expressed, 477 of which were downregulated and 198 of which were upregulated; in contrast, genes involved in GO categories such as Growth Factors, Angiogenesis, LTP, MAPK Cascade and Gene Silencing were down-regulated. A total of 94 genes whose expression was altered by epilepsy and modified by LEV were identified. The RT-qPCR confirmed that LEV treatment reversed the increased expression of Hgf mRNA and decreased the expression of the Efcab1, Adam8, Slc24a1, and Serpinb1a genes in the DG. These results suggest that LEV could be involved in nonclassical mechanisms involved in Ca²⁺ homeostasis and the regulation of the mTOR pathway through Efcab1, Hgf, SLC24a1, Adam8, and Serpinb1a, contributing to reduced hyperexcitability in TLE rats.

MODULATION OF GLUTAMATE UPTAKE BY THE SUBACUTE ACTIVATION OF THE HISTAMINE H₃ RECEPTOR IN RAT CEREBRO-CORTICAL ASTROCYTES IN PRIMARY CULTURE

Yrving Daniel Díaz-De Lucio¹, José Antonio Arias-Montaño¹

Center for Research and Advanced Studies (Cinvestav) of the National Polytechnic Institute
Av. IPN, 2508, 07360 Mexico City, Mexico.
E-mail: yrving.diaz@cinvestav.mx

Glutamate, as the main excitatory neurotransmitter in the central nervous system (CNS), plays a critical role in various physiological functions, including neurotransmission and synaptic plasticity. However, dysregulation of the glutamatergic transmission leads to excitotoxicity and neurodegeneration¹. Therefore, after glutamate is released neurotransmitter levels should be restored to low nanomolar concentrations, a function performed by transporters located on glutamatergic nerve terminals and astrocytes.

Histamine regulates CNS function via the activation of G protein-coupled receptors (H₁, H₂ and H₃)². A high density of histamine H₃ receptors (H₃Rs) is found in several CNS regions, including the cerebral cortex, and H₃Rs inhibit glutamate release from nerve terminals³. H₃Rs are also expressed by astrocytes but the information on their function in these cells is still scarce. In this work we studied the effect of the acute (15 min) or subacute (1-4 days) activation of H₃Rs on glutamate uptake by rat cerebro-cortical astrocytes in primary culture.

The sodium-dependent uptake by astrocytes of a mixture of glutamate (5 μM)/[³H]-glutamate (250 nM) was potently inhibited (IC₅₀ 21 nM) by the inhibitor TFB-TBOA, but modestly by dihydrokainic acid (DHKA), indicating that neurotransmitter uptake is mediated mainly by the excitatory amino acid transporter 1 (EAAT1), unlike the adult CNS where EAAT2 is mostly involved. Acute (15 min) H₃R activation with the selective agonist immepip (100 nM) did not modify glutamate uptake, but the subacute activation (1, 2 or 3 days) significantly increased uptake to 139.6 ± 5.1, 133.7 ± 4.7 and 127.0 ± 5.7 % of control values, respectively. The effect of immepip (1 day) was prevented by the selective H₃R antagonist iodophenpropit (1 μM).

These results indicate that via H₃R activation histamine regulates the expression of glutamate transporters, and the cellular mechanisms are currently being studied.

References

1. Martínez-Lozada Z, Ortega A (2023) *Journal of Neurochemistry*, 165(4), 457-466.
2. Nieto-Alamilla G et al., (2016) *Molecular Pharmacology*, 90 (5), 649-673.
3. Molina-Hernández A et al. (2001) *Neuropharmacology*, 41(8), 928-934.

COGNITIVE STIMULATION REDUCES NEUROINFLAMMATION AND INCREASES KLOTHO LEVELS IN THE HIPPOCAMPUS OF MOUSE ALZHEIMER'S DISEASE MODEL

Gustavo J. Fabian- Rodríguez¹, David F. Aguilar-Ávila¹, Héctor E. López-Valdés¹, Isabel Arrieta-Cruz², Mónica Torres-Ramos³, Brian I. Morales-López, Raúl Librado-Osorio² and Hilda Martínez-Coria^{1*}

¹Departamento de Fisiología, Facultad de Medicina, UNAM, CDMX. ²Departamento de Investigación Básica, INGER, CDMX. ³Ciencia Traslacional. Centro de Investigación sobre el Envejecimiento del CIVESTAV -Instituto Nacional de Neurología y Neurocirugía Manuel Velasco Suárez.

*hildamcoria@gmail.com

Klotho reduction is implicated in the activation of the brain's innate immune cells, and neuroinflammation has been associated with cognitive decline processes leading to neurodegenerative diseases. Furthermore, cognitive stimulation improves cognitive decline and delays the pathogenesis of AD. We previously showed that cognitive stimulation decreases the Iba-1 signal in the brain and cognitive decline in 3xTg-AD mice. We sought to investigate the effects of cognitive stimulation by Enrichment Environmental (EE) on microglia immunophenotype and klotho levels.

Males and females 12-month-old 3xTg-AD were housed in EE that changed every 7 days and trained for 5 days every month for 3 months. At the end of the stimulation program, they were tested in a Barnes Maze paradigm and compared with 3xTg-AD of the same age and sex without stimulation. After the memory test, we obtained the brain, and thirty-micrometer frozen sections were prepared as described previously and used Iba-1, TNF α , and klotho antibodies to identify inflammation and klotho levels in different areas.

Our results showed that cognitive stimulation improved memory, decreased inflammation, and increased klotho levels in 3xTg-AD mice.

ANTIOXIDANT EFFECT OF *PSACALIUM DECOMPOSITUM* IN AN ALZHEIMER'S DISEASE MOUSE MODEL

Paulina Flores Galicia¹, Raúl Librado-Osorio¹, Hilda Martínez-Coria², Héctor E. López-Valdés², Ricardo Reyes-Chilpa³, Jorge Iván Castillo-Arellano³, Armando Luna-López¹, Isabel Arrieta-Cruz^{1*}

¹Departamento de Investigación Básica, Instituto Nacional de Geriátria, CDMX. ²Departamento de Fisiología, Facultad de Medicina, UNAM, CDMX. ³Laboratorio de productos Naturales, Instituto de Química, UNAM, CDMX.

*arrieta777@mail.com, Telephone 5549504345

Redox reactions, involving both reduction and oxidation of relevant biomolecules, are important for physiological functions, including ATP generation, cell signaling, and antioxidant defense. Conversely, their perturbations have been implicated in pathological events such as oxidative stress or mitochondrial dysfunction. In advanced aging, altered redox homeostasis participates in the development of Alzheimer's disease (AD) by promoting A β deposition, tau hyperphosphorylation, and the subsequent loss of synapses and neuronal damage.

On the other hand, *Psacalium decompositum* (Pd) is one of many Mexican plant species employed mainly for their hypoglycemic properties. Our group has previously shown that the extract of this plant prevents cognition decline. Most of the identified compounds from their roots are sesquiterpenes, such as cacalol and cacalone. This study analyzed the effect of the hexane extract, which is rich in sesquiterpenes, on the antioxidant effect in an AD mouse model fed with a high-fat diet.

We used 9-month-old 3xTg-AD, which were divided into 4 groups: 1) Normal diet (ND), 2) Normal diet plus extract (ND+E), 3) High-fat diet (FD) with 34% of fat, and 4) High-fat diet plus extract (FD+E). The treatment with the hexane extract of Pd was given for one month after 2 months on a high-fat diet. Oxidative damage was determined in the brain cortex, hippocampus, and liver. The concentration of reduced and oxidized glutathione (GSH and GSSG) was analyzed by high-performance liquid chromatography (HPLC) as previously described.

Our data showed that the high-fat diet increased oxidative stress, while the Pd hexane extract significantly decreased oxidized glutathione and increased reduced glutathione.

STUDY OF MORPHOLOGICAL ALTERATIONS CAUSED BY *PLPP3* DEFICIENCY IN THE CEPHALIC NEURAL CREST OF MOUSE EMBRYOS

Flores-Oliva, Alejandro Elliot; Sheyla Cruz-Cruz; Martínez-Silva, Ana Valeria; Escalante-Alcalde, Diana

Instituto de Fisiología Celular, División de Neurociencias (UNAM), Ciudad de México, C.P.04510
descalan@ifc.unam.mx

The Phospholipid phosphatases constitute a group of integral membrane enzymes able to dephosphorylate bioactive lipids such as sphingosine-1-phosphate (S1P) and lysophosphatidic acid (LPA)¹. Within this family, we find the phospholipid phosphatase 3 (*Plpp3*), which stands out as the only essential during development, as its deletion causes embryonic lethality around E9.5 in mice. The causes of this lethality are associated with a series of defects, including abnormal vascularization, alterations in the allantois, defects in the establishment of the body axis, and closure of the neural tube².

Research from our group has suggested that the behavior of neural crest cells (NCC), a multipotent cell population exclusive to vertebrates, may be affected when *Plpp3* is deleted throughout the embryo. In this model, an expansion in domains of a premigratory NCC marker, *Wnt1*, was observed. Likewise, immunofluorescence against caspase 3 in mutant embryos showed that cell populations undergo an abnormal cell death process in regions through which NCC migrate³. Other assays revealed defects in the migratory capacity of NCC when explants of the neural tube were cultured *in vitro*. Additionally, single-cell transcriptomic analyses conducted by external groups have allowed us to confirm that, in normal development, *Plpp3* as well as bioactive phospholipid receptors are expressed in this population⁴. Therefore, we have proceeded to use scanning electron microscopy techniques, which have allowed us to discern in great detail the structural differences in the areas where NCC emerge and migrate.

These results prompted us to use the *Cre/loxP* system to selectively delete *Plpp3* in premigratory NCC. For this purpose, we acquired the *Wnt1::Cre2⁵* transgenic mouse line and crossed them with our conditional *Plpp3^{ff}* mouse line⁶. Directed deletion of *Plpp3* in premigratory NCC produces offspring with cardiac and craniofacial abnormalities, with a considerable percentage of postnatal lethality in the early days. Furthermore, using the reporter lineage tracing allele *ROSA26^{mTmg/mTmg}*, we found that at E11.5, NCC in the cephalic region exhibit abnormal cellular behavior in embryos lacking *Plpp3*. We have set out to morphologically characterize this population in this model, through scanning electron microscopy techniques. Our results suggest that *Plpp3* is a regulator of NCC migratory behavior. Through cryofractures⁷, we aimed to study whether *Plpp3* influenced the degrees of compaction or collective migration of branchial arches' NCC-derived cells.

Reference

1. Kok, B. P., Venkatraman, G., Capatos, D., & Brindley, D. N. (2012). Unlike two peas in a pod: lipid phosphate phosphatases and phosphatidate phosphatases. *Chemical reviews*, 112(10), 5121–5146. <https://doi.org/10.1021/cr200433m>

2. Escalante-Alcalde, D., Hernandez, L., Le Stunff, H., Maeda, R., Lee, H. S., Jr-Gang-Cheng, Sciorra, V. A., Daar, I., Spiegel, S., Morris, A. J., & Stewart, C. L. (2003). The lipid phosphatase LPP3 regulates extra-embryonic vasculogenesis and axis patterning. *Development (Cambridge, England)*, 130(19), 4623–4637. <https://doi.org/10.1242/dev.00635>
3. Ramírez-Corona, A. (2018). El papel de la fosfatasa de fosfolípidos 3 en el desarrollo del cerebro medio y posterior [Tesis de Licenciatura, Universidad Nacional Autónoma de México]. <http://132.248.9.195/ptd2018/junio/0775641/Index.html>
4. Pijuan-Sala, B., Griffiths, J.A., Guibentif, C. et al. A single-cell molecular map of mouse gastrulation and early organogenesis. *Nature* 566, 490–495 (2019). <https://doi.org/10.1038/s41586-019-0933-9>
5. Dinsmore, C. J., Ke, C. Y., & Soriano, P. (2022). The Wnt1-Cre2 transgene is active in the male germline. *Genesis (New York, N.Y. : 2000)*, 60(3), e23468. <https://doi.org/10.1002/dvg.23468>
6. Escalante-Alcalde, D., Sánchez-Sánchez, R., & Stewart, C. L. (2007). Generation of a conditional Ppap2b/Lpp3 null allele. *Genesis (New York, N.Y. : 2000)*, 45(7), 465–469. <https://doi.org/10.1002/dvg.20314>
7. Paganos, P., Caccavale, F., Cocurullo, M., D'Aniello, E., Arnone, M., Benvenuto, G. (2023). Whole animal freeze-fracture scanning electron microscopy: an easy-to-use method to investigate cell type morphology of marine embryos and larvae. *Frontiers in Ecology and Evolution*. 11. 10.3389/fevo.2023.1146749.

EFFECT OF SULFORAPHANE OR DASATINIB + QUERCETIN ON COGNITIVE IMPAIRMENT AND NEUROINFLAMMATION IN AN EXPERIMENTAL MODEL OF CHRONIC OBESITY IN MIDDLE-AGE FEMALE WISTAR RATS

Rosa Pamela Flores-Torres*; Marisol De la Vega-Tinajar; Verónica Salas-Venegas; Roberto Santín-Márquez; Jessica Aviléz-Aviléz; Fernanda Medina-Flores; Anahí Chavarría-Krauser; Beatriz Gómez-González, Mina Königsberg

Universidad Autónoma Metropolitana, Unidad Iztapalapa

*Av. San Rafael Atlixco 186, Leyes de Reforma 1ra Secc, Iztapalapa, 09340, Ciudad de México.

Contact: 5536620683. pamefltorres@gmail.com

Obesity is a worldwide health problem. In Mexico, the highest prevalence is observed at 59 years of age, particularly in women. This disease induces neuroinflammation and accumulation of senescent cells in the brain, processes that have been related to cognitive deficit during aging. Senolytics are molecules that eliminate senescent cells (such as dasatinib+quercetin, D+Q) and senomorphics are molecules that modulate SASP (such as sulforaphane, SFN). The effect of these molecules on neuroinflammation and cognitive decline related to chronic obesity in females is unknown. We fed female Wistar rats with a hypercaloric diet (HD) from weaning until middle age (14 months). SFN (0.5 mg/kg, subcutaneously 5 days a week) and D+Q (5 mg/kg and 50 mg/kg respectively, by nasogastric tube once a month) were administered from 12 to 14 months of age. Memory and learning at 5, 10, and 14 months of age were evaluated using the Novel Object Recognition test (NOR) and the Barnes maze test. The expression of inflammation markers in cerebral cortex, hippocampus, and serum was determined by ELISA assay. HD-fed rats showed mild cognitive impairment from 10 months on, which became evident at 14 months of age compared to the group fed with a standard diet (SD). Senolytic treatment did not prevent the cognitive deficit, while SFN improved the performance in both tests. The cognitive deficit in HD rats was associated with inflammation, which were reversed with senotherapy, with sulforaphane being more effective. In conclusion, chronic obesity is associated with cognitive deficit and neuroinflammation in middle-aged female rats. These effects could be prevented by modulating the SASP with the senomorphic, but not by eliminating senescent cells with the senolytics.

We thank Dr. María de los Ángeles Guerrero-Aguilera from UAM-I for providing the necessary animals needed for this project. This work was supported by FORDECYT-PRONACES/263957/2020. FTRP, RSM, SVV, SMR, AAJ and MFF are CONAHCyT scholarship holders.

MAQUI BERRY DECREASES OXIDATIVE STRESS AND INFLAMMATION IN THE HIPPOCAMPUS OF OZONE-EXPOSED RATS

Christian Guerra-Araiza¹, Carlos Orozco-Barríos², Emily L. Castillo-García¹, Angélica Coyoy-Salgado², Cristina García-Viguera³, Diego A. Moreno³

¹Unidad de Investigación Médica en Farmacología, Centro Médico Nacional SXXI, IMSS.

²CONAHCYT-Unidad de Investigación Médica en Enfermedades Neurológicas, Centro Médico Nacional SXXI, IMSS.

³CEBAS-CSIC, Food Science and Technology Department, Phytochemistry and Healthy Foods Lab, Campus Universitario de Espinardo – 25E-30100, Espinardo, Murcia, Spain.

*Address: Av. Cuauhtémoc 330, Colonia Doctores, Delegación Cuauhtémoc; C.P. 06720. Mexico City, Mexico. Tel.: +52 55 56 27 69 00 ext. 21367. E-mail: christianguerre2001@gmail.com

As ozone is one of the main pollutants worldwide, different studies indicate that prolonged exposure can produce a state of oxidative stress, which in turn causes alterations in the dynamics of the brain and affects processes such as memory and learning. Moreover, different investigations have shown that berries, especially those with a high flavonoid content, show great antioxidant activity. The relationship between the protective effect of the maqui berry extract and its antioxidant and anti-inflammatory properties in the brain has not been studied in depth. The objective of this study was to evaluate the effect of maqui berry on markers of oxidative stress and inflammation in the hippocampus of rats exposed to ozone. Sprague Dawley rats were exposed to 0.25 ppm ozone 4 hours daily for 30 days, and 1 hour after each exposure session were administered with maqui berry extracts.

Histological assays were performed on brains embedded in paraffin blocks. Tissue sections 5 µm wide were obtained and mounted on poly-L-lysine-coated slides. Immunohistochemistry assay was performed to confirm whether maqui berry modified the total positive cells to 4HNE, NT3, IL-6 and IL-1. We observed that ozone exposure increases lipid (4HNE) and protein (nitrotyrosine NT3) oxidation, as well as the content of inflammation markers (IL-6 and IL-1), while the administration of 100 mg/kg body weight of maqui berry aqueous extract decreases these markers in the dentate gyrus and CA3 region of the hippocampus. These results show an antioxidant and anti-inflammatory effect of maqui berry aqueous extract on the dentate gyrus and CA3 region of the hippocampus and suggest a neuroprotective effect against damage produced by chronic ozone exposure.

References

1. Bribiesca-Cruz I, Moreno DA, García-Viguera C, Gallardo JM, Segura-Urbe JJ, Pinto-Almazán R, Guerra-Araiza C. Maqui berry (*Aristotelia chilensis*) extract improves memory and decreases oxidative stress in male rat brain exposed to ozone. *Nutr Neurosci*. 2021 Jun;24(6):477-489
2. Castillo-García EL, Cossio-Ramírez AL, Córdoba-Méndez ÓA, Loza-Mejía MA, Salazar JR, Chávez-Gutiérrez E, Bautista-Poblet G, Castillo-Mendieta NT, Moreno DA, García-Viguera C, Pinto-Almazán R, Almanza-Pérez JC, Gallardo JM, Guerra-Araiza C. In Silico and In Vivo Evaluation of the Maqui Berry (*Aristotelia chilensis* (Mol.) Stuntz) on Biochemical Parameters and Oxidative Stress Markers in a Metabolic Syndrome Model. *Metabolites*. 2023 7;13(12):1189

HYPOTHALAMIC TRANSCRIPTOMICS UNDER DIFFERENT HIGH-FAT DIETS

Ruth Gutiérrez-Aguilar^{1,2,3,*}, Yael Efrén Díaz-López², Ekta Pathak³, Montserrat Torres-Vanda²,
Cristina Mencias-Castillejo³, Paul Pfluger³

¹Facultad de Medicina, Universidad Nacional Autónoma de México, México. ²Laboratorio de Investigación en Enfermedades Metabólicas: Obesidad y Diabetes, Hospital Infantil de México Federico Gómez, México.

³Helmholtz Zentrum München, Germany. *Corresponding author: ruthgutz@unam.mx
Dr. Márquez No. 162, Col. Doctores, Delegación Cuauhtémoc, Ciudad de México, C.P. 06720
Tel. +52.55.52289917 ext. 4509

Obesity is a consequence of an increased high-fat diet (HFD) consumption. HFD has been shown to modulate the expression of genes involved in the energy balance. The hypothalamus is the main regulator of energy homeostasis by controlling food intake and energy expenditure, leading to control of body weight. The hypothalamus is composed of various nuclei, including the arcuate (ARC), paraventricular nucleus (PVN), ventromedial (VMH), dorsomedial (DMH), and lateral hypothalamic area (LH).

In the ARC, the energy balance is regulated by the AgRP/NPY-coexpressing neurons, and the POMC/CART-coexpressing neurons. POMC is cleaved into α -melanocyte stimulating hormone and binds to MC4R in the PVN triggering an anorexic response. However, AgRP binds to MC4R, provoking the opposite effect, by enhancing food intake.

Besides the genes codifying these main peptides, expression of other genes, not yet described, could be involved in the regulation of energy homeostasis and in the adaptation to HFD. Therefore, it is important to study the broad spectrum of genes regulated by different type of HFD using a transcriptomic approach.

Methods. We performed a systematic review using the terms: “hypothalamus, high-fat diet, diet-induced obesity, RNAseq, transcriptomics, microarray,” from papers published between 2013-2023. From the selected papers, we obtained the list of differential expressed genes in at least 2 studies. Then, we compared the gene expression among areas of hypothalamus, percentage of high-fat diet, and time of exposure to the diet.

Results. We found 6 papers that analyzed the whole hypothalamus, obtaining 133 genes differentially expressed under different high-fat diets, highlighting genes involved in pathways such as hormone activity, feeding behavior, signaling receptor binding, double stranded DNA binding, among others. Moreover, we found 4 papers analyzing ARC gene expression, finding that over 2000 genes were differentially expressed, mainly involved in lipid binding, synaptic signaling, behavior, etc. From VMH, we found 2 papers, with 106 differential expressed genes, that were involved in presynapse and synapse. For PVN and LH, we only found one study per region. Interestingly, AgRP/NPY expression was lower under 60% than 45% HFD in the whole hypothalamus, contrary to what we observed in the ARC.

Conclusions. Transcriptomic analysis of different areas of the hypothalamus under different HFD could lead to identify new genes and pathways involved in the regulation of the energy balance.

EFFECT OF A HIGH-CARBOHYDRATE DIET ON INFLAMMATION AND MICROGLIAL PHENOTYPE IN THE ADENOHYPOPHYSIS OF POSTPUBERTAL WISTAR RATS

Hugo Hernández Fragoso¹, Samuel Treviño Mora²

¹Faculty of Chemical Sciences, BUAP, Puebla, Puebla, Mexico. ²Faculty of Chemical Sciences, BUAP, Av. San Claudio, Col. San Manuel, Puebla, Puebla, Mexico. C.P. 72570. Phone: +52 222 118 1729
E-mail: samuel_trevino@hotmail.com

The pituitary gland is the central endocrine regulator of growth, reproduction, metabolism, and stress response. The presence of microglia, immune cells of the central nervous system, has been revealed in the adenohypophysis. Microglia modulate the pituitary's local immune response and tissue homeostasis, releasing nitric oxide (NO), cytokines, chemokines, and reactive oxygen species (ROS). The aim was to evaluate the effect of consuming a high-carbohydrate diet for two months on microglial differentiation, inflammatory, and redox balance in the adenohypophysis of postpubertal Wistar rats.

Adenohypophysis cell differentiation was performed using hematoxylin and eosin staining. Microglial phenotypes were characterized using Iba1, CD16, and CD206 by immunofluorescence. In the tissue, IL-1 β , IL-10, IL-17, IL-6, IL-4, and TGF- β were quantified by chemiluminescence. Redox balance was analyzed, measuring catalase and superoxide dismutase activity, nitrite, malondialdehyde, 4-hydroxy-2-nonenal, and total lipoperoxidation concentration. Serum luteinizing hormone, follicle-stimulating hormone, and adrenocorticotrophic hormone were assayed.

The results showed that consuming a high-carbohydrate diet for two months alters the serum concentration of pituitary hormones. Furthermore, it induces an anti-inflammatory adenohypophysis microglial phenotype (M2), establishing an anti-inflammatory status and an antioxidant redox balance.

References

1. Ormel, P.R., Vieira de Sá, R., van Bodegraven, E.J. et al. Microglia innately develop within cerebral organoids. (2018). *Nat Commun* 9, 4167
2. Lenz K.M., Nugent B.M., Haliyur R., McCarthy M.M. Microglia are essential to masculinization of brain and behavior. (2013) *J. Neurosci.* 2013; 33:2761-2772.

ROLE OF SODIUM PHENYLBUTYRATE ON HIPPOCAMPAL MORPHOLOGICAL CHANGES OF HYPERAMMONEMIC RATS INDUCED WITH CCl₄

Brenda Hernández Juárez¹, Alfonso Daniel Díaz Fonseca³, Addí Rhode Navarro Cruz²,
Obdulia Vera López², Anna Bella Handal Silva³, Ivan Cesar Arteaga²

¹Facultad de Ciencias Químicas, BUAP; ²Departamento de Bioquímica-Alimentos, FCQ-BUAP; ³Instituto de fisiología BUAP;

⁴Posgrado en Ciencias Químicas, BUAP; Bioquímica Alimentos, BUAP. Av. San Claudio S/N CP:72595, Puebla, México.

Cel: 2271059868

Correo: ivan.cesar@correo.buap.mx

The ammonia (NH₃) is important nitrogen source, essential for amino acid synthesis; but, in high concentrations in the blood, is toxic and causes diverse metabolic pathologies. In physiological conditions, the NH₃ is regulated for urea cycle, various diseases hereditary and acquired can deregulate this process generating an accumulation of NH₃ in blood. The hyperammonemia is defined as elevation of NH₃ levels on blood plasma >110µmol/L. Although of hepatotoxicity carbon tetrachloride (CCl₄) is well known, there are few reports about its effects on the brain, especially in motor and cognitive areas such as the hippocampus. Sodium phenylbutyrate (NaPB) is a drug indicated for disorders of urea cycle, since it reduces the elevated plasma NH₃ concentrations, facilitating its renal excretion, although now is exploring its neuroprotective capacity. The objective of this project is evaluate the effect of NaPB on the hippocampal morphology of rats with hyperammonemia induced with CCl₄. 21 male Wistar rats of 3-month-old obtained from Claude Bernard vivarium of Benemérita Universidad Autónoma de Puebla. Rats were separated in different groups: 1) Control group: untreated rats; 2) CCl₄ group: manage group with CCl₄ for 10 weeks and 3) CCl₄ + NaPB group: manage group with CCl₄ for 10 weeks and 8 weeks with treatment with NaPB. After administration, animals were euthanized, the brains were extracted and cut into 200-µm-thick coronal sections using a vibratome. Subsequently the Golgi-Cox technique was performed, revealing the complete three-dimensional neuronal morphology of hippocampal regions (CA1, CA3 y GD). The results showed that treatment with NaPB allows the preservation of total dendritic length, number of dendritic spines and ramification order of three regions analyzed compared to those that did not receive the treatment. We results show that CCl₄ causes changes on neuronal hippocampal morphology, we suggest to NaPB like effective therapeutic alternative to improve the quality of life of the population with hyperammonemia.

GH TREATMENT DECREASE ASTROGLIOSIS IN THE POST STROKE

Martin Hernández Lucas¹, Isabel Arrieta-Cruz², Maricela Luna Muñoz³, Carlos Arámburo de la Hoz³,
Hilda Martínez-Coria¹, Emilio E. Muñiz Reynoso¹, Nadia F. Esteban Román¹,
Renata Jordá Chávez¹ and Héctor E. López-Valdés^{1*}

¹Departamento de Fisiología, Facultad de Medicina, UNAM, CDMX. ²Departamento de Investigación Básica, INGER, CDMX.

³Laboratorio de Bioquímica de hormonas, INB, UNAM, QRO.

*helopezv@gmail.com, teléfono: 5522165594

After a stroke, two areas of brain damage are observed. One is an area of necrosis and the other is the penumbra area. In the penumbra, there is a decrease in blood perfusion and a gradient of damage is observed. The site furthest from the necrotic zone shows the least damage. Astrocytes located in the penumbra are exposed to various substances that induce morphofunctional changes known as astrogliosis. The intensity of astrogliosis also presents a gradient, the closer the astrocyte is to the necrotic zone, the greater the intensity of the response. Assessment of the extent of astrogliosis reflects the magnitude of the functional alteration.

In this work, we analyzed the effect of growth hormone (GH) on the astrogliosis response in three-month-old male mice with stroke. GH treatment started 24 hours after stroke and lasted seven days. Astrogliosis was assessed by immunohistochemistry and image analysis of glial fibrillary acidic protein (GFAP) immunoreactivity in brain slices from control and GH-treated animals.

The results show that GH treatment decreases the extent of astrogliosis, suggesting a protective effect of this hormone.

RESVERATROL AS A MODULATOR OF THE GLUTATHIONE SYSTEM AND ITS NEUROPROTECTIVE EFFECT ON THE PREFRONTAL CORTEX

Brenda Hernández Pérez¹, Norma Judith Cruz Ortega³, José Luis Morán Perales⁴, Daniel Juárez Serrano², Ivan Cesar Arteaga³, Alfonso Daniel Díaz Fonseca⁴

¹Facultad de Ciencias Químicas, BUAP; ²Posgrado en Ciencias Químicas, BUAP; ³Departamento de Bioquímica – Alimentos, BUAP;

⁴Instituto de Fisiología, BUAP, Instituto de Fisiología, BUAP. 14 Sur 6301, CP 72592, Puebla, México.

Telefono: 22295500 Ext:7361

Correo: alfonso.diaz@correo.buap.mx

Resveratrol (trans-3,5,4-trihydroxystilbene) is a molecule belonging to the group of polyphenols. It has been shown to be a potent modulator of oxidative damage related to aging. This molecule consists of two phenolic rings joined by a styrene double bond, which is responsible for the different isomeric forms, with the *trans* isoform being the most abundant. The objective of this research project is to determine the effect of resveratrol on the activity of the enzymes of the glutathione system. In the methodological part, a total of 45 three-month-old male Wistar strain rats were used. They were maintained in standard vivarium conditions under 12-hour light/dark cycles with ad libitum access to water and food. The rats were randomly divided into three groups: Control (CTRL) (no treatment), Vehicle (VEH) (given 7.5% ethanol), and Resveratrol + Vehicle (RSVL + VEH) (given 10 mg/kg/day of resveratrol + 7.5% ethanol). Evaluations were conducted at the ages of 5, 14, and 24 months. The total content of GSH and GSSG was determined by the enzymatic recycling technique based on the use of the enzyme glutathione reductase. The results showed a greater increase in GSSG at 14 months, while GSH had a lower concentration in this same age group, as glutathione is reduced in GSH and to a lesser extent in its GSSG oxidation state. The significance lies in the glutathione system (L-γ-glutamyl-L-cysteinyl-glycine; GSH), which is the most important antioxidant defense protecting cellular function at the brain level. Aging is accompanied by a decrease in GSH levels, often associated with impairments in the prefrontal cortex, resulting from the loss of neurons, leading to a decline in cognitive performance. Oxygen radicals play an important role in age-related changes and in the pathogenesis of several neurodegenerative diseases. Enzymes of the antioxidant glutathione/thioredoxin system inactivate ROS and NOS, while glutathione peroxidase detoxifies peroxides, including H₂O₂ and the peroxides generated during the oxidation of membrane lipids. The enzymatic oxidation of GSH to the GSSG disulfide determines the degree of reduction of peroxide levels. To maintain the cellular balance of GSH and GSSG, GSH can be regenerated from GSSG by glutathione reductase. Thus, the use of resveratrol aims to reduce the negative physiological impact of oxidative stress derived from aging on the prefrontal cortex, highlighting its importance for potential clinical applications.

SEX-DEPENDENT EFFECTS ON NEURODEVELOPMENT OF PERINATAL EXPOSURE TO GLYPHOSATE HERBICIDES IN THE RAT

Isela Hernández-Plata, Jair Santiago Salazar, María Soledad Mendoza-Trejo,
Jonathan Sánchez-Yepéz, Verónica M. Rodríguez Córdova

Departamento de Neurobiología Conductual y Cognitiva. Instituto de Neurobiología, Universidad Nacional Autónoma de México. Blvd. Juriquilla 3001. C.P. 76230. Querétaro, Qro. México. Tel. (442) 2381061 ext. 34061.
E- mail: vmrodriguez@comunidad.unam.mx

Faena® is a herbicide used in Mexico that contains Glyphosate (Glyph) as the active ingredient. Glyphosate exposure during the perinatal period has been associated with neurodevelopmental alterations in human and animal models. Also, it has been suggested that Glyph exposure produces traits similar to the valproic model, like autism. In this study, our research group determined the Glyph/Faena® perinatal treatment on neurodevelopmental in male and female pups rats. Also, we compared our results with the valproic model in rats. The following groups were obtained (n=6-7 dams per group). A) Glyphosate: Pregnant rats drank 50 mg Glyph/L in distilled water (DW) during gestation and lactation. B) Faena®: Pregnant dams were exposed to Faena (50 mg Glyph/L) in DW during gestation and lactation. C) Valproic (Val): Pregnant dams received an intraperitoneal injection of 500 mg valproic/kg at 14.5 gestational days. D) The control group drank DW. Two male and two female pups per litter were assessed in several neurodevelopmental tests from postnatal day (PND) 5 to 21: negative geotaxis (PND 5, 7, 9), righting reflex (PND 5-10), cliff avoidance (PND 5, 7, 9), sniff test (PND 8), forelimb grip strength (PND 12, 14, 16), inverted screen test (PND 17, 19, 21), and hindlimb suspension test (PND 9, 11, 13). Female pups exposed to Glyph and Faena® showed delayed performance in turns to face upward in the negative geotaxis test at PND 9. In the same way, female pups treated with Val delay performance in the geotaxis negative at PND 5 and 9. At the same time, only male pups exposed to Val had difficulties noticing the mom's sawdust during the sniff test. Negative geotaxis is a sensible test to reveal the subtle effects of Glyph and Faena® herbicide on female pups. Glyphosate as the active ingredient and the commercial mix modify motor development and vestibular function in female pups. These effects were similar to the alterations caused by Val exposure, a known model of autism in rodents. The effects of Glyph and Faena® herbicide are sexually dimorphic in early development. More studies are necessary to unravel the neurotoxic mechanism of glyphosate in the developing brain.

Supported by UNAM-DGAPA-PAPIIT IN 212522; IHP received a fellowship from CONAHCyT 164300, and Ing. Luis Antonio Prado Loeza.

EVALUATION OF THE EFFECT OF METFORMIN ON LEARNING AND LONG-TERM MEMORY IN TYPE 2 DIABETIC MICE

Hernandez Poblano Joan Aldae¹, Luis Enrique Cobos Puc¹, Aguayo Morales Hilda¹, Tamayo Ordoñez María Concepción¹, Rodríguez Salazar María del Carmen¹, Silva Belmares Sonia Yesenia¹

¹Facultad de Ciencias Químicas, Universidad Autónoma de Coahuila. Boulevard V. Carranza S/N esq con Ing. José Cárdenas Valdés, República Oriente, CP 25280, Saltillo, Coahuila.
E-mail: joanhernandezpoblan@uadec.edu.mx

Diabetes mellitus and Alzheimer's disease share common mechanisms that warrant investigation of the effects of hypoglycemic agents on learning.¹ This study focused on evaluating the effects of metformin at different concentrations on long-term memory in mice with type 2 diabetes mellitus. (DM2).

The study was conducted on 27 female mice of the BALB/c strain, between 8 and 12 weeks old, divided into four groups: a control group without treatment and three groups treated with metformin at doses of 50 mg/kg, 150 mg/kg and 250 mg/kg. Metformin was administered orally once daily for one month. The study was carried out in accordance with the NOM-062-ZOO-1999 standard and approved by the Ethics Committee of the Faculty of Chemical Sciences (UAdeC, TMC-22-09-23-2).

Long-term memory was assessed using the object recognition test.² In addition, blood glucose levels and body weight were monitored weekly throughout the study.

Initial results showed that untreated DM2 mice had poorer long-term memory than those treated with metformin. Among the treated groups, the group receiving the highest dose (250 mg/kg) showed the greatest improvement in learning. After four weeks, the control group continued to show long-term learning problems associated with elevated blood glucose levels. The metformin-treated groups showed improvements in long-term learning that were proportional to the dose administered.

References

1. Samaras, K., et al, (2020). Metformin Use Is Associated With Slowed Cognitive Decline and Reduced Incident Dementia in Older Adults With Type 2 Diabetes, 43(11), 2691-2701.
2. Lueptow, L. M. (2017). Novel Object Recognition Test for the Investigation of Learning and Memory in Mice. Journal of Visualized Experiments, 126. Memory and Ageing Study. Diabetes Care, 43(11), 2691-2701.

LEPTOMERIN REDUCES LOCOMOTOR AND SPATIAL MEMORY DEFICITS CAUSED BY INTRAHIPPOCAMPAL INJECTION OF $A\beta_{1-42}$

Dulce Hernández Sacramento¹, Alfonso Díaz⁴, Víctor Gómez Calvario⁵, Diana M. Torres Cifuentes⁶, Ivan Cesar Arteaga², Daniel Juárez Serrano³

¹Facultad de Ciencias Químicas, BUAP; ²Departamento de Bioquímica – Alimentos, FCQ – BUAP; ³Posgrado en Ciencias Químicas, BUAP; ⁴Instituto de fisiología, BUAP; ⁵Instituto de ciencias – BUAP; ⁶Departamento de microbiología, FCQ – BUAP
Posgrado en Ciencias Químicas, BUAP. Av San Claudio S/N CP. 72595, Puebla, México.

Cel: 2211343520

Email: danieljuarezserrano@viep.com.mx

Alzheimer's disease is a neurodegenerative disorder characterized by a progressive and irreversible decline in motor and cognitive ability. One of the toxic changes that occur is the formation of beta-amyloid ($A\beta$) peptide plaques between neuronal connections, causing their death. Leptomerin is one of the acetylcholinesterase enzyme activity inhibitors (AChEI) indicated as a therapeutic treatment for Alzheimer's disease given that it increases the availability of acetylcholine (ACh), thus slowing cognitive deterioration. The objective of this study was to evaluate the effect of leptomerin on neurotoxicity induced by intrahippocampal injection of $A\beta_{1-42}$ on motor behavior and declarative memory. 30 male rats of the Wistar strain, 3 months old, obtained from the Claude Bernard bioherium of the Benemérita Universidad Autónoma de Puebla (BUAP), which were divided into five experimental groups: Group 1: Control group (no treatment); Group 2: Simulated surgery was performed without administering $A\beta$ -peptide or leptomerin (SHAM) treatment; Group 3: Only received an intrahippocampal injection of $A\beta_{1-42}$; Group 4: only received leptomerin treatment; Group 5: received an intrahippocampal injection of $A\beta_{1-42}$ and 30 days later received leptomerin treatment for 30 days. At the end of treatment, all groups were realized the Novel Object Recognition (NOR) test. The results showed that the control, SHAM and leptomerin-treated groups did not present significant differences in motor and cognitive activity, on the other hand, the group that was administered with $A\beta$ did show a great deterioration in motor and memory capacity, finally the $A\beta$ + leptomerin group presented significant improvements in terms of exploration time and motor capacity. In conclusion, results indicate that leptomerin treatment produces significant improvements in short-term and long-term declarative memory in Alzheimer rats without affecting their motor activity.

EFFECT OF AGOMELATINE ON THE CPF 5-HT_{2c} RECEPTOR IN A RAT MODEL OF MAJOR DEPRESSION

Daniel Juárez Serrano¹, Ivan Cesar Arteaga², Alfonso Daniel Díaz Fonseca³

¹Posgrado en Ciencias Químicas, BUAP; ²Departamento de Bioquímica-Alimentos, FCQ-BUAP; ³Instituto de Fisiología, BUAP
Instituto de Fisiología, BUAP. Intituto de Fisiología, BUAP. 14 Sur 6301, CP 72592, Puebla, México.
Telefono: 22295500 Ext:7361; Correo: alfonso.diaz@correo.buap.mx

Major depressive disorder (MDD) is a condition affecting individuals of all ages, with a higher prevalence among females. The monoamine theory suggests that a decrease in serotonin concentration is the primary cause of MDD, also implicating the levels of dopamine and norepinephrine. Among various antidepressant medications, selective serotonin reuptake inhibitors (SSRIs) like fluoxetine are prominent for increasing serotonin levels in the synaptic cleft. However, SSRIs are associated with adverse effects, necessitating the exploration of new treatments for MDD. One promising candidate is agomelatine, which acts as an antagonist of 5-HT_{2c} receptors and an agonist of melatonin receptors. Agomelatine has demonstrated the ability to restore dopamine and norepinephrine levels within hours of administration, though it does not affect serotonin levels. Consequently, further research is needed to fully understand its potential in treating MDD. The aim of this work is to evaluate the effect of agomelatine on the 5-HT_{2c} receptor of prefrontal cortex (CPF) in a model of major depression in rats. 120 male rats of the Wistar strain weighing between 250-300g, 6 animals were randomly assigned to the following 5 groups: Sham, OBX, OPBX-V, OBX-F and OBX-A. The depression model included bilateral rat bulbectomy surgery (OBX) which was characterized by the olfactory discrimination test, the open field test and the Light/dark test, and the forced swim test. The administration of drugs was carried out daily for periods of 0, 7, 14, 21 days. Once the treatment was finished, the samples were kept at -80°C. Subsequently, the quantification of serotonin in the supernatant was performed using the Serotonin ELISA kit ADI-900-175. The results of the behavioral tests showed an improvement in the motor behavior of the animals after the treatment. In addition, an increase in the concentration of serotonin in the prefrontal cortex occurs after prolonged treatment with agomelatine. In conclusion, both antidepressants reduced the hyperactivity of the animals with OBX after the different periods, as well as the regulation of neurotransmitters.

CATALASE AS A BIOMARKER OF OXIDATIVE STRESS RELATED TO DEPRESSIVE DISORDERS

Francisco Javier Lievanos-Ruiz¹ and Bertha Fenton-Navarro²

^{1,2}Universidad Michoacana de San Nicolás de Hidalgo, Laboratorio de Glicobiología y Farmacognosia. División de estudios de Posgrado. Facultad de Ciencias Médicas y Biológicas "Dr. Ignacio Chávez". Av. Dr. Rafael Carrillo S/N, Cuauhtémoc, 58020 Morelia, Michoacán México.

E-mail: ¹francisco.lievanos@umich.mx, ²bertha.fenton@umich.mx

Catalase (CAT) is an enzyme crucial in protecting cells from oxidative damage. It accelerates the decomposition of hydrogen peroxide in cellular metabolism into water and oxygen. This reaction prevents hydrogen peroxide from accumulating and causing oxidative stress, which can lead to cellular damage and disease. Altered levels of CAT may be associated with an increased risk of depressive disorders due to high levels of oxidative stress in the brain. The study aimed to conduct a systematic review of the evidence from the past ten years on CAT activity as a biomarker of oxidative stress in depressive disorders in humans. Method: A systematic review followed the PRISMA guidelines, searching six principal scientific article databases, including original scientific articles that evaluated CAT in participants with depressive disorders. Results. Ten articles meet the selection criteria. Half (50%) of the articles reported an increase in serum and plasma CAT in patients with depressive disorders such as major depressive disorder (MDD) with acute episodes, bipolar disorder (BD), treatment-resistant bipolar disorder (TRBD), and depressive severity without specific diagnosis. One article (10%) showed a negative correlation between CAT and depressive symptomatology in athletes without a specific depressive disorder diagnosis. Three of the four articles that did not yield significant results were conducted on erythrocyte samples in patients with MDD, BD, and unspecified depressive disorder. Conclusion: There is evidence that a significant increase in CAT in plasma and serum, but not in erythrocytes, is associated with a diversity of depressive disorders. The evidence on the relationship between CAT and depressive symptoms without a specific psychiatric diagnosis is controversial, possibly associated with variables such as health condition, duration of the disorder, and treatment. Due to limited evidence, further research is required to corroborate these relationships.

RNA SEQ AND CYTOKINE SECRETION COMPARISON IN PRIMARY RAT CEREBRAL CORTEX ASTROCYTES INDUCED TO CELLULAR SENESCENCE OR GLIOSIS WITH PALMITATE

Michel López-Teros^{1,2}, Karla Estephania Ávila-Galicia^{1,2}, Raúl Librado-Osorio³, Adriana Alarcón Aguilar², Armando Luna-López³, Alejandro Sánchez-Flores⁴, Veronica Jiménez-Jacinto⁴, Jorge Antonio García-Álvarez⁵, Mina Königsberg²

¹Posgrado en Biología Experimental, División de Ciencias Biológicas y de la Salud, Universidad Autónoma Metropolitana Unidad-Iztapalapa, Ciudad de México. ²Laboratorio de Bioenergética y Envejecimiento Celular, División de Ciencias Biológicas y de la Salud, Universidad Autónoma Metropolitana Unidad-Iztapalapa, Ciudad de México. ³Instituto Nacional de Geriátria, San Jerónimo Lídice, Ciudad de México. ⁴Instituto de Biotecnología, Cuernavaca, Morelos. ⁵Facultad de Ciencias, Universidad Nacional Autónoma de México, Ciudad de México. Ferrocarril San Rafael Atlixco, Núm. 186, Col. Leyes de Reforma 1 A Sección, Alcaldía Iztapalapa, C.P. 09310, CDMX. Tel: 555804-4600 ext 3383.
E-mail: michellopez@gmail.com

Astrocytes are the most abundant cells of the Central Nervous System (CNS) and show an adaptive plasticity that defines the functional maintenance of said system. When a stressful stimuli exist in the CNS, astrocytes can enter in a state of cellular senescence (SC) or be activated into gliosis. Obesity is a global health problem, so in this study we used palmitate to induce astrocytes cellular senescence or gliosis, simulating a pathological environment caused by the effects of obesity on neuroinflammation, due to the secretion of proinflammatory cytokines and contributing to changes in the redox state in the brain.

To achieve this, primary cultures of neonatal Wistar rat astrocytes (3-5 days) were performed. They were exposed to a Palmitate-Albumin conjugate (1:6) at different concentrations: 200µM for senescence induction, and 40µM for gliosis during 24 h. Senescent and gliosis markers were evaluated, as well as viability, proliferation and SA-β-Gal. Cytokine secretion was evaluated using Bio-Plex Cytokine Rat 23-Plex and RNA-Seq was performed. Finally, the redox state of both inductions was evaluated by means of GSH/GSSG ratio. Using the Oily Red technique, we confirmed that palmitate (PA) was introduced into the cells. Significant differences were found in the treated cells proliferation with respect to the control, both in senescent and gliosis. The SA-β-Gal and safranin assay showed that the treatment with PA increased the number of senescent cells, but not the control or gliotic cells. The senescence state and gliosis were confirmed using immunofluorescence tests with markers of senescence (γH2AX, Lamin B1 and B-gal) and gliosis (C3, S100A10 and GFAP). The of cytokines evaluation showed significant differences in the secretion of GM-CSF, GRO-a, IFN-g, IL-1a, IL-1b, MCP-1, IL-18, MIP-1a, MIP-3a, RANTES, TNF-a, VEGF, IL-6 among senescence, gliosis and control. RNA-Seq showed significant differences in the expression levels of genes related to the inflammation processes and cytokine secretion among the phenotypes. Finally, the redox state of both was evaluated and differences were found between both phenotypes.

We thank to Dr. Ángeles Guerrero-Aguilera for the animals. This Project is financed by-FORDECYT-PRONACES/263957/2020. MLT and KEAG are CONAHCyT scholarship holders

DEPRESSION-LIKE BEHAVIOUR IS COMORBIDITY IN A MODEL OF HEART FAILURE WITH PRESERVED EJECTION FRACTION INDUCED BY HIGH-FAT DIET AND L-NAME

Roger Alexis Maldonado Ruiz¹, Victoria Palafox Sánchez¹, Judith Bernal-Ramírez¹, Omar Lozano García¹, Gerardo de Jesús García Rivas¹

¹Av. Eugenio Garza Sada 2501 Sur, Tecnológico, 64849 Monterrey, N.L., C.P. 64700, tel. 88882161
E-mail: gdejesus@tec.mx.

Heart failure with preserved ejection fraction (HFpEF) is a syndrome with a 35% 2-year rate of hospitalization and 14% 2-year mortality. HFpEF patients have a prevalence of depression 20%¹. Several studies report that HFpEF and depression, share proinflammatory cytokines, including IL-33, which plasma concentration has a positive correlation with left ventricle diastolic dysfunction² and depressive symptoms³. In addition, downregulation of the astrocyte derived IL-33 in the amygdala prevents the development of depression in mice⁴. Even though, there is an under-recognition of depression in 50% of the HFpEF cases, that may be due to the absence of benefit from standard pharmacotherapy⁵. Thus, we propose that a mice model of HFpEF develops depression-like behavior and have an increases expression of IL-33 in the amygdala. To achieve this, we employ eight-week-old male C57BL/6 mice that were randomly assigned to the following groups. HFpEF group: mice fed with ad libitum HFD (60% Fat) and drinking water with L-NAME (0.5g/L) for 12 weeks, control: mice fed with ad libitum chow diet and tap water for 12 weeks. To determine cardiac hypertrophy, we measure the left ventricle cardiomyocyte cross-sectional area and, to identify the presence of depression we employ the open field test (OF), sucrose preference (SP) and novelty suppressed feeding (NSF). To determinate inflammation at the amygdala, we measure IL-33, IL-10, IL-6 and IL-1 β expression by qPCR. Also, we measure GFAP gene expression as an indirect marker of gliosis. Finally, knowing that cholesterol homeostasis is critical for normal brain function, we quantify the ABCA1 expression. We found that the HFpEF group presents a 26% increase cardiomyocyte area, a reduction of 11% (SP), a latency to eat of 53s (NSF) and spend 30% less time at the center and 20% more time at the edges of the arena in the OF test. Finally, the HFpEF group present an increased expression of ABCA1 (1.6-fold), GFAP (1.7-fold), IL-1 β (1.9-fold) and IL-33 (1.8-fold) as well as a 50% downregulation of IL-10 in the amygdala. With this evidence we can conclude that, an exposure to HFD+ L-NAME for 12 weeks promotes cardiac hypertrophy and depression-like behavior accompanied by an increased expression of IL-33 in the amygdala.

References

1. Dunlay, S. M., Roger, V. L. & Redfield, M. M. (2017). *Nat Rev Cardiol* 14, 591–602.
2. Yu Song, Fuhai Li, Yamei Xu, Yuan Liu, Yanyan Wang, Xueting Han, Yuyuan Fan, Juan Cao, Jie Luo, Aijun Sun, Kai Hu, Jingmin Zhou, Junbo Ge (2020). *Int J Cardiol* 304, 95–100.
3. Ewa A Ogłodek, Marek J Just (2018). *Pharmacol Rep*. 70(6):1065-1072.
4. Xiao Zhuang , Bing Zhan, Yufeng Jia, Chaoze Li, Nan Wu, Ming Zhao, Nuo Chen, Yaxin Guo, Yingxin Du, Yi Zhang, Baihui Cao, Yan Li, Faliang Zhu, Chun Guo, Qun Wang, Yuan Li, Lining Zhang (2022). Volume 102, Pages 98-109.
5. Haider J Warraich, Dalane W Kitzman, David J Whellan, Pamela W Duncan, Robert J Mentz, Amy M Pastva, M Benjamin Nelson, Bharathi Upadhyya, Gordon R Reeves (2018). *Circ Heart Fail*. Nov;11(11):e005254.

MUTATION ANALYSIS IN THE RCBTB1 GENE CAUSING RETINAL DYSTROPHY: A MODEL FOR MAPPING RELATED GENES IN MEXICO

Claudia Mancilla Simbro^{1*}, Fernando I. Ramírez Luna¹, Julio C. Cordero Olivares¹, Fritz D. Herrera Salcedo¹, Brandon Ortiz Pérez¹, Saúl A. Ríos Cano², Leisy M. Santiago-Hernández³, Sandra Reyes Carmona⁵, Laura G. Hernández Aragón⁴, Citalli Gamboa Esteves⁶ and Alberto Ramírez-Mata⁵

¹Hybrid.Lab Fisiología y Biología Molecular de Células Excitables. Instituto de Fisiología. Benemérita Universidad Autónoma de Puebla. 14 sur 6301 CU. Sn. Manuel. Puebla México. *claudia.mancilla@correo.buap.mx; ²Lab. Fisiología Humana, Facultad de Medicina, Complejo Nororiental Campus Teziutlán. Benemérita Universidad Autónoma de Puebla. C. Bravo 15-17, C.P. 73800, Teziutlán, Puebla México. ³Lingüística y Literatura Hispánica-Preparatoria Benito Juárez García. Benemérita Universidad Autónoma de Puebla. 14 Sur 5500, C.P. 72550, Puebla México. ⁴Lab. Cáncer y Comunicación Intercelular-Instituto de Fisiología. Benemérita Universidad Autónoma de Puebla, México. ⁵Lab. de la Interacción bacteria-planta. ICUAP. Benemérita Universidad Autónoma de Puebla. CU. Sn. Manuel, Puebla México, C.P. 72000. ⁶Lab. Neurofisiología Sensorial. Instituto de Fisiología. Benemérita Universidad Autónoma de Puebla. 14 sur 6301 CU. Sn. Manuel. Puebla México.

Injuries affecting the retina or the optic nerve cause manifestations in the homolateral visual field. Visual field defects usually range from scotomas to anopsias, and are due to total damage to the retina or the optic nerve. When alterations affect the macular fibers, loss of visual acuity is common and significant. The main causes for vision loss and the onset of inherited retinal diseases (IRD) are genetic defects or environmental changes, affecting around 2 million people, impacting 1 in every 2000 individuals. IRDs are characterized by the progressive degeneration of the retina, leading to the progressive degeneration of photoreceptors. In the RetNet database (<https://web.sph.uth.edu/RetNet/>), more than 260 genes related to these diseases have been identified. RCBTB1 is a guanine nucleotide exchange, it is related to cell cycle regulation through chromatin remodeling. It possesses BTB domains and RCC1 domains. RCBTB1 can function as a substrate adapter for the Cullin E3 ligase complex (CUL3), which regulates ubiquitination for proteasomal degradation of proteins. Additionally, it has been associated with angiogenesis through Norrin-induced β -catenin signaling. Our aim is to investigate the trajectory of mutations present in RCBTB1 reported to date and identify the presence of these mutations within the Mexican population. Develop and validate a methodology for the identification and analysis of genes involved in the gradual vision loss process. Our conclusion is a compilation of mutations reported in RCBTB1 causing vision impairment was conducted. This information enabled the design of 2 pairs of primers aimed at identifying them within the Mexican population. The methodology that showed the best results was with blood samples. Therefore, the interpretation and simplicity of our method make it efficient for healthcare professionals.

References

1. Ascari, G. (2020). CIL-EYE : functional characterization of potential ciliary genes involved in syndromic inherited retinal diseases. <http://hdl.handle.net/1854/LU-8665762>
2. Coppieters F, Ascari G, Nikopoulos K, Xu M, Langmann T, Chen R, et al. Hypomorphic mutations in RCBTB1 cause autosomal recessive isolated and syndromic inherited retinal dystrophy. *Invest Ophthalmol Vis Sci*. 2016 Sep 26;57(12):637–637.
3. Nuzbrokh, Y., Ragi, S. D., & Tsang, S. H. (2021). Gene therapy for inherited retinal diseases. *Annals of translational medicine*, 9(15), 1278. <https://doi.org/10.21037/atm-20-4726>
4. Huang, E. (2022). Disease modelling and treatment development for RCBTB1-associated retinopathy. [Doctoral Thesis, The University of Western Australia].
5. Chaharbakhshi, E., & Jemc, J. C. (2016). Broad-complex, tramtrack, and bric-à-brac (BTB) proteins: Critical regulators of development. *Genesis (New York, N.Y. : 2000)*, 54(10), 505–518. <https://doi.org/10.1002/dvg.22964>
6. Catomeris, A. J., Ballios, B. G., Sangermano, R., Wagner, N. E., Comander, J. I., Pierce, E. A., Place, E. M., Bujakowska, K. M., & Huckfeldt, R. M. (2022). Novel RCBTB1 variants causing later-onset non-syndromic retinal dystrophy with macular chorioretinal atrophy. *Ophthalmic Genetics*, 43(3), 332–339. <https://doi.org/10.1080/13816810.2021.2023196>
7. Ung, T. T., Starr, C. R., Zhylykibayev, A., Saltykova, I., & Gorbatyuk, M. (2024). Development of TRIB3-Based Therapy as a Gene-Independent Approach to Treat Retinal Degenerative Disorders. *International journal of molecular sciences*, 25(9), 4716. <https://doi.org/10.3390/ijms25094716>
8. Mancilla SC, Contreras BCF, Ramírez MA, López ALE, Ríos CSA, Santiago HLM, Hernández ALG. (2024). Creación de encuestas para el diseño de un jardín sensorial: un enfoque etnobotánico y bioinformático en la BUAP. *Körperkultur Science* 2(4): 37-40.

FUNCTIONAL CHARACTERIZATION OF HUMAN EMBRYONIC STEM CELL DERIVED-CHROMAFFIN CELLS

H. Angélica Martínez-Becerril¹, Óscar J. Parada-Parra¹, Alejandro Salazar-Salgado¹
Diana A. Millán-Aldaco¹ and Arturo Hernández-Cruz¹

¹ Instituto de Fisiología Celular UNAM. Ciudad de México. México. C.P 04530.
hmartinez@ifc.unam.mx

Chromaffin cells (CCs) of the adrenal medulla secrete catecholamines adrenaline and noradrenaline, which act as neurohormones regulating multiple functions, including blood pressure, cardiovascular homeostasis and adaptive responses to stress conditions. These cells have been extensively studied and characterized in different species, however, comparatively little is known in humans due to difficulties in obtaining and/or availability. In this regard, cells from pheochromocytoma or from patients undergoing kidney removal (transplantation) have been studied. In this context, it is relevant to expand research in the development of *in vitro* models of CCs, since there are also notable species differences in terms of their physiological properties, catecholamine secretion patterns and responses to stimuli. Considering the above, the main objective of this project is to develop an experimental protocol capable of generating human embryonic stem cell derived-chromaffin cells as an *in vitro* model for functional studies of the autonomic nervous system.

OBESITY INDUCED BY A DIET RICH IN FATS AND SUGARS AFFECTS SPATIAL MEMORY IN OVARIECTOMIZED FEMALE CD1 MICE

Martínez-Pacheco H.*, V.E. Bolado-García, Mendoza-Calles C.A. and Zepeda R.C.

Centro de Investigaciones Biomédicas, Universidad Veracruzana

Email: heydimartinez@uv.mx

During the climacteric, 40% of women experience a significant increase in visceral adipose tissue. This stage has the highest prevalence of obesity and the risk of developing metabolic syndrome. The combined effects of obesity and estrogen deficiency on cognitive decline in postmenopausal women are not well studied. Therefore, this rigorously conducted study, which meticulously investigated the impact of a high-fat and high-sugar diet (HF-HSD) with ingredients commonly used in the Mexican population on spatial memory in 8-week-old ovariectomized female CD1 mice (OVX), provides robust evidence. OVX or sham-operated (SHAM) mice were fed HFD (44% fat) or a standard diet (3% fat) for 16 weeks. The HF-HSD-OVX group showed a 60% weight gain since week 1 ($P < 0.001$), high blood glucose levels compared to the control group (mean CTL 118.33 vs. mean HF-HSD 166.33, $P < 0.001$), significant accumulation of visceral, abdominal and perinephric adipose tissue ($P < 0.001$) and impairment in the consolidation of spatial memory ($P < 0.05$). These findings suggest that low levels of estrogen do not impair spatial memory. However, combining low estrogen levels and high-calorie food intake impairs hippocampal-dependent memory.

Acknowledgments: Laboratorio de Investigacion Medicina Traslacional of the Universidad Veracruzana for supporting us in the use of its facilities.

Supported by: Estancias Posdoctorales por México 2022, CONACYT.

ALTERATIONS IN SPATIAL MEMORY OF CD-1 MICE STRAIN FEED WITH MEXICAN CAFETERIA DIET

M. en C. César A. Mendoza Calles^{1,2}, Dra. Heidy Martínez Pacheco², Dr. Aleph A. Corona Morales³, Dra. Mónica Flores Muñoz⁴, Dra. Victoria E. Bolado García³, Dra. Rossana C. Zepeda²

¹Programa de Doctorado en Ciencias Biomédicas, UV, ²Centro de Investigaciones Biomédicas, UV.

³Facultad de Nutrición, UV, ⁴Instituto de Ciencias de la Salud, UV.

E-mail: mendoza.camc@outlook.com

The excessive consumption of diets high in certain types of fats, such as saturated and trans fats, as well as sugars like fructose and sucrose, leads to obesity. These dietary changes significantly impact the brain, particularly spatial memory, which relies on the hippocampus. Our study aimed to assess metabolic and cognitive differences after 16 weeks of exposure to a high-fat and high-sugar diet in female and male CD-1 mice. We used eight-week-old female and male CD-1 mice, dividing them into four groups: control (CTL) groups received a standard diet plus water, and high-fat and high-sugar diet (HF-HSD) groups were fed a high-fat diet plus 40% sucrose. We monitored body weight, glucose levels, and caloric intake for 16 weeks. Spatial memory was evaluated using the T-maze and novel place and object recognition tasks. Our findings show that both male and female mice fed with HF-HSD experienced significant weight gain and elevated blood glucose levels compared to mice fed a standard diet. The HF-HSD groups also exhibited increased abdominal, visceral, perirenal, and gonadal adipose tissue and elevated serum cholesterol levels. However, their locomotor and exploratory activities were similar to those of the control group. Spatial tasks such as the T-maze and novel place recognition were challenging for the HF-HSD groups. Unlike other studies that use commercial food, we utilized a standardized and cost-effective handmade diet with ingredients commonly found in Mexico. Our model using CD-1 mice resulted in more significant weight gain compared to other mouse strains commonly used to study diet-induced obesity. Our study using commonly found Mexican ingredients revealed alarming results. Female and male mice fed with HF-HSD doubled their weight and experienced significant challenges in spatial memory tasks. These findings emphasize the health risks associated with dietary habits and lifestyle, highlighting the need for immediate attention and action.

SENSITIVITY OF TUBB4A TO MICROTUBULE STABILIZATION AND DEPOLYMERIZATION

Aurora Montes^{1,2}, Andrés Silva¹, Valeria Piazza², Víctor Henández¹

¹Laboratorio de Neurociencias, Universidad de Guanajuato, Loma del Bosque 103 CP 37150.

²Centro de investigaciones en Óptica, A.C. León, Gto.

E-mail: ac.montescenteno@ugto.mx

Hypomyelination with Atrophy of the Basal Ganglia and Cerebellum (H-ABC) is a neurodegenerative disease caused by mutations in *TUBB4A*. Is characterized by hypomyelination and wide range of neurological symptoms such as motor and cognitive developmental delays, rigidity, involuntary movements, dystonia, speech problems, ataxia, epilepsy, and spasticity (Van Der Knaap et al., 2002).The *TUBB4A* gene encodes for Beta-4A tubulin, a component of the microtubules in the myelinating oligodendrocytes in the early stages of myelination (Terada et al., 2005).

Understanding the correlation between *TUBB4A* mutations and their cellular, tissue, and organic effects is of paramount importance. For instance, the role of *TUBB4A* is the dynamics and organization of microtubules, a crucial aspect of cellular function, is yet to be fully elucidated.

We hypothesize that *TUBB4A* mutations disrupt the stability of the cytoskeleton, potentially leading to demyelination. This study aims to investigate the microtubule's stability in cells expressing *TUBB4A*. To this end, we transfected HeLa cells with a plasmid containing the *TUBB4A* gene sequence and a fluorescent reporter gene (TurboGFP). Then, we treated the cells with paclitaxel, a microtubule-stabilizing agent that inhibits their depolymerization. On the other hand, to produce depolymerization, we lowered the temperature of the medium. Confocal images were analyzed using super-resolution algorithms to look for changes in the tubulin cytoskeleton and its effects on the cells.

Currently, there is no treatment for H-ABC disease, so knowledge of the effects of mutations in *TUBB4A*, could contribute to properly treating these diseases in the future.

References

1. Terada, N., Kidd, G. J., Kinter, M., Bjartmar, C., Moran-Jones, K., & Trapp, B. D. (2005). *GLIA*, 50(3), 212–222.
2. Van Der Knaap, M. S., Leegwater, P. A. J., Könst, A. A. M., Visser, A., Naidu, S., Oudejans, C. B. M., Schutgens, R. B. H., & Pronk, J. C. (2002). *Annals of Neurology*, 264–270

MITOCHONDRIAL DYNAMICS IN THE RETINA AT THE EARLY ONSET OF DIABETES IN A RAT MODEL OF TYPE I DIABETES

Frida Paulina Muñiz-Ruvalcaba¹, Rocío Salceda-Sacanelles¹

¹Instituto de Fisiología Celular, UNAM, Circuito Exterior s/n Ciudad Universitaria, Coyoacán, 04510, Ciudad de México.
E-mail: paulinam@ifc.unam.mx

Diabetic Retinopathy is a major complication of diabetes, characterized by vascular alteration. The retina is a highly metabolic tissue, and its energetic requirements are achieved mainly through glycolysis, although OXPHOS is required¹. Mitochondrial dynamics are the processes of fusion, fission and mitochondrial biogenesis. Mitochondrial fusion is accomplished mainly by Mfn 1 & 2 on the OMM and by OPA1 at the IMM². Fission is modulated by cytoplasmic DRP1 which translocates to adaptor proteins of the OMM constricting the mitochondria for excision³. It is assumed that failure in maintaining mitochondrial dynamics influence OCR, $\Delta\Psi_m$ and promote apoptosis leading to diseases progression⁴. Previous results from our laboratory have demonstrated that in the retina of STZ-induced diabetic rats there is no change in OCR, ATP, and ERO production from 7 up to 45 days post injection⁵. Hence, we aimed to evaluate the expression levels of proteins involved in mitochondrial dynamics in diabetic rats.

Diabetes was induced in 200 g, Long Evans, female rats with a single injection of STZ (98 mg/Kg). Rats were sacrificed at 7, 20 and 45 days post STZ injection. Eyes were enucleated and retinas were collected and processed for WB analysis. Anti-DRP1, Anti-FIS1, anti-OPA1, anti-Mfn2 and Anti- β -Actin were used.

Results: We observed that there was an increase in Fis1 with a reduction of OPA1 at all the evaluated times of diabetes. We did not identify changes in protein levels of DRP1 and Mfn2 at the evaluated times of diabetes.

Conclusion: The increase in Fis1 levels, in addition to the reduction of OPA1 are indicative of an increase of mitochondrial fission in the retina at early STZ induced diabetes. These results are indicative of mitochondrial dynamic changes caused by hyperglycemia.

Acknowledgements: The authors thanks to Sánchez-Chávez G. for technical assistance. This study was supported by DGAPA/UNAM, Project IN-204523. Muñiz-Ruvalcaba F. P. is a doctoral student from Programa de Doctorado en Ciencias Biomédicas, Universidad Nacional Autónoma de México and received fellowship (919709) from CONAHCYT.

References

1. Wang, W., & Lo, A. C. Y. (2018). *Int J Mol Sci*, 19(6).
2. Song, Z., Ghochani, M., Mccaffery, J. M., Frey, T. G., & Chan, D. C. (2009). *Mol Biol Cell*, 20(15), 3525-32.
3. Kalia, R., Wang, R. Y. R., Yusuf, A., Thomas, P. V., Agard, D. A., Shaw, J. M., & Frost, A. (2018). *Nature*, 558(7710), 401-405. 20, 3525-3532.
4. Chan, D. C. (2020). *Annu Rev Pathol*. 15, 235-259.
5. Osorio-Paz, I., Uribe-Carvajal, S., & Salceda, R. (2015). *PLoS ONE*, 10(5), 1-15.

UNRAVELING THE CORTICAL ORIGINS AND DOPAMINERGIC MODULATION OF BETA OSCILLATIONS IN PARKINSON'S DISEASE MODELS

Aidán Alejandro Ortega García*, Antonio Laville-Conde, Miguel Serrano-Reyes, Yeshua-Eliezer González-Jiménez, Montserrat Padilla-Orozco, Yohana Parrado Quintana, Héctor Aaron Vázquez-Vázquez, Janintzitic López-Niño, Dagoberto Tapia-Ramírez, Elvira Galarraga-Palacio and José Bargas Díaz

División de Neurociencia Cognitiva, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Mexico City, 04510.

*aidan.ortega@ifc.unam.mx

The primary motor cortex (M1) is essential for the planning and execution of motor activities, receiving inputs from the premotor cortex and dispatching excitatory glutamatergic signals to both the basal ganglia (BG) and spinal cord. Dopamine (DA) plays an indirect yet significant role in regulating motor functionality via its effects on BG activities. In patients with Parkinson's disease (PD) and corresponding mouse models, beta oscillations (13–40Hz) are prominent in BG local field potentials (LFPs). Employing a unilateral 6-OHDA lesion model of Parkinsonism in mice, which depletes BG dopamine while maintaining cortical DA modulation through an intact ventral tegmental area (VTA), revealed key insights. Microelectrode array (MEA) evaluations of in vitro brain slices showed atypical beta synchronization, likely due to impaired interactions between the BG and cortex. Notably, cortical beta oscillations persisted even in the absence of BG dopamine influence, but ceased following levodopa treatment. Furthermore, applying dopaminergic antagonists to acute Parkinsonian mouse brain slices disrupted beta oscillations in M1, pointing to a possible cortical source within the BG-cortex circuit. These observations underscore the cortical components of beta oscillations in the absence of DA, suggesting novel perspectives on the origins within the BG-cortex loop and informing potential therapeutic avenues for PD.

Keywords. *Parkinsonism, Neural Ensembles, Oscillatory Dynamics.*

CHARACTERIZATION AND DIFFERENTIATION OF HUMAN INDUCED PLURIPOTENT STEM CELLS INTO MIDBRAIN DOPAMINERGIC NEURONS

Andrés Paredes-Rocha^{1,2}, Xóchitl Flores-Ponce^{1,2}, Iván Velasco-Velázquez^{1,2}

¹Instituto de Fisiología Celular–Neurociencias, Universidad Nacional Autónoma de México. Circuito Exterior S/N, Ciudad Universitaria, CDMX 04510, México. ²Laboratorio de Reprogramación Celular, Instituto Nacional de Neurología y Neurocirugía. Insurgentes sur 3877, CDMX 14269, México.
E-mail: aparedes@ifc.unam.mx. Tel: +52 (55) 5556225649

Induced pluripotent stem cells (iPSCs) are produced by somatic cells that are reprogrammed to pluripotency, and therefore regain the ability to differentiate into all intraembryonic tissues. There are protocols allowing the *in vitro* differentiation of pluripotent cells into midbrain dopaminergic neurons (mDANs) by exposure to *Shh* activating molecules, FGF8, and *Wnt* signaling, along with dual inhibition of SMAD. The death of mDANs in the *substantia nigra* leads to a decrease in dopamine, the cause of motor dysfunction in Parkinson's disease (PD). With the aim of differentiating a healthy individual's iPSC line into mDANs, the 24L2 cell line were characterized through immunofluorescence assays with pluripotency markers, prior to the dopaminergic differentiation protocol (defined as day 0 of differentiation) and with dopaminergic lineage markers at an early neuronal maturation stage (day 28). Before the initiation of dopaminergic differentiation, iPSCs exhibit large nuclei and reduced cytoplasm with a spherical morphology, forming numerous colonies, characteristic of pluripotency. After differentiation, the newly formed neurons show axonal projections that emanate from somata that assemble into clusters, which interconnect among them. In the expression of immunofluorescent markers, the cells show an abundant amount of SOX2, OCT4, and NANOG for pluripotency, and of TH, FOXA2, and LMX1A for neuronal and dopaminergic phenotype. The differentiation of iPSCs derived from healthy individuals into mDANs serves as a controlled and effective tool for studying molecular aspects of this specific cellular phenotype, thereby contributing to the use of cells obtained from patients with PD to model the disease *in vitro* and, thus, investigate its etiology. This positions cellular reprogramming as a plausible method to have pathological models of neurodegenerative diseases.

Keywords. induced pluripotent stem cells; midbrain dopaminergic neurons; Parkinson's disease.

Acknowledgements. This study was supported by PAPIIT IN219122 and Conahcyt CF-2023-1668. We thank the technical support of Dr. Itzel Escobedo-Avila.

MUSCARINIC MODULATION OF CALCIUM CURRENTS IN STRIATAL CHAT'S INTERNEURONS: THE IMPACT OF M1 RECEPTOR

Lesly Yohana Parrado Quintana¹, Aidán Alejandro Ortega García¹, Montserrat Padilla Orozco¹,
Juan Antonio Laville Conde¹, Fabiola Hernández Vázquez², Víctor Hugo Avilés Rosas¹,
María Elvira Galarraga¹ and José Bargas Díaz^{1*}

¹Instituto de Fisiología Celular. División de Neurociencias. Universidad Nacional Autónoma de México. Circuito Exterior s/n Ciudad Universitaria, Col. Coyoacán, 04510 México D.F. México. ²Departamento de Fisiología, Facultad de Medicina, Universidad Nacional Autónoma de México (UNAM), PO Box 70250, Ciudad de México, 04510, Mexico.

Corresponding author: Dr. José Bargas at the above address

Tel: +52 55 5622 5615; fax: +52 55 5622 5615

Cholinergic interneurons (CIN) represent a small subpopulation (approximately 1 to 3%) of the total neuronal population in the striatum (Str). They are the only non-GABAergic neurons in the Str and express Choline Acetyltransferase (ChAT). These neurons are responsible for most of the acetylcholine (ACh) concentration in the Str, indicating their critical role in regulating striatal circuitry. Previous studies have shown that CIN express muscarinic M2-class receptors (M2/M4), which downregulate calcium current through CaV2.1 and 2.2 channels (N and P/Q). However, about 30% of them also express mRNA encoding for the M1 receptor, whose function in CIN remains unknown. This study investigates the role of the M1 receptor in CIN on whole-cell calcium current and neuronal excitability.

Patch-clamp whole-cell recordings of calcium current in acutely dissociated CIN from ChAT-Cre transgenic animals, using a specific muscarinic receptor agonist, muscarine, an M2-class receptor antagonist (AF-DX116), and an M1 receptor antagonist (MT7), show that the M1 receptor is functional in a subset of these cells, where it upregulates calcium current. Further recordings in brain slices using the same technique demonstrate that activation of this receptor increases neuronal excitability. Additional research is needed to determine whether the M1 receptor regulates the same or different calcium channels as those regulated by M2-class receptors in CIN.

Keywords. Cholinergic interneurons, Muscarinic receptors, Ca²⁺-currents, M2-class receptors, M1 receptor, Muscarinic modulation, Striatum.

EPIGENETIC CONTROL OF NEURON ACTIVATION AND MEMORY IN AGING

G. Aleph Prieto*, Citlali A. Suárez-Rangel, Yunuen Moreno-López

Laboratory of Molecular Codes of Memory, Institute for Neurobiology, UNAM Campus Juriquilla.
Blv. Juriquilla 3001, Juriquilla, Querétaro, 76230. Tel. 442 238 1059.

*E-mail: aleph.prieto@inb.unam.mx

Recent evidence suggests that epigenetic mechanisms contribute to memory deficits in aging. Specifically, in the hippocampus, the accumulation of the repressive epigenetic mark H3K9me3 (tri-methylated lysine 9 on histone H3) has been implicated in the silencing of genes essential for memory. Yet, the impact of the age-related increased H3K9me3 levels on neuronal function remains unclear. We hypothesized that H3K9me3 accumulation renders neurons refractory to stimulation. First, using immunohistochemistry (IHC) in brain slices and flow cytometry in isolated cell nuclei from the hippocampus of male mice, we found that H3K9me3 is mainly expressed in neurons compared to non-neuron cells and, notably, the H3K9me3 epigenetic mark increases with age selectively in the dorsal dentate gyrus (DG). Next, to assess the influence of H3K9me3 accumulation on neuronal activation in aged mice (18-month-old), we reduced H3K9me3 levels by silencing SUV39H1, the major histone methyltransferase responsible for producing H3K9me3. SUV39H1 was knocked down by stereotaxic delivery of shRNA-containing adeno-associated virus (AAV) in the dorsal DG. For stimulation, aged mice were exposed to a novel environment for 15 min, and neuronal activation was assessed 1-hour later by measuring the presence of the Arc protein. IHC and flow cytometry data confirmed that SUV39H1 knockdown reduces H3K9me3 levels in transfected neurons (tagged with EGFP) compared to controls. Notably, we observed increased levels (%) of activated neurons in the AAV-shRNA-SUV39H1 transfected population relative to controls, indicating an enhanced capacity for neuronal activation following H3K9me3 downregulation. Finally, we tested whether direct inhibition of SUV39H1 in the dorsal DG of aged mice suffices to restore cognitive function. Mice injected with SUV39h1-shRNA particles showed better performance when tested in the object location memory task (OLM) compared to mice injected with control particles. Our data show that SUV39H1 knockdown facilitates neuronal activation by decreasing the repressive H3K9me3 mark, thus supporting the concept that epigenetic regulation through H3K9me3 plays a role in the decline of hippocampal-dependent memory with age.

ROLE OF TESTOSTERONE IN THE GAIN OF STEMNESS CHARACTERISTICS IN CELLS DERIVED FROM HUMAN GLIOBLASTOMA

Quintero-Gallegos, Juan Carlos¹; Ignacio Camacho-Arroyo¹

¹Unidad de Investigación en Reproducción Humana, Instituto Nacional de Perinatología–Facultad de Química, Universidad Nacional Autónoma de México, Mexico City 11000, Mexico.
E-mail: jc.chimiste@gmail.com

Glioblastoma is the most common and lethal malignant brain tumor. Recently, the poor prognosis of this cancer has been associated with a cellular subpopulation present in the tumor called glioma stem cells, which are highly related to neural stem cells. Furthermore, this neoplasm occurs more frequently in men than in women, in a ratio of 3:2, suggesting that sex hormones, such as testosterone, may play an important role in tumor maintenance. In this work, the role that testosterone plays in the maintenance of glioma stem cells is studied. Using testosterone treatments in human glioblastoma cells of the U251 cell line, we analyzed the effect of this hormone on the expression of stemness factors, such as Sox2, Nestin and CD133. In addition, the action of testosterone on cell proliferation and generation of neurospheres is analyzed to study its self-renewal capacity. With this work we aim to understand the possible role of testosterone in the generation and maintenance of glioma stem cells.

References

1. Rodríguez-Lozano DC, Piña-Medina AG, Hansberg-Pastor V, Bello-Alvarez, C, Camacho-Arroyo, I (2019). Testosterone Promotes Glioblastoma Cell Proliferation, Migration, and Invasion Through Androgen Receptor Activation. *Front Endocrinol*, 10:16.
2. Zhao N, Wang F, Ahmed S, Liu K, Zhang C, Cathcart S.J, DiMaio D.J, Punsoni M, Guan B, Zhou P, et al (2021). Androgen Receptor, Although Not a Specific Marker For, is a Novel Target to Suppress Glioma Stem Cells as a Therapeutic Strategy for Glioblastoma. *Front. Oncol*, 21, 616625.
3. Kim HJ, Kim TJ, Kim YG, Seong C, Cho JH, Kim W, Lee KH, Kim DY (2021). Antioxidant and Antiproliferative Activity of Finasteride against Glioblastoma Cells. *Pharmaceutics*, 13, 1410.
4. Quintero JC, Días NF, Rodríguez-Dorantes M, Camacho-Arroyo, I (2023). Cancer Stem Cells and Androgen Receptor Signaling: Partners in Disease Progression. *International Journal of Molecular Sciences*, 24(20):15085.

THE CALCIUM HYPOTHESIS IN ALZHEIMER'S DISEASE

¹Ramirez Cipriano Luis Angel, ²Salas Casas Andres, ³Figueroa Gutierrez Ana Hilda

¹Universidad Autónoma del Estado de Hidalgo, Instituto de Ciencias de la Salud, ¹Área Académica de Nutrición, ²Área Académica de Gerontología, ³Área Académica de Medicina. Circuito Ex Hacienda la Concepción s/n Carretera, Pachuca Actopan, San Agustín Tlaxiaca, Hidalgo. C.P. 42160. Tel. 5529525019.
Correo: andres_salas15@yahoo.com.mx

Calcium (Ca^{2+}) is an essential micronutrient belonging to the group of minerals that should be an integral part of our diet at all times. Ca^{2+} is a cation with diverse functions as a second messenger in different types of cells of the immune system, intervening in numerous processes being crucial in these, as well as in the release of neurotransmitters and postsynaptic activity, excitability, second messenger cascades, gene expression and apoptosis (1).

The aim of this work is to analyze the relationship between the calcium hypothesis and Alzheimer's disease, which is considered a neurodegenerative disease, predominantly affecting people aged 65 years and older. 50 million people live with Alzheimer's type dementia worldwide; due to the aging of the population, it is expected that the number of patients will triple by 2050, in Mexico there are between 800,000 and 900,000 people, it is expected that by 2050 this number will triple (2), which increases the risk of disability, disease burden and health care costs. The interaction between Alzheimer's disease and Ca^{2+} is that the higher the amount of the cation, the greater the neurodegeneration, the lower the amount, the lesser the neurodegeneration. In aging an interaction of slow AHP (Ca^{2+} -dependent afterhyperpolarization), Ca^{2+} -dependent or Ca^{2+} -mediated AHP (sAHP) is seen.

By means of bioinformatics techniques, we used molecular docking to analyze the interaction of STIM and ORAI, as main Ca^{2+} reservoirs in the endoplasmic reticulum with the tau protein, we studied the interaction with the purpose of finding out if STIM and ORAI let more or less Ca^{2+} through during Alzheimer's disease, in addition; we analyzed their physicochemical characteristics during the interaction (3).

In conclusion, STIM and ORAI alter calcium homeostasis by increasing its concentration in the endoplasmic reticulum, increasing the probability of Alzheimer's disease.

References

1. Izquierdo J-H, Bonilla-Albadia F, Cañas CA, Tobon GJ (2014). *Reumatol clin*, vol 10, 43-7.
2. Arias C, del Carmen Cardenas M, (2020). *Internet UNAM*.
3. Thibault O, Gant JC, Landfield PW, (2007). *Journal*, vol 6, 307-17

METABOLIC AND EPIGENETIC EFFECTS OF β -HYDROXYBUTYRATE DURING THE DIFFERENTIATION OF HUMAN PLURIPOTENT CELLS TO DOPAMINERGIC NEURONS

Margareht Reyes Aldana¹, Juan Jair Santillán-Cigales¹, Iván Velasco-Velázquez^{1,2}

¹Instituto de Fisiología Celular–Neurociencias, Universidad Nacional Autónoma de México. Circuito Exterior S/N, Ciudad Universitaria, CDMX 04510, México. ²Laboratorio de Reprogramación Celular, Instituto Nacional de Neurología y Neurocirugía. Insurgentes sur 3877, CDMX 14269, México.
E-mail: margareht@ifc.unam.mx Tel: +52 (55) 5556225649

Glucose supply in mammals does not cover energy requirements in several physiological states, such as the neonatal period, pregnancy, lactation, fasting, and prolonged exercise. Specifically, during brain development, it has been proposed that there is a change in metabolic substrate utilization, with ketone bodies (KB) serving as the main energy source during these fetal periods. However, the effects of KB during these early stages of human neurodevelopment have not been described. The most abundant KB is β -hydroxybutyrate (BHB); this KB participate in multiple signaling functions, both on the cell surface and intracellularly, affecting gene expression, lipid and protein metabolism, as well as neuronal functions and metabolic rates through both direct and indirect mechanisms. Therefore, BHB might play a physiologically important role in neural differentiation. However, BHB has not been used in protocols for *in vitro* neuronal differentiation of human pluripotent cells. The objective of this study was to evaluate the inductive and epigenetic changes induced by BHB during the differentiation of human embryonic stem cells into mesencephalic dopaminergic neuron. We found neural progenitors exposed to different concentrations of BHB exhibit significant changes in the expression level of markers related to dopaminergic differentiation. Additionally, BHB was observed to induce global changes in epigenetic marks H3K27me3 and H3K27ac. The next step is to understand how BHB treatment contributes to the observed changes in the induction of this neuronal type.

Keywords. β -hydroxybutyrate (BHB), stem cells, dopaminergic differentiation.

Acknowledgements: This study was supported by PAPIIT IN219122 and Conahcyt CF-2023-1668. We thank the technical support of Dr. Itzel Escobedo-Avila.

INACTIVATION OF *PLPP3* IN NEURAL CREST CELLS LEADS TO NEONATAL CARDIAC FAILURE

Erick Rojas-Pacheco¹, Elliot Flores-Oliva¹, Sheyla Cruz-Cruz¹, Ana Valeria Martínez-Silva¹, Diana Escalante-Alcalde^{1*}

¹Instituto de Fisiología Celular, División de Neurociencias (UNAM), Ciudad de México C.P. 04510

*descalan@ifc.unam.mx

Phospholipid phosphatases (PLPPs) are integral membrane enzymes broadly expressed in animal tissues. They dephosphorylate bioactive lipids such as lysophosphatidic acid (LPA) and sphingosine-1-phosphate (SIP), among others¹. These enzymes are dynamically localized on cell membranes and participate in various cellular processes².

Our group studies *Plpp3*, a member actively expressed and essential during development. *Plpp3* gene inactivation in mice causes embryonic lethality around E9.5. These embryos fail to form a chorio-allantoic placenta, and defects in yolk sac vasculature and neural tube closure³. Moreover, the absence of this enzyme produces an expansion of *Wnt1* expression domains at 8-somites. *Wnt1* is a molecular marker for premigratory neural crest cells (NCC), a multipotent cell population. Also, an increase in cell death markers was observed on putative regions of NCC migratory paths⁴. In addition, *in vitro* culture of neural tube explants suggested defects in their migratory behavior. These observations in conjunction with transcriptomic analysis that reveals *Plpp3* expression in NCC⁵ and neural crest-derived structures⁶, suggested a potential role in the development of these cells for this enzyme.

To test this hypothesis we conditionally inactivated *Plpp3* in premigratory NCC using the *Wnt1::Cre2* driver, commonly used to study NCC and its derivatives^{7,8}. We recovered homozygous mutants in an expected frequency, however, mutant pups regularly died a few hours after birth, with very few individuals found at weaning. Mutant pups showed evident craniofacial and cardiovascular defects. Heart abnormalities are thought to be the leading cause of lethality in the offspring. We analyzed heart morphology and found normal cardiac structure, although atrioventricular myocardium and coronary sinus walls vary in thickness. These features cannot explain mortality. Electrophysiological analysis exhibited a diminished heart rate in mutant pups (~276 bpm, bradycardia), whereas 386 bpm was registered in wild type. Bradycardia and lineage tracing experiments hinted at alterations in sympathetic components, which are neural crest-derived structures. Fasciculation impairment in postganglionic nerves and changes in epicardial axonal thickness can be seen in mutant hearts.

In summary, these results suggest that *Plpp3* is a novel regulator of cardiovascular function through NCC derivatives. We propose it as a candidate gene for neurocristopathies, syndromes that affect a percentage of the population⁹.

References

- Kok, B. P., Venkatraman, G., Capatos, D., & Brindley, D. N. (2012). Unlike two peas in a pod: lipid phosphate phosphatases and phosphatidate phosphatases. *Chemical reviews*, 112(10), 5121–5146. <https://doi.org/10.1021/cr200433m>
- Tang, X., & Brindley, D. N. (2020). Lipid Phosphate Phosphatases and Cancer. *Biomolecules*, 10(9), 1263. <https://doi.org/10.3390/biom10091263>
- Escalante-Alcalde, D., Hernandez, L., Le Stunff, H., Maeda, R., Lee, H. S., Jr-Gang-Cheng, Sciorra, V. A., Daar, I., Spiegel, S., Morris, A. J., & Stewart, C. L. (2003). The lipid phosphatase LPP3 regulates extra-embryonic vasculogenesis and axis patterning. *Development* (Cambridge, England), 130(19), 4623–4637. <https://doi.org/10.1242/dev.00635>
- Ramírez-Corona, A. (2018). El papel de la fosfatasa de fosfolípidos 3 en el desarrollo del cerebro medio y posterior [Tesis de Licenciatura, Universidad Nacional Autónoma de México]. <http://132.248.9.195/ptd2018/junio/0775641/Index.html>
- Pijuan-Sala, B., Griffiths, J.A., Guibentif, C. et al. A single-cell molecular map of mouse gastrulation and early organogenesis. *Nature* 566, 490–495 (2019). <https://doi.org/10.1038/s41586-019-0933-9>
- Escalante-Alcalde, D., Sánchez-Sánchez, R., & Stewart, C. L. (2007). Generation of a conditional Ppap2b/Lpp3 null allele. *Genesis* (New York, N.Y. : 2000), 45(7), 465–469. <https://doi.org/10.1002/dvg.20314>
- Dinsmore, C. J., Ke, C. Y., & Soriano, P. (2022). The *Wnt1*-*Cre2* transgene is active in the male germline. *Genesis* (New York, N.Y. : 2000), 60(3), e23468. <https://doi.org/10.1002/dvg.23468>
- Lewis, A. E., Vasudevan, H. N., O'Neill, A. K., Soriano, P., & Bush, J. O. (2013). The widely used *Wnt1*-*Cre* transgene causes developmental phenotypes by ectopic activation of *Wnt* signaling. *Developmental biology*, 379(2), 229–234. <https://doi.org/10.1016/j.ydbio.2013.04.026>
- Erhardt, S., Zheng, M., Zhao, X., Le, T. P., Findley, T. O., & Wang, J. (2021). The Cardiac Neural Crest Cells in Heart Development and Congenital Heart Defects. *Journal of cardiovascular development and disease*, 8(8), 89. <https://doi.org/10.3390/jcdd808089>

IMPACT OF DUAL INHIBITION OF PAK1 AND CAMK2 KINASES ON GLIOBLASTOMA ONCOGENESIS

Héctor Iván Saldívar Cerón^{1,2}, Genaro Patiño-López³, Marlene Areli Gaytán Gómez⁴, Claudio Adrián Ramos Cortés⁴, Xochitl Lucia Ramirez Hernandez⁴, Olga Villamar Cruz¹, Luis E Arias-Romero¹

¹UBIMED, Facultad de Estudios Superiores-Iztacala, UNAM, Tlalnepantla, México. ²Carrera de Médico Cirujano, Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México, Tlalnepantla 54090, México. ³Laboratorio de Investigación en Inmunología y Proteómica, Hospital Infantil de México, México. ⁴Laboratorio de Medicina de Conservación, Escuela Superior de Medicina, Instituto Politécnico Nacional, Ciudad de México, 11340, México.

Glioblastoma is the most common and aggressive malignant brain tumor, characterized by limited survival and poor response to conventional therapies. The kinases PAK1 and the CaMK2 have been identified as critical factors in tumor progression and treatment resistance. This study explores the efficacy of combined inhibition of these kinases in glioblastoma cell models.

Glioblastoma cell lines U251, U87, and TN98 were employed in this study. Kaplan-Meier survival analyses indicated a correlation between high expression of *PAK1* and *CaMK2* and unfavorable prognosis in patients, suggesting that these kinases could play critical roles in glioblastoma aggressiveness. Western blot analysis showed variability in the expression of PAK1 and CaMK2 among cell lines, which may reflect the genetic and phenotypic diversity found in primary tumors. Indirect immunofluorescence confirmed that the proteins do not directly interact, implying that their pro-tumor effects might be independent but complementary.

In MTT cell viability assays, results revealed that the combined inhibition of PAK1 with G5555 and CaMK2g with KN93 produces a synergistic reduction in cell viability. This finding suggests that the interaction of pathways modulated by these kinases might be more effective in disrupting essential cellular functions of glioblastoma than interventions targeting a single kinase. Additional assays, including wound healing, transwell migration, cell proliferation, and clonogenic assays, consistently showed that the combination of inhibitors enhances the restriction of cell growth and migration compared to monotherapies, indicating a potential increase in therapeutic efficacy through combined treatments.

The results suggest that dual inhibition of PAK1 and CaMK2 could represent a promising therapeutic strategy for glioblastoma treatment, overcoming some limitations of current therapies. This approach not only effectively impedes tumor progression but also offers an advantage over the use of monotherapy inhibitors, opening new avenues for future research on therapeutic combinations in glioblastomas.

ACTIVITY MODULATION OF STRIATAL CHOLINERGIC INTERNEURONS MEDIATED BY ACTIVATION OF DOPAMINE D1-LIKE RECEPTORS

M. Lizbeth Saucedo Bello, Yohana Parrado Quintana, José Bargas Díaz

Instituto de Fisiología Celular

José Bargas Díaz. Dirección postal: 04519; Correo: jlbargas@ifc.unam.mx; tel: +52 55 1654 7430

The striatum (Str) is the primary nucleus of the basal ganglia in the midbrain, playing a central role in motor function and reward processes. It serves as the input stage of the basal ganglia, receiving glutamatergic signals from the cortex (Ctx) and thalamus (Th), and dopaminergic input from the substantia nigra pars compacta (SNc). The Str is predominantly composed of spiny projection neurons (SPNs, GABAergic neurons), which constitute 90%-95% of its neuronal population. SPNs are divided into direct pathway SPNs (dSPNs) with D1 receptors that promote movement, and indirect pathway SPNs (iSPNs) with D2 receptors that suppress movement. Additionally, the Str contains GABAergic and cholinergic interneurons (CINs), comprising approximately 1%-3% of its neuronal population. CINs are the main source of acetylcholine (ACh) in the Str, and along with dopamine (DA) from SNc terminals, they modulate striatal circuitry.

There are two classes of dopamine receptors: D1-like (D1 and D5 receptors) and D2-like (D2, D3, and D4 receptors). CINs possess both D1-like and D2 receptors. The effect of D2 receptors in CINs involves N-type Ca^{2+} channels, Gi/q protein activation, and subsequent PKC signaling pathway activation. The role of D1-like receptors in CINs under control conditions remains unclear. Due to the opposing effects of these receptors, it is crucial to elucidate the impact of D1-like receptor activation in CINs of the Str under control conditions. This is the focus of our investigation. This study aims to investigate this question employing patch-clamp in whole-cell recording in brain slices and dissociated acutely neurons using ChAT-Cre transgenic mice.

This work was supported by grants from DGAPA-UNAM IN 205023 (JB) and CONAHCYT (México) F003-154039 to JB.

MOLECULAR CLONING, FUNCTIONAL CHARACTERIZATION, AND DIFFERENTIAL EXPRESSION OF TWO NOVEL GABA_AR-LIKE SUBUNITS FROM RED SWAMP CRAYFISH *PROCAMBARUS CLARKII*

Iván Uriel Valladares-Hernández¹, Juan Manuel Hernández-Martínez¹, José Miguel Cuaxospa¹, Eric Nahum Jiménez-Vázquez³, Edith Sánchez-Jaramillo⁴, Juan Manuel Arias², Ubaldo García¹

¹Departamento de Fisiología, Biofísica y Neurociencias, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Ciudad de México 07360, México. ²Proyecto de Neurociencias, Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México, Estado de México 54090, México. ³Department of Pharmacology, University of Michigan Medical School, Ann Arbor, MI, United States. ⁴Laboratorio de Neuroendocrinología Molecular, Dirección de Investigaciones en Neurociencias, Instituto Nacional de Psiquiatría Ramón de la Fuente Muñiz, Ciudad de México 14370, México.
Ubaldo García: ugarcia@fisio.cinvestav.mx and Juan Manuel Arias: jmarias@comunidad.unam.mx

In this work, we cloned and functionally expressed two novel GABA_A receptor subunits from *Procambarus clarkii* crayfish. These two new subunits, PcGABA_A-α and PcGABA_A-β2, revealed significant sequence homology with the PcGABA_A-β subunit, previously identified in our laboratory. In addition, PcGABA_A-α subunit also shared a significant degree of identity with the *Drosophila melanogaster* genes DmGRD (GABA and glycine-like receptor subunits of *Drosophila*) as well as PcGABA_A-β2 subunit with DmLCCH3 (ligand-gated chloride channel homolog 3). Electrophysiological recordings showed that the expression in HEK cells of the novel subunits, either alone or in combination, failed to form functional homo- or heteromeric receptors. However, the co-expression of PcGABA_A-α with PcGABA_A-β evoked sodium- or chloride-dependent currents that accurately reproduced the time course of the GABA-evoked currents in the X-organ neurons from crayfish, suggesting that these GABA subunits combine to form two types of GABA receptors, one with cationic selectivity filter while the other preferentially permeates anions. On the other hand, PcGABA_A-β2 and PcGABA_A-β co-expression generated a chloride current that does not show desensitization. Muscimol reproduced the time course of GABA-evoked currents in all functional receptors, and picrotoxin blocked these currents; bicuculline did not block any of the recorded currents. RT-PCR amplifications and FISH revealed that PcGABA_A-α and PcGABA_A-β2 are predominantly expressed in the crayfish nervous system. Altogether, these findings provide the first evidence of a neural GABA-gated cationic channel in the crayfish, increasing our understanding of the role of these new GABA_A receptor subunits in native heteromeric receptors.

PROGESTERONE TREATMENT RESTORES NORMAL BLOOD-BRAIN BARRIER FUNCTION IN A RAT MODEL OF CHRONIC CEREBRAL HYPOPERFUSION

Maydelín Espadín¹, Claudia Espinosa-García², Jose M. Cervantes³, Beatriz Gómez-González⁴, Gabriela Moral⁵

¹Universidad Autónoma Metropolitana, Posgrado en Biología Experimental, Ciudad De México, México. ²School of Medicine, Yale University, New Haven, USA. ³Facultad de Ciencias Médicas y Biológicas "Dr. Ignacio Chávez", Universidad Michoacana de San Nicolás de Hidalgo, División de estudios de Posgrado, Morelia, Michoacan, Mexico. ⁴Universidad Autónoma Metropolitana. Unidad Iztapalapa Biología de la reproducción, Mexico City, Mexico. ⁵Centro Médico Nacional Siglo XXI, IMSS, Unidad de Investigación Médica en Farmacología, Ciudad De México, México.
amiv540@gmail.com

Cerebral blood flow supplies oxygen and nutrients to support normal brain function. Aging is associated with chronic cerebral hypoperfusion (CCH). These blood flow age-related alterations might be attenuated by progesterone (P4), a neurosteroid which has been proven to exert pleiotropic neuroprotective effects in several models of brain injury. The aim of this study was to evaluate the effects of P4 on the blood-brain barrier (BBB) in the short term and on spatial learning and memory in the long term in rats subjected to CCH. Male Sprague-Dawley rats (12-14 months old) were randomly distributed in the following groups: CCH+vehicle; CCH+P4 (8 mg/kg/day) and sham procedure as a control. At seven and fourteen days after CCH, the function of the BBB was evaluated through permeability assays by systemic administration of a cocktail containing Evans blue and Na-fluorescein tracers. In addition, the expression of BBB tight junction proteins and inflammation factors was evaluated by western blot. At one hundred and eighty days later, memory and learning were evaluated using the Barnes maze and the novel object recognition test. CCH induced BBB dysfunction, decreased tight junction protein expression, increases inflammation factors and impairment in memory and learning. Treatment with P4 improved the BBB function, restoring the expression of the tight junction proteins, decreased inflammation factors, and preserves learning and memory. These results suggest that, in old male rats with disrupted blood-flow, P4 plays an important role in restoring BBB function, which may contribute to the neuroprotective effects that have been previously reported.

IMPACT OF INTESTINAL LOW-GRADE INFLAMMATION ON PURINERGIC-CALCIUM SIGNALING IN ENTERIC GLIAL CELLS

Irving Israel Vega-Juárez¹, Edgar Octavio Bonales-Alatorre¹, Julio Guerrero-Castro¹,
Fernando Ochoa-Cortes², Andrómeda Liñán Rico¹

¹Centro Universitario de Investigaciones Biomédicas, Universidad de Colima, Colima, México. ²Escuela Superior de Huejutla, Universidad Autónoma del Estado de Hidalgo, Hidalgo, México.
E-mail: andromeda_linan@ucol.mx

Enteric glial cells (EGCs) play a critical role in the regulation of motility, secretion, epithelial barrier function, and intestinal homeostasis. These cells express purinergic receptors and connexins (Cx43), which are involved in the propagation of Ca²⁺ signals (calcium waves) and gliotransmission. Intestinal inflammation induces a “reactive phenotype” in EGCs, which results in neuroinflammation, enteric neuropathy, intestinal motor dysfunction, “leaky gut” and visceral pain¹. Our goal was to assess the impact of intestinal low-grade inflammation associated with obesity on the induction of a reactive phenotype in EGCs and evaluate its effect on purinergic and mechanosensitive Ca²⁺ wave propagation. For this purpose, we incubate EGCs (CRL-2690) with palmitate (PA, 400µM) or BSA (Control) for 48 hours prior to induction with LPS (1 and 10µg/ml +PA)². After 24 hours, total RNA was extracted, followed by RT-qPCR to analyze the expression of GFAP, s100β, Sox10, IL6, iNOS2, TNFα, Cx43, TLR4, and P2X, P2Y and A1 receptors. We also tested cell viability and ATP release. Fluo-4AM Ca²⁺ imaging was conducted to evaluate the impact of LPS+PA treatment on Ca²⁺ wave propagation evoked by UTP 100µM (P2Y2/4/6) and mechanical stimulation. Treatment induced an increase in the expression of the inflammatory markers TNFα, IL6, iNOS2, and the purinergic genes P2Y1,4,6, P2X7, A2A and A2B. Cx43 expression was significantly reduced in PA+LPS 10ug/ml treatment. Cell viability and mechanosensitivity were not affected (PA +LPS 1 and 10µg/ml vs control), however, EGCs showed an increase in ATP release (PA+LPS 10µM vs control). The rate of propagation (µm/s) of UTP-induced Ca²⁺ waves as well as the number of cells responding were significantly reduced in PA+LPS-treated EGCs. In conclusion, intestinal low-grade inflammation induces a reactive phenotype in EGCs, characterized by the activation of pro-inflammatory signals and functional disturbances in the purinergic network that in turn affect the properties of calcium waves, with potential implications for the pathophysiology of gut dysmotility associated with obesity.

References

1. Linan-Rico A, Ochoa-Cortes F, Schneider R, Christofi FL. (2023). *Neurosci Lett*, 24, 812:137395.
2. Antonioli L, D'Antongiovanni V, Pellegrini C, Fornai M, Benvenuti L, di Carlo A, van den Wijngaard R, Caputi V, Cerantola S, Giron MC, Németh ZH, Haskó G, Blandizzi C, Colucci R. (2020). *FASEB J*, 34(4), 5512-5524.

POSSIBLE INFLAMMATION MEDIATED BY NF- κ B P65 IN THE RETINA ON SHORT-TERM INDUCED-DIABETES IN A RAT MODEL

Alejandro Canizales-Ontiveros¹, Rocío Salceda-Sacanelles¹

¹Instituto de Fisiología Celular, UNAM, Cto. Exterior s/n, C.U., Coyoacán, 04510, CDMX.

E-mail:canizales@ifc.unam.mx

Diabetes mellitus is a chronic metabolic disease characterized by high glucose levels. Diabetic retinopathy (DR) is a major complication in diabetic patients, being one of the main blindness causes worldwide¹. Although DR is clinically classified as a microvascular complication, recent evidence suggests that neuronal alterations may occur before. Several cellular mechanisms have been reported to be altered during DR including inflammation, oxidative stress and AGEs production². Inflammation is a highly regulated response to damage or stress which aims to recover a particular tissue from potential damage. In long-term induced diabetes, an increase in NF- κ B p65 (p65) and its phosphorylation has been reported, along with the production of cytokines like IL-1 β and TNF- α ³. Thus, we aim to determine whether NF- κ B p65 protein expression and IL-1 β increase in the retina on a short-term streptozotocin-induced diabetic rat model.

We induced diabetes to female 200g Long-Evans rats by administration of Stz (98mg/kg i.p.). Rats were considered diabetic if glucose was ≥ 250 mg/dL. After 7, 20 and 45 days of hyperglycaemia, the retinas were isolated and proteins extracted for Western blot assays (WB), were transferred into PVDF Immobilon. Densitometry of chemiluminescence using chemiluminescent HRP substrate was analysed. We measured the protein expression of NF- κ B p65, phospho- NF- κ B p65, IL-1 β , TLR-4 and Iba1 and obtained the relative expression (actin as the control).

We found a significant increase (90.10% \pm 17.72) in NF- κ B p65 expression at 20d, and slight but not statistically significant increase of IL-1 β at 45d. We did not observed changes in TLR-4 and Iba1 expression compared to the normal. These results suggest that NF- κ B p65 may start an inflammatory response at early STZ diabetes induction.

Acknowledgments: The authors thank Sánchez-Chávez, Gustavo for expert technical assistance. This project was supported in part by DGAPA/UNAM, Project IN204523. Alejandro Canizales Ontiveros is a PhD student from the Programa de Doctorado en Ciencias Biomédicas, Universidad Nacional Autónoma de México (UNAM) and has received CONAHcyT fellowship 1180490.

References

1. DF DIABETES ATLAS 9th ed. (2019). Bruselas, Bélgica; p. 1-176.
2. Lynch SK, Abràmoff MD. (2017). *Vision Res.* 139:101-7.
3. RübSam A, Parikh S, Fort PE. (2018). *Int. J. Mol. Sci.*, 19.

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex of atoms, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

REACTIVE OXYGEN SPECIES

YWQN ENCODES A NADP(H)/FMN-DEPENDENT QUINONE REDUCTASE THAT PROTECTS *BACILLUS SUBTILIS* FROM OXYGEN RADICAL GENOTOXICITY

Karen Abundiz-Yáñez¹, Norma Ramírez-Ramírez¹, Beatriz Rufina González-Pérez¹,
Víctor M. Ayala-García² and Mario Pedraza-Reyes¹

¹Department of Biology, Division of Natural and Exact Sciences, University of Guanajuato, Guanajuato, Mexico. Noria Alta S/N, Guanajuato, Gto; C.P. 36050, Tel 47320006 ext. 8151. ²Juárez University of Durango State. Faculty of Chemical Sciences, Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur, Durango, Dgo. C.P. 34120. E-mail:pedrama@ugto.mx

YwqN is a predicted protein with an unassigned function that possesses partial structural homology with *B. subtilis* YhdA, *Pseudomonas putida* ChrR and *Escherichia coli* YeiF. These proteins belong to a family of NADP(H)/FMN-dependent oxidoreductases that reduce Cr(VI) to Cr(III) avoiding the generation of partially reduced species that promote oxidative stress. Here, we report that a purified His₆-YwqN recombinant protein lacks chromate reductase activity but retains the ability to reduce distinct synthetic AZO dyes. Remarkably, His₆-YwqN also exhibited a potent quinone reductase activity that catalyzed the efficient reduction of menadione and 1,4 naphthoquinone. The individual and combined roles of YwqN and YhdA in protecting *B. subtilis* from ROS-promoting agents was further tested. The simultaneous loss of YwqN and YhdA was necessary to sensitize *B. subtilis* to the oxidizing agent H₂O₂. In contrast, strains deficient for *ywqN* or *yhdA* exhibit similar but higher susceptibilities to the superoxide-producer agent menadione (Md) than the WT strain. Therefore, YwqN and YhdA seem to participate in a common pathway that protects *B. subtilis* from the deleterious effects of oxygen radicals. In agreement with this contention, in reference to the WT strain, the loss of *ywqN* and *yhdA* increased the H₂O₂-promoted Rif^R mutagenesis in *B. subtilis*. Finally, the overexpression of YwqN counteracted the hyper mutagenic effects derived from accumulation of 8-OxoG in a *B. subtilis* strain deficient for the prevention/repair guanine oxidized (GO) system. In summary, our results indicate that YwqN counteracts the cytotoxic and mutagenic effects promoted by ROS in *B. subtilis* and unveil its potential role in remediating soils and effluents contaminated with carcinogenic Azo dyes.

Work supported by CONAHCYT (Grants A-1S-27116 and CBF2023-2024-708); Universidad de Guanajuato (CIIC-029-2024). K.A.Y and B.R.G.P. were supported by doctoral and masters' scholarships from CONAHCYT, respectively.

ENZYMATIC ANTIOXIDANT RESPONSE DURING PHYSIOLOGICAL CARDIAC HYPERTROPHY INDUCED BY PREGNANCY

José Bruno Apodaca Fuentes¹, Jesús Alfredo Rosas Rodríguez¹, Ana Paola Balderrama Carmona¹, Enrico Alejandro Ruiz Castillo¹, José Guadalupe Soñanez Organist¹

¹Universidad de Sonora. Blvd. Lázaro Cárdenas No.100, Col. Francisco Villa, Navojoa, Sonora. CP. 85880.
Tel. 6428534044. A217205710@unison.mx

During pregnancy, the physiological generation of ROS is involved in several cellular processes, allowing for a successful pregnancy¹. However, abnormal overproduction of ROS disrupts these processes, resulting in reproductive failure. When the generation of ROS increases, the heart is affected, causing cellular damage that leads to heart failure². In cells, ROS are counteracted by antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx)³. Therefore, in this work, we will evaluate the gene expression and enzymatic activity of SOD, CAT, and GPx in rat hearts during physiological cardiac hypertrophy induced by pregnancy and its reversible postpartum process. The mRNA levels for SOD, CAT, and GPx decreased during pregnancy and postpartum. In contrast, the activity for SOD and GPx increased during late pregnancy but not in postpartum, while CAT activity decreased during pregnancy and postpartum. Our results suggest that SOD and GPx participate in the protection of the heart during physiological cardiac hypertrophy induced by pregnancy.

References

1. Iorga, A., Dewey, S., Partow-Navid, R., Gomes, A. v., & Eghbali, M. (2012). *PLoS ONE*, 7(11).
2. Chiarello, D. I., Abad, C., Rojas, D., Toledo, F., Vázquez, C. M., Mate, A., Sobrevia, L., & Marín, R. (2020). *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease*, 1866(2).
3. Hafstad, A. D., Nabeebaccus, A. A., & Shah, A. M. (2013). *Basic research in cardiology*, 108(4).

DETERMINATION OF ANTIOXIDANT ACTIVITY AND PHENOLIC COMPOUNDS IN FERMENTED COCOA BEAN SHELL INFUSION

Sebastián Cervera-Pereyra¹, Carlos Javier López-Victorio¹, Adelma Escobar Ramírez,
Ivonne María Olivares-Corichi² and José Arnold González-Garrido¹

¹Centro de Investigación de Ciencia y Tecnología Aplicada de Tabasco (CICTAT), División Académica de Ciencias Básicas. Laboratorio de Bioquímica y Biología Molecular, Universidad Juárez Autónoma de Tabasco, Carretera Cunduacán-Jalpa KM. 1 Colonia la Esmeralda, Tabasco, C.P. 86690, México. ²Sección de Estudios de Posgrado e Investigación, Escuela Superior de Medicina del Instituto Politécnico Nacional. Laboratorio de Oncología Molecular y Estrés Oxidativo, Instituto Politécnico Nacional, Plan de San Luis y Díaz Mirón, C.P. 11340 Ciudad de México, México.
E-mail: 232A27001@alumno.ujat.mx, C.P: 86250, Tel:(993) 1532184

The Symbiotic Culture of Bacterial and Yeast (SCOBY) is used for the preparation of kombucha, a fermented beverage made from black tea and sucrose, thus obtaining a functional food rich in bioactive substances, especially phenolic compounds. These represent the main group of antioxidants present in kombucha and are responsible for the health benefits of the beverage. Recently, the use of alternative substrates for kombucha preparation has been described. Therefore, SCOBY was used in combination with cocoa bean shell infusion, a byproduct of the cocoa industry, which has recently been proposed as a functional ingredient mainly due to its polyphenols content, for the preparation of a fermented beverage with antioxidant activity. This ferment was evaluated for 10 days to establish the evolution of antioxidant activity and polyphenols content. Antioxidant capacity was assessed using the ABTS and DPPH methods. The highest antioxidant activity was observed on day 6 of fermentation with average values of 26% and 38%, respectively. Total phenols were quantified using the Folin-Ciocalteu method, finding similar concentrations of phenolic compounds throughout fermentation, around 2 milligrams of gallic acid equivalents per mL. In conclusion, significant differences were established with a p-value <0.05 between the antioxidant activity by the ABTS and DPPH methods between cocoa bean shell infusion and its fermentation on days 3 and 6. Additionally, cocoa bean shell is a good source of polyphenols, specifically flavanols, secondary metabolites responsible for the health benefits associated with the consumption of this food. For this reason, and within the framework of a circular economy, cocoa bean husk has recently been proposed as a low-cost ingredient for functional foods.

EFFECT OF VITAMIN C ON RENAL MARKERS OF OXIDATIVE STRESS INDUCED BY METHOTREXATE IN PSORIASIS

Elodia Nataly Díaz-De la Cruz¹, Walter Ángel Trujillo-Rangel², Grecia Elena Hurtado-Nuñez¹, Sandra Guadalupe Sánchez-Ceja¹ and Martha Estrella García-Pérez¹

¹Laboratory of Pharmacognosy and Experimental Pharmacology, Faculty of Químico Farmacobiología, Universidad Michoacana de San Nicolás de Hidalgo, Morelia, México ²Laboratory of Pharmacology, Tonalá University Centre, Universidad de Guadalajara, Guadalajara, México.
E-mail: nataly-diaz@live.com

Psoriasis is an incurable autoimmune dermatological disease that affects more than 3 million Mexicans. Current therapies are aimed at managing its symptoms, the methotrexate (MTX) being considered as the standard of systemic treatment in moderate and severe psoriasis. Despite its therapeutic usefulness, abandonment of therapy with this medication has been reported due to the induction of nephrotoxicity, which is related to an increase in oxidative stress in the kidneys. In this work, the impact of oral vitamin C on renal markers of oxidative stress (OS) induced by MTX is evaluated in the context of a murine model of psoriasis induced with imiquimod (IMQ). For this, C57BL/6 mice were used, in which psoriasis was induced with IMQ. Animals received MTX (20 mg/kg/day) and vitC (175 mg/kg/day) and were compared with their respective controls for 7 days. The psoriasis severity index and the animals' body weight evolution were determined. After sacrifice, the impact of the treatments on the activity of some OS biomarkers such as malondialdehyde, nitrates/nitrites, superoxide dismutase (SOD), and total antioxidant capacity in kidney homogenates were evaluated. The results showed that the concomitant treatment of MTX with vitC improves the severity of psoriasis on day 7 compared to the IMQ group, being able to restore the renal SOD activity, which was severely damaged by MTX. Given that no significant improvement was observed in the evolution of body weight or in the production of lipoperoxides at the renal level in the groups that received VitC, it is considered that this vitamin slightly counteracts the deleterious action of MTX. New investigations should be performed on other markers of oxidative stress and inflammation in the kidney to precisely estimate the protective effects of VitC on MTX-induced nephrotoxicity during psoriasis.

EFFECT OF POTENTIAL HYDROGEN ON HYDROGEN SULFIDE PRODUCTION IN *SACCHAROMYCES CEREVISIAE*

Mariana Michell García-Reyes, Elías Nieto Zaragoza, Francisco Torres-Quiroz
Instituto de Fisiología Celular, UNAM. Cto. Exterior s/n, C.U., Coyoacán, 04510, CDMX.
ftq@ifc.unam.mx, 56225703.

The vacuolar ATPase (V-ATPase) enables the vacuole to maintain a pH of approximately 5. This enzyme functions to generate electrochemical and proton gradients across the membranes of the vacuolar system. Researchers have identified a link between the vacuole and the degradation of cysteine, which releases H₂S. The elimination of each subunit that makes up the V1 subcomplex of V-ATPase resulted in low H₂S production from cysteine. However, the mechanism by which V-ATPase regulates this is still unknown. Recent results suggest that one of the factors involved is pH. Since the 1990s, it has been determined that deleting any of the genes encoding V-ATPase (VMA genes) results in the strain's inability to grow at pH levels above 7.5, similar to Δ cys4 strains.

The *VMA1* gene encodes the A subunit of V-ATPase, which contains a cysteine residue (C261) related to the catalytic activity of this multiprotein complex. We demonstrated that there are two genetic interactions between the *CYS4* gene, which encodes cystathionine β -synthase and is involved in the transsulfuration pathway, and the *VMA1* gene.

We evaluated the ability of some *Saccharomyces cerevisiae* strains to grow in media with different pH values, supplemented with cysteine or glutathione. We also performed a qualitative analysis of vacuolar acidification and staining to observe the morphology of this organelle.

The results obtained in this study indicate that there are two genetic interactions between *CYS4* and *VMA1*: one affecting the growth phenotype and the other affecting the H₂S production phenotype. We conclude that changing the pH of the medium results in the inability of strains with blocked transsulfuration pathways to grow. However, the addition of cysteine and glutathione restores the growth phenotype of these strains, suggesting a disruption in the transsulfuration pathway. Finally, we determined that deletion of the *CYS4* gene results in the complete formation of the vacuole in yeast, which is not only present but also active from the start of the cell's life.

This project was financed by UNAM-DGAPA-PAPIIT through the research program IN208922 and by the Consejo Nacional de Humanidades, Ciencias y Tecnologías through the Ciencia de Fronteras 2023 program CF-2023-1884.

EFFECT OF OXIDATIVE STRESS ON THE MRNA LEVELS OF *IL-1 β* , *IL-6*, AND *IL-10* IN THE INTESTINE OF RATS EXPOSED TO OZONE

Denisse García Sánchez, Marlen Valdez Fuentes, Lorena De la Cruz-Mendoza, Selva Rivas Arancibia

Departamento de Fisiología, Facultad de Medicina. Universidad Autónoma de México (UNAM). C.P. 04510.

Tel:+52 55 56 23 25 00; e-mail: srivas@unam.mx

Human activities emit gases, polluting particles, and ozone at concentrations higher than the World Health Organization recommends daily, which has become a public health problem. Exposure to ozone increases the formation of reactive oxygen species (ROS) in the body, leading to oxidative stress and the development of certain diseases, such as degenerative diseases. Some studies suggest that ROS, resulting from ozone exposure, may play a role in the cellular and molecular mechanisms that lead to intestinal permeability loss, activating inflammatory cascades. The chronic state of oxidative stress generates a loss of regulation of the intestinal inflammatory response, which promotes a gradual degenerative process. This work aimed to evaluate the expression of interleukins *IL-1 β* , *IL-6*, and *IL-10* in the intestine of rats. Thirty-six male Wistar rats were used and divided into six random groups: control group (exposed to ozone-free air for 30 days) and exposed for 7, 15, 30, 60, and 90 days to 0.25 ppm of ozone, respectively. The animals were anesthetized and sacrificed, and samples were collected to later be analyzed using the qPCR technique. The results were evaluated using the $2^{-\Delta\Delta CT}$ method with respect to the constitutive gene *Rps18*. The study's results revealed an increase in the control group in the expression of *IL-1 β* in the jejunum at 7, 15, and 60 days of exposure, as well as in the colon from 7 to 90 days. The *IL-10* gene in the jejunum showed significant increases from 15 to 90 days, while the same gene in the colon showed an increase after 15 days, but at 30 and 60 days it decreased compared to the control group. The *IL-6* gene in the jejunum significantly increases from 30 to 90 days. There is also an increase in the colon from 7 days to 90 days compared to the control group ($p \leq 0.05$). These results indicate the activation of inflammatory processes leading to greater intestinal permeability.

The results suggest that the activation of inflammatory processes creates a vicious cycle. This cycle includes increased intestinal permeability, leading to a loss of regulation of the inflammatory response, which disrupts intestinal balance and contributes to the development of intestinal diseases. Therefore, we conclude that repeated exposure to ozone-induced oxidative stress increases pro- and anti-inflammatory proteins. These proteins lose their regulation and promote the presence and growth of intestinal inflammatory conditions.

Acknowledgments. Financed by PAPIIT-IN204324 UNAM to S.R-A.

HYDROGEN PEROXIDE DETOXIFICATION THROUGH OF THE THIOREDOXIN SYSTEM IN THE CYSTICERCI OF *TAENIA*

Guevara-Flores, A.^{1*}; Martínez-González, J. J.¹; Vásquez-Lima, C.¹; Nava Balderas, G.²; Rendón, J.L.¹; del Arenal Mena, I.P.¹

¹Departamento de Bioquímica, FACMED, UNAM, ²Departamento de Microbiología y Parasitología, FACMED, UNAM.

*E-mail: guevarafa@bq.unam.mx

Cestodes are an exclusively parasitic class of flatworms, as *Taenia solium*, which causes significant health and economic problems. These parasites are exposed to reactive oxygen species (ROS) generated by their own metabolism and those released by the host as a defense mechanism. On the other hand, this group has in common the absence of antioxidant enzyme such as catalase (CAT), glutathione reductase (GR) and thioredoxin reductase (TrxR). Additionally, it is well known that peroxiredoxins (Prxs) and glutathione peroxidases (GPxs), the main enzymes of the thiol-dependent antioxidant systems are responsible for reducing the H₂O₂. These antioxidant systems maintain a proper redox state in cells. *Taenia crassiceps* cysticerci tolerate millimolar concentrations of this peroxide, in vivo. For this reason, we evaluate the role of Prx in its detoxification. Two genes for Prxs, identified in the genome of *Taenia solium* (TsPrx1 and TsPrx3), were cloned, the sequence suggests that both isoforms belong to the class of typical Prxs2-Cys. In addition, TsPrx3 harbors a mitochondrial localization signal peptide and two motifs (-GGLG- and -YP-) associated with overoxidation. Our kinetic characterization assigns them as thioredoxin peroxidases (TPxs). While TsPrx1 and TsPrx3 exhibit the same catalytic efficiency, instead, we found that the thioredoxin-glutathione reductase from *T. crassiceps* (TcTGR) is also involved in H₂O₂ reduction, with a lower affinity (>30-fold) in comparison with TsPrx1 and TsPrx3. The TcTGR contains a Sec residue in its C-terminal, which confers additional peroxidase activity. The aforementioned information implies that TsPrx1 and TsPrx3 are catalytically active at low H₂O₂ concentrations, and the TcTGR acts at high H₂O₂ concentrations. These results may explain why the *T. crassiceps* cysticerci can tolerate high H₂O₂ concentrations.

Acknowledgments. This work was financially supported by the DGAPA-PAPIIT IN215223.

PARTIAL PURIFICATION AND BIOCHEMICAL CHARACTERIZATION OF THIOREDOXIN REDUCTASE (TRXR) FROM THE INSECT *SHELFORDELLA TARTARA*

Ibarra-Estrada, I. E.¹; Martínez-González, J. J.; Nava Balderas, G.²; del Arenal Mena, I.P.; Rendón, J.L.¹; Guevara-Flores, A.^{1*}

¹Departamento de Bioquímica, FACMED, UNAM, ²Departamento de Microbiología y Parasitología, FACMED, UNAM.
*E-mail: guevarafa@bq.unam.mx

Worldwide, there are ~ 4,000 species of cockroaches, and although some can be considered annoying pests for humans (because they can transmit some diseases), others can also be beneficial in some aspect, for example: food for some types of pets due to their high nutritional value as is the case of *Shelfordella tartara* or as it is commonly known, the “Runner cockroach”. These insects, like other aerobic organisms are exposed to reactive oxygen species (ROS) and their enzymatic antioxidant machinery is generally robust. However, studies carried out on insects mainly focus on the biological model *Drosophila melanogaster*, where it is known that it lacks glutathione reductase (GR), but it does have the enzymatic machinery for the synthesis of glutathione (GSH). Atypically, the glutathione disulfide (GSSG) reduction necessarily relies on the thioredoxin system. In insects, thioredoxin reductase (TrxR) is responsible of the reduction of thioredoxin (Trx-S₂) and indirectly GSSG, thus maintaining redox homeostasis. In insects, TrxR is cysteine-dependent (TrxR-Cys) contrary to the isoforms present in mammals that are Sec-dependent (TrxR-Sec). In this sense, it was important to corroborate this information, and for that we were to use the insect *S. tartara* because of the amount of biomass it offers us and purify TrxR to characterize it biochemically which would be an important step in obtaining its basic information. In contrast to the purification schemes described, we got a low yield of < 10%. The electrophoresis showed a band of ~ 57 kDa, its analysis by mass spectrometry revealed a sequence of peptides that identified a TrxR. Interestingly, this TrxR can use NADH as a reducing substrate with a K_m of 2.8 μ M. In contrast, NADPH is 5 times better as a reductant with a K_m of 0.5 μ M comparable to that reported for *D. melanogaster* and for DTNB it is 700 μ M comparable to that reported for *A. gambiae*.

Acknowledgments. This work was financially supported by the DGAPA-PAPIIT IN215223.

ANTIOXIDANT EFFECT OF THE WATER: METHANOL FRACTION OF THE ETHYL ACETATE EXTRACT OF *POTENTILLA INDICA* ON RENAL MITOCHONDRIA OF RATS WITH TYPE 2 DIABETES MELLITUS

Cinthia Itzel Landa Moreno¹, Marina Murillo Villicaña¹, María de los Ángeles Rangel Rosales¹,
Jenaro Lemus De La Cruz¹, Salvador Manzo Avalos¹, Rafael Salgado-Garciglia¹ and Alfredo Saavedra Molina¹

¹Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo,
Francisco J. Múgica S/N, Morelia 58030, Michoacán, Mexico.

E-mail: 1419561g@umich.mx; 0850421k@umich.mx; 1642374d@umich.mx; 1315680c@umich.mx; smanzo@umich.mx;
rsalgado@umich.mx; francisco.saavedra@umich.mx

Multiple studies have suggested dysfunctional renal mitochondria as the main sources generating reactive oxygen species (ROS) under diabetic conditions, thus compromising the endogenous antioxidant system, which eventually leads to an oxidation-reduction imbalance, culminating in oxidative stress. This has been proposed as a determining factor in the development of diabetic kidney disease¹, a diabetic microvascular complication with marked mortality figures worldwide. An alternative therapeutic approach is the use of exogenous antioxidants from medicinal plants. *Potentilla indica* is a plant native to East Asia, which has been widely used in traditional Asian medicine due to its pharmacological effects. Previously, we determined the presence of phenolic compounds in the crude ethyl acetate extract of *Potentilla indica*², which is notable for their potent antioxidant activity. Therefore, the polar fraction of the crude extract could represent a promising treatment for the reduction of oxidative damage caused by ROS and thus improving renal mitochondria function in diabetic rats.

Firstly, the extract was obtained by cold maceration with ethyl acetate for 7 days. Subsequently, the fraction was obtained from a liquid-liquid extraction with water:methanol. The *in vitro* antioxidant activity of the fraction was evaluated through its ferric reducing potential (FRP), its anti-lipid peroxidation activity and its anti-ABTS^{•+} activity. Male Wistar rats were used as a biological model. The experimental model of type 2 diabetes was obtained from a high-fat diet and a single intraperitoneal low dose of streptozotocin. Then, the extract was administered orally for 60 days. Blood glucose levels and body weight were recorded every 15 days. At the end of the treatment, the rats were sacrificed and the renal mitochondria were isolated to evaluate oxygen consumption and mitochondrial membrane potential.

References

1. Jha, J.C.; Banal, C.; Chow, S.M.; Cooper, M.E.; Jandeleit-Dahm, K. (2016), *Redox Signal*, 25, 657–684.
2. Landa-Moreno, C.I.; Trejo-Hurtado, C.M.; Lemus-de la Cruz, J.; Peña-Montes, D.J.; Murillo-Villicaña, M.; Huerta-Cervantes, M.; Montoya-Pérez, R., . & Saavedra-Molina A. (2023), 12, 3196.

ASSESSMENT OF OXIDATION MARKERS, MAPK ACTIVATION AND THEIR POTENTIAL ASSOCIATION IN CERVICAL CANCER

Karen Larrauri^{1,2}, Lourdes Pérez Peña², Bertha León², Paola Maycotte¹

¹Centro de Investigación Biomédica de Oriente (CIBIOR), Instituto Mexicano del Seguro Social (IMSS), Atlixco, Puebla, México, C.P. 74360, ²Centro de Química, Instituto de Ciencias, Posgrado en Ciencias Químicas, Benemérita Universidad Autónoma de Puebla, México, C.P. 72000.

E-mail: paola.maycotte@imss.gob.mx, Tel: 2441023048.

Among the types of cancer (Ca) affecting women, cervical cancer (CC) is a public health problem with high global incidence and mortality rates. It is currently classified into two main subtypes according to their histological type: squamous cell carcinoma (SCC) and adenocarcinoma (AC); but there are also other less frequent cancers such as adenosquamous (AdSq) and neuroendocrine. There are several risk factors associated with the development of this pathology, being the Human Papilloma Virus (HPV) the main one. HPV infection leads to an increase in reactive oxygen species (ROS), associated with malignant transformation in several cancer types. At basal levels, ROS can function as second messengers in signalling pathways, and elevated concentrations have been linked to the overactivation of pathways such as ERK. The ERK pathway is a signalling cascade implicated in cell proliferation and differentiation, often found dysregulated in different cancer types, thus promoting malignant transformation. Consequently, several studies have proposed antioxidant supplementation or ERK inhibitors as targeted therapies. In this project, we examined basal levels of oxidation and ERK activation in patient databases and CC cell lines with varying histological types, HPV genotypes, and integrated viral genome copy number. We also explored their potential association with histological subtype and assessed the impact of antioxidant or ERK inhibition on proliferation, survival, and cell migration. Our findings demonstrated that antioxidant treatment reduced viability, migration, and ROS levels in CC cells. However, no significant differences in ROS levels were observed between histological subtypes, although a potential correlation with HPV copy number emerged. ERK activation exhibited variance among histological subtypes, but its role in regulating cell proliferation and migration varies according to cellular characteristics. Furthermore, ERK inhibition appeared to differentially affect ROS levels across CC cells. In summary, our results suggest that ROS regulate migration and viability in CC, with no discernible variance based on histological subtype. ERK activation, however, differs according to CC histological subtype and regulates cellular migration. Notably, a defined synergistic mechanism between ROS and ERK was not observed in CC cells, indicating independent pathways and variations dependent on subtype and characteristics.

CYANIDIN EFFECTS ON OXIDATIVE STRESS AND MITOCHONDRIAL BIOGENESIS IN PAE CELLS, DURING HYPOXIA-REOXYGENATION

Melani León Martínez, Ariadna Zelanda Salazar and Vicente Castrejón Téllez*

*Instituto Nacional de Cardiología Ignacio Chávez, Physiology Department. Mexico City, 14080. Mexico.
(55) 55-73-29-11 Ext. 25402. e-mail: vcastrejn@yahoo.com.mx

Diets rich in fruits and vegetables have beneficial effects on health. Anthocyanins are a group of water-soluble pigments that give the blue, purple and red color to many fruits. More than 600 anthocyanins have been described, of which 30 are the most abundant, and cyanidin represents one them. Some beneficial properties of anthocyanins include being antioxidants, anti-inflammatory, preventing mutations, cancer development, and regulating cellular enzyme functions. Has been seen a regulation on the control of free radicals in the cell. In cardiac cells, exposed to anthocyanins, it has been observed that a reduction in cytosolic cytochrome c released from the mitochondria, preventing apoptosis. Anthocyanins can participate as substrates in complex I of the mitochondrial respiratory chain, or as uncoupling agent of oxidative phosphorylation. Anthocyanins may protect the heart against ischemia-reperfusion injury. Pathways that control mitochondrial biogenesis are potential therapeutic targets for the amelioration of different diseases or process as endothelial dysfunction. We study the effect of cyanidin on oxidative stress and mitochondrial biogenesis during the hypoxia-reoxygenation process. Expression of antioxidants proteins as catalase, SOD2, UCP2, GPX4 and proteins of mitochondrial biogenesis, fusion and fission such as, PGC1 α , SIRT1, NRF2, PPAR γ , Mfn1, Opa1, Fis1 and Drp; will be determined by western blot. Free radical levels were determined by flow cytometry using the CellRox and Mitosox fluorescent indicators and, the mitochondrial membrane potential was determined by MitoTracker indicator. Preliminary results shows, increase in the expression of mitochondrial antioxidant enzymes, decrease in the levels of free radicals, indicating a regulation in mitochondrial activity, this stability in the mitochondria, was also observed by microscopy with MitoTracker, an indicator of mitochondrial transmembrane potential, and, increased in the expression of protein related with mitochondrial biogenesis. All experiments were made in two different groups, control group and hypoxia/reoxygenation group, in the presence of different concentration of cyanidin. This work may contribute in the search for natural product treatments that can replace or synergize with current pharmaceutical products and minimize pharmaceutical side effects. The PAE cell line is a well-differentiated endothelial cell line, that provides a good in vitro system to test the effects of natural compounds, on process as oxidative stress or in intracellular effects as in mitochondria.

CHARACTERIZATION OF THE CYTOSOLIC AND MITOCHONDRIAL ANTIOXIDANT SYSTEM OF *USTILAGO MAYDIS* UNDER OXIDATIVE CONDITIONS

Aranzazú López-Jaime¹, Marisol Ubaldo-Aguilar¹, Rodrigo Martínez-Acosta¹, Miguel Ángel Rosas-Paz², James González², Juan Pablo Pardo-Vázquez^{3b} and Lucero Romero-Aguilar^{3a}

¹Facultad de Ciencias Biológicas-BUAP, Av San Claudio s/n, Cd Universitaria, La Hacienda, 72592, Puebla, ²Facultad de Ciencias-UNAM, ³Facultad de Medicina-UNAM Ciudad Universitaria, Av. Universidad 3000 C.P. 04510, Ciudad de México.

E-mail: alusromaguila@bq.unam.mx, bpardov@bq.unam.mx

Ustilago maydis is a phytopathogenic fungus that infects the corn plant, causing the disease known in Mexico as *huitlacoche*. The cytosolic antioxidant systems have been studied in *U. maydis* under conditions of nutritional stress by carbon source. However, it has not been investigated the behavior of mitochondria in cells exposed to oxidizing agents. In this work, we studied the response of mitochondria in cells treated with H₂O₂. We determined the specific activity and the gene expression of the superoxide dismutase (SOD), catalase-peroxidase (CAT-Px), glutathione peroxidase (GPx), glutathione reductase (GR), and thioredoxin reductase (TrxR) in cytosolic and mitochondrial extracts in the presence and absence of H₂O₂. Compared to cytosolic activities, a decrease in the activity of SOD, GPx, GR, and TrxR in mitochondria was observed. Regarding the expression of antioxidant enzyme genes, it was determined that at low concentrations of H₂O₂, expression increased for SOD (mitochondrial and cytosolic) and peroxidase enzymes (CAT-Px and GPx). However, at higher concentrations, expression decreased for all genes, suggesting that the cell reaches an oxidative stress impossible to alleviate.

References

1. Romero-Aguilar L, Vázquez-Meza H, Guerra-Sánchez G, Luqueño-Bocardo OI, Pardo JP. The Mitochondrial Alternative Oxidase in *Ustilago maydis* Is Not Involved in Response to Oxidative Stress Induced by Paraquat. *J Fungi (Basel)*. 2022 Nov 19;8(11):1221. doi: 10.3390/jof8111221. PMID: 36422042; PMCID: PMC9693204.

EFFECT OF SELENIUM SUPPLEMENTATION ON ASEXUAL REPRODUCTION OF TAENIA CRASSICEPS CISTICERCI

Martínez-González, J. J.^{1*}; Guevara-Flores, A.¹; Herrera-Garduño, C.P.¹ & del Arenal Mena, I.P.^{1*}

¹Departamento de Bioquímica, FACMED, UNAM.

*E-mail: darenal@bq.unam.mx

Cysticercosis is caused by the larval form (cysticerci) of *Taenia solium* and is currently a public health problem in developing countries. Due to the difficulty of working directly with this parasite, the cysticerci of *T. crassiceps* has been used as a reference. This organism offers the possibility of obtaining large quantities of biological material under laboratory conditions, the only limitation being the use of mice for its propagation. Experimentally, it is possible to cultivate the cysticerci in RPMI 1640 medium (INVITROGEN); under these conditions they are maintained but do not reproduce. Supplementation of the medium with various compounds, such as hormones or other metabolites, has been tested, but it has not been possible to obtain more individuals. On the other hand, since selenium is essential for the activity of thioredoxin-glutathione reductase (TGR), which is a key enzyme in the synthesis of deoxyribonucleotides by ribonucleotide reductase (RR), and since this element is not reported in the RPMI medium, we theorize that selenium is a limiting factor in the in vitro reproduction of this parasite. Therefore, the aim of this work is to evaluate the effect of selenium supplementation on the reproduction of *T. crassiceps* cysticerci. Our results showed that the administration of sodium selenite to the medium had no effect on either cysticerci reproduction or enzyme activity. However, after oral and intraperitoneal administration of sodium selenite and selenomethionine to mice previously parasitized with the cysticerci, an increase in parasite load and TGR activity was observed. This suggests that there are other elements that the parasite incorporates from its host that are key to its reproduction.

This work was carried out with the support of DGAPA- PAPIIT IN215223.

EXPLORATORY ANALYSIS OF ANTIOXIDANT AND DETOXIFICATION ENZYMATIC SYSTEMS IN DENDROCTONUS GENUS (COLEOPTERA CURCULIONIDAE)

Martínez-González, J. J.^{1*}; Guevara-Flores, A.; Nava Balderas, G.²; Burgos Dueñas, O.³; Rendón, J.L.¹; Burgos Solorio, A.³; Armendáriz-Toledano, F.⁴; & del Arenal Mena, I.P.^{1*}

¹Departamento de Bioquímica, FACMED, UNAM, ²Departamento de Microbiología y Parasitología, FACMED, UNAM,

³Laboratorio de Parasitología Vegetal, CIB, UAEM. ⁴Departamento de Zoología, I Biología, UNAM.

*E-mail: jjmtz@bq.unam.mx; darenal@bq.unam.mx

Bark beetles of the genus *Dendroctonus* naturally inhabit pine forests in Mexico. These organisms, together with the trees they inhabit, form a complex ecological network in which cellular detoxification processes are key to the establishment and proliferation of the former and the defense of the latter. For example, Liu et al. have described the development of resistance to (+)-alpha-pinene by *D. armandi* through the activation of reactive oxygen species (ROS)-dependent signaling pathways¹. Therefore, it is of utmost importance to know the antioxidant system of these organisms as a starting point to understand the complex ecological-chemical interaction between *Dendroctonus* spp. and their hosts. Through an exploratory analysis of various databases containing the genomes available for the genus *Dendroctonus* (NCBI, WORMBASE, KEGG), a search was performed for key genes or proteins associated with redox homeostasis and detoxification processes, specifically in *D. ponderosa* and *D. valens*. The information collected was integrated to determine the architecture of the enzymatic antioxidant system of *Dendroctonus* spp. compared with that observed in a model insect (*Drosophila melanogaster*) and an endoparasitic flatworm (*Taenia solium*). Preliminary results showed that *Dendroctonus* spp., like other insects, has a peculiar antioxidant system architecture in which the thioredoxin-dependent system plays a fundamental role in the redox balance as well as its xenobiotics detoxification mechanisms.

References

1. Liu B, Tang M, & Chen H. (2022). *Int J Mol Sci*, 23, 11578.

8-OXOG LEADS TO STRESS SURVIVAL AND EVOLUTION IN *BACILLUS SUBTILIS*

Lisett Esther Martínez Magaña¹, Maria Jose Toledo Ramírez¹, Norma Ramírez¹,
Eduardo A. Robleto² and Mario Pedraza-Reyes¹

¹Department of Biology, Division of Natural and Exact Sciences, University of Guanajuato, ²Guanajuato, Mexico.
Noria Alta S/N, Guanajuato, Gto; C.P. 36050, Tel 47320006 ext. 8151.
School of Life Science, University of Nevada, Las Vegas, NV 89557, USA;
email:pedrama@ugto.mx

The guanine oxidized (GO) system of *Bacillus subtilis*, composed of the YtkD (MutT), MutM and MutY proteins, counteracts and prevent the cytotoxic and hypermutagenic effects (HPM) of the oxidized nucleobase 8-OxoG. The mechanism(s) that connect the accumulation of the mutagenic lesion 8-OxoG with the ability of *B. subtilis* to evolve and survive the noxious effects of oxidative stress were dissected in this work. Genetic and biochemical evidence indicated that the synthesis of KatA was exacerbated, in a PerR-independent manner, and that the transcriptional coupling repair factor, Mfd, contributed to H₂O₂-hypersistance (HPHR) and HPM in a Δ GO *B. subtilis* strain. In addition, these phenotypes are associated with wider pleiotropic effects, as revealed by a global proteome analysis. The inactivation of the GO system results in the upregulated production of KatA, and it reprograms the synthesis of the proteins involved in distinct types of cellular stress; this has a direct impact on (i) cysteine catabolism, (ii) the synthesis of iron-sulfur clusters, (iii) the reorganization of cell wall architecture, (iv) the activation of AhpC/AhpF-independent organic peroxide resistance, and (v) increased resistance to transcription-acting antibiotics. In summary, the results presented in this work strongly suggest that beyond promoting mutagenesis, the genetic lesion 8-OxoG impacts the ability of *B. subtilis* to adapt to HPM and withstand peroxide and antibiotic stress.

Work supported by CONAHCYT (Grants A-1S-27116 and CBF2023-2024-708); Universidad de Guanajuato (CIIC-029-2024). L.E.M.M was supported by doctoral scholarship from CONAHCYT.

EVALUATION OF THE ANTIOXIDANT ACTIVITY OF THE DAMAGE SUPPRESSION PROTEIN (DSUP) OF THE TARDIGRADE RAMAZZOTTIUS VARIEORNATUS

Mariel Alexis Quiven Feria¹, Rosa María Bermúdez Cruz¹

¹Center for Research and Advanced Studies of the National Polytechnic Institute. Department of Genetics and Molecular Biology. ¹Av. Instituto Politécnico Nacional 2508, Col. San Pedro Zacatenco, Alcaldía Gustavo A. Madero, Ciudad de México, Código Postal 07360. Apartado Postal: 14-740, 07000. Tel 55 5747 3800 ext. 5334, mariel.quiven@cinvestav.mx roberm@cinvestav.mx

Tardigrades are an extremophile species capable of withstanding various extreme environments. One of the most important discoveries of these microorganisms is the damage suppression protein (Dsup) from *Ramazzottius varieornatus*. This protein has been studied in HEK293 cells where DNA damage caused by gamma radiation was reduced with respect to the transfected control. In *E. coli* cells expressing Dsup, ionizing radiation viability was less affected when compared with empty plasmid control. However, the mechanism by which this protein can confer this protection to bacteria is unknown. It has been shown that Dsup binds DNA, so it may act as a physical barrier. In order to shed light on the mechanism by which this protein can promote bacterial viability, we searched for conserved motifs in Dsup amino acid sequence, predicted tertiary structure, and identified a HMNG nucleosomal binding site and several regions with a disorder. For the in vivo experiments, the viability of the bacteria expressing Dsup and the control under oxidative stress was carried out by quantification of colonies and a growth curve using the optical density of the bacteria culture. These results revealed that bacteria expressing the Dsup protein were able to cope better under oxidative stress conditions compared to control cells transformed with an empty plasmid. In addition, to measure oxidative environment, levels of carbonylated proteins were determined. The bacteria expressing Dsup had a much lower quantity of carbonylated proteins compared to the control. In addition to this, recombinant Dsup protein was purified and its ability to bind DNA was tested by electrophoretic mobility shift assay using a 22-bp fluoresceinated. Our results show that Dsup protein has both an antioxidant activity and a DNA binding activity, thus protecting directly from damage as well as proteins to help bacteria survive under oxidative conditions.

RESVERATROL AS AN ANTIOXIDANT POTENTIAL IN THE REGULATION OF OXIDATIVE STRESS IN THE PREFRONTAL CORTEX DURING AGING IN WISTAR RATS

Raúl Vladimir Ramírez Olmos¹, Ivan Cesar Arteaga², Lourdes Millán Pérez Peña³, José Luis Morán Perales⁴, Addí Rhode Navarro Cruz², Alfonso Díaz⁴, Daniel Juárez Serrano³

¹Facultad de Ciencias Químicas, BUAP; ²Departamento de Bioquímica – Alimentos, FCQ – BUAP; ³Posgrado en Ciencias Químicas, BUAP; ⁴Instituto de fisiología, BUAP; ⁵Instituto de Ciencias, BUAP, Posgrado en Ciencias Químicas, BUAP.

Av San Claudio S/N CP.72595, Puebla, México. Teléfono: 2211343520

Correo: danieljuarezserrano@viep.com.mx

Aging is a natural process of multifactorial origin. In recent decades, research has focused on studying physiological mechanisms such as oxidative stress, which is an imbalance between the antioxidant and oxidative systems. This stress, although essential for life, causes damage when produced in excess. It has been shown that aging increases several markers of oxidative stress in various regions of the brain. The prefrontal cortex (PFC) is a key region of study due to its executive functions: decision-making, attention, cognition, and motor functions. Resveratrol is a natural antioxidant that has demonstrated several beneficial effects; however, its role in various brain regions, such as the PFC, remains unknown. The aim of this work is to elucidate the role of resveratrol on markers of oxidative stress during aging in the prefrontal cortex of Wistar rats. Forty-five three-month-old male Wistar rats were used. The rats were divided into three groups: Control (CTRL) (no treatment), Vehicle (VEH) (7.5% ethanol), and Resveratrol + Vehicle (RSVL + VEH) (10 mg/kg/day resveratrol + 7.5% ethanol), with 15 animals in each group. A further subdivision was performed into three groups of 15 animals each, corresponding to the evaluated ages of 5, 14, and 24 months. At the end of the administrations, the animals were euthanized, the brain was removed, and the prefrontal cortex was dissected, followed by biochemical tests (MDA + 4-HDA, ROS, and NO₂). Our results showed that resveratrol treatment significantly improved oxidative stress markers (MDA, ROS, NO₂) compared to the control group and vehicle group in all three periods evaluated, demonstrating its antioxidant capacity. In conclusion, resveratrol reduces oxidative stress markers produced during aging in the prefrontal cortex of Wistar rats. Taken together, these results suggest that resveratrol could be a useful molecule to prevent damage caused by oxidative stress associated with brain aging.

IN VITRO STUDIES OF IRON CHELATING CAPACITY AND ANTIOXIDANT ACTIVITY OF THE ETHANOLIC EXTRACT OF *ERYNGIUM CARLINAE*

María de los Ángeles Rangel Rosales, Marina Murillo Villicaña, Cinthia Itzel Landa Moreno, Jenaro Lemus De La Cruz, Rafael Salgado-Garciglia, Alfredo Saavedra Molina

Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Morelia 58030, Mich, México.

Alfredo Saavedra-Molina, saavedra@umich.mx

Iron overload is a group of heterogeneous diseases that can be either hereditary or acquired, characterized by an excessive accumulation of iron in the body. This condition is linked to an increase in the generation of free radicals, and the development of serious complications in vital organs, such as liver, heart, kidney, neurodegenerative diseases, leading to cell death among others. Currently, the management of iron overload primarily relies on therapeutic approaches such as phlebotomy, which involves the removal of blood, dietary iron restriction, and iron chelation therapy. Although effective, these therapies can have limitations, including side effects and inadequate targeting of tissues, highlighting the need for alternative and more specific therapeutic options. Flavonoids have demonstrated the ability to form stable complexes with iron ions, as well as antioxidant activity, creating a synergy. This ability makes flavonoids and other polyphenols promising candidates as natural iron chelators, known as phyto-chelators, for potential therapeutic use in iron overload disorders. Studies conducted by our research team have shown that extracts from the plant *Eryngium carlinae* contain a rich variety of secondary metabolites, such as flavonoids, phenolic acids, and terpenoids. In traditional medicine, *Eryngium carlinae* has been widely used by various cultures due to its therapeutic properties. The maceration was performed at a ratio of 1:10 plant to 96% ethanol and kept at 4°C for 5 days. The chelating activity was evaluated using the ferrozine colorimetric assay and the iron-reducing activity using the ferric reducing power (FRP) assay of the ethanolic extract of *Eryngium carlinae*. The IC₅₀ was also determined using the DPPH radical and the ABTS cationic radical. The ethanolic extract of *Eryngium carlinae* exhibited significant iron-chelating activity. Additionally, its antioxidant activity was confirmed by an effective IC₅₀ determination.

EFFECT OF PCL/F-68 NANOPARTICLES LOADED WITH CURCUMIN ON THE VIABILITY OF TAENIA CRASSICEPS CYSTICERCI (CESTODA)

Reyes-García, L.; Guevara-Flores, A.¹; Nava Balderas, G.²; del Arenal Mena, I.P.^{1*}, Leyva-Gómez, G.³; Del Prado-Audelo, M.L.⁴; & Martínez-González, J. J.^{1*}

¹Departamento de Bioquímica, FACMED, UNAM, ²Departamento de Microbiología y Parasitología, FACMED, UNAM, ³Departamento de Farmacia, Facultad de Química, UNAM. ⁴Departamento de Tecnologías Sostenibles e Ingeniería Civil, Escuela de Ingenierías y Ciencias, TEC de Monterrey.
*E-mail: jjmtz@bq.unam.mx; darenal@bq.unam.mx

Curcumin (CUR) is a complex and unstable molecule whose antiparasitic use has started to be studied due to its ability to interfere with key physiological processes in parasites. Previously, we demonstrated that CUR exerts a dose- and time-dependent lethal effect on *Taenia crassiceps cysticerci* under *in vitro* conditions¹. This effect is attributed partly to the inhibition of the enzyme thioredoxin glutathione reductase (TGR) which results in an increase in the generation of reactive oxygen species (ROS). This leads to the death of the parasite due to oxidative stress. As part of this mechanism, it has been observed that the metabolites responsible for the biological effect of CUR appear by autooxidation of this molecule when it passes into an aqueous medium². This would make its pharmaceutical administration to be difficult. The objective of this study is to evaluate the efficacy of a nanoformulation designed to stabilize CUR in aqueous media, thereby facilitating its pharmaceutical administration. For this purpose, nanoparticles (NPs) of the biopolymer polycaprolactone (PCL) plus the surfactant Pluronic-F68, loaded with CUR (PCL/F68-CUR), were prepared³. The NPs were administered to *T. crassiceps cysticerci* under maintenance conditions for 24, 48, 72, and 96 h. The results indicated that PCL/F68-CUR significantly delayed the onset of the lethal effect of CUR on the parasites compared to *in vitro* administration of CUR alone. This opens the possibility of evaluating the effect of pharmacological administration of PCL/F68-CUR on the parasite load of parasitized hosts.

References

1. Martínez-González JJ, Ríos-Morales SL, Guevara-Flores A, Ramos-Godínez MDP, López-Saavedra A, Rendón JL, Del Arenal Mena IP. (2022) *Exp Parasitol*, **239**, 108319.
2. Guevara-Flores A, Martínez-González JJ, Herrera-Juárez ÁM, Rendón JL, González-Andrade M, Torres Durán PV, Enríquez-Habib RG, Del Arenal Mena IP. (2019) *PLoS One*, **14**, e0220098.
3. Del Prado-Audelo ML, Caballero-Florán IH, Meza-Toledo JA, Mendoza-Muñoz N, González-Torres M, Florán B, Cortés H, Leyva-Gómez G. (2019) *Biomolecules*, **9**, 56.

EVALUATION OF ANTIOXIDANT CAPACITY OF VARIOUS MORINGA OLEIFERA BASED HERBAL PRODUCTS USING CYTOCHROME C

Carlos Alejandro Rocha Martinez¹, Mónica Andrea Valdez Solana¹, Miguel Aguilera Ortiz¹, Erick Sierra Campos¹

¹Universidad Juárez del Estado de Durango, Av. Artículo 123 s/n Fracc, Filadelfia, 35019 Gómez Palacio, Dgo. Mexico.

E-mail: ericksier@ujed.mx

Determining the antioxidant capacity of natural compounds and formulations is fundamental to understanding their biological impact. Although ABTS and DPPH are widely used, their lack of physiological relevance limits the interpretation of results. Therefore, it is necessary to look for other redox alternatives such as oxidized cytochrome c, a key protein in the mitochondrial transport chain, is proposed as a redox probe to evaluate the antioxidant capacity of food, pharmaceutical and natural products, something not previously reported despite knowledge of its redox properties. *Moringa oleifera* has demonstrated high antioxidant capacity due to its phytochemicals, flavonoids, and polyphenols including chlorogenic acid, syringic acid, luteolin, rutin, isoquercetin, and astragalin¹. In this study, the cytochrome c assay against ABTS and DPPH was compared. To prepare the extract, 1 g/mL of herbal product based on *M. oleifera* was mixed with 80% ethanol, sonicated, filtered and concentrated in a rotary evaporator. In DPPH, Vidanat showed the highest percentage of inhibition (89.9%), followed by Akuanandi (77.6%), Natura CE (65.3%), VitHerbal (74.6%), and Herbonaturista (74.3%). In ABTS, Akuanandi showed the highest inhibition (91.8%), followed by Vidanat (91.65%) and BroncoGol (85.49%). The ABTS and DPPH assays showed a correlation greater than 0.965 (R^2) for the tests with Akuanandi and BroncoGol. To validate bovine cytochrome c as a probe, its response to reference antioxidants was evaluated by comparing the oxidized spectrophotometric profile with the reduced one, showing spectral changes indicative of heme iron reduction (Fe^{3+} to Fe^{2+}). With trolox and ascorbic acid, a shift in the Soret peak (410 to 415nm) and an increase in the absorbance of characteristic Q bands of reduced cytochrome c were observed, increasing proportionally to the concentration used. The extracts also showed a reduction profile, with Vidanat and Akuanandi inducing the greatest spectral changes, consistent with their high antioxidant capacity demonstrated in ABTS and DPPH assays. The Akuanandi extract (70 μ g/mL) was equivalent to 0.214 mM trolox and 1.71 mM ascorbic acid. Although the cytochrome c results do not directly correlate with those obtained by DPPH and ABTS, it was shown that cytochrome c can be used as a redox probe to analyze antioxidants that can be directed to the mitochondria, complementing the general information from ABTS and DPPH.

DIFFERENTIAL CHANGES IN REDOX STATE INDUCED BY SENOLYTIC AND SENOMORPHIC TREATMENTS IN THE SERUM OF MIDDLE-AGED FEMALE RATS DURING CHRONIC OBESITY

Verónica Salas-Venegas¹, Roberto Santín-Márquez¹, Pamela Flores-Torres², Ángeles Fortis-Barrera¹, Raúl Librado-Osorio³, Norma E. López-Díazguerrero¹, Armando Luna-López³, Mina Königsberg^{1*}

¹Departamento de Ciencias de la Salud. Universidad Autónoma Metropolitana Unidad-Iztapalapa. ²Posgrado en Biología Experimental, Universidad Autónoma Metropolitana Unidad Iztapalapa. ³Departamento de Investigación Básica, Instituto Nacional de Geriátría.

*mkf@xanum.uam.mx

Obesity is an increasing problem worldwide linked to various comorbidities, with a higher prevalence in women. Recently, adipose tissue has been recognized not just as a simple energy storage tissue, but also as an active endocrine organ releasing a range of molecules known as cytokines. Due to the complex interactions between cytokines, obesity is also marked by chronic low-grade inflammation with consistently elevated oxidative stress. Cellular senescence is a response to oxidative stress levels, and recent evidence shows that various cell types are highly susceptible to becoming senescent. Senescence is linked to inappropriate expansion (hypertrophy) of adipocytes, insulin resistance, dyslipidemia, and various diseases. Significant efforts have been made to identify methods to eliminate senescent cells, including the use of “senolytic and senomorphic” compounds. The most established senolytic treatment so far is the combination of dasatinib and the antioxidant quercetin (D+Q). Sulforaphane (SFN), one of the most potent phase II enzyme inducers isolated from edible cruciferous vegetables, is a strong activator of the Nrf2-Keap1 signaling pathway. This activation allows Nrf2 to escape Keap1-dependent degradation, leading to the stabilization and nuclear accumulation of Nrf2.

Our aim was to determine how the treatments with D+Q and SFN can affect the systemic redox state in an obesity model using middle-aged female Wistar rats. In this study, we used female Wistar rats fed a hypercaloric diet (HD) from 21 days after birth until they were euthanized at 14 months of age. SFN 5 days a week for two months, while D+Q were once a month for two months. At 14 months of age, the rats were euthanized, and we collected blood samples, separating the serum and plasma. In the serum samples, we evaluated oxidized glutathione and reduced glutathione (GSH/GSSG). We found an increase in GSH levels with SFN administration, and the administration of D+Q and SFN decreased the oxidized glutathione levels in obese animals.

We thank Dr. Guerrero-Aguilera from UAM-I for animal supply. This work was supported by CONACyT grant FORDECYT-PRONACES/263957/2020T.

EVALUATION OF THE PHOTO-PROTECTION AND REPAIR EFFECTS EXERTED BY THE CAROTENOIDS (LUTEIN, ZEAXANTHIN AND β -CAROTENE) AGAINST PHOTODAMAGE CAUSED BY UV RADIATION IN HUMAN KERATINOCYTES”

Judith Santos-Ceseña¹, Laura C. Berumen¹, Eloy Rodríguez-de-Leon¹,
Guadalupe García-Alcocer¹, Irasema Mendieta¹

¹Posgrado en Ciencias Químico-Biológicas, Facultad de Química, Universidad Autónoma de Querétaro, Las Campanas S/N, C.P. 76010, Santiago de Querétaro, Qro.

E-mail: jsantos30@alumnos.uaq.mx, alicia.mendieta@uaq.mx

The skin, is the largest organ in our body, fulfills multiple functions of great importance, including being the biggest physiological barrier; it help us to reduce the risks of the environment, such as: cold, heat, shocks, chemical products, pollution, UV radiation, among others. However, the chronic exposure to any of these elements may increase the risk of skin diseases; including photodamage, cutaneous melanoma and nonmelanoma skin cancer (NMSC). NMSC is a global health problem, it ranks 5th place in incidence and 22nd in mortality worldwide in both sexes¹.

Chronic exposure to UV radiation is one of the biggest risk factors for the skin damage, the effects includes DNA damage by UVB and lipid and protein peroxidation by reactive oxygen species². Many authors have tried to find a new alternative to protect the skin from damage caused by UV radiation. For example, the photoprotective effect of donkey milk³, *Saussurea involucreta* polysaccharide⁴, and modified Qing'e formula⁵, have been studied. In addition, the carotenoids such as zeaxanthin, lutein, lycopene, β -carotene, and astaxanthin have been shown to have a photoprotective effect when administered orally.

The aim of this project is to evaluate the photoprotective and reparative effect carotenoids (lutein, zeaxanthin and β -carotene) on human skin keratinocytes (HaCaT) once exposed to UVB radiation. To this end, three methodologies are used. The first methodology is MTT, in order to observe cell viability, the second methodology is cell-tox, this one with the purpose of evaluating cell apoptosis, and finally DCFH-DA is used with the aim of determining the reactive oxygen species present in cell cultures with treatments once exposed to UVB radiation.

The results showed a decrease in the viability of HaCaT cells after 30 min of UVB radiation. However, an increase in cell viability was observed when exposed to lutein while the cells were irradiated with UVB. This could indicate a photoprotective effect of the carotenoid lutein. On the other hand, our results showed that when adding β -carotene after exposure, an increase in cell viability was observed. This suggests that β -carotene may have restorative properties against the damage caused by UVB radiation.

These data suggest that the photoprotective effect of carotenoids, studies to determine the mechanisms involved in this process are carried out.

References

1. Ferlay J, Ervik M, Lam F, Laversanne M, Colombet M, Mery L, Piñeros M, Znaor A, Soerjomataram I, Bray F. (2024). GLOBOCAN
2. Brand RM, Wipf P, Durham A, Epperly MW, Greenberger JS, Faló LD Jr. (2018). *Frontiers in Pharmacology*, vol. 9.
3. Li, A., He, H., Chen, Y., Li, F., Tang, J., Li, L., Fan, Y., Li, L., & Xiong, L. (2023). *Frontiers In Nutrition*, vol. 10.
4. Wang, L., Yang, K., Jing, R., Zhao, W., Guo, K., Hu, Z., Liu, G., Xu, N., Zhao, J., Li, L., & Gao, S. (2023). *Frontiers In Pharmacology*, vol. 14.
5. Zhu, S., Qin, W., Liu, T., Liu, T., Ma, H., Hu, C., Yue, X., Yan, Y., Lv, Y., Wang, Z., Zhao, Z., Wang, X., Liu, Y., Xia, Q., Zhang, H., & Li, N. (2022). *Frontiers In Pharmacology*, vol. 13

XANTHINE OXIDASE-REGULATION OF 8-OXOG-PROMOTED *BACILLUS SUBTILIS* FITNESS

Maria Jose Toledo Ramírez¹, Lissett Esther Martínez Magaña¹ and Mario Pedraza-Reyes¹

¹Department of Biology, Division of Natural and Exact Sciences, University of Guanajuato, Guanajuato, Mexico. Noria Alta S/N, Guanajuato, Gto; C.P. 36050, Tel 47320006 ext. 8151.
email:pedrama@ugto.mx

Reactive oxygen species (ROS) impact the nucleic acids generating a diversity of lesions, including DNA oxidized bases and strand breaks; however, guanine is more prone to oxidation due to its low redox potential. The resulting 8-hydroxyguanine (8-OxoG) lesion, which can also be incorporated into DNA from the 8-Oxo-dGTP precursor, is highly problematic for cells due to its mutagenic and cytotoxic properties. In *Bacillus subtilis*, the mutagenic effect of this lesion is counteracted by the proteins MutT(YtkD), MutM, and MutY, altogether called the guanine oxidized (GO) system. A recent report revealed that the genetic disruption of the GO system and the consequent accumulation of 8-OxoG, confers adaptive advantages to this bacterium, including hyperresistance to peroxides (HPRP) and transcription-acting antibiotics. Of note, the HPRP phenotype exhibited by the GO-deficient strain was independent of PerR, a repressor which regulates the oxidative response in *B. subtilis*. Therefore, this work was aimed to explore if the oxidative status produced during hydroxylation of hypoxanthine and xanthine by the NAD-dependent oxidoreductase Xanthine Oxidase (XO) underlies the HPRP phenotype of the Δ GO strain. To this end, the entire *pUC-ABCD* operon, encoding XO, was deleted from genetic backgrounds proficient and deficient for the GO system. The impact in mutagenesis and survival to oxidizing agents of the resulting strains was further tested. Results revealed that the genetic inactivation of the XO operon impacted negatively the HPRP phenotype and hypermutagenesis of the GO deficient strain. Overall, our results strongly suggest that the catabolism of xanthine and hypoxanthine are implicated in the ability of 8-OxoG to promote stress survival and evolution in *Bacillus subtilis*.

Work supported by CONAHCYT (Grants A-1S-27116 and CBF2023-2024-708); Universidad de Guanajuato (CIIC-029-2024). L.E.M.M was supported by doctoral scholarship from CONAHCYT.

PARTIAL PURIFICATION OF THIOREDOXIN REDUCTASE (TRXR) FROM *DENDROCTONUS VALENS*

Vásquez-Lima, C.¹; F. Castellanos López¹; Martínez-González, J. J.¹; Armendáriz-Toledano, F.²; Burgos Dueñas, O.³; Burgos Solorio, A.³; del Arenal Mena, I.P.¹; Guevara-Flores, A.^{1*}

¹Departamento de Bioquímica, FACMED, UNAM. ²Departamento de Zoología, Instituto de Biología, UNAM. ³Laboratorio de Parasitología Vegetal, CIB, UAEM.

*E-mail: darenal@bq.unam.mx; guevarafa@bq.unam.mx

In Mexico, one of the main agents of forest degradation is caused by bark beetles, among them we find *Dendroctonus valens*, which in areas of Mexico and Central America has been able to kill trees in perfect health. Among the strategies used to combat these beetles is the use of natural insecticides, including Neem, however, biochemical strategies have not been used to combat beetles. Insects, like other aerobic organisms, are exposed to reactive oxygen species (ROS), and it is inferred that they may have an antioxidant system similar to that of *Drosophila melanogaster* and *Dendroctonus ponderosae*. Performing a search in the PUBMED database (<https://pubmed.ncbi.nlm.nih.gov/>), we found that two insects phylogenetically close to bark beetles, *D. melanogaster* and *D. ponderosae*, lack glutathione reductase (GR) and thioredoxin glutathione reductase (TGR), but thioredoxin reductase (TrxR) is present. Therefore, this last enzyme is responsible for keeping thioredoxin (Trx-S₂) and indirectly glutathione disulfide (GSSG) in their reduced state, maintaining their redox homeostasis. In this work we aim to find a biochemical strategy to combat the beetle, in this sense, TrxR could be a target enzyme for the use of drugs, due to the role it plays within insects (taking *D. melanogaster* as a model insect), comparable to what occurs with TGR present in parasites of the Cestoda and Trematoda classes within the group of flatworms. Therefore, we consider that purifying and characterizing TrxR from *D. valens* would allow us to find, in the future, a treatment based on the inhibition of this enzyme without affecting its counterpart, the tree. The purification of the enzyme shows a low yield < 0.3 %, and an optimal pH of 7.0. A bioinformatic analysis between the TrxR of *D. valens* and that of *Pinus spp.*, its host, showed considerable differences such as i) a low identity (~ 22.4 %) in its sequences, ii) a molecular weight per subunit of ~ 57 kDa and 35 kDa, respectively, iii) two catalytic redox sites in the former, and only one in the latter; and by bibliographic information iv) different kinetic models. Therefore, we consider that TrxR from *D. valens* is an enzyme that could be a pharmacological target against bark beetles.

Acknowledgments. This work was financially supported by the DGAPA-PAPIIT IN215223.

ELUCIDATION OF A DISA-INDEPENDENT CHECKPOINT MECHANISM DURING GERMINATION/OUTGROWTH OF *BACILLUS SUBTILIS* SPORES

Alejandra Rangel-Mendoza¹, Rocío del Carmen Barajas-Ornelas¹,
Estefanía Ramírez Meraz¹ & Mario Pedraza-Reyes¹

¹Department of Biology, Division of Natural and Exact Sciences, University of Guanajuato, Guanajuato, Mexico.
Noria Alta S/N, Guanajuato, Gto; C.P. 36050, Tel 47320006 ext. 8151;
E-mail: pedrama@ugto.mx

Under conditions inappropriate for growth, *Bacillus subtilis* activates the synthesis of spores, which can remain dormant for undefined periods of time until environmental conditions support vegetative growth, through a process termed germination/outgrowth (G/O). During this developmental stage, the entrance of water and the activation of aerobic metabolism activates the production of oxygen radicals (ROS) that impact the spore's nucleic acids. The BER system processes oxidative DNA damage during spore G/O employing enzymes with AP-endonuclease (APE) activity. *B. subtilis* possesses distinct APEs, including Nfo, ExoA, and Nth. Interestingly, disruption of *nfo* and *exoA* delays G/O, and this phenotype is suppressed following *disA* disruption, thus unveiling a checkpoint role for the DNA damage-scanning protein DisA. This work is aimed to advance our understanding regarding the downstream repair processes following DisA-induced halt of transcription/replication during spore's return to vegetative growth. Employing a combination of molecular, genetic and microscopy analyses, we report here that: 1) In reference to wild type (WT) spores, during outgrowth, the spontaneous and H₂O₂-promoted mutagenesis increase in spores deficient for Nfo/ExoA/Nth but decrease following disruption of *disA* in this genetic background. 2) During outgrowth, the number of the ROS-promoted lesion 8-OxG decreased in WT spores, whereas spores lacking AP endonucleases and DisA, retained a greater number of such lesions, being more abundant in *nth/nfo/exoA/disA*-deficient spores. These results strongly suggest that i) DisA promotes low-fidelity repair to eliminate ROS-promoted lesions; ii) Oxidative DNA damage activate the checkpoint function of DisA; iii) DisA together with Nfo, ExoA and Nth process these genetic lesions, and iv) DisA may recruits repair proteins to eliminate DNA lesions of oxidative nature. 3) Further results revealed that the genetic inactivation of *nth* in the *nfo/exoA* genetic background generates a delay in the germination/outgrowth process and disruption of *disA* does not alleviate this phenotype. Kinetics of G/O and analyses of chromosome replication during G/O, in spores deficient for APs and DisA, suggested that intermediates products of Nth repair activate DisA-independent checkpoint mechanism(s). Future work is aimed to identify the factors, in addition to DisA, that inspect the status of the chromosome before granting transcription/replication and an efficient spore's return to vegetative growth.

Work supported by CONAHCYT (Grants A-1S-27116 and CBF2023-2024-708); Universidad de Guanajuato (CIIC-029-2024). A.R.M. and R.C.B.O were supported by doctoral and postdoctoral scholarships from CONAHCYT, respectively.

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex of molecules, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

SIGNAL TRANSDUCTION

IDENTIFICATION OF NOVEL C-DI-AMP BINDING PROTEINS THAT REGULATE MUTAGENESIS AND DNA REPAIR IN *BACILLUS SUBTILIS*

Karen Abundiz-Yáñez¹, Víctor M. Ayala-García² and Mario Pedraza-Reyes¹

¹University of Guanajuato, Division of Natural and Exact Sciences, Noria Alta, s/n., col. Noria Alta, Guanajuato, Gto., 36050.

²Juárez University of Durango State, Faculty of Chemical Sciences.

Av. Veterinaria S/N. Circuito Universitario. Col. Valle del Sur, Durango, Dgo. C.P. 34120.

E-mail: pedrama@ugto.mx

To deal with stressful conditions in highly competitive environments, bacteria activate distinct stress responses frequently modulated by signaling pathways. Cell signaling in Gram-positive bacteria employs the second messenger cyclic di-adenosine monophosphate (c-di-AMP) to regulate processes like ion transport, cell wall homeostasis, antibiotic resistance, DNA damage detection, and protection against genotoxic compounds. Most recently, our group reported that during growth, the c-di-AMP synthetases (DACs) CdaA and DisA, counteract *B. subtilis* spontaneous and mitomycin-C-induced mutagenesis, whereas a divergent function was attributed to these proteins in hydrogen peroxide-induced mutagenesis. In contrast, under starving conditions, CdaA and DisA, were found to promote mutations that allowed *B. subtilis* to escape from growth-limiting conditions. These results unveiled a novel function for c-di-AMP in modulating mutagenesis during the life cycle of *B. subtilis*. However, the downstream factors that c-di-AMP employs to exert these effects have remained elusive. Currently, the described targets for c-di-AMP are limited to potassium and magnesium transporters that cannot explain the multiple physiological roles attributed to this signaling molecule, including mutagenesis and DNA repair. Based on structural comparisons and molecular docking analyses, we report here the identification of twelve novel *B. subtilis* proteins that possess a specialized domain (CBS) to interact with c-di-AMP. Based on their predicted functions, three of these proteins playing presumptive roles on, *i*) the general stress response, *ii*) transcriptional regulation, and *iii*) oxidoreduction processes, were selected to investigate its contribution to growth-associated (GAM) and stationary phase-associated (SAM) mutagenesis as well as to corroborate its ability to interact with c-di-AMP. In summary, this work will advance our understanding regarding the mechanisms employed by c-di-AMP in modulating mutagenic processes and promote genetic diversity in the spore-former bacterium *B. subtilis*.

Work supported by CONAHCYT (Grants A-1S-27116 and CBF2023-2024-708); Universidad de Guanajuato (CIIC-029-2024). K.A.Y was supported by a doctoral scholarship from CONAHCYT.

REGULATION OF THE ACTIVITY OF INSULIN-LIKE GROWTH 1 RECEPTOR BY ESTRADIOL IN BREAST CANCER CELL MCF-7

Esteban Acosta-Ramos¹, Judith Hernández-Aranda¹,
Mariana Segovia-Mendoza², Jesús Alberto Olivares-Reyes¹

¹Laboratory of Signal Transduction, Department of Biochemistry, CINVESTAV-IPN,
San Pedro Zacatenco, 07360, Mexico City, MEXICO.

²Department of Pharmacology, School of Medicine, UNAM, Coyoacan, 04510 Mexico City, MEXICO.

Breast cancer (BC) is the most common neoplasia in women around the world. In 2022 it registered 2.3 million new cases and 670,000 deaths by BC. In Mexico the national incidence was 27.64 for 100,000 women in 2023, and 23790 deaths, setting as a health issue (Arnold et al. 2022). BC is a heterogeneous disease with diverse histological and molecular characteristics (Harbeck et al. 2019). Molecular classification is of particular interest for the establishing each patient treatment and prognosis survival of (Horvath 2021). One of the most common subtypes of BC is the positive estrogen receptor characterized by the overexpression of the nuclear estrogen receptor alpha (ER α), which has been well studied. However, a second type of estrogen receptor belonging to the G protein-coupled receptor superfamily, named G protein-coupled estrogen receptor 1 (GPER), has been identified. The actions and functions of GPER have not been fully understood and they represent a new field of research interested. One example is the work of Sukocheva and colleagues in 2006 (Sukocheva et al. 2006), which reported that estradiol (E2) could activate the epidermal growth factor receptor (EGFR), through his receptors ER α and GPER, in the breast cancer cell line MCF-7, but the molecular mechanism has not fully elucidated. In the laboratory we observed that E2 can activate the activation of the insulin-like growth factor 1 receptor (IGF-1R), a central receptor that mediates different roles in the development, progression and resistance in BC positive to ER. The stimulus of E2 10 nM during 15 min led to the activation of the IGF-1R; we also detected ERK-1/2 and Akt activation, proteins involved in the signaling cascade of the receptor. Even, we found the participation of ER α and GPER in activating the IGF-1R, ERK1/2, and Akt, using the antagonism ICI182,780 and G15 for each receptor. We determined that protein Src is involved in the molecular mechanism of the activation of the IGF-1R by E2. However, the full mechanism has not been elucidated and is in continuous research.

Funding. This project was supported by CINVESTAV-IPN, and EA-R hold a CONAHCYT graduate scholarship (1073659).

Reference

1. Arnold, Melina, Eileen Morgan, Harriet Rumgay, Allini Mafrà, Deependra Singh, Mathieu Laversanne, Jerome Vignat, et al. 2022. "Current and Future Burden of Breast Cancer: Global Statistics for 2020 and 2040." *Breast* 66 (December):15–23. <https://doi.org/10.1016/j.breast.2022.08.010>.
2. Harbeck, Nadia, Frédérique Penault-Llorca, Javier Cortes, Michael Gnant, Nehmat Houssami, Philip Poortmans, Kathryn Ruddy, Janice Tsang, and Fatima Cardoso. 2019. "Breast Cancer." *Nature Reviews Disease Primers* 5 (1). <https://doi.org/10.1038/s41572-019-0111-2>.
3. Horvath, Eleonora. 2021. "Molecular Subtypes of Breast Cancer-What Breast Imaging Radiologists Need to Know." *Rev Chil Radiol.* Vol. 27.
4. Sukocheva, Olga, Carol Wadham, Andrew Holmes, Nathaniel Albanese, Emily Verrier, Feng Feng, Alex Bernal, et al. 2006. "Estrogen Transactivates EGFR via the Sphingosine 1-Phosphate Receptor Edg-3: The Role of Sphingosine Kinase-1." *Journal of Cell Biology* 173 (2): 301–10. <https://doi.org/10.1083/jcb.200506033>.

α_{1A} -ADRENERGIC RECEPTOR FUNCTION: ROLES OF IL3 AND CTERM PHOSPHOSITES

Rocío Alcántara Hernández; Hernández-Espinosa, David A.;
García-Merino, Gabriela; Solís, K. Helivier and García-Sáinz, J. Adolfo

Instituto de Fisiología Celular, Circuito exterior, Ciudad Universitaria.
Universidad Nacional Autónoma de México, CDMX 04510.

Alpha-1A-adrenergic receptors (α_{1A} -ARs) are G-protein coupled receptors (GPCR) that participate in many responses, such as vasoconstriction, proliferation, and metabolism, to mention a few. α_{1A} -ARs activate the phospholipase C, generating diacylglycerol and IP3, which releases calcium from the endoplasmic reticulum, activates protein kinase C (PKC), and the MAP Kinase pathway (ERK1/2). Human α_{1A} -ARs have more than 18 phosphorylation sites at the intracellular loop 3 (IL3) and the carboxyl terminus (Cterm). Interestingly, in silico analysis indicates that IL3 phosphorylatable residues are mainly GRK substrates, whereas Cterm residues are mainly PKC substrate sites. In this work, we obtained four inducible cell lines expressing similar receptor amounts with all these receptor mutants (substituting found phosphosites by non-phosphorylatable amino acids) (IL3, Cterm, and IL3/Cterm) and the wild-type. We studied different cellular responses to determine the role of phosphosites in receptor function, including phosphorylation, desensitization, changes in intracellular calcium concentration, internalization, β -arrestin colocalization, and ERK1/2 activation. Data showed that IL3, Cterm, and IL3/Cterm mutants showed markedly reduced agonist-induced phosphorylation compared to the wild-type receptor. Preliminary evidence indicates that some sites modulate intracellular calcium concentration and receptor internalization, whereas others seem involved in ERK 1/2 activation.

Research partially supported by CONAHCYT (Fronteras 6676) and DGAPA (IN201924)

THROMBIN-INDUCED NF κ B ACTIVATION IN RETINAL PIGMENTED EPITHELIUM CELLS (RPE)

Daniela Alejandra Alvarado Fernández, Irene Lee Rivera,
Edith Catalina López Hernández, Ana María López Colomé

Instituto de Fisiología Celular, UNAM, Circuito Exterior s/n, Ciudad Universitaria, México City. C.P. 04510, México,
Tel.: +52 55 5622 5517; acolome@ifc.unam.mx

Thrombin is a serine/threonine protease, which contributes to the inflammatory process and induces the transformation, proliferation, and migration of retinal pigmented epithelium cells (RPE). Previous work in the lab suggests that the nuclear factor kappa light chain enhancer of activated B cells (NF κ B) plays a major role in these processes. This is an ubiquitous transcription factor known for its role in the regulation of inflammation, proliferation, differentiation, and development. In particular, since thrombin is a pro-inflammatory factor, this work aims to determine thrombin-induced molecular mechanisms involved in NF κ B activation in RPE cells.

The NF κ B activation and translocation from the cytoplasm to the nucleus is dependent on the removal of I κ B, which is bound to NF κ B dimers. In response to stimulation, I κ B is phosphorylated and releases NF κ B, which upon translocation to the nucleus acts as a transcription factor. In previous studies, it has been shown that thrombin induces NF κ B activation, via PI3K/Akt, cSrc and G α q(PLC/MAPK) pathways in epithelial-like 293T cells, lung epithelial cells A549 and human traqueal smooth muscle cells, respectively. In RPE cells, however, these mechanisms have not been elucidated. As this transcription factor is crucial for the inflammatory processes such as cytokine secretion and cell proliferation in response to injury, the objective of this work is to determine the thrombin-activated signaling pathways that induce NF κ B activation in the retinal pigmented epithelium cells (RPE). For this purpose, the Western Blot technique was used. We observe the maximum level of I κ B phosphorylation at 15 minutes, and degradation appears to peak 30 minutes after stimulation. We determined that thrombin activates PAR1 specifically, since inhibition by SCH 79797, a PAR 1 agonist inhibitor, ablates thrombin-induced I κ B phosphorylation. We also found that I κ B phosphorylation is dependent on phospholipase C, PI3K activation and requires intracellular calcium, by using pharmacological tools.

In conclusion, thrombin induces I κ B phosphorylation, promotes its degradation, and possibly promotes NF κ B activation in a specific manner through phospholipase C, PI3K and intracellular calcium in retinal pigmented epithelium cells (RPE). The determination of the molecular mechanism regulating NF κ B activation, can be important for developing future therapeutic treatments for diseases involving this transcription factor and its subsequent role in inflammation of the retina.

This work was funded by PAPIIT-UNAM project IN201223.

EFFECT OF PROINFLAMMATORY CYTOKINES ON THE EXPRESSION OF MOLECULAR MARKERS OF THE CANCER STEM CELL PHENOTYPE IN PC3 CELLS

Osiris Evelyn Aparicio Carrillo, Rubén Ávalos López, María Julieta López Cuevas, Eduardo Monjaraz Guzmán

Laboratorio de Neuroendocrinología. Instituto de Fisiología-BUAP.

Prolongación de la 14 sur 6301 Ciudad Universitaria Puebla Puebla.

Cel:2212734263

Correo: carrilloosiris@gmail.com

All body tissues are derived from stem cells. These cells are defined by their ability to undergo self-renewal, as well as to differentiate in a controlled way. Cancer stem cells are neoplastic cells with an indefinite potential for self-renewal, and therefore, oncogenic capacity. Recent investigations report that a fraction of these neoplastic cells are considered cancer stem cells, which explains the continuous resistant to the treatment and tumoral recurrence. The cancer stem cell hypothesis has fundamental implications for understanding the biology of carcinogenesis as well as for developing new strategies for cancer prevention and therapies for advanced disease.

The purpose of the present project was to investigate whether proinflammatory cytokines (IL-6, IL-1 β , IL-8 and IL-11) modify the expression of mRNA of biomarkers of the CSC phenotype (CD44, CXCR4, ALDH1 and OCT4), in castration-resistant prostate cancer cells (PC3).

The expression of cancer stem cell biomarkers (CD44, CXCR4, ALDH1 and OCT4) increased in a manner dependent on the treatment concentration. The results indicate that proinflammatory interleukins (IL-6, IL-1 β , IL-8 and IL-11) participate in increasing the characteristics of cancer stem cells (CSC) in the castration-resistant prostate cancer cell line. PC3.

UNLOCKING THE POTENTIAL OF APPROVED DRUGS FOR THE INHIBITION OF PTP1B IN CANCER THERAPY

Luis Enrique Arias Romero^{1,*}, Ximena Valeria Sánchez Nava¹, Janisse Michelle Ramírez García¹, Karen Pelcastre Guzmán², Héctor Iván Saldívar Cerón³, Angélica Samantha Santos Reyes¹, Marco Antonio Loza Mejía² and Olga Villamar Cruz¹

¹Unidad de Investigación en Biomedicina (UBIMED), Facultad de Estudios Superiores-Iztacala, UNAM. Tlalnepantla, Estado de México 54090, Mexico. ²Design, Isolation and Synthesis of Bioactive Molecules Research Group, Departamento de Ciencias Químicas, Universidad La Salle-México, Mexico City, Mexico. ³Carrera de Médico Cirujano. Facultad de Estudios Superiores-Iztacala, UNAM. Tlalnepantla, Estado de México 54090, Mexico.

Corresponding author: larias@unam.mx, Phone: 55-5623-1333 x39785

PTP1B is a ubiquitous protein tyrosine phosphatase that regulates insulin and leptin signaling, and plays a key role in the development of different types of cancer. Recent evidence indicates that PTP1B promotes cell proliferation and tumor growth by modulating several pro-survival signaling pathways. The development of PTP1B-targeted therapeutics is an active area of research. Single-target and multitarget small molecule competitive inhibitors, peptide-based inhibitors and allosteric modulators have been reported. However, only a few PTP1B inhibitors have reached the clinical trials stage. Therefore, this study aimed to identify approved drugs that may serve as PTP1B inhibitors for cancer therapy by using *in silico* and *in vitro* approaches. First, we performed a screening of 2056 drugs from the e-Drug3D FDA-approved drug database, in order to assess their repurposing potential using molecular docking. The five drugs with higher theoretical affinity were selected for performing molecular dynamic simulations, confirming their stable interactions with PTP1B, and their ligand binding energies were estimated by using MM-PBSA calculations. Next, we tested the ability of these drugs to inhibit PTP1B catalytic activity in an *in vitro* phosphatase assay, where we observed that two FDA-approved drugs, Naloxone and Isradipine, blocked in more than 80% the ability of PTP1B to dephosphorylate an Insulin Receptor-derived phosphopeptide. Finally, we assessed the effect of both drugs in cell proliferation and in the modulation of some signaling pathways regulated by PTP1B. Our results indicate that both drugs have an anti-proliferative effect in human breast cancer cells, and have that ability to prevent the activation of pro-survival signaling pathways modulated by PTP1B. Altogether, our results may serve as theoretical guidance for further conducting experimental-based preclinical studies necessary for repurposing therapeutic agents targeting PTP1B.

LEPTIN MODIFIES THE METASTATIC POTENTIAL IN CASTRATION-RESISTANT PROSTATE CANCER CELLS (PC3) THROUGH THE TRPM7 CHANNEL

Rubén Ávalos López, Julieta López-Cuevas, Eduardo Monjaraz Guzmán

Instituto de Fisiología, BUAP.

14 sur 6301, CU, San Manuel, Puebla, Pue. CP 72570. (222) 229-55-00 Ext. 7311,7340
ral603@hotmail.com

Several studies show that leptin, obesity-associated adipokine, can activate the TRPM7 channel. TRPM7 is a divalent cation channel characterized by its permeability to Ca^{2+} , Mg^{2+} , and trace metal ions and, notably, its kinase activity. TRPM7 exhibits abnormal expression in the majority of cancer types and is widely reported to potentially impact carcinogenesis. Additionally, TRPM7 is associated with cell migration and invasion in lung cancer, prostate cancer, and gastric adenocarcinoma.

The central objective of the present work is to determine if the presence of leptin is capable of modulating the metastatic potential of tumor cells by influencing the functional expression of TRPM7. For this purpose, the PC-3 cell line, representative of castration-resistant prostate cancer, was used as an experimental model, which was maintained under standard culture conditions.

Initially, it was observed by RT-PCR that PC-3 cells express mRNA encoding TRPM7, and in the presence of leptin (100ng/mL) significantly increased TRPM7 expression. To evaluate the proliferative capacity, we performed a cell count in the presence and absence of Leptin, 2APB (2 μM) (TRPM7 Inhibitor), observing a significant increase in proliferation in the presence of leptin compared to the control, and a significant decrease in the presence of 2APB. The migratory capacity was evaluated by assays in transwell chambers, observing that the activation of TRPM7 by leptin significantly favors the migratory capacity of the cells, while the presence of 2APB significantly decreases migration in PC3 cells. In a complementary manner, we evaluated whether the increase in the migration capacity of PC-3 cells was accompanied by a change in the expression levels of the mRNA that codes for metalloproteins -7, -9 and -13. RT-PCR assays showed that PC-3 cells in the presence of leptin overexpress the mRNA encoding MMP7, MMP9 and MMP13, a condition that favors cell invasion.

These results propose TRPM7 as a new pharmacological target for the treatment of prostate cancer.

EVALUATION THE ROLE OF SER176 PHOSPHORYLATION IN THE ACTIVITY AND LOCALIZATION OF SNRK1

Beatriz Alejandra Ávila-Castañeda, Carmen Aitana López Zequeira and Patricia Coello-Coutiño*

Departamento de Bioquímica, Facultad de Química, UNAM. México, CDMX.
pcoello@unam.mx

The evolutionarily conserved SnRK1 kinase complex is a key regulator of cellular metabolism during starvation, stress, and growth-promoting conditions. SnRK1 is a heterotrimeric complex composed of two regulatory subunits (SnRK1 β and SnRK1 $\beta\gamma$) and an α catalytic subunit (SnRK1 α 1/2). Activation of SnRK1 α 1 requires phosphorylation of the canonical T-loop by upstream kinases SnAK1 and SnAK2. Our research focused on understanding the activation and regulation of SnRK1 α 1. We discovered that Ser-176, adjacent to the T-loop, can be phosphorylated by both SnAK kinases. Subsequent analysis of SnRK1 activity in non-phosphorylatable versions of SnRK1 α 1, both *in vitro* and *in vivo*, demonstrated that Ser-176 is essential for full activation of SnRK1. Furthermore, the critical role of Ser-176 in SnRK1 function was evident, as different SnRK1 mutants failed to complement the yeast *snf1 Δ* strain, while only the wild-type SnRK1 could do so.

We are now investigating the role of Ser-176 phosphorylation in the SnRK1 complex in planta. Using an Arabidopsis CRISPR-Cas *snrk1 α 1* mutant line, complementation analysis with different non-phosphorylatable forms of SnRK1 α 1 will be discussed.

PAPIIT: IN201922, PAIP 5000-9126

INITIATION OF INTRACELLULAR TRAFFICKING OF THE LPA3 RECEPTOR AFTER ITS STIMULATION

Elias Uriel Avila-García, K. Helivier Solís-González, Ma. Teresa Romero-Ávila, J. Adolfo García Sáinz

Department of Cell Biology and Development; Institute of Cellular Physiology, National Autonomous University of Mexico. University City. Ap. Postal 70-600, Mexico City 04510. Mexico.
Corresponding author: urielavila@ifc.unam.mx

The receptor for lysophosphatidic acid type 3 (LPA₃) belongs to the family of G protein-coupled receptors (GPCRs) that are structurally constituted by seven transmembrane domains (7TMD), with an extracellular amino-terminal region and an intracellular carboxyl-terminal region. These receptors are coupled to two types of G proteins ($G\alpha_{q/11}$ and $G\alpha_{i/o}$), promoting the activation of downstream signaling pathways, which favor various functions, such as migration, proliferation, invasion, cell contact. -cell, among others, processes that until now are not known how they are regulated by this receptor. Experimental studies have shown that the LPA₃ receptor, when activated by its natural ligand, lysophosphatidic acid (LPA), promotes phosphorylation processes that affect it, which induces the receptor to be internalized, to begin its journey through the cytoplasm. through intracellular trafficking, which is a crucial mechanism to ensure proper signaling function. The small GTPase, Rab5, plays a central role in the initiation of this mechanism, regulating protein trafficking between the plasma membrane and the early endosome. Therefore, the objective of this work was to study the process of internalization of the LPA₃ receptor by its agonist and to observe if the Rab5 protein is involved in this process. To study this process, the GFP-bound LPA₃ receptor was overexpressed in HEK293 cells of the T-REx™-293 system. Determining its functionality by measuring the concentration of intracellular calcium, to subsequently be transfected with the cDNA of the Rab5 m-Cherry protein, to observe and determine by confocal microscopy whether this receptor was internalized, and by means of the FRET technique, determine if there was an association of the Rab5 protein with the receptor. This same study was performed with cDNA of the dominant negative Rab5 protein. The results obtained show that the LPA₃ receptor is functional, promoting the release of calcium after being stimulated. Furthermore, it was observed that LPA₃ receptors are internalized when activated by LPA, with a maximum of internalization observed after 5 minutes, but they do not present a clear association with the Rab5 protein. However, the dominant negative Rab5 protein decreased internalization. In summary, the LPA₃ receptor is internalized after being activated by its ligand LPA but we were unable to define an association with the Rab5 protein, which indicates that LPA₃ trafficking could be mediated by another Rab protein or that the determination of association by FRET does not reflect this properly. This will require new approaches to its definition.

ROLE OF HPV16 E1 PROTEIN IN THE ACTIVATION NF- κ B SIGNALING PATHWAY

Alicia Beltrán-Soto¹, Joaquín Manzo-Merino¹, Imelda Martínez-Ramírez¹,
Lizano Soberón M^{1,2} and Leonardo Josué Castro-Muñoz³

¹Instituto Nacional de Cancerología, CP14180 ²Instituto de Investigaciones Biomédicas, UNAM CP 04510.

³The Wistar Institute, Philadelphia, USA

Introduction. The nuclear factor kappa B family of transcriptional factors are key regulators of the inflammatory response, as well as the progression to cancer. Human papillomavirus (HPV) is known to modulate the NF- κ B pathway. Particularly, it has been shown that the HPV E1 protein can regulate NF- κ B. Nakahara et al. demonstrated that HPV16 E1 activates NF- κ B, increasing the levels and nuclear translocation of p65 and decreasing the levels of the inhibitor I κ B α . Furthermore, Castro-Muñoz et al. demonstrated that the expression of the E1 protein of high- and low-risk HPVs decreases I κ B α levels and increases p52 levels.

Objective. Our interest in this work was to analyze the molecular mechanisms by which HPV16 E1 protein modulate the NF- κ B pathway.

Methods. The expression of E1 HPV16 in HaCaT cells was evaluated by RT-qPCR; protein levels and cellular localization of canonical and non-canonical NF- κ B elements were assessed by Western blotting and immunofluorescence. Through in silico analysis, we determined the cellular processes and genes associated with the NF- κ B pathway that were differentially regulated by E1. The effect of E1 on the expression of NF- κ B target genes was evaluated.

Results. HPV16 E1 protein induces the activation of the NF- κ B pathway by increasing the levels of p50, p52 and p65 proteins and inducing the nuclear translocation of p50 and p52. Furthermore, E1 induces an increase in the activating kinases, NIK and IKK. In silico analysis indicated that E1 induces the deregulation of 35 NF- κ B target genes associated with cellular processes involved in cancer, such as immune response evasion, epithelial-mesenchymal transition, and apoptosis.

Conclusions. The E1 proteins of HPV16 modulate NF- κ B by regulating p50, p52, p65 IKK α and NIK, which favors gene expression by modulating cellular processes that can favor viral replication and/or evasion of the immune system and transition epithelial-mesenchymal processes involved in carcinogenesis.

This work was partially supported by CONAHCYT PRONAI-7-Virus y Cáncer 303044, and a scholarship to A. Beltrán (CVU956879)

ROLE OF AHR IN CELL MORPHOLOGY DURING NEURONAL DIFFERENTIATION OF SH-SY5Y CELLS

Jimena Cris Campos-Arce¹, Jorge Humberto Limón Pacheco¹, Guillermo Elizondo*

¹Departamento de Biología Celular, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional.
Av. Instituto Politécnico Nacional 2508, 07360 Ciudad de México.
Tel: +52 (55) 5747 3800, Ext. 5555
E-mail: jimena.campos@cinvestav.mx, gazuela@cinvestav.mx

The aryl hydrocarbon receptor (AhR) is a ligand-dependent transcription factor known primarily for its role as a Xenosensor. It can be activated by a wide variety of ligands, including exogenous compounds from environmental pollution and endogenous compounds such as tryptophan and its metabolites.

In *C. elegans* and *D. melanogaster*, it has been reported that AhR interact with other factors to guide the differentiation process towards different neuronal subtypes, as well as dendrite morphogenesis. On the other hand, in murine models, it has been determined that exposure to contaminants such as TCDD and its interaction with AhR induce hypothalamic memory loss, as well as other associated cognitive impairments.

Due to this evidence, it is suggested that AhR might be a potential transcriptional regulator of neurogenesis and cognitive function; however, the physiological functions of AhR in the human brain remain poorly defined.

The present study aims to evaluate the influence of the AhR activity on SH-SY5Y cell morphology during the neuronal differentiation process.

OUABAIN PROMOTES CLAUDIN-1, -2, AND -4 AUTOPHAGIC DEGRADATION THROUGH OXIDATIVE STRESS AND AMPK ACTIVATION IN MDCK CELLS

Jessica P. Campos-Blázquez¹; Catalina Flores-Maldonado¹; Juan M. Gallardo²; José Bonilla Delgado³; Alan A. Pedraza-Ramírez¹; Octavio López-Méndez¹; Enoc M. Cortés-Malagón⁴ and Rubén G. Contreras¹

¹Department of Physiology, Biophysics and Neurosciences, Center for Research and Advanced Studies of the IPN (Cinvestav-IPN), Mexico City, Mexico, ²Unidad de Investigación Médica en Enfermedades Nefrológicas, Hospital de Especialidades, Centro Médico Nacional "Siglo XXI" Instituto Mexicano del Seguro Social, Mexico City, Mexico, ³Research Unit, Hospital Regional de Alta Especialidad de Ixtapaluca, Ixtapaluca 56530, and Department of Biotechnology, Escuela de Ingeniería y Ciencias, Instituto Tecnológico y de Estudios Superiores de Monterrey, Toluca 50110, Mexico, ⁴Research Division, Hospital Juárez de México, Mexico City 07760, Mexico.
E-mail: jp.camposblazquez@hotmail.com

Epithelial cells transport substances through the cellular and paracellular pathways. The last one depends on tight junctions, particularly on claudins, the family of integral membrane proteins responsible for the permeability and selectivity of these junctions. 300 nM ouabain induces endocytosis and lysosomal degradation of claudin-2 and -4 in a c-Src and ERK1/2 kinases-dependent manner. Here we investigated whether ouabain-induced lysosomal degradation of claudins implicates autophagy in renal epithelial MDCK cells. During autophagy, LC3 protein binds phosphatidylethanolamine and incorporates, together with protein p62, into the phagophore. Subsequently, the autolysosome degrades both LC3 and p62 proteins. Ouabain occupancy of its site in the Na⁺/K⁺-ATPase (300 nM, 10 h) increases autophagic flux because of degradation of LC3 and p62; and an increase of the number of autophagosomes, as detected by fluorescent LC3 and p62 *puncta* and the rise in autolysosomes seen by the GFP-LC3-RFP probe. Finally, ouabain increases the colocalization of claudin-1, -2, or -4 with p62 in these *puncta*. Ouabain induces autophagy increasing reactive oxygen species (ROS) generation that activates AMPK, phosphorylating ULK1 at S555. The autophagy inducer rapamycin causes a degradation of the studied claudins comparable to the one generated by ouabain. Furthermore, the autophagy inhibitor dorsomorphin blocks ouabain-induced autophagy and claudin-1, -2, and -4 degradation. These results demonstrated that ouabain induces claudin-1, -2, and -4 autophagy through oxidative stress.

TESTOSTERONE INCREASES THE EXPRESSION OF K_v CHANNELS AND ENHANCES THE AIRWAY SMOOTH MUSCLE RELAXATION INDUCED BY $P2Y_4$ AND ADENYLYL CYCLASE SIGNALING

Abril Carbajal-García¹, Jorge Reyes-García¹, Verónica Díaz Hernandez² and Luis M. Montaña^{*}

¹Departamento de Farmacología, Facultad de Medicina, U.N.A.M. ²Departamento de Embriología, Facultad de Medicina, U.N.A.M. Circuito interior, Ciudad Universitaria. Av. Universidad 3000. C.P.04510.

*E-mail: lmmr@unam.mx

The evidence shows that adenosine 5'-triphosphate (ATP) and other nucleotides are implicated in the pathogenesis of asthma. It is thought that androgens, such as testosterone (TES), may relieve the symptoms of asthma in young males. Airway smooth muscle (ASM) relaxes in response to purinergic $P2Y_2$ and $P2Y_4$ receptors stimulation by ATP and uridine-5'-triphosphate (UTP). Also, this signaling causes the activation of K^+ channels. Previous research carried out by our group has shown that TES increases the expression of voltage-dependent K^+ channels (K_v) in ASM. This study investigated how TES may enhance ASM relaxation induced by ATP and UTP. In organ bath studies, we found that guinea pig ASM treated with TES (40 nM for 48 hours) showed increased relaxation in response to ATP and UTP. This effect was reversed by tetraethylammonium, a nonspecific K^+ channel blocker. TES also amplified ATP- and UTP-induced K^+ currents (IK^+) in tracheal myocytes, an effect abolished by the androgen receptor antagonist, flutamide. The selective blocker of K_v channels (4-aminopyridine) annulated the effect of TES on the IK^+ . Moreover, RB2, which antagonizes most $P2Y$ receptors, including $P2Y_4$ (except $P2Y_2$), N-ethylmaleimide (an uncoupler of G proteins), and SQ 22,536 (inhibitor of adenylyl cyclase), attenuated the TES-induced enhancement of the IK^+ . Finally, $P2Y_4$ expression was not augmented by TES, whereas $K_v1.2$ and $K_v1.5$ are increased in the ASM treated with this androgen. In conclusion, TES improves $P2Y_4$ signaling and K_v channel expression in guinea pig ASM, enhancing ATP and UTP relaxation responses and potentially reducing bronchospasm severity in young males. These findings hold exciting potential for the future of respiratory medicine.

This work was partially funded by the Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica (PAPIIT), UNAM (IN200522) to L. M. Montaña, and (IA203924 and IA201322) to J. Reyes-García and. Additionally, L. M. Montaña and J. Reyes García received a grant from CONAHCYT, México (CF -137725 and CBF2'23-24-1074, respectively).

THYROID HORMONE T3 EXERTS DISTINCT REGULATORY EFFECTS ON METABOLISM IN WHITE, BROWN, AND BRITE ADIPOCYTES

Lidia Itzel Castro Rodríguez¹, Ma. Cristina Amada Vélez del Valle¹,
Claudia Patricia Hernández Mosqueira¹ and Walid Kuri Harcuch¹

¹Department of Cell Biology, Center of Research and Advanced Studies- IPN (CINVESTAV), Mexico City, 07360, Mexico.
E-mail: walidkuri@cinvestav.mx Tel. 55 57473353

White (WAT), brown (BAT), and brite (BRT) adipocytes exhibit functional and metabolic differences. WAT stores energy, while BAT and BRT engage in heat generation through thermogenesis. The impact of thyroid hormones, such as T3 and T4, on the metabolic activities of these cells remains poorly understood due to their wide-ranging effects across various tissues. Recently, we isolated embryonic preadipocytes EB5 and EB7 that differentiate to BAT and BRT fat cells, respectively. In this work, our objective was to study the direct action of T3 on adipose differentiation, gene expression related to lipid and carbohydrate metabolism, and thermogenesis in F442A (WAT), EB5 (BAT), and EB7 (BRT) cultured cells. We induced adipogenesis using a specific cocktail. Subsequently, we exposed the cultures with or without 10 nM T3. Gene expression was analyzed using RT-PCR.

Changes in intracellular ligand availability and/or modulation of hormone receptor expression constitute a critical mechanism in regulating thyroid hormone receptor (TR)-mediated activity. Our results show that T3 increases the expression of *Thrb1* and *Dio2* in brown and brite, but not in white adipocytes. This suggests a possible regulatory mechanism, which involves the positive regulation of *Thrb1* transcription by T3, subsequently influencing gene expression associated with lipid and carbohydrate metabolism, as well as thermogenesis. Our findings indicate that BRT tissue also responds to T3 action, which is significant because previously BRT tissue was not known to be a major target of T3.

We found that T3 increases the expression of genes linked to thermogenesis (*Ppargc1a* and *Ucp1*) and lipid/carbohydrate metabolism (*Lpl*, *Fasn*, *Mlxipl*, *Gpd1*, *Slc2a4*, *Fabp4*, *Cd36* and *Cpt1b*) in EB5 and EB7 cells, while suppressing the expression of most of them in F442A cells.

Our results suggest that if replicated in *in vivo* animal models, T3 could promote lipid mobilization from WAT and lipid utilization in BAT and BRT to support thermogenesis. T3 also decreased leptin and adiponectin gene expression in all adipocyte types, that may prevent hormone resistance. Despite similar responses in BAT and BRT, significant gene differences suggest specific physiological responses to T3. This research highlights that T3 has a differential effect on metabolism regulation across different types of adipocytes, and suggests that thyroid hormones promote a balanced metabolic state, contributing to weight loss or preventing weight gain.

PARTICIPATION OF E6 AND E7 HPV16 ONCOPROTEINS IN CELL INVASION AND MIGRATION MEDIATED BY THE DOWN REGULATION OF RHOE/RND3 GTPASE

Yesenia Cid Cruz^{1,2*}, Marcela Lizano Soberón^{2,3}, Omar Muñoz Bello²

¹Posgrado en Ciencias Bioquímicas, UNAM; ²Instituto Nacional de Cancerología, Av. San Fernando No. 22, Col. Sección 16, C.P. 14080, ³Instituto de Investigaciones Biomédicas, UNAM, C.P. 04510 CdMx
*Cel: 5559437697. E-mail: yesenia.cid.cr@gmail.com

Introduction. Persistent infection with high-risk HPV (HR) is responsible for approximately 4.5% of cancer cases worldwide. One of its main characteristics in developing this transforming phenotype is the presence of oncoproteins encoded in its viral genome, which not only endow the virus with the ability for uncontrolled proliferation but also provide significant plasticity, allowing cancer cells to migrate and invade other tissues, causing what is known as metastasis. These oncoproteins, named E6 and E7, have various functions and interactions at the molecular level, and it has been reported that they suppress the expression of multiple genes, including those encoding proteins that regulate cellular plasticity such as RhoE/Rnd3, an atypical small GTPase that negatively regulates the formation of actin filaments, thereby granting the cancer cell the ability to move and invade. Although it is known that the RhoE transcript is markedly reduced in the presence of E6 and E7 from high-risk HPV16, it has not been described how this GTPase is involved in cell migration and plasticity in cervical cancer. Given that RhoE has been associated with both tumor suppressive and promoting functions, determining its role in this type of cancer would be highly relevant.

Objective. To establish the effect of the oncogenes E6 and E7 from HPV16 on the activity of the GTPase RhoE and its involvement in cell migration and invasion.

Methods. RT-qPCR assays and Western blot were performed to analyze RhoE expression and protein levels in cervical cancer cells, HaCaT cell line and CaSki cell line with silencing or E6 and E7. Expression of mesenchymal and epithelial markers vimentin, fibronectin, E-cadherin, and ZO-1 were analyzed. RND3 was overexpressed in cervical cancer cells following the previous analyzes.

Results. The gene encoding RhoE is downregulated in the context of cervical cancer without HPV infection; however, when the E6 and E7 oncoproteins from HPV16 are present, this expression decreases even further. Despite this being anticipated based on transcriptomic data from our study model, it was also found that E6 and E7 reduced protein levels of vimentin, a mesenchymal marker involved in migration that is generally found at high levels in cancer. Therefore, it is important to further investigate the cellular plasticity mediated by these oncoproteins.

This work was partially supported by CONAHCYT PRONAI-7-Virus y Cáncer 303044.

We acknowledge CONAHCYT support with the scholarship to Y. Cid Cruz (CVU 1321593)

[Ca²⁺]_i DISTRIBUTION IN SPERMATOZOA FROM TWO POPULATIONS OF *SCELOPORUS GRAMMICUS* ALONG AN ALTITUDINAL GRADIENT

Gabriela Contreras González¹, Daniel Uriostegui Escoto², Gihovani Ademir Samano Barbosa³, Ahiezer Rodríguez-Tobón⁴, Fausto Roberto Méndez de la Cruz⁵, Edith Arenas-Ríos⁶

¹Maestría en Biología de la Reproducción Animal; ²Laboratorio de Biología e Ingeniería Tisular Neuroinmunoendocrina; ³Laboratorio de Biología y Ecología de Mamíferos; ⁴Laboratorio de Morfofisiología y Bioquímica del Espermatozoide, Universidad Autónoma Metropolitana-Iztapalapa. ⁵Laboratorio de Investigación en Reproducción Animal, Universidad Autónoma Benito Juárez de Oaxaca y; ⁶Laboratorio de Herpetología, Universidad Nacional Autónoma de México. Tel. 55 4957 2974 E-mail: COGG961028_bio@hotmail.com

The increase in temperature due to climate change affects the biological functions of ectothermic vertebrates such as lizards, it has been proposed that these organisms could adapt to this increase through physiological adaptations related to altitudinal migration, in which lizards move to higher altitudes in search of lower temperatures. *Sceloporus grammicus* is the appropriate model since it is distributed from 80-4600m where two populations with different body temperatures can be studied. The increase in temperature alters the signaling pathways involved in the epididymal sperm maturation, the search for lower temperatures through altitudinal migration could be accompanied by adaptations in these processes to ensure the survival of the gametes. The present study aims to know the thermal tolerance of the cells as a reflection of the temperature increase, establishing the distribution and [Ca²⁺]_i in epididymal spermatozoa of two populations of *S. grammicus*. For this purpose, ten organisms were collected from two populations (2250 and 3500 m); identification of species, morphological data collection, organ dissection and spermatozoa collection from the three regions of the epididymis (*caput*, *corpus* and *cauda*) were performed to determine sperm parameters. Subsequently, the spermatozoa were exposed to different temperatures (21, 25, 29, 33 and 37°C) to evaluate the effect of increased temperature on motility. Organisms from the low population presented greater snout-vent length and weight concerning to the high population (6.70±0.22 vs 5.12±0.14; 11.85±0.29 vs 5.04±0.21, respectively). Cell viability was ~70% for both populations. The percentage of cytoplasmic droplet was higher in cells of the low population at the *corpus* and *cauda* level (17.50±7.0 vs 8.001.68±5.3; 14.20±6.7 vs 7.00±5.1). DNA compaction was greater than 40% for both populations. We found that the optimum temperature for maintaining sperm motility was 25 and 21°C for the low and high populations, respectively. Four distribution patterns of [Ca²⁺]_i were determined, with calcium being predominant in the acrosomal region as well as in the cytoplasmic droplet for the low population, while in the population at 3500 a higher percentage was found in the acrosomal region and in the middle piece of the spermatozoa. We suggest that exposure to high temperatures could be intervening in the signaling pathways involved in [Ca²⁺]_i transport, concentration and distribution.

EFFECT OF CORTICOTROPIN-RELEASING FACTOR (CRF) IN THE ACTIVATION OF MAPK AND PI3K/AKT PATHWAYS INDUCED BY INSULIN-LIKE GROWTH FACTOR-1 (IGF-1) IN CHO-K1 CELLS

Carlos De Jesus-Quiroz¹, Judith Hernández-Aranda¹ and Jesús Alberto Olivares-Reyes¹

¹Department of Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav). Av. Instituto Politécnico Nacional 2508, Col. San Pedro Zacatenco. C.P. 07360, Mexico City, Mexico. Tel: (52) (55) 5747-395. E-mail: carlos.dejesus@cinvestav.mx; jolivare@cinvestav.mx

Insulin-like growth factor-1 (IGF-1) is a peptide hormone important in cellular processes such as proliferation, differentiation, survival, and metabolism. These cellular responses are carried out through the MAPK and PI3K/Akt pathways, which are activated after the binding of IGF-1 to its receptor (IGF-1R); this is a relevant member of the receptor tyrosine kinases family (RTKs). Several reports indicate a reduction in the levels and effects of IGF-1 in psychiatric disorders such as depression and anxiety, as well as in metabolic disorders such as anorexia nervosa¹. On the other hand, high concentrations of corticotropin-releasing factor (CRF) have been found in these pathologies²; CRF is an important neuropeptide for the activation of the hypothalamic-pituitary-adrenal axis (HPA), an important system that coordinates the neuroendocrine stress response. Currently, the molecular mechanism between the interaction of both factors is not clear; however, it is known that high concentrations of CRF change the signaling and regulatory pathways mediated by the CRF type 1 receptor (CRF1R), a member of the G protein-coupled receptors (GPCRs) family. Therefore, it has been suggested that chronic activation of GPCRs can desensitize the signal in the receptors themselves (homologous desensitization) or in members of the RTK family (heterologous desensitization) like IGF-1R³. Considering the above, this work aims to determine the molecular mechanism of CRF through CRF1R, which regulates the MAPK and PI3K/Akt pathways activated by IGF-1. CHO-K1 cells (Chinese hamster ovary) were transfected with a plasmid that encodes HA-hCRF1R using the liposome transfection method. Transfected cells were pre-incubated with CRF at different times and subsequently stimulated with IGF-1. Our results indicated that CRF through the CRF1R reduces IGF-1R, Shc, and ERK1/2 phosphorylation induced by IGF-1, while Akt phosphorylation was not affected; therefore, we suggest that CRF is regulating differentially MAPK and PI3K/Akt pathways activated by IGF-1. Furthermore, the use of rapamycin and SP600125, inhibitors of mTOR and JNK, respectively, prevents the reduction of ERK1/2 phosphorylation induced by CRF. Thus, we suggest that these kinases participate in the regulatory mechanism induced by CRF, possibly increasing phosphorylation on serine/threonine residues at the level of IGF-1R or the scaffold proteins Shc and IRS-1, reducing its activation.

Funding: This work was supported by CINVESTAV-IPN, and CDJ-Q held a CONAHCYT graduate scholarship (CVU: 1002001).

References

1. Zegarra-Valdivia, J.A. (2017). *Medwave*. 17(7). 7031–7031.
2. Arborelius et al. (1999). *Journal of Endocrinology*. 160 (1). 1-12.
3. Di Liberto, V., Mudò, G., & Belluardo, N. (2019). *Neuropharmacology*. 152. 67–77.

PTP1B MODULATES ENDOTHELIAL-TO-MESENCHYMAL TRANSITION DURING TNF-INDUCED ENDOTHELIAL DYSFUNCTION

José Esparza-López¹, Jorge Romo-Tena^{1,2}, Mariana J. Kaplan², María de Jesús Ibarra-Sánchez¹

¹Unidad de Bioquímica Dr. Guillermo Soberón Acevedo, Laboratorio de Señalización en Cáncer, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán, Vasco de Quiroga 15, Belisario Domínguez Sección XVI, Tlalpan, 14080, CDMX, Mexico.

²Systemic Autoimmunity Branch, NIAMS/NIH, 10 Center Drive, 12N248C, Bethesda, MD 20892, USA.

E-mail: maria.ibarras@incmnsz.mx

Chronic inflammation is one of the critical steps for the induction of endothelial dysfunction associated with several diseases like cardiovascular disease, diabetes, and cancer. Activation of endothelial cells induces several changes augmenting endothelial permeability, pro-thrombotic, and proliferative state. Activated endothelial cells display morphological and biochemical modifications known as an endothelial-mesenchymal transition (EndMT). There is evidence associating the protein tyrosine phosphatase 1B (PTP1B) with the development of endothelial dysfunction; however, the role of PTP1B in an inflammation-mediated EndMT is not fully understood. To study the participation of PTP1B in tumor necrosis factor (TNF)-induced EndMT in endothelial cells, we established an EndMT model induced by TNF using Human Aortic Endothelial Cells. Western blot was performed to evaluate EndMT markers, PTP1B expression, and nuclear factor- κ B (NF- κ B) activation. A wound-healing assay was performed to assess cell migration. We employed an IKK β inhibitor, BMS-345541, to inhibit NF- κ B activation. PTP1B knockdown was performed by transfecting siRNA targeting PTP1B. TNF triggered EndMT, with concomitant PTP1B protein expression increase and endothelial cell migration through the Akt/NF- κ B pathway. IKK β inhibition using BMS-345541 prevented NF- κ B activation and Akt phosphorylation. Moreover, BMS-345541 attenuated the TNF-induced mesenchymal markers N-cadherin, Snail, TWIST1, PTP1B upregulation, and cell migration. PTP1B silencing prevented TNF-induced endothelial markers decrease and upregulation of mesenchymal markers. Furthermore, PTP1B knockdown reduced TNF-induced Akt activation but not Erk and p38, impairing the TNF-induced cell migration. Our results demonstrate that PTP1B is essential in TNF-induced EndMT. These data suggest that targeting PTP1B could represent a putative target to ameliorate endothelial dysfunction and associated diseases such as cardiovascular, diabetes, and cancer.

EFFECT OF VPH16 E6 AND E7 ON MAST CELL MIGRATION

Zyanya Patricia Espinosa Riquer^{1*}, Omar Muñoz Bello¹ y Marcela Lizano Soberón^{1,2}

¹Instituto Nacional de Cancerología, CP 14080, ²Instituto de Investigaciones Biomédicas, UNAM, CP 04510 CdMx

*E-mail: espinosariquer@gmail.com

Introduction. Mast cells (MC) are recognized as part of the host's first defense line and play relevant roles in cancer. MC are part of the tumor microenvironment where they have both, anti- and pro-tumoral effects. Pro-tumoral effects include the promotion of proliferation, migration, and metastasis. The anti-tumor effects of MC include proliferation inhibition, phagocytosis and apoptosis induction, and pro-inflammatory immune cell attraction. Cervical cancer (CC) is usually developed by the persistent infection with high-risk human papillomavirus 16 (HPV16). HPV-dependent CC establishment and progression requires two early expressed genes E6 and E7, known as oncogenes. Little information exists regarding the effects of MC on CC. Some studies demonstrated that when MC abundantly infiltrate CC cells, there is a reduction in the overall survival of patients. In mice studies, MC number increased in epithelial tissues expressing HPV16 early genes, which augmented when epithelial tissue transformed into cancer, suggesting pro-tumoral effects of MC.

Objective. This work aimed to study the effect of HPV16 oncoproteins E6 and E7 on the migration of MC to establish the relationship between these immune cells, CC, and HPV.

Methods. Using a transwell assay, we analyzed MC migration toward C33 cells stably expressing E6 and E7 oncoproteins. To test the specific effect of E6 and E7 on MC migration, we silenced E6 and E7 on the VPH16+ Ca Ski cell line and measured the number of migrated cells in the transwell assay. To determine the mediators that could attract MC, we analyzed VEGFA, VEGFB, VEGFC, CXCL16, and TGFBI mRNA expression by qPCR in C33 cells expressing E6 and E7.

Results. E7, but not E6, promotes MC migration, and E7 silencing prevented this effect. In addition, E6 and E7 increased VEGFA, VEGFC, and CXCL16 and decreased VEGFB, while only E7-expressing cells increased TGFBI mRNA. In conclusion, E7 promotes MC migration through a mechanism that could involve changes in the secretory profile of CC cells.

This project is partially supported by PAPIIT IN208924 and a postdoctoral fellowship for Z. Espinosa Riquer by CONAHCyT (CVU 487483).

ROLE OF THE IRE1-MEDIATED ENDOPLASMIC RETICULUM STRESS PATHWAY DURING THE DEVELOPMENT OF THE TRACHEAL SYSTEM IN *DROSOPHILA MELANOGASTER*

María Fernanda Frutis Osorio¹, Berenice Otero Díaz¹, Daniel Ríos Barrera¹

¹Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, UNAM. Ciudad Universitaria, 04510. Ciudad de México. México.

*Contacto: 55562-29250 ext. 46804, fernandafrutis@iibiomedicas.unam.mx

The accumulation of unfolded proteins inside the endoplasmic reticulum (ER) induces a phenomenon referred to as ER stress, which has critical consequences for the cells. Therefore, organisms have evolved mechanisms that function to restore protein homeostasis and ensure cell survival. Normally, the appearance of ER stress is associated with pathological conditions. Nevertheless, this phenomenon is also seen in homeostatic conditions, such as embryonic development. In this work, we aim to determine whether the activation of the Ire1-mediated endoplasmic reticulum stress pathway is important for the formation of the tracheal system (TS) in *Drosophila melanogaster*. Ire1, a transmembrane protein that resides in the ER, is activated by the accumulation of unfolded proteins. With its endoribonuclease activity, it cleaves specific mRNAs to lower protein synthesis and induce the expression of Xbp1, a transcription factor that also acts to alleviate the stress. Preliminary results from the lab suggest that this pathway is activated in the TS during its development. The TS of *Drosophila* is a ramified network of tubes that transports oxygen to all body tissues. Throughout its formation, high levels of membrane and proteins are synthesized and secreted to the apical membrane to stabilize lumen formation. This implies that there is an elevated protein production that could naturally induce ER stress. To study the role of Ire1 in this process, we will use mutant embryos (Ire1⁻) to evaluate the embryonic lethality and the morphology of the TS. Furthermore, we are developing a machine learning-based protocol for analyzing the dynamics of secretion in the TS *in vivo*, using control and Ire1⁻ embryos.

ROLE OF STAT5 IN ENERGY MITOCHONDRIAL METABOLISM GENES REGULATION IN CERVICAL CÁNCER CELLS STIMULATED WITH INTERLEUKIN 2

Rubén-Alejandro Fuentes-Pascacio, Arturo Valle-Mendiola, Adriana Gutiérrez-Hoya, Isabel Soto-Cruz

Laboratorio de Oncología Molecular, FES Zaragoza, Universidad Nacional Autónoma de México,
Batalla 5 de mayo s/n Col. Ejército de Oriente, CP 09230, Ciudad de México, México.
Tel: 5556230796. arzad1029@gmail.com; sotocruz@unam.mx.

Cervical cancer (Cacu) is the fourth most common cancer in women globally, being a public health problema. An important risk factor in Cacu is the chronic infection with human papillomavirus (HPV), this infection can change the regulation on the cell cycle and normal cells transforms into cancer cells and with this acquire new characteristics. One of these characteristics is an aberrant signal transduction and protein synthesis one of this proteins is the synthesis of Interleukin 2 (IL-2) and its receptor (IL-2R). Our research group have demonstrated that cervical cancer cell lines produce IL-2, and the IL-2 receptor is present in these cell lines. Also, the JAK/STAT pathway is active in Cacu cell lines since JAK3, STAT3, and STAT5 increase their phosphorylation in response with 10 UI/mL of IL-2, thus increasing cell proliferation. Another hallmark of cancer cells is metabolic reprogramming, the mitochondria plays a key role on this regulation specifically, in genes relationated with the electron transport chain (ETC). We demonstrate in cervical cáncer cell lines that STAT5 is localized in mitochondria in a basal form and the 10 UI/mL stimulation of IL-2 increases activation and the translocation of STAT5 to mitochondria. Consequently, this treatment of IL-2 decreases the transcription of ETC nuclear related genes, specifically, UQCRC1, NDUFB1 and ATP5FB1.

Furthermore a transient transfection with a RNA interference (RNAi) against STAT5 demonstrated this IL-2 stimulation did not decrease the transcription of this ETC related genes comparation with the wild type Cacu cells.

The results of STAT5 translocation to the mitochondria and the regulation of the gene expression of the proteins that includes the complexes of ETC could suggest STAT5 as a negative regulator in nuclear genes related to ETC and contribute with the switch metabolic in cancer cells.

ANALYSIS OF THE RELATIONSHIP BETWEEN THE JAK/STAT PATHWAY AND THE DEATH RECEPTOR CD95 TO INDUCE PROLIFERATION AND SURVIVAL OF CERVICAL CANCER CELLS

Armando García-Velasco, Adriana Gutiérrez-Hoya, Maria Isabel Soto-Cruz

Universidad Nacional Autónoma de México, FES Zaragoza
Av. Nuevo Leon 275^a CDMX, México, 555551729, armando_jkd@hotmail.com

Cervical cancer is a public health problem in Mexico, it represents the second most common type of cancer in women and, together with breast cancer, is the main cause of death in women. The cervical carcinoma cell lines CALO and INBL have been shown to express all three subunits of the IL-2 receptor. Furthermore, the activation of the JAK-STAT pathway has been analyzed in response to the binding of IL-2 to its receptor. It has been shown that both JAK3, STAT3 and STAT5 are constitutively phosphorylated and that they respond to treatment with 10 IU/ml of IL-2, increasing their phosphorylation. This induces an increase in the proliferation of cervical carcinoma cells. Furthermore, we have found that JAK3 kinase inhibition induces a decrease in STAT5 phosphorylation, but does not inhibit it, however, it does decrease tumor cell proliferation. On the contrary, high concentrations of IL-2 (100 IU/mL) induce a decrease in the proliferation of cervical cancer cells. Furthermore, it has been reported that this pathway has important implications in survival and anti-apoptotic stimulation in normal cells and some types of tumor cells, so we consider that it is a pathway that participates significantly in the cellular transformation that cells undergo. cervical cancer cells. An interesting finding that we observed is the overexpression of the death receptor CD95 (FAS) in the tumor cells obtained from the ATCC and in those derived from biopsies of Mexican patients. Theoretically, this overexpression should contribute to the induction of cell death, however, apparently this function is lost or blocked. In addition to the different activities of CD95 and CD95L in inducing apoptosis, mostly in the context of an immune response, it has already been established that CD95 has multiple non-apoptotic functions. It was recently shown that phosphorylation of CD95 on different tyrosines (Y232 and Y291) in the intracellular domain can modulate signaling pathways. Tyrosine phosphorylation turns off the pro-apoptotic signal and turns on pro-survival signals that lead to cell proliferation and migration. These data agree with recent data obtained in different cancer lines, which show that CD95 has an important role during tumorigenesis since it can apparently promote tumor growth.

ABERRANT EPITHELIAL REPROGRAMMING IN THE LUNG FROM MMP8-MMP13 DOUBLE KNOCKOUT MOUSE IMPAIRS FIBROSIS RESOLUTION

Ángeles García-Vicente¹, Miguel Gaxiola², Moisés Selman², Annie Pardo¹ and Sandra Cabrera¹

¹Laboratorio de Biopatología Pulmonar, Facultad de Ciencias, UNAM, Circuito Interior S/N, Ciudad Universitaria, Coyoacán, 04510, CDMX. ²Instituto Nacional de Enfermedades Respiratorias "Ismael Cosío Villegas", Calz. de Tlalpan 4502, Sección XVI, Tlalpan, 14080, CDMX.
E-mail: scb@ciencias.unam.mx

Current evidence suggests that Idiopathic Pulmonary Fibrosis (IPF) is the result of repetitive epithelial injury and a deregulated wound-healing process in aged individuals. This abnormal wound healing response is characterized by fibroblast proliferation and extensive deposition of extracellular matrix (ECM). Matrix metalloproteinases (MMPs) and their inhibitors (TIMPs) regulate ECM homeostasis. ECM homeostasis is dysregulated through processes that are not fully understood. In this study, we use a double knockout mouse model deficient in Mmp8 and Mmp13 proteases to evaluate lung fibrosis development and resolution after the bleomycin challenge. We observed severe fibrosis response associated with increased proliferation and migration rates in Mmp8-Mmp13 dKO fibroblasts compared to WT and Mmp13 single KO fibroblasts, along with increased CTHRC1 and α -SMA mesenchymal cells in Mmp8-Mmp13 dKO fibrotic lungs compared to WT lungs. Moreover, at the resolution stage, we found significantly extended areas of keratin 8 (KRT8)- positive stained epithelial cells in Mmp8-Mmp13 dKO fibrotic lungs, coexisting with permanent fibrotic foci.

In contrast, WT lungs have reached a typical tissue architecture, and KRT8-positive cells were scarce at 56 days post-bleomycin treatment. Next, we analyzed the non-ECM substrates from both MMP8 and MMP13 enzymes in silico, using the PANTHER platform; substrates were classified according to the Gene Ontology (GO) enrichment into molecular function, biological process, protein class, and pathway categories. Among common substrates for both enzymes, we found fibronectin 1, CCL2, and CCL17, which promote an aberrant epithelial phenotype. These proteins could be targets to be analyzed in our Mmp8-Mmp13 dKO mouse model for future experiments. Additionally, we found significantly increased nuclear p21-positive epithelial cells in Mmp8-Mmp13 dKO fibrotic lungs compared to WT lungs at the resolution stage. Our results show that the Mmp8-Mmp13 dKO mouse phenotype is recapitulating the aberrant basaloid state and impaired fibrosis resolution observed in human IPF. This work was funded by CONAHCYT 235891 SEP-Ciencia Básica Grant. Ángeles García Vicente is a doctoral student from Programa de Doctorado en Ciencias Biomédicas, Universidad Nacional Autónoma de México (UNAM) and received fellowship 742879 from CONAHCYT.

TOLL-LIKE RECEPTOR (TLR)-4 ACTIVATION INDUCES MAST CELL MIGRATION AND PARTICIPATES ON THEIR INCORPORATION TO SOLID TUMORS

Daniel Guerrero-Morán¹, Alfredo Ibarra-Sánchez¹, Marcela Lizano Soberón^{2,3}, Omar Muñoz Bello^{2,3}, Claudia González-Espinosa¹

¹Pharmacobiology Department and Center for Research in Aging, Center for Research and Advanced Studies (Cinvestav) of the National Polytechnic Institute; ²Genomic Medicine and Environmental Toxicology Department, Biomedical Research Institute, National Autonomous University of Mexico (UNAM). ³Unit of Biomedical Research in Cancer, National Cancer Institute (Mexico). Mexico City, Mexico.

Mast cells (MC) are important stromal components distributed throughout nearly all human tissues. These cells possess a broad repertoire of receptors that allow them a surprising recognition capacity. An important group of receptors expressed in mast cells are Toll-like receptors (TLRs). They enable the recognition of pathogen-associated molecular patterns (PAMPs) and damage-associated molecular patterns (DAMPs), among other molecules. In the last years, the TLR4 receptor, a member of this family, has been identified as a molecule that actively participate in the inflammatory microenvironment observed in solid tumors, modulating tumor initiation/growth through vascular activation and recruitment of endothelial and immune cells. In this study we aimed to investigate if the activation of TLR4 receptor could induce MC chemotaxis and recruitment of these cells to solid tumors. For this goal, we evaluated the chemotaxis to conditioned media from B16-F1 malignant melanoma cell line in a Boyden's chamber assay using bone marrow-derived mast cells (BMMCs) of WT and TLR4-deficient (*Lps-del*) mice. Then, we evaluated the participation of mast cell's TLR4 receptor in the recruitment of MCs to tumors *in vivo*, generating B16-F1 melanoma tumors in mice deficient in mast cells (*Wsh*) and *Wsh* mice reconstituted with BMMCs from WT or TLR4-deficient animals, with the respective controls. Our results indicate that activation of TLR4 receptor leads to a significant concentration-dependent migration of BMMCs and it was also as potent as the well-characterized mast cell chemoattractant sphingosine 1-phosphate (S1P). *In vivo*, defective recruitment of MC and diminished tumor growth was observed in *Wsh* mice reconstituted with TLR4-deficient BMMCs and this was opposite to the observed in *Wsh* animals reconstituted with WT BMMCs. Finally, characterizing the molecular process involved in TLR4-dependent chemotaxis of MC, we found that c-Abl, a Src-related kinase, associates to TLR4 receptor and actin cytoskeleton, contributing to TLR4-dependent chemotaxis to B16-F1 conditioned media. Our results strongly suggest that TLR4 receptor present in MC participates in the incorporation of those immune cells to tumors with the participation of a novel TLR4-c-Abl-actin interaction.

Supported by Conacyt Grant CF-2019-51488 to Claudia González and Marcela Lizano and scholarship 1008896 to Daniel Guerrero.

EFFECT OF INSULIN ON VASCULAR ENDOTHELIAL GROWTH FACTOR-INDUCED ACTIONS IN ENDOTHELIAL CELLS EA.HY926

Maikel González Matos¹, Jesús Alberto Olivares-Reyes²

^{1,2}Laboratory of Signal Transduction, Department of Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute, Av. Instituto Politécnico Nacional No.2508, Colonia San Pedro Zacatenco, C.P 07360, CDMX, México.

¹E-mail: jolivare@cinvestav.mx

Insulin is a hormone that regulates systemic carbohydrate and lipid metabolism, but it also plays an important role in cellular processes such as vascular function, promoting vasodilation and tissue perfusion. Insulin also stimulates the secretion of vascular endothelial growth factor (VEGF) in several cell types, and this growth factor is known to promote endothelial cell migration and angiogenesis¹. Insulin could influence VEGF-induced signaling pathways in endothelial cells by activating protein kinases and other signaling factors. Insulin activates PI3K/Akt and mitogen-activated kinase (MAP kinase) signaling pathways, which play an important role in cell survival, growth, and proliferation. In some cases, activation of the PI3K/Akt pathway may lead to the activation of negative feedback mechanisms that inhibit VEGF signaling. This could include phosphorylation and degradation of key components of the VEGF signaling pathway. On the other hand, it has been reported that the MAPK pathway can influence the modulation of the balance between cellular responses. If signaling through this pathway becomes dominant, it could inhibit or counteract VEGF signaling, thereby decreasing the cellular response to VEGF. Therefore, the present project aims to determine the molecular mechanism by which insulin affects VEGF-induced responses in EA.hy926 endothelial cells. To meet this objective, the insulin and VEGF responses of the EA.hy926 cell line were initially characterized. Subsequently, the potential molecular mechanisms underlying the interaction between insulin and VEGF pathways were explored, and the influence of insulin signaling on VEGF-induced angiogenic responses, such as cell proliferation and migration, was analyzed. Our results showed that insulin mediates the phosphorylation and activation of Erk1/2 in EA.hy926 endothelial cells with an EC50 value of 4.254×10^{-11} M, with this effect being maximum at 5 min. In addition, insulin mediates the phosphorylation and activation of Akt at concentrations greater than 1nM with an EC50 value of 1.880×10^{-10} M from 2-15 min. On the other hand, VEGF activates Erk1/2 with an EC50 value of 1.757×10^{-12} M from 5 min, with a maximum effect at 30 min, and Akt with EC50 value of 6.240×10^{-10} M from 2 min and maintained up to 30 min. Interestingly, prestimulation of 100nM insulin for 5 min can negatively regulate the phosphorylation and activation of the VEGF-mediated MAPK pathway, an effect that is functionally reflected in the decrease of cell migration and proliferation mechanisms at times of 12-24 and 48 hours respectively.

Funding. This project was supported by CINVESTAV-IPN and MG-M hold a CONAHCYT scholarship (1233952).

References

1. Belfiore A, et al. (2009). *Endocr Rev.* 30(6): 586-623.

STAT3 ROLE IN THE AUTOPHAGY ACTIVATION IN CERVICAL CANCER CELLS TREATED WITH HIGH DOSES OF IL-2 AND DDP

Romero Hernández C.B.¹, Ortiz Garrido I.¹, Soto Cruz M.A.¹, Gutierrez Hoya A.^{1,2}

¹Laboratorio de Oncología Molecular, Unidad de Diferenciación Celular e Investigación del Cáncer, FES Zaragoza, Universidad Nacional Autónoma de México, Batalla 5 de Mayo s/n Col. Ejército de Oriente, Ciudad de México 09230, México. agutierrezho@gmail.com. 56230796.

²Investigador CONACYT, CONACYT, Avenida Insurgentes Sur 1582, Col. Crédito Constructor Del. Benito Juárez, Ciudad de México 03940, México

Autophagy is activated in front of stress stimuli for cells, it's a process highly associated with survival, tumorigenesis and metastasis in cervical cancer cells, these cells responds in dual manner with IL-2 stimuli which activates JAK/STAT3 via, meanwhile low doses cell proliferation increments, at high doses the cell proliferation decreases and they showed it's because of arrest of cells in the G1 phase of cell cycle so, they administrated DDP after IL-2 high doses and determinate apoptosis, but the co-stimulus increase the percentage of viable cells. This work pretends to determine the role of STAT3 in the autophagy that is a via involucrate in the chemoresistance develop in cancer.

We determine DDP IC50 for both cell-lines HeLa 6.9 μ M and SiHa 5.1 μ M and observed same behavior of HeLa and SiHa cell-lines before IL-2 treatment low doses (10UI/mL) increment proliferation, and high doses (100UI/mL) decrease it, while high doses of IL-2 with DDP treatment augment HeLa proliferation in SiHa doesn't show the same answer.

Also determine IC50 HO-3867 that is an inhibitor of STAT3 phosphorylation in HeLa cells 3.7 μ M and increase with IL-2 low doses treatment 9.2 μ M.

LC3B was determined by confocal microscopy, and we observed the signal augment when we treated HeLa cells with high doses of IL-2 and DDP, meanwhile SiHa we didn't observe the same behavior.

We also determine apoptosis in HeLa cells and we observed an retardment of apoptosis process when we administrated high doses of IL-2 and DDP in regard to a IC50-DDP treated control and then apoptosis reestablished with administration of HO-3867 what suggest pSTAT3 is the responsible of activation of autophagy via and its chemoresistance mechanism.

ROLE OF MEOX2 IN MODULATING THE EXPRESSION AND/OR ACTIVATION OF MEKK1/2 PROMOTING CELL PROLIFERATION IN LUNG CANCER

Brenda Hernández¹ and Federico Ávila²

^{1,2}Facultad de Estudios Superiores Iztacala. Laboratory 12 of Pulmonary Diseases and Cancer Epigenomics. UNAM-INER. Biomedicine Research Unit (UBIMED), FES, Iztacala, UNAM.
E-mail: brendana0400@gmail.com

The complexity of non-small cell lung cancer (NSCLC) stems from a multifactorial issue where the signaling pathways of mitogen-activated protein kinases (MAPK) and the PI3K/AKT/mTOR pathway play a very significant role in regulating cancer cells. In the oncological context, these pathways are reprogrammed and regulate cell differentiation, proliferation, cell growth, survival and angiogenesis, contributing to the pathogenesis and malignancy of cancer. It has been demonstrated that the transcription factor mesenchyme homeobox 2 (MEOX2) participates in the regulation of these signaling pathways by promoting a significant increase in the protein AKT, epidermal growth factor receptor (EGFR), and the activated protein p-AKT, members of the aforementioned pathways. Additionally, it has been identified that MEOX2 can induce the phosphorylation of ERK. The project aims to elucidate whether MEOX2 regulates the MAPK and PI3K/AKT/mTOR pathways by modulating the expression and/or phosphorylation activation of MEKK1/2, subsequently affecting the phosphorylation of ERK1/2, contributing to cell proliferation, malignancy, and resistance to lung cancer therapy.

Methodology. Lung cancer cell models were established under genetic silencing of MEOX2 using shRNAs. The expression levels of the genes MEK1/2, ERK1/2, and AKT were analyzed in the presence and absence of genetic silencing of MEOX2. Additionally, the phosphorylation levels of the protein members of these signaling pathways were analyzed in the presence and absence of MEOX2 genetic silencing. Finally, the participation of MEK1/2 in lung cancer cell proliferation was determined using its inhibitor (Selumetinib) in the presence and absence of MEOX2 through MTT assays and clonogenic assays.

Results. Genetic silencing of MEOX2 promotes a significant increase in the expression of the ERK and AKT genes in the A549 cell line. Additionally, it induces an increase in total ERK protein but a decrease in phosphorylated p-ERK protein, demonstrating that MEOX2 is related to the modulation at the expression or activation level of the signaling pathways towards the ERK protein. Furthermore, it has been demonstrated that inhibition of MEKK through the drug Selumetinib and under genetic silencing of MEOX2 significantly decreases lung cancer cell proliferation.

EFFECT OF CAFFEINE ON METASTATIC POTENTIAL IN NON-SMALL CELL LUNG CANCER (A549)

Yarely Rocio Herrera Sánchez, Max Alejandro Maximino Rojas, María Julieta López Cuevas, Eduardo Monjaraz Guzmán, David Eduardo Pérez Calderas

Benemérita Universidad Autónoma de Puebla
Prolongación de la 14 sur 6301. Cd Universitaria, Puebla, Pue.
yarerocioherrera@gmail.com Cel. 2223831618

Lung cancer has a high global incidence and is one of the main death causes worldwide. Paclitaxel is one of the main drugs used against cancer, however, it has been observed that such treatment can generate chemoresistance on tumor cells. Caffeine has been an object of study due to its capacity to enhance the effect of anti-cancer drugs, by inhibiting DNA repair, inducing cell apoptosis and improving its antitumor capacity. Therefore, developing strategies to overcome paclitaxel resistance and improve its therapeutic efficacy is crucial for lung cancer treatment. Combination of both caffeine and paclitaxel might be a promising option.

The hypothesis raised in our work suggests that caffeine could enhance the effects of paclitaxel in lung cancer treatment, especially with regard to reduction of metastatic potential. The main objective of our investigation was to determine whether caffeine, when combined with paclitaxel, can improve its therapeutic efficacy in A549 cell line.

A series of *in vitro* experiments were carried out which evaluated the effect of caffeine and paclitaxel, both individually and in combination, on cell proliferation, cancer cell migration capacity and their resistance to treatment. The results of these experiments provide valuable information on the potential of caffeine as an adjuvant in cancer lung treatment and its ability to improve paclitaxel efficacy, possibly allowing the reduction of necessary dose for achieving therapeutic results without compromising efficacy of the treatment. The results of our study are quite detailed and show a significant caffeine effect on several aspects of A549 cells molecular biology. Caffeine significantly reduced cells migratory and proliferative capacity in comparison to control, in addition, both caffeine and paclitaxel significantly increased the expression of molecular elements associated with apoptosis and cell survival.

Our study suggests that when paclitaxel and caffeine are combined, an increase in the expression of ABCG2 transporter is not generated, an associated element to drug extrusion that we can relate to the reduction in its effectiveness. These findings suggest that caffeine might have significant effects on the migratory and proliferative capacity of A549 cells, just as well as on the expression of molecular markers associated with apoptosis and chemotherapy resistance, resulting in a promising candidate as adjuvant in non-small cell lung cancer.

JASMONIC ACID REGULATES CELL DEATH AND REGENERATION IN THE ARABIDOPSIS ROOTS OF A MUTANT WITH COMPROMISED CELL VIABILITY

Huerta-Venegas Pedro Iván^{1,2}, Raya-González Javier¹, Ortiz-Castro Randy², López-Bucio José¹

¹Universidad Michoacana de San Nicolás de Hidalgo. Av. Francisco J. Múgica, CP 58030, Morelia, Mich.

²Instituto de Ecología, A. C.; Carretera Antigua a Coatepec 351, CP 91073, Xalapa, Ver.

jbucio@umich.mx

Cells within the root and shoot meristems are highly susceptible to programmed cell death (PCD) upon exposition to biotic and abiotic stressors. This trait helps plant survival and maintains genome integrity, unlike necrosis with uncontrolled cell death. Jasmonic acid (JA) is a phytohormone often associated with plant immunity by regulating the hypersensitive response, a type of PCD involved in plant resistance. In roots, JA increases the expression of *ERF115* to induce cell regeneration when cell damage occurs^[1]. But how JA regulates PCD in roots is an open question. Here, we report that JA inhibits the PCD in *med18* mutants (*med18-1* and *med18-2*) of the MEDIATOR complex. Previously, the *med18* alleles were identified with the presence of cell death in root meristems, which correlated with the increase of root width, root hairs, and the overexpression of *ERF115*^[2]. Upon JA treatments, the root phenotype of *med18* was similar to that in wild-type (WT), and the *ERF115* factor was expressed in the vasculature of WT and the meristem of *med18*, indicating that JA repressed the PCD and promotes cell regeneration. The JA responses are controlled by the COI1 receptor. Assays with the double mutant *coi1-1 med18-1* showed that the repressing effect of JA on PCD depends on the COI1 receptor. Foliage wounding stimulates JA biosynthesis and activates *JAZ* expression. The JA signals are transmitted from shoot-to-roots where it influenced root growth. Time-dependent analysis revealed that wounds increased *JAZ1* expression and decreased PCD in the root of *med18* mutants. This suggests that JA signals are naturally transmitted from the shoot to protect root cells from environmental stress cues.

References

1. Zhou W, Lozano-Torres JL, Blilou I, Zhang X, Zhai Q, Smant G, Li C, Scheres B (2019). A Jasmonate Signaling Network Activates Root Stem Cells and Promotes Regeneration. *Cell* 177(4):942-956.e14
2. Raya-González J, Oropeza-Aburto A, López-Bucio JS, Guevara-García AA, de Veylder L, López-Bucio J, Herrera-Estrella L (2018). MEDIATOR18 influences Arabidopsis root architecture, represses auxin signaling and is a critical factor for cell viability in root meristems. *Plant J.* 96(5):895-909

CELL SENESCENCE MODIFIES PHENOTYPE AND FUNCTION OF MAST CELLS

Alfredo Ibarra-Sánchez¹, Iris K. Madera-Salcedo², Diana L. Esparza-Reyes¹, Paola Mendoza-Montiel¹, Alejandro Padilla-Trejo³ and Claudia González-Espinosa^{1,4,#}

¹Pharmacobiology Department, Center for Research and Advanced Studies (Cinvestav) of the Polytechnic Institute (IPN), South Campus. Calzada de los Tenorios # 235 Col. Rinconada las Hadas, Tlalpan, 14330, Mexico City, Mexico. ²Rheumatology Department, National Institute of Medical Sciences and Nutrition "Salvador Zubirán". Vasco de Quiroga 15, Belisario Domínguez Secc. 16, Tlalpan, 14080, Mexico City, Mexico. ³Departamento de Microbiología and Parasitología Faculty of Medicine, National Autonomous University of Mexico, Circuito Exterior S/N, Ciudad Universitaria, 04510, Mexico City, Mexico. ⁴Center for Research on Aging (CIE), Cinvestav. South Campus. Calzada de los Tenorios # 235, Col. Granjas Coapa, Tlalpan, 14330, Mexico City, Mexico. #Correspondence: cgonzal@cinvestav.mx.

Mast cells (MC), derived from myeloid stem cells, play a critical role in both innate and adaptive immunity. As occurs with other immune cells, MCs can become senescent due to replicative processes, which can impact their functionality and responses to different stimuli. In this study, we investigated how cell senescence affects the phenotype and responsiveness of MCs. Bone marrow-derived mast cells (BMMCs) were derived from 6 to 12 week-old WT C57BL/6J mice and cultured over several weeks to monitor the expression of main senescence markers and responsiveness to FcεRI receptor crosslinking. Analysis included cell cycle arrest, senescence-associated β-galactosidase expression, key proteins such as p16 and p21, and the appearance of SASP (Senescence-Associated Secretory Phenotype) markers like IL-6, TNF, VEGF, IL-1α, IL-1β, IL-10, CXCL-1, and CCL-2. Our findings reveal that replicative senescence is established and sustained in BMMCs after 13 weeks in culture, and that senescence modifies FcεRI-induced responses, including signaling protein activation, intracellular calcium mobilization and β-hexosaminidase secretion. This study sheds light on senescence's modulation of MC function.

Project supported by the grant Conahcyt CF-2019-51488 to CGE and Scholarships No. 1230115 to DLER and 1148727 to PMM

SPERMATOZOA ADHESION TO AN IMMOBILIZED FIBRONECTIN MATRIX ALTERS THEIR PHYSIOLOGY AND INCREASES THEIR SURVIVAL

Coral Y. Jorge-Cruz, Ana L. Roa-Espitia, Enrique O. Hernández-González

Departamento de Biología Celular, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional.
Av. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, Municipio Gustavo A. Madero, México City 07360, México.
Email: enrique.hernandez@cinvestav.mx

Isthmus is the region of the oviduct considered a reservoir for spermatozoa, where they are retained and then released synchronously with ovulation. Integrins mediate this interaction, and it is suggested that they regulate the viability and capacitation of spermatozoa. Spermatozoa retained in the oviductal epithelial cells show specific characteristics: normal morphology, intact acrosome and plasma membrane, no DNA fragmentation and low intracellular Ca^{2+} and protein phosphorylation levels at Tyr. This work aimed to define spermatozoa's ability to adhere to an immobilized fibronectin matrix and its effects on their viability and capacitation. We found that guinea pig spermatozoa showed a high affinity to adhere to an immobilized fibronectin matrix but not to those made up of type I collagen or laminin. This interaction was mediated by integrins that recognize the RGD domain. Spermatozoa adhered to an immobilized fibronectin matrix were maintained in a state of low capacitation: low levels of intracellular concentration of Ca^{2+} , protein phosphorylation in Tyr and F-actin. Also, they kept their plasma membrane and acrosome intact, flagellum beating, and showed low activation of caspases 3/7. The spermatozoa adhered to the immobilized fibronectin matrix, gradually detached, forming rosettes and did not undergo a spontaneous acrosomal reaction but were capable of experiencing a progesterone-induced acrosomal reaction. In conclusion, the adhesion of spermatozoa to an immobilized fibronectin matrix alters the physiology of the spermatozoa, keeping them in a steady state of capacitation, increasing their half-life in a similar way to what was reported for spermatozoa adhered to oviductal epithelial cells.

CONAHCYT: CB-284183 y CBF2023-2024-1894.

POSSIBLE INVOLVEMENT OF PAK1 AND CAMKII IN INSULIN SECRETION BY PANCREATIC BETA CELLS

Nely Gisela López Desiderio^{1,4}, Olga Villamar Cruz², Luis Enrique Arias Romero²,
Jazmín García Machorro⁴, Héctor Iván Saldívar Cerón^{1,2}

¹Carrera de Médico Cirujano, Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México, Tlalnepantla 54090, México. ²Laboratorio 14, Unidad de Biomedicina (UBIMED), Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México, Tlalnepantla 54090, México. ³Red de Medicina para la Educación y Desarrollo y la Investigación Científica de Iztacala (Red MEDICI), Carrera Médico Cirujano, Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México, Tlalnepantla 54090, México. ⁴Laboratorio de Medicina de Conservación, Escuela Superior de Medicina, Instituto Politécnico Nacional, Ciudad de México, 11340, México. Facultad de estudios Superiores Iztacala, UNAM y Escuela Superior de Medicina, IPN. Laboratorio 14, Unidad de Biomedicina (UBIMED), Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México, Tlalnepantla 54090, México.
Correo electrónico: oivansaldi@iztacala.unam.mx

Type 2 diabetes, a persistent condition marked by heightened blood sugar levels stemming from the body's resistance to insulin and reduced insulin output from pancreatic beta cells, is the subject of ongoing investigation into its underlying mechanisms. Insulin release hinges on intricate intracellular mechanisms governed by an array of proteins and enzymes.

This study delves into the potential collaboration between two enzymes, Pak1 and CaMKII, pivotal players in various stages of insulin secretion within pancreatic beta cells. Employing indirect immunofluorescence techniques, we observed the co-localization and formation of a protein complex involving Pak1 and CaMKII exclusively when stimulated with 20 mM glucose, contrasting with lower glucose levels at 4 mM glucose.

Furthermore, we conducted pharmacological inhibition experiments using Frax-1036 to target Pak1 and KN-93 to inhibit CaMKII. The assessment of insulin secretion via ELISA revealed a notable reduction under both inhibition conditions, with a suggestive trend towards further suppression upon simultaneous inhibition of both enzymes.

Additionally, mRNA expression analysis of Pak1 and CaMKII in beta cells sourced from healthy individuals, those with pre-diabetes, and diabetic patients uncovered heightened expression levels in pre-diabetic and diabetic cohorts. Notably, expression elevation initially observed in pre-diabetes potentially serves as a compensatory mechanism, though it normalizes in established T2DM, likely due to beta cell destruction, a hallmark of T2DM pathophysiology.

These findings propose the involvement of Pak1 and CaMKII in insulin secretion dynamics by pancreatic beta cells. Their overexpression in pathological states hints at their potential as early indicators for pre-diabetes and T2DM detection. Experimental data corroborated a substantial reduction in insulin secretion upon pharmacological inhibition of Pak1 and CaMKII, with implications for clinical practice. However, further research is essential to precisely delineate their roles in insulin secretion and assess their clinical relevance comprehensively.

LIPID DROPLETS IN PLANT CELLS: INTERACTION WITH NUCLEI AND THE ENDOPLASMIC RETICULUM

Alejandro López Hernández, Jesús Salvador López-Bucio, José López-Bucio*

Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Edificio B 3,
Ciudad Universitaria, C. P. 58030, Morelia Michoacán, México.
e-mail: jlbucio@umich.mx*

Lipid droplets are critical vesicles in animal cells that enables energy conversions. Instead, in plants, photosynthesis mostly drives the production of sugars and starch as energetic reserves, whereas lipid vesicles have been scarcely reported. In this report, we show the dynamic behavior of lipid droplets in onion epidermal cells, in which these vesicles are more mobile than in other cell types. Noteworthy, lipid droplets were found to move along cytoplasmic currents and interact with the nuclear and endoplasmic reticulum membranes in response to auxin and cytokinin treatments, indicating the importance of these small organelles not only in energetic metabolism but also in organellar maintenance.

Still not a lot is known about their functions in vegetative tissues, but work recently is revealing interesting similarities and differences in how the lipid droplets are formed in cells of different plant parts and among other diverse organisms, indicating the ubiquity of their function in eukaryotic cells.

References

1. Ischebeck T, Krawczyk HE, Mullen RT, Dyer JM, Chapman KD. 2020. Lipid droplets in plants and algae: Distribution, formation, turnover and function. *Semin Cell Dev Biol.* 108:82-93.

INSIGHTS INTO THE REGULATION OF SNRK1 THROUGH PHOSPHORYLATION BY UPSTREAM KINASES

Carmen Aitana López Zequeira, Beatriz Alejandra Ávila-Castañeda and Patricia Coello-Coutiño*

Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México, México City. 04510.
E-mail: pcoello@unam.mx

The Snf1-related protein kinase 1 (SnRK1) is the plant ortholog of AMP-activated protein kinase (AMPK) in mammals and sucrose non fermenting (SNF1) from yeast. SnRK1 orchestrates the response to unfavorable conditions such as nutrient deficiency. It consists of three subunits: one catalytic subunit (α) and two regulatory subunits (β and $\beta\gamma$). In *Arabidopsis thaliana*, the α subunit has two main isoforms, $\alpha 1$ and $\alpha 2$. A key regulatory mechanism of SnRK1 is phosphorylation by upstream kinases. SnAK1 and SnAK2 phosphorylate a canonical threonine residue within the T loops of catalytic subunits ($\alpha 1/\alpha 2$ T175/T176 respectively), which is essential for SnRK1 kinase activity. Our research has shown the importance of phosphorylating the residue adjacent to the canonical threonine (Ser 176/177 of $\alpha 1/\alpha 2$) both *in vitro* and *in vivo*. However, the impact of this serine phosphorylation on global SnRK1 activity is not yet fully understood. To monitor the conditions and developmental stages at which threonine/serine phosphorylation occurs *in vivo* we are employing two main approaches. First, we are developing a genetically encoded biosensor based on Förster Resonant Energy Transfer (FRET) to detect kinase activity on the $\alpha 1$ subunit. This biosensor is modeled after a previously described MAPK activity biosensor. Our goal is to create different versions of the biosensor containing various regions around the T-loop, to analyze their behavior upon phosphorylation. Second, we are using specific phospho-antibodies against T175 and Ser176 to gain a comprehensive and detailed understanding of how phosphorylation occurs and regulates SnRK1 activity.

PAPIIT: IN201922, PAIP 5000-9126. Conahcyt scholarship funding 1278275

CANNABIDIOL INCREASES ADIPOGENESIS ON 3T3-L1 PRE-ADIPOCYTES VIA PPAR γ

Helen Yarimet Lorenzo-Anota¹, Silvia López^{1,2}, Luis Luevano¹, Eduardo Vázquez-Garza¹, Gerardo García-Rivas^{1,2}, Jorge Benavides^{1,3}, Karla Mayolo-Deloisa^{1,3}, Marco Rito-Palomares^{1,3}, Omar Lozano García^{1,2*}

*Correspondence

¹Tecnológico de Monterrey, Institute for Obesity Research, Av. Eugenio Garza Sada 2501 Sur, Monterrey NL 64849, México.

²Tecnológico de Monterrey, Escuela de Medicina y Ciencias de la Salud, Av. Morones Prieto 3000 Pte, Monterrey, NL, Mexico.

³Tecnológico de Monterrey, Escuela de Ingeniería y Ciencias, Centro de Biotecnología-FEMSA, Av. Eugenio Garza Sada 2501 Sur, Monterrey, NL 64849, Mexico.

Cannabidiol (CBD), a major non-psychoactive phytocannabinoid of *Cannabis sativa*, has demonstrated a wide medical application as anti-inflammatory, immunomodulator, anxiolytic, anti-psychotic, and anti-emetic. Recently, anti-obesity effect through adipocyte browning was reported. However, phenotype, effector function and metabolic changes are not yet demonstrated. This project evaluated the effect of CBD on 3T3-L1 pre-adipocytes during differentiation into white-like adipocytes. Intracellular lipid droplets were assessed by oil red stain and analyzed on ImageJ. Triglycerides content was analyzed by colorimetric assay and flow cytometry using Bodipy stain. To evaluate molecular phenotype, relative RNA expressions were quantified by qPCR, and to evaluate immunophenotype, the surface markers CD40 and Eva-1 were tested by flow cytometry. Results indicate that CBD does not modify cellular metabolism of pre-adipocytes 3T3-L1 at 24, 48 and 72 h. Nevertheless, during adipogenesis, CBD increased the frequency of lipid droplets in the range 5-20 μm^2 (3.9 ± 0.8 -fold) at day 10 and day 14 (1.6 ± 0.02 -fold). Also, CBD increased triglycerides at day 10 (1.81 ± 0.5 -fold) and day 14 (1.97 ± 0.45 -fold). Interestingly CBD did not modify oxygen consumption, and relative RNA expression of CIDEA, FGF21, and UCP-1 on white-like adipocytes. In contrast, its administration increased gene expression of PPAR γ (1.87-fold), PGC-1 α (1.2-fold) and adiponectin (2.6-fold) at day 10, which additionally correlated with high adiponectin release in supernatant. Also, it decreased the relative gene expression of Pref-1 (0.18-fold), suggesting enhanced adipocyte differentiation. Preliminary data demonstrates that CBD promoted surface markers associated with browning adipocytes. In conclusion, CBD enhanced adipogenesis on differentiated adipocytes, suggesting that pathways of PPAR γ and PGC1 α are associated with lipogenesis.

THROMBIN-INDUCED ACTIVATION OF FOCAL ADHESION PROTEINS IN THE RETINAL PIGMENT EPITHELIUM (RPE)

Katya Manzanita Quintero, Irene Lee Rivera, Edith Catalina López Hernández, Ana María López Colomé*

*Instituto de Fisiología Celular, UNAM, Circuito Exterior s/n, Ciudad Universitaria, México City. C.P. 04510, México, Tel.: +52 55 5622 5517; acolome@ifc.unam.mx

The retinal pigment epithelium (RPE) is a monolayer of pigmented cells that, along with Bruch's membrane, constitute the outer blood-retinal barrier; which, when damaged, promotes photoreceptor death. Among the diseases affecting the RPE, proliferative vitreoretinopathy (PVR) involves the transformation and migration of RPE cells into the vitreous cavity and underneath the retina. These cells, along with Müller glial cells form membranes capable of detaching the retina.

In previous studies, it has been shown thrombin's activity increases in the vitreous cavity of patients with PVR, which can induce cell migration. This process involves the assembly and disassembly of focal adhesion proteins such as focal adhesion kinase (FAK), Paxillin and proline-rich tyrosine kinase 2 (Pyk2). Previous work in the laboratory has shown that thrombin activates FAK, resulting in increased RPE cell migration. This suggests that other proteins involved in focal adhesions, such as Paxillin and Pyk2 are also activated by this protease. Therefore, the aim of this project is to determine the effect of thrombin on the phosphorylation of these proteins. Paxillin and Pyk2 activation was assessed by Western Blot in RPE primary cultures. Using thrombin concentration curves, we determined that the activation peak corresponds to a 5U/ml concentration for both proteins. Also, we evaluated activation from 2 to 30 minutes of stimulation and found maximum phosphorylation at 5 minutes for Paxillin and 2 minutes for Pyk2. Finally, we evaluated the specificity of the phosphorylation of both proteins using Hirudin and PPACK, specific thrombin inhibitors, and the PAR1-specific antagonist SCH79797, finding that the activation of Paxillin and Pyk2 is specific for thrombin, through PAR1.

In conclusion, thrombin promotes the assembly of focal adhesions by inducing the phosphorylation of key proteins, which may result in increased RPE cell transformation and migration. The determination of optimal timing and thrombin concentration for the induction of these proteins, can be of importance to develop future therapeutic treatments for diseases such as PVR or to prevent the deleterious effects of retinal detachment on photoreceptor death.

THE ROLE OF APOPTOSIS IN THE DEVELOPMENT OF THE DROSOPHILA TRACHEAL SYSTEM AND ITS RELATIONSHIP WITH PROTRUDING CELLS OF THE TRACHEAL DORSAL TRUNKS

Pedro Alberto Marmolejo Cruz¹, Daniel Ríos Barrera¹

¹Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, UNAM. Ciudad Universitaria, 04510. Ciudad de México. México.

*Contacto: 55562-29250 ext. 46804, daniel.rios@iibiomedicas.unam.mx

The *Drosophila* respiratory or tracheal system is a network of interconnected tubes that brings oxygen to target tissues throughout the body of the animal. This tissue is formed by the invagination of ectodermal cells that form tubes that branch and fuse to form a continuous network directly below the epidermis. Our lab has identified a subpopulation of tracheal cells (referred to as “protruding cells”), that interact with the extracellular matrix underneath the epidermis. These cells are important mediators of coordinated development between the tracheal system and the epidermis. Other works have shown that tracheal cells that match the position of protruding cells undergo apoptosis and are extruded from the tissue towards the end of embryogenesis. The aim of this work is to define if protruding cells and the previously identified apoptotic cells correspond to the same cell population. Using an *in vivo* apoptosis fluorescent sensor, and immunostainings for apoptosis and protruding cell markers, our results suggest that the two cell populations overlap at least partially. We have also observed that the process of cell death coincides in time with the maturation of the epidermis, and that blocking apoptosis results in an aberrant tracheal morphogenesis. We speculate that these defects are a consequence of inadequate detachment from the epidermis towards the end of embryonic development.

SPECIFIC TEMPORAL REQUIREMENT OF PROX1 ACTIVITY DURING PANCREATIC ACINAR CELL DEVELOPMENT

Angelica S. Martinez-Ramirez¹, Thomas L. Borders¹, Leena Paul², Matthew Schipma³, Xinkun Wang³, Farida Korobova⁴, Christopher V. Wright⁵ and Beatriz Sosa-Pineda¹

¹Department of Medicine, Feinberg Cardiovascular and Renal Research Institute, Northwestern University Feinberg School of Medicine, Chicago, Illinois; ²Department of Genetics, St. Jude Children's Research Hospital, Memphis, Tennessee; ³Center for Genetic Medicine, Northwestern University Feinberg School of Medicine, Chicago, Illinois; ⁴Center for Advanced Microscopy, Northwestern University Feinberg School of Medicine, Chicago, Illinois; and ⁵Department of Cell and Developmental Biology, Vanderbilt University, Nashville, Tennessee.

Pancreatic acinar cell maturation requires a complex regulatory network of transcription factors involving their expression and downregulation throughout the organogenesis process.

Deciphering the transcriptional mechanisms involved in this process will allow us to design protocols to induce acinar cell differentiation.

Prox1 is a transcription factor highly expressed in murine pancreatic progenitors, later its expression is conserved along several mature pancreatic cell types, however it is no longer present in mature acinar cells. Here we investigated expression Prox1 pattern along the acinar cell maturation and its consequences in the organogenesis process. By using murine models of Prox1 ectopic expression and Prox-1 null embryos we also studied how this transcription factor can be involved in acinar cells specification and differentiation.

We found that Prox-1 is expressed in pancreatic progenitors and in newly committed embryonic acinar cells. In Prox-1 null embryos (Prox1^{GFPCre/Δ}) we found precocious expression of "late" acinar genes by RNA-sequencing, suggesting that Prox-1 expression in pancreatic progenitors delay acinar maturation to allow proper sequential gene expression. However, in the Prox1 ectopic expression model (Prox1^{AcOE}) we found that adult mice had pancreatic alterations including reduced acinar genes expression, acinar atrophy, abnormal secretory granules, increased endoplasmic reticulum stress and mild chronic inflammation. All together these results suggest that Prox1 expression is necessary in early acinar cells to allow correct sequential gene expression, nevertheless Prox1 expression must be terminated during developing acinar cells to allow complete maturation and maintain pancreas homeostasis.

ACTIVATION OF THE IGF-1/IGF-1R INCREASES CHEMORESISTANCE TO PACLITAXEL OF NON-SMALL CELL LUNG CANCER CELL LINE (A549)

Max Alejandro Maximino Rojas, Natalia Herrera Castillo, María Julieta López Cuevas,
Eduardo Monjaraz Guzmán, Fabián Galindo Ramírez

Benemérita Universidad Autónoma de Puebla
Prolongación de la 14 sur 6301. Cd Universitaria, Puebla, Pue.
max.mr0911@gmail.com Cel. 2229260920

Insulin-like growth factors (IGF) are a set of elements of protein nature, with high sequence similarity to insulin, which physiologically participate in the normal development of an individual. However, its participation in the progression of various types of cancer has been widely documented. Lung cancer is the leading cause of cancer mortality worldwide. It is classified into two types: a) non-small cell lung carcinoma (NSCLC) and, b) carcinoma of the small cell lung. The NSCLC represents 75% of the total lung cancer cases.

In this context, the aim of the present work is to determine if the IGF signal transduction system exerts the chemoresistance of non-small cell lung cancer cell line (A549). The A549 cell line was maintained under standard culture conditions. A549 cells were treated for 72 hours with IGF-1 at different concentrations (100 or 200 ng/mL) and paclitaxel 500nM for 48 hours. At the end of the treatment, it was evaluated whether the activation of IGF-1R, as well as its signaling pathways: PI3K/AKT pathway and MAPK pathway affects the chemoresistance of these cells. With the interest of being able to discriminate which of the two intracellular pathways is involved in the chemoresistance, stimulated by IGF-1, we used specific antagonists for each of the pathways, as well as the IGF-1R. Experimental tests were carried out to evaluate the role of IGF-1/IGF-1R complex on the chemoresistance to paclitaxel capacity. At the end of these tests, the evaluation of molecular markers associated with chemoresistance was carried out.

Our results indicate that the chronic presence of IGF-1 exacerbates the chemoresistance to paclitaxel of A549 cells, positively associated with an increase in the level of expression of Ki67, Birc3 and survivin 3. On the other hand, we show that the blocking either of the two pathways and IGF1R has a negative impact on their chemoresistance. With the results obtained, IGF-1/IGF-1R complex could be proposed as new pharmacological targets for the treatment of NSCLC.

INTERFERON-STIMULATED GENE 15 AND ISGYLATION LEVELS ARE MODULATED BY IFN-GAMMA IN MEDULLOBLASTOMA CELLS

Medina-Abreu Karen H., Velasco-Loyden Gabriela, Palacios-Serrato Eva, Oropeza-Martinez Enrique, Sosa-Garrocho Marcela, Macías-Silva Marina, Ángeles Tecalco Cruz*

Posgrado en Ciencias genómicas, Universidad Autónoma de la Ciudad de México (UACM) Apdo, 03100, Ciudad de México, México. Instituto de Fisiología Celular, UNAM, Apdo, 04510, Ciudad de México, México.

ISG15 (interferon-stimulated gene 15) is a small ubiquitin-like protein that can be detected as a protein of 15 kDa (free) or conjugated to its target proteins to modify them through a system denominated ISGylation similar to ubiquitination. ISG15 and ISGylation levels have been reported to increase in some cancer types. Furthermore, an upregulation of ISG15/ISGylation by IFN-gamma occurs in breast cancer and glioblastoma. However, the *ISG15* expression and modulation in pediatric nervous system tumors is unknown. Medulloblastoma is an embryonal tumor grade 4 that develops in the cerebellum and occurs mainly in children. Molecular studies are required to explore *ISG15* expression and its modulation by IFN-gamma in this cancer. We analyzed *ISG15* expression in human medulloblastoma samples using databases (OncoPrint, UALCAN-CBTTC) and examined the abundance of ISG15 protein in a tissue microarray containing medulloblastoma samples by immunohistochemistry. Moreover, we used the DAOY cell line with or without IFN-gamma treatment to determine *ISG15* expression through RT-PCR assays and the abundance of ISG15/ISGylation by immunoblot, whereas the subcellular distribution of ISG15/ISGylation was evaluated by subcellular enrichment and immunofluorescence. Our results indicate that the expression and abundance of ISG15 are reduced in samples from patients with medulloblastoma compared to normal tissue. IFN-gamma signal increases ISG15 levels in the nuclear and cytoplasmic compartment of DAOY medulloblastoma cells. Thus, IFN-gamma/ISG15 axis may be critical in the medulloblastoma context.

MECHANISMS OF ACTION AND SECRETION OF THE TGF- β CYTOKINE IN COLORECTAL CANCER

Christian Daniel Meza-Juárez¹, Marcela Sosa-Garrocho¹, David Toledo-Padilla¹, Cristina Castañeda Patlán², Martha Robles Flores², Gloria Soldevila³, Alfredo Ibarra-Sánchez⁴, Tzindilú Molina Muñoz⁴, Claudia González-Espinosa⁴ and Marina Macías-Silva¹

¹Instituto de Fisiología Celular, ²Facultad de Medicina and ³Instituto de Investigaciones Biomédicas de la UNAM, Mexico City 04510. ⁴Departamento de Farmacobiología del CINVESTAV-IPN, Mexico City 07360. E-mail: cdmeza@ifc.unam.mx

Colorectal cancer (CRC) is one of the most lethal cancers worldwide, ranking third in incidence and the second leading cause of cancer-related mortality. A critically important process in the development of CRC involves intercellular communication within the tumor microenvironment (TME), which comprises a diversity of non-cancerous cells and their interactions within the tumor. Additionally, it includes components such as the extracellular matrix and soluble factors like chemokines, cytokines, growth factors, and extracellular vesicles (EV) that enable intercellular communication¹. Among the cytokines involved in this cellular communication, transforming growth factor-beta (TGF- β) stands out as a cytokine with multiple biological effects, both in homeostatic conditions and in various pathologies, including CRC. It has been observed that, in the early stages of CRC, this cytokine exerts antitumor effects, while in advanced stages it promotes tumor progression². There is evidence that some CRC cells secrete TGF- β primarily in an active state and to a greater extent in EVs. Therefore, we were interested in studying how CRC T84 and SW620 cells secrete TGF- β , classified as CMS2 and CMS4 cells, respectively, both molecular subtypes that represents CRC cases. In fact, information on TGF- β secretion in CRC cells classified as CMS2 and CMS4 is limited. Therefore, in this study, we experimentally examined TGF- β secretion by T84 and SW620 cells cultured both in monolayer (2D) and spheroids (3D). The results show that T84 cells cultured in 2D secrete TGF- β in low amounts, mostly in a latent form, while 3D cultures have high TGF- β secretion both in latent and active forms, mainly in a soluble state with a small fraction found in microvesicles. SW620 cells were cultured in spheroids (25,000 cells vs. 200,000 cells) and apparently secrete larger amounts of TGF- β , which promotes the migration of mast cells. We also are investigating which components in the MC affect mast cell migration or also what promotes a synergism with TGF- β .

Our work is supported by grant No. IN211024 from PAPIIT, DGAPA, UNAM. CDMJ receives a fellowship from CONAHCyT for his M. Sc. studies.

References

1. De Visser, K.E, Joyce, J. A. (2023). *Cancer cell*, 41(3), 374-403.
2. Toledo-Padilla, D. (2023). *Revista de Educación Bioquímica (REB)* 42(3), 141-151.
3. Lara-Salas, M. A, García-Díaz, P. O., Mendoza-Lara, D. F., (2023). *Revista Especializada en Ciencias Químico-Biológicas*, 26, 1-18

EVALUATION OF THE EFFECT OF AMIODARONE AND DESETHYLAMIODARONE ON CELL PROLIFERATION AND MIGRATION IN LUNG CANCER CELLS, A549

Zurisadai Morales Herrera¹, María Julieta López Cuevas¹, José Everardo Avelino Cruz¹, Miguel Froylan Gómez Laguna², Eduardo Monjaraz Guzmán¹

¹Benemérita Universidad Autónoma de Puebla, Prolongación de la 14 sur 6301. Cd Universitaria, Puebla, Pue.

²Facultad de Medicina, Universidad Nacional Autónoma de México.

zuri943@hotmail.com

Lung cancer is one of the most commonly diagnosed cancers and remains the leading cause of cancer-related death worldwide. Non-small-cell lung cancer (NSCLC) is a heterogeneous group of tumors, accounting for roughly 75% of all new lung cancer diagnoses. Unfortunately, NSCLC is often characterized by reduced responsiveness to chemotherapy and radiotherapy, highlighting the critical need for the development of novel therapeutic targets to effectively combat this disease.

Emerging evidence suggests that a wide range of tumor cells overexpress hERG potassium channels, which play a crucial role in regulating various cellular processes, including cell proliferation, apoptosis, angiogenesis, cell survival, and migration. These findings underscore the potential of hERG channels as promising therapeutic targets for targeted therapy.

This study aimed to investigate whether blocking hERG channels using amiodarone (AD) and desethylamiodarone (DEA) could effectively reduce the metastatic potential of tumor cells and consequently impede lung cancer progression, thereby improving patients' life expectancy in the short, medium, or long term.

The A549 cell line served as the experimental model. To assess the impact of treatment on cell proliferation and migration, A549 cells were exposed to 10 mM AD and 2 mM DEA. Additionally, RT-PCR was employed to quantify the mRNA expression levels of molecular markers associated with cell proliferation and survival, including Ki-67, Bcl-2, cyclin B, cyclin E, and Bax.

Treatment with AD and DEA resulted in a significant reduction in the proliferative capacity of A549 cells, exhibiting a decrease of approximately 20% and 23%, respectively, compared to the control condition. This effect was attributed to a marked decrease in Bcl-2 mRNA expression levels and a trend towards increased Bax expression.

Our findings suggest that blocking hERG channels with AD and DEA enhances the apoptotic capacity of A549 cells, providing preliminary evidence for the potential of hERG channels as therapeutic targets in NSCLC treatment.

RANKL DIFFERENTIALLY REGULATES BREAST CANCER STEM CELLS ACCORDING TO RANK OR LGR4 ACTIVATION

Alejandro Ordaz Ramos¹, Jorge Diaz Blancas², Rosario Castro Oropeza², Aketzalli Martinez Cruz², Jorge Meléndez Zajgla³, Vilma Maldonado Lagunas² and Karla Vazquez Santillan¹

Núcleo B de innovación en medicina de precisión¹, Laboratorio de Epigenética² and Laboratorio Genómica Funcional del Cáncer³. Instituto Nacional de Medicina Genómica. Periférico Sur 4809, Arenal Tepepan, Tlalpan, 14610 Ciudad de México, CDMX
5553501900 ext. 1224; kivazquez@inmegen.gob.mx

Breast cancer stem cells (BCSCs) are tumor cells with the capacity of self-renewal and the ability to differentiate. Some studies have demonstrated that BCSCs are responsible for resistance and relapse of breast cancer. BCSCs are regulated by signaling pathways that induce self-renewal maintaining undifferentiated phenotype and avoiding differentiation.

The receptor activator of nuclear factor kappa- β ligand (RANKL) is a cytokine that can act through two receptors: RANK and LGR4. It has demonstrated that RANKL/RANK signaling induces BCSC characteristics. However, the effect of RANKL through LGR4 has not been elucidated.

Recent evidence in osteoclasts has shown that RANKL/LGR4 blocks RANK signaling and generates a cellular opposite response. Since RANKL/RANK signaling rapidly expands the pool of BCSCs, anti-RANKL inhibitors have increasingly been proposed to eradicate BCSCs. Nevertheless, RANKL-inhibitor therapies fail to consider the biological effects of disrupting RANKL binding to LGR4.

Here, we evaluated the impact of RANKL inhibition on breast cancer cells according to LGR4 expression. Further, we analyzed the signaling pathways that RANKL activates when acts through LGR4 or RANK.

We analyzed the survival of breast cancer patients according to RANKL expression and found that RANKL represented a good prognosis factor in patients with high levels of LGR4. In LGR4-low cell lines, RANKL inhibition decreased BCSC properties. However, in LGR4-high cells, RANKL inhibition does not affect BCSCs. Moreover, we found that RANKL treatment activates NF- κ B signaling and promotes BCSC properties on RANK-overexpressed breast cancer cells. However, in LGR4-overexpressed cells, RANKL failed to activate NF- κ B, blocked the Wnt/ β -catenin pathway, and decreased BCSC activity.

In conclusion, RANKL regulates differentially BCSCs according to RANK or LGR4 activity. RANKL/RANK promotes BCSC characteristics and through LGR4, blocks BCSC properties. RANKL inhibitors have different responses according to LGR4 levels in breast cancer. LGR4 levels need to be evaluated before propounding RANKL therapies to treat breast cancer patients.

DIASTOLIC DYSFUNCTION, EXCITATION-CONTRACTION COUPLING DEFECTS AND ARRHYTHMIAS IN ABSENCE OF MITOCHONDRIA Ca^{2+} UPTAKE

Marilen Federico¹, Facundo Barraque¹, Yuexing Yuan¹, Sergio De La Fuente², Hui-Ying Tsai¹, Shey-Shing Sheu¹, Julieta Palomeque³

¹Center for Translational Medicine, department of medicine, Thomas Jefferson University. ²Departamento de bioquímica biología molecular y fisiología, universidad de Valladolid. ³Institute for Obesity Research-Tec de Monterrey, Quetzales 2968, Guadalupe CP 67178, +52 8182 74 6115, jpalomeque@tec.mx

Mitochondrial Ca^{2+} is important in promoting mitochondrial ATP production, however mitochondrial Ca^{2+} uniporter (MCU) knock-out (KO) mice did not show significant bioenergetics changes unless exposed to stress. This leads to whether the MCU is rescindable in physiological conditions. We hypothesize that when mitochondrial Ca^{2+} uptake is abolished, such as MCU global KO, the heart undergoes adaptations that prevent bioenergetic breakdown under physiological conditions but are prone to develop cellular arrhythmias due to abnormal ECC.

We used wild-type (WT) and MCU global KO (KO) mice to measure diastolic and systolic function by echocardiography, electrical rhythm by electrocardiogram (ECG), and ECC in isolated cardiomyocytes. T-test was used for groups comparison, $p < 0.05$ (*) was considered significant difference.

Echocardiographic results showed no difference in hypertrophy parameters or ejection fraction in KO vs WT mice. Diastolic function analysis of KO revealed a decrease in the reverse longitudinal strain rate (**rLSR**, 3.5 ± 0.86 vs $5.5 \pm 0.4^*$ KO vs WT) and global longitudinal strain (**GLS**, -13.2 ± 2.8 vs $-19.7 \pm 1.4^*$ KO vs WT). KO also presented a higher isovolumetric relaxation time (**IVRT**, 14.3 ± 1.4 vs $11.1 \pm 0.5^*$ KO vs WT), ejection time (**ET**, 43.33 ± 1.4 vs $38.8 \pm 1.0^*$ KO vs WT) and myocardial performance index (**MPI**, 0.57 ± 0.04 vs $0.47 \pm 0.02^*$ KO vs WT) than WT. At the ECC level, KO showed reduced fractional shortening (4.8 ± 0.8 vs $7.4 \pm 0.8^*$ KO vs WT) with an increment in relaxation time (0.21 ± 0.01 vs $0.19 \pm 0.006^*$ KO vs WT). Moreover, KO showed an increased Ca^{2+} Transient amplitude (**CaIT**, 50.8 ± 5.4 vs $38.2 \pm 2.2^*$ KO vs WT) and peak height (0.32 ± 0.03 vs $0.26 \pm 0.01^*$ KO vs WT) with a reduced basal Ca^{2+} (0.68 ± 0.02 vs $0.73 \pm 0.01^*$ KO vs WT). No differences were found in Ca^{2+} re-uptake in WT vs KO. Thus, to unmask a possible NCX hyperactivity in the KO we used an inhibitor of NCX (Sea400). KO Ca^{2+} relaxation time at 10%, was increased vs WT (0.06 ± 0.003 vs $0.05 \pm 0.001^*$ KO vs WT). At the ECG level, KO animals showed a higher frequency of arrhythmias normalized by the beats per minute and the total recording time vs WT (0.0005 ± 0.0001 vs $0.0002 \pm 0.00003^*$ KO vs WT).

The results showed that while the systolic function is preserved in animals lacking MCU, the diastolic function is impaired under non-stress conditions. This data is supported by abnormal ECC where there is a decrease in myofilament sensitivity to Ca^{2+} and impaired relaxation. Cytosolic Ca^{2+} is greater in KO mice and systolic Ca^{2+} removal is associated with an NCX hyperactivity which could predispose to cardiac arrhythmias.

ROLE OF INDOLEAMINE SEROTONIN IN MODULATING CELL DEATH IN THE ROOT MERISTEM OF *ARABIDOPSIS THALIANA*

Ramón Pelagio-Flores¹, Javier Raya-González¹ and José López-Bucio²

¹Facultad de Químico Farmacobiología, Universidad Michoacana de San Nicolás de Hidalgo, Tzintzuntzan 173, Matamoros, C. P., 58240 Morelia, Michoacán, México. ²Instituto de Investigaciones Químico-Biológicas, Universidad Michoacana de San Nicolás de Hidalgo Edificio B3, Ciudad Universitaria. C. P. 58030 Morelia, Michoacán. México.

Serotonin (5-hydroxytryptamine) is a natural molecule that activates diverse processes in these organisms, including growth and development, as well as plant defense and has important protective effects against biotic and abiotic stresses. We have previously reported on the effects of serotonin on Arabidopsis root architecture and its possible mechanisms of action. To better understand its natural functions in plants, we studied the effects of serotonin on cell death in the Arabidopsis root meristem in this work. Two cell death induction systems were used, and the serotonin effect was evaluated. An Arabidopsis mutant with spontaneous cell death in the root meristem shows tolerance to the primary root growth inhibition induced by serotonin. This response correlated with an active meristem and a reduction or complete abolition of the cell death in the root meristem, as evidenced by the gene expression of *CycB1:uidA* and *pERF115:GUS-GFP*. Interestingly, not all mutants with spontaneous root meristem cell death showed the same response, and serotonin could not protect or prevent the cell death induced by the genotoxic agent zeocin on root meristem; on the contrary, this seems to exacerbate the zeocin effects, resulting in shorter roots and morphological alterations in the root under serotonin treatments. Our data indicates that serotonin is involved in cell death regulation, but it differentially modulates this process in Arabidopsis root meristem in a specific manner.

PXD1 AND ORGANELLE TRAFFICKING: IMPLICATIONS FOR SEXUAL DEVELOPMENT IN *PODOSPORA ANSERINA*

Matías Ramirez-Noguez¹, Beatriz Aguirre-Lopez¹,
Sebastián Palacios-Martinez¹, Leonardo Peraza Reyes^{1*}

¹Instituto de Fisiología Celular, Universidad Autónoma de México. Cto. Exterior s/n, C.U., Coyoacán, 04510
Ciudad de México, México.

*lperaza@ifc.unam.mx

Given the polarized growth of filamentous fungi cells, the intracellular transport system is characterized by directed polarized movement of organelles along hyphae. This movement relies on a network of cytoskeletal elements and molecular motors that propel various cargoes. In some instances, organelles hitchhike on others, achieving indirect transport. PxdA, a coiled-coil domain protein, serves as a key mediator of peroxisome hitchhiking on early endosomes in *Aspergillus nidulans*. In *Podospora anserina*, Pxd1 exhibits broader role, influencing somatic development, growth, peroxisome motility, and senescence. During sexual development, precise spatial and temporal organization of organelles is critical for proper meiotic-spore (ascospore) formation. This study investigates the role of Pxd1 in *P. anserina*, through the knock-out and partial deletion of *PXD1*. We showed that Pxd1 contributes to the correct distribution of peroxisomes and the endoplasmic reticulum. Additionally, we observed its influence on spore formation, spindle body distribution, and the time frame of sexual development. Our results disclose an important role for peroxisome mobility regulation during the sexual phase of *P. anserina*. This research was supported by grant IN227823 from PAPIIT- DGAPA.

Keywords: Intrahyphal trafficking, molecular motors, meiosis, sexual development, ascospore formation, peroxisome distribution, Pxd1.

PARTICIPATION OF THE RAC1 GTPASE IN PROGRESSIVE MOTILITY IN GUINEA PIG AND MOUSE SPERM

Danelia Ramírez Ramírez¹, Monica Lizbeth Salgado Lucio¹, Ana Lilia Roa Espitia¹,
Joaquín Cordero Martínez² y Enrique Othón Hernández González¹

¹Departamento de Biología Celular, Centro de Investigación y Estudios Avanzados del Instituto Politécnico Nacional. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, Ciudad México, México. ²Departamento de Bioquímica, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional. Prolongación Manuel Carpio y Plan de Ayala s/n Col, Santo Tomás, Del. Miguel Hidalgo, México City, México.
Email: enrique.hernandez@cinvestav.mx

The sperm is a highly differentiated cell, and its primary function is to fertilize the egg to combine its genome and initiate the development of a new organism. Sperm motility has an essential role in fertilization. Mammalian spermatozoa show two types of physiological motility: 1) activated motility, observed in freshly ejaculated sperm, and 2) hyperactivated motility, observed in sperm recovered from the site of fertilization. Hyperactivated motility is crucial for sperm to propel themselves through the viscous medium of the oviduct and penetrate the envelopes that cover the egg. Mammalian sperm express different Rho proteins (Cdc42, RhoA and Rac1) in the flagellum, and recently, it suggested that Rac1 is essential by regulating progressive sperm motility, which is necessary for sperm to reach the egg. Sperm competitiveness appears to depend on an optimal level of active Rac1 since both a reduced or an excessive Rac1 activity interferes with progressive motility. Therefore, this work aimed to determine the role of Rac1 in the motility of guinea pig and mouse sperm. The presence of Rac1 was determined in guinea pig and mouse sperm. Inhibition of the GTPase Rac1 induces a morphological alteration of the flagellum in both guinea pig and mouse sperm. Inhibition of Rac1 modifies the factors associated with hyperactivated motility in mice and guinea pigs: VCL and ALH, LIN and WOB. It was determined that Rac1 does not regulate motility through the actin cytoskeleton. It was also observed that the Rac1 inhibition changed the phosphorylation of tyrosine, serine and threonine residues. Therefore, Rac1 could regulate motility through the phosphorylation of flagellar cytoskeletal structures.

CONAHCYT: CB-284183 y CBF2023-2024-1894.

LOSS-OF-FUNCTION OF MEDIATOR 12 OR 13 SUBUNITS CAUSES THE SWELLING OF ROOT HAIRS IN RESPONSE TO SUCROSE AND ABSCISIC ACID IN ARABIDOPSIS

Javier Raya-González¹, León Francisco Ruiz-Herrera², José López-Bucio²

¹Facultad de Químico Farmacobiología, Universidad Michoacana de San Nicolás de Hidalgo, Morelia Michoacán, México.

²Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Morelia Michoacán, México.

E-mail: javier.raya@umich.mx

Root hairs are epidermal cell extensions that increase the root surface for water and nutrient acquisition. Thus, both the initiation and elongation of root hairs are critical for soil exploration and plant adaptation to ever changing growth conditions. Here, we describe the critical roles of two subunits of the Mediator complex, MED12 and MED13, in root hair growth in response to sucrose and abscisic acid, which are tightly linked to abiotic stress resistance. When compared to the WT, *med12* and *med13* mutants showed increased sensitivity to sucrose and ABA treatments on root meristem and elongation zones that were accompanied with alterations in root hair length and morphology, leading to the isodiametric growth of these structures. The swollen root hair phenotype appeared to be specific, since *med8* or *med16* mutants did not develop rounded hairs when supplied with 4.8% sucrose. Under standard growth medium, MED12 and MED13 were mainly expressed in root vascular tissues and cotyledons, and their expression was repressed by sucrose or ABA. Interestingly, *med12* and *med13* mutants manifested exacerbated levels of nitric oxide under normal growth conditions, and upon sucrose supplementation in trichoblast cells, which coincided with root hair deformation. Our results indicate that MED12 and MED13 play non-redundant functions for maintenance of root hair integrity in response to sucrose and ABA and involve nitric oxide as a cellular messenger in *Arabidopsis thaliana*.

EFFECT OF 17 β -ESTRADIOL ON SALBUTAMOL-INDUCED AIRWAY SMOOTH MUSCLE RELAXATION

Jorge Reyes-García¹, Abril Carbajal-García¹, María F. Casas-Hernández and Luis M. Montaña¹

¹Departamento de Farmacología, Facultad de Medicina, U.N.A.M.
Circuito interior, Ciudad Universitaria. Av. Universidad 3000. CP.04510.

*E-mail: lmmr@unam.mx

During childhood, boys experience asthma symptoms more often than girls. This trend reverses at puberty, with females experiencing more severe symptoms. Around one-third of women with asthma suffer from worsening symptoms during pregnancy, while another third experience improvement, possibly due to fluctuations in 17 β -estradiol (E2) levels. In young and adult women, plasmatic E2 levels fluctuate between 0.08 and 1.3 nM, but during pregnancy, these levels increase to 1-150 nM. Estrogens may affect various mechanisms of airway smooth muscle (ASM) relaxation. One of the primary mechanisms through which the common bronchodilator, salbutamol, induces ASM relaxation is the activation of K⁺ channels. The main subtypes of K⁺ channels involved in ASM relaxation are voltage-dependent K⁺ channels (K_v) and high-conductance Ca²⁺-activated K⁺ channels (BK_{Ca}). This study investigated whether E2 promotes the activation of K⁺ channels in ASM and enhances salbutamol-induced relaxation. It also explored which subtypes of K⁺ channels are involved in the E2-mediated effect. In organ baths, this study evaluated salbutamol-induced relaxation in guinea pig tracheal rings exposed to different concentrations of E2. The findings showed that E2 at concentrations of 0.32 and 1 nM potentiated salbutamol-induced ASM relaxation. This potentiation was eliminated by tetraethylammonium (3 mM, a non-specific K⁺ channel blocker). Furthermore, 4-aminopyridine (3 mM, a blocker of K_v channels) and charybdotoxin (100 nM, a blocker of BK_{Ca} channels) partially reduced the E2-mediated potentiation of salbutamol-induced ASM relaxation. Conversely, exposure to higher concentrations of E2 (10, 32, and 100 nM) decreased salbutamol-induced ASM relaxation. In conclusion, E2 at 0.32 and 1 nM activates K_v and BK_{Ca} channels, enhancing the ASM relaxation elicited by salbutamol. This mechanism may contribute to the reduction of asthma symptoms in a percentage of pregnant women.

This work was partially funded by the Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica (PAPIIT), UNAM (IA203924) to J. Reyes-García and (IN200522) to L. M. Montaña. In addition, L. M. Montaña and J. Reyes García received a grant from CONAHCYT, México (CF -137725 and CBF2023-24-1074, respectively).

EVALUATION OF A TRIAZASPIRANE-TYPE DERIVATIVE ON CELL VIABILITY, PROLIFERATION AND MIGRATION IN MDA-MB-231 BREAST CANCER CELLS

Bryan Rivera¹, Jesús Medina¹, J. Gustavo Vázquez¹, Octavio Galindo¹

¹Universidad Autónoma de Baja California, Facultad de Medicina Mexicali,

Dr. Humberto Torres Sanginés S/N, Centro Cívico, 21000 Mexicali, B.C.

E-mail: bryan.rivera@uabc.edu.mx¹, jmedina18@uabc.edu.mx², gustavo.vazquez@uabc.edu.mx³, octavio.galindo@uabc.edu.mx⁴

Breast cancer is the most common carcinoma in women worldwide, being the first cause of death from neoplasia in women, exceeding 500,000 cases annually. In Mexico, an incidence of 29,929 cases was reported in 2022. Several therapeutic options are currently available, however, in patients with triple negative breast cancer (TNBC) these are limited and have multiple side effects. Furthermore, this type of cancer is very aggressive, with a poor prognosis and survival [2]. Therefore, the design and generation of novel treatments that improve the prognosis of patients with TNBC are necessary. **Aim.** Evaluate the effect of a triazaspirane (1,3,8-triazaspiro-[4,5]-decane-2,4-dione) on cell viability, proliferation and migration processes in TNBC cell line MDA-MB-231. **Materials and methods.** By using MDA-MB-231 cell line, the effect of a triazaspirane in cell viability was evaluated with MTT cell viability assay; cell proliferation was assessed with clonogenic assay/colony formation assay during four days; the inhibitory effect of triazaspirane on cell migration was evaluated by wound healing assay. The data obtained was analyzed with a one-way ANOVA test compared with Dunette's multiple test and statistical probability $P < 0.05$ was considered significant. **Results.** The cell viability and proliferation were not affected by triazaspirane, but the molecule demonstrated an inhibitory effect in cell migration in MDA-MB-231 breast cancer cells.

References

1. Globocan. (n/d). <https://gco.iarc.who.int/today/>
2. Yin, L.; Duan, J.; Bian, X.; Yu, S. (2020). Breast Cancer Research, 22.

TMEM16A REGULATES SPERM CELL VOLUME THROUGH RHOA AND THE ACTIN CYTOSKELETON

¹Ana Lilia Roa Espitia, ¹Tania Reyes Miguel, ¹Aideé Saray López Torres, ¹Monica Lizbeth Salgado Lucio, ²Joaquín Cordero Martínez, ¹Enrique Othón Hernández González

¹Departamento de Biología Celular, Centro de Investigación y Estudios Avanzados del Instituto Politécnico Nacional. Av. IPN No. 2508 Col. San Pedro Zacatenco C.P. 07360. México DF. Tel. 55-57473352. ²Departamento de Bioquímica, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional. Prolongación Manuel Carpio y Plan de Ayala s/n Col, Santo Tomás, Del. Miguel Hidalgo, México City, México.
Email: enrique.hernandez@cinvestav.mx

During the journey through the oviduct to reach the egg and fertilize it, mammalian sperm experience different changes in their environment that imply osmolar modifications, and for which the spermatozoa have mechanisms that regulate cellular osmolarity since these changes alter the physiology of the sperm and would prevent fertilization. Also, on this journey, the sperm experience different physiological and biochemical changes, called capacitation, which enable them to carry out acrosome reaction and hyperactivated motility, which prepare sperm to fertilize the eggs. One of the main changes during capacitation is increased polymerization and remodeling of the actin cytoskeleton. Also, during capacitation, different ion transporters are activated, including different Cl⁻ channels such as Ca²⁺-activated Cl⁻ channels (TMEM16A), the cystic fibrosis transmembrane conductance regulator (CFTR) and the chlorine 3 (ClC-3), which are related to the regulation of cell volume. In the case of TMEM16A, it has been associated with regulating the actin cytoskeleton, although the mechanism involved is unknown. Therefore, the present work aims to investigate the role of TMEM16A in regulating sperm cell volume and whether RhoA protein activity and actin polymerization are involved. The results show that inhibition of TMEM16A significantly increased sperm volume and inhibited capacitation and actin polymerization, effects that were not observed when CFTR and ClC-3 were inhibited. When RhoA activity was assessed, inhibition of TMEM16A significantly reduced RhoA-GTP levels obtained by pulldown. In conclusion, the results show that TMEM16A is related to regulating sperm cell volume through a mechanism that involves the activation of RhoA and leads to the inhibition of actin polymerization.

CONAHCYT: CB-284183 y CBF2023-2024-1894.

BUB 1 IS A NODULAR KINASE THAT PREDICTS OVERALL SURVIVAL IN OSTEOSARCOMA, LIPOSARCOMA, SYNOVIAL. SARCOMA, AND LEIOMYOSARCOMA

Frida Rodríguez-Izquierdo^{1,2}, Mercedes Olvera-Valencia^{2,3}, Oliver Millan Catalan², Carlos Pérez-Plasencia^{2,4}, Eloy-Andrés Pérez Yépez²

¹Experimental Biology Ms Program, Universidad Autónoma Metropolitana unidad Iztapalapa, Av. San Rafael Atlixco 186, Leyes de Reforma 1ra Secc, Iztapalapa, Ciudad de México. ²Genomics Laboratory, Instituto Nacional de Cancerología, Av. San Fernando 22, Belisario Domínguez Secc 16, Tlalpan, Ciudad de México. ³Doctorate in Biotechnology Science, Instituto Politécnico Nacional. Av. Luis Enrique Erro S/N, Unidad Profesional Adolfo López Mateos, Zacatenco, Alcaldía Gustavo A. Madero, Ciudad de México ⁴Functional Genomics Laboratory, Unidad de Biomedicina, FES-Iztacala, UNAM, Av. de los Barrios 1, Hab Los Reyes Ixtacala Barrio de los Árboles/Barrio de los Héroes, 54090 Tlalnepantla, Méx. E-mail: eperezy2306@gmail.com // 55 2563 7936

Sarcomas is a heterogeneous group of neoplasms derived from mesenchymal cells. There are approximately 50 different histological varieties divided in soft tissue sarcoma and bone tissue sarcoma, which represent 80% and 20% of these tumors respectively¹. For the past 40 years, cytotoxic agents led by anthracyclines, alkylating agents, and taxanes have remained as the standard treatment for sarcomas. However, it has been known that a problem in the treatment of sarcomas is the resistance to conventional treatments. Kinases drive protein phosphorylation and the dysregulation of this process plays a fundamental role in cell proliferation, migration, and differentiation during cancer progression³. Recently, several clinical trials using Tyrosine Kinase Inhibitors (TKI) or antibodies to block the kinase activity of receptors have been carried out for sarcomas. However, few TKIs such as imatinib, and pazopanib have been approved by the FDA for use in sarcoma patients⁴. Therefore, the analysis of the axis phosphorylation/dephosphorylation through the establishment of nodular enzymes that drive it in sarcomas may be key in the search for new therapeutic schemes. For this work, we used gene expression databases and bioinformatic tools to establish the overexpressed kinases in the four most frequent types of sarcomas: osteosarcoma, synovial sarcoma, liposarcoma, and leiomyosarcoma comparing with expression in healthy tissues. Interestingly, only the overexpression of BUB1 kinase was shared in the fourth sarcomas analyzed. BUB1 expression was predicted to be regulated by transcription factors, RNA binding protein, and ncRNAs, all of them involved in signaling pathways that regulate sarcoma progression through proliferation and invasion. Enrichment assays were performed and showed signaling pathways that regulate malignancy and the progression of sarcomas were the most represented. The experimental validation was performed on cell lines derived from each type of sarcoma analyzed to determine BUB1 expression and protein levels. Also, loss of function assays showed that BUB1 knockdown inhibits proliferation. Finally, we found that high levels of this kinase correlate with poor overall survival of sarcoma patients. All this evidence allows us to propose BUB1 such as a nodular kinase in the progression of osteosarcoma, liposarcoma, leiomyosarcoma, and synovial sarcoma. Its inhibition using target therapy could provide a pharmacologic strategy in sarcoma treatment.

This research was supported by Consejo Nacional de Humanidades Ciencia y Tecnología, CONAHCyT (CF-2023-I-2071)

References

1. Ferrari A, Dirksen U, Bielack S. Sarcomas of Soft Tissue and Bone. *Progress in Tumor Research*. 2016;43:128-141. doi:10.1159/000447083
2. Ferguson FM, Gray NS. Kinase inhibitors: the road ahead. *Nature Reviews. Drug Discovery*. 2018;17(5):353-377. doi:10.1038/nrd.2018.21
3. Pollack SM, Ingham M, Spraker MB, Schwartz GK. Emerging Targeted and Immune-Based Therapies in Sarcoma. *Journal of Clinical Oncology*. 2018;36(2):125-135. doi:10.1200/JCO.2017.75.1610

ROLE OF STAT3 ON THE REGULATION OF CRITICAL GENES FOR METABOLIC REARRANGEMENT IN CERVICAL CANCER CELLS AFTER IL-2 TREATMENT

Rodrigo Rojas-Mercado, Arturo Valle-Mendiola, Adriana Gutiérrez-Hoya**,
Benny Weiss-Steider, Isabel Soto-Cruz

Laboratorio de Oncología Molecular, FES Zaragoza, Universidad Nacional Autónoma de México, Batalla 5 de mayo s/n
Col. Ejército de Oriente, CP 09230, Ciudad de México, México.

**Investigador CONACYT. Tel: 5556230796. rodrigo.rm521@hotmail.com; sotocruz@unam.mx

Worldwide, cervical cancer is the fourth leading cause of cancer death in women. Cervical cancer is associated with human papillomavirus infection, which modifies cells to acquire transformed characteristics such as uncontrolled proliferation and reprogramming their energy metabolism to maintain rapid and uncontrolled growth. This characteristic of tumor cells showing a high glycolysis ratio while growing even under normoxic conditions correlates with increased expression of glycolytic enzymes and glucose transporters. It is important to note that the JAK/STAT pathway links to proliferation in cervical cancer cells, so activating the STAT3 and STAT5 proteins by tyrosine phosphorylation is essential to initiate their function. Therefore, in this work, we evaluated the role of STAT3 on the expression of critical genes that are characteristic of the metabolic change in tumor cells.

We demonstrate that treatment with 10 IU/mL of IL-2 increases the activation of STAT3 by phosphorylation on Y705 and S727 and cell proliferation. Thus, its activity as a transcription factor, increasing the transcription of the PDK1, HIF-1 α and GLUT1 genes. On the other hand, we also observed that mitochondrial genes UQCRC1 and NDUFV1 decreases after the treatment with 10 IU/mL of IL-2, suggesting that STAT3 can also have a role of negative transcription regulator.

Altogether, these results suggest the fundamental role of STAT3 as a transcription factor on some critical genes in the Warburg effect necessary to continue the cell cycle and increase cell proliferation. Furthermore, the increase in PDK1, HIF-1 α and GLUT1, and the decrease on UQCRC1 and NDUFV1 demonstrates the importance of these genes for tumor maintenance and progression and that these genes are the central regulators of the energy-obtaining pathways of the transformed cell. The information generated in this project suggests that STAT3 may be a target for treating this cancer.

SPERM VIABILITY IS REGULATED BY INTRACELLULAR CALCIUM LEVELS THROUGH THE CASR/PI3K/AKT PATHWAY, WHICH PROBABLY REGULATES TRPV CHANNELS IN GUINEA PIG SPERM

Monica Lizbeth Salgado Lucio^{1,2}, Danelia Ramírez Ramírez¹, Humberto González Márquez², Ana Lilia Roa Espitia¹ y Enrique Othón Hernández González¹

¹Departamento de Biología Celular, Cinvestav-IPN, Av. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, México City, México 07360. ²Departamento de Ciencias de la Salud, Universidad Autónoma Metropolitana- Iztapalapa. Email: enrique.hernandez@cinvestav.mx

Mammalian sperm acquire their fertilizing capacity within the female reproductive tract by experiencing the processes of capacitation and acrosomal reaction. These processes are dependent on Ca²⁺. Calcium sensor receptor (CaSR) is related to mobility, phosphorylation on tyrosine residues and acrosome integrity. Recent reports suggest that the PI3K/Akt pathway is essential in sperm survival by preventing the activation of caspases. Therefore, the project's objective was to determine the presence and participation of the CaSR/PI3K/Akt pathway in sperm survival and its relationship with the TRPV1 and TRPV2 channels in guinea pig sperm. The role of these proteins was determined using specific inhibitors, and their inhibition was analyzed to determine how they affect physiological processes and sperm viability. The results suggest the presence of the CaSR/PI3K/Akt pathway and the TRPV1 and TRPV2 channels in guinea pig sperm. We observed the presence and activation of the CaSR/PI3K/Akt pathway, which depends on extracellular Ca²⁺. We also determined that inhibiting the CaSR/PI3K/Akt pathway accelerated and increased the spontaneous acrosomal reaction. This effect was also dependent on extracellular calcium, which could be explained by inhibiting this pathway, increasing the speed of Ca²⁺ uptake and the concentration of intracellular Ca²⁺, probably due to a deregulation of the TRPV1 or TRPV2 channels. Finally, we observed that inhibiting the CaSR/PI3K/Akt pathway increases the activation of caspases 3/7 during capacitation in a Ca²⁺-dependent manner. The intracellular Ca²⁺ increase could be related to the activation of caspases since chelating intracellular Ca²⁺ prevented the activation of caspases that occurs by inhibiting CaSR. The results suggest that the CaSR/PI3K/Akt pathway regulates the acrosomal reaction and viability in guinea pig sperm by regulating intracellular Ca²⁺. CONAHCYT: CB-284183 y CBF2023-2024-1894.

EXPRESSION AND ASSOCIATED EPIGENETIC MECHANISMS OF DSC3 AND DSG3 DURING EPITHELIAL-MESENCHYMAL TRANSITION INDUCED BY TGF- β AND EGF IN BREAST CANCER CELL LINES

Ángel Salmerón Hernández, Pablo Sinuhé Gómez González, Ana Cecilia Sánchez Trujillo, Gabriela Rodríguez Rodríguez, Ángel Zarain Herzberg*

Departamento de Bioquímica, Facultad de Medicina. Universidad Nacional Autónoma de México, UNAM. Av. Universidad 3000, 04510, Mexico City, Mexico.

*Correspondence: zarain@unam.mx

Background. Calcium ion (Ca^{2+}) is a molecule that participates as a second messenger in cellular processes such as muscle contraction, metabolism, phagocytosis, apoptosis, signaling, division, and cell motility. In cancer, increased calcium signaling favors cell migration, a process related to the epithelial-mesenchymal transition (EMT)¹⁻³. In our laboratory, we found that one of the principal families that decrease their expression in breast cancer cells and tissue is the cadherin family, among which are desmocollin 3 (DSC3) and desmoglein 3 (DSG3), and that this decrease was due to increased DNA methylation and decreased euchromatin-related histone marks⁴.

Objective. To analyze the epigenetic mechanisms associated with the expression of DSC3 and DSG3 during the epithelial-mesenchymal transition in breast cancer cells.

Methods. We cultured the non-tumor breast cell line MCF10A and the luminal A breast cancer cell line MCF7 to induce an epithelial-mesenchymal transition (EMT) model with 5ng/ml of TGF- β and 50ng/ml of EGF; we analyzed morphological changes by microscopy and expression of E-cadherin/fibronectin as EMT markers. We studied the expression of DSC3 and DSG3 by RT-qPCR in cells treated with the DNA methyltransferase inhibitor (DNMTi) 5-azacytidine and with the histone deacetylase inhibitor (HDACi) Panobinostat (LBH589).

Results. In MCF10A and MCF7, the EMT decreases the expression of E-cadherin, a marker of epithelial phenotype, and increases fibronectin, a marker of mesenchymal phenotype, we observed morphological changes associated with the acquisition of mesenchymal phenotype. The EMT model decreases the expression of DSC3 and DSG3 in MCF10A, and treatment with DNMTi and HDACi increases their expression. Importantly, we observed that in MCF7 cells, the expression of DSC3 was not detectable by RT-qPCR, but when 5-azacytidine and/or Panobinostat were used, the expression increased to detectable levels. In the case of the triple-negative MDA-MB-231 cells with mesenchymal phenotype, we found that the two inhibitors increased DSC3 and DSG3 gene expression.

Conclusions: These findings not only demonstrate the involvement of DSC3 and DSG3 in the EMT process but also underscore the crucial role of epigenetic mechanisms, specifically DNA methylation and histone acetylation, in regulating their expression. This research holds promising potential in advancing our understanding of breast cancer biology and potentially paving the way for novel therapeutic interventions.

References

1. Skrypek, N., et al. (2017). *Trends in Genetics*, 33(12), 943–959.
2. Li, Y., et al. (2016). *Cold Spring Harbor Perspectives in Medicine*, 6(10).
3. Vietri, M. T et al. (2021). *International Journal of Oncology*, 59(5), 98.
4. Hernández-Oliveras, A., et al. (2022). *J. Cell Commun. Signal*. 16, 461–474

DISCOVERY OF PTP1B LIGANDS THROUGH DRUG REPURPOSING FOR TYPE 2 DIABETES THERAPY: *IN SILICO* AND *IN VITRO* STUDIES

Ximena V. Sánchez-Nava¹, Olga Villamar Cruz¹, Karen Pelcastre Guzmán², Héctor Iván Saldívar Cerón³, Marco A. Loza-Mejía² and Luis Enrique Arias Romero^{1*}

¹Unidad de Investigación en Biomedicina (UBIMED), Facultad de Estudios Superiores-Iztacala, UNAM, Tlalnepantla, Estado de México 54090, Mexico. ²Facultad de Ciencias Químicas, Universidad La Salle-México, Benjamin Franklin 45 06140, Mexico City, Mexico. ³Carrera de Médico Cirujano, Facultad de Estudios Superiores Iztacala, UNAM, Tlalnepantla 54090, Mexico.

* Corresponding author: larias@unam.mx Tel. +52-55-5623-1333 ext. 39785

Type 2 Diabetes Mellitus (T2DM) results from the reduced sensitivity of target organs to the actions of insulin combined with failure of pancreatic beta cells to produce and secrete this hormone, ultimately leading to high blood glucose levels. Therefore, therapeutic strategies to improve insulin action and/or insulin secretion are an active area of research for the treatment of T2DM. Estimating the cost of developing a new drug is a complex and multifaceted task, and the figures can vary widely depending on the source, the therapeutic area, the complexity of the drug, and the stage of development. In this challenging landscape, the concept of drug repurposing has emerged as a transformative strategy to accelerate the development of novel therapeutic interventions.

In this sense, the modulation of the activity of PTP1B, an enzyme involved in insulin and leptin signaling, has been proposed as a strategy to enhance insulin sensitivity; therefore, PTP1B inhibitors have been investigated as potential therapeutics for T2DM. The development of PTP1B-targeted therapeutics is an active area of research. Single-target and multitarget small molecule competitive inhibitors, peptide-based inhibitors and allosteric modulators have been reported. However, only a few PTP1B inhibitors have reached the clinical trials stage. Therefore, this study aimed to identify approved drugs that may serve as PTP1B inhibitors for cancer therapy by using *in silico* and *in vitro* approaches. First, we performed a screening of 2056 drugs from the e-Drug3D FDA-approved drug database, in order to test their repurposing potential using molecular docking. The five drugs with higher theoretical affinity were selected for performing molecular dynamic simulations, confirming their stable interaction with PTP1B catalytic domain, and their ligand binding energies were estimated. Next, we tested the ability of these drugs to inhibit PTP1B catalytic activity in an *in vitro* phosphatase assay, where we observed that two FDA-approved drugs, Naloxone and Isradipine, reduced in more than 80% the ability of PTP1B to dephosphorylate an Insulin Receptor-derived peptide. Finally, we tested the effect of these drugs to modulate the Insulin Receptor signaling pathway in a cellular system. Altogether, our results suggest that repurposing therapeutic agents targeting PTP1B could be a successful strategy for the treatment of T2DM.

TRACHEA-EPIDERMIS INTERACTIONS AS A MODEL OF COORDINATED MORPHOGENESIS IN *DROSOPHILA* DEVELOPMENT

Eduardo Sánchez-Cisneros^{1*}, Daniel Ríos Barrera¹

¹Instituto de Investigaciones Biomédicas, Universidad Nacional Autónoma de México, UNAM.

Ciudad Universitaria, 04510. Ciudad de México. México.

*Contacto: 55562-29250 ext. 46804, lesanchez@iibiomedicas.unam.mx

During development, the formation and organization of different tissues depends on the correct communication and coordination between them to form functional structures. Halfway in *Drosophila* embryogenesis, two lateral sheets of epidermis extend and migrate to the dorsal side to fuse and form one continuous sheet of epithelium. At the same time, the main tubes of the respiratory system also move to the dorsal side. The mechanisms that coordinate these processes remain unknown. Since the respiratory system forms just below the epidermis, it is possible that the displacement of both tissues is mechanically coupled. In this work we study this phenomenon using genetics and microscopy tools. We found that the main tubes of the respiratory system have specialized cells that extend filopodia to the basement membrane of the epidermis. These contact points mediate the coordinated displacements between both tissues. Inhibiting adhesion by knocking down the integrin complex in the respiratory system results in defects in the two tissues without affecting tissue coordination. Similarly, when epidermal dorsal closure is perturbed, the coordination is preserved but the respiratory system is misshapen, supporting the mechanic relationship between these processes. We are currently characterizing the subpopulation of tracheal cells that mediate the interaction with the epidermis using loss and gain of function tools expressed specifically in this cell type.

THE EXPRESSION OF TAZ TRANSCRIPTIONAL CO-REGULATOR IS INDUCED BY TGF- β /SMAD PATHWAY IN HEPATIC CANCER CELLS

Marcela Sosa-Garrocho¹, D.G. Ríos-Lopez¹, Ángeles Tecalco Cruz², D. Martínez-Pastor¹, G. Tapia-Urúza¹, Y. Aranda-López³, B. Domínguez-Ortega¹, F. Recillas-Targa¹, Genaro Vázquez-Victorio³ and Marina Macías-Silva¹

¹Instituto de Fisiología Celular, UNAM. ²Programa en Ciencias Genómicas, UACM. ³Departamento de Física and Lansbiodyt, Facultad de Ciencias, UNAM. Correspondence: M. Macías-Silva. Instituto de Fisiología Celular, UNAM. CDMX 04510, México. E-mail address: mmacias@ifc.unam.mx

The transcriptional cofactor TAZ, also named WWTR1, is a downstream effector of Hippo pathway that plays a key role in the maintenance of liver physiological functions. In the liver, the TGF- β and Hippo pathways are critical for organ size control, tissue regeneration, and cancer progression. However, the upregulation of TAZ expression has been associated with the progression of liver cancer. Recent evidence shows crosstalk of TGF- β and Hippo pathways since TGF- β modulates TAZ expression through different molecular mechanisms in a cellular context-dependent manner. Here, we evaluate the molecular interplay between the TGF-beta/Smad canonical pathway and TAZ expression. We analyzed the TAZ gene promoter sequence, identifying SMAD binding elements. Our results suggest that TGF- β /SMAD canonical pathway induces the activity of the TAZ gene promoter, increasing its mRNA levels and, subsequently, its protein levels in HepG2 cells. In addition, TGF- β signaling promotes TAZ protein nuclear localization. Thus, this work provides evidence that the TGF- β /SMAD canonical pathway exerts crosstalk with the Hippo pathway by enhancing TAZ levels and its nuclear accumulation in liver cancer cells.

This work is supported by grant No. 304023 from CONACyT and grant No. IN208118 from PAPIIT/DGAPA, UNAM.

ESTUDIO DEL PAPEL QUE JUEGA LA VÍA WNT/CA++ NO CANÓNICA EN LA QUIMIORRESISTENCIA DE CÉLULAS DE CÁNCER DE COLON

Andrea Terán Ramos, Paola Briseño Díaz, Cristina Castañeda Patlán,
Víctor Hugo Rosales García, Martha Robles Flores

Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México. Facultad de Medicina,
Circuito Interior, Ciudad Universitaria, Av. Universidad 3000, CP. 04510
Tel. 56232258, rmartha@unam.mx

Colorrectal cancer (CRC) is the third most common neoplasia worldwide, and the fourth cause of death associated with cancer. Wnt signaling pathway controls intestinal homeostasis and is considered a main regulator of self-renewal and differentiation in stem cells. Consequently, several activating mutations of the Wnt pathway have been identified in CRC cases, and many of its components are considered promoters of this disease. Therefore, CRC is considered a disease with an abnormal Wnt pathway. Cancer stem cells (CSC) constitute a tumoral cell subpopulation with the ability to self-renew and initiate a tumor. They play a key role in resistance to therapy and cancer relapse, since not only do they have effective mechanisms of DNA repair and elimination of cytotoxic agents, but because they exhibit great genetic plasticity and under stress conditions, they can change into a state of quiescence which allows them to survive and adapt to adverse conditions. It has been observed that eliminating these cells can make a tumor sensitive towards therapy. Recently, it was demonstrated that non-canonical Wnt Pathways are active in CSC, and particularly, Wnt/Ca⁺⁺ is key in the maintenance of their self-renewal capacity. Unlike the canonical Wnt Pathway, little is known regarding the participation of non-canonical Wnt pathways in the development of chemoresistance of cancer cells, and for this reason, the main goal of this research project is focused on exploring the role Wnt/Ca⁺⁺ pathway play in the development of chemoresistance in CRC. Spheroid cultures and monolayer cell cultures have been characterized using Western Blot and Flow Cytometry techniques to evaluate the expression of stem cell markers CD133, CD44, LGR5 and CD44v6. Additionally, the expression of NFATc1, NFAT1, NFATc4 and NFATc3 (target of the Wnt/Ca⁺⁺ pathway) has been evaluated in four different CRC cell lines. Also, sensitivity towards CCI-779 has been evaluated in resistant and sensitive CRC cell lines through cell viability assays in monolayer and spheroid cultures. It has been observed that there is a differential expression of NFAT proteins between CRC cell lines, just as there are differences in said proteins nuclear localization. These results hint toward changes and differences in Wnt/Ca⁺⁺ signaling, considering that NFATs are targets of this pathway. There were also changes in the expression of stem cell markers among different cell lines in spheroid cultures, as a study model for CSC. Particularly, CD133 was detected in a resistant cell line whilst in its sensitive counterpart it was absent. Meanwhile, there was a shared expression of CD44 and LGR5. These markers are considered characteristic of CRC. On the other hand, evaluation of sensitivity towards CCI-779 in spheroid cultures has shown that consecutive culture generations reduce their sensitivity towards this drug, therefore acquiring chemoresistance.

THE IL-6 INDUCES METASTATIC PROPERTIES IN LUMINAL BREAST CANCER CELLS THROUGH GPR30

Ana Carolina Tirado-Garibay¹, Alejandra Ochoa-Zarzosa¹ and Joel Edmundo López-Meza¹

¹Centro Multidisciplinario de Estudios en Biotecnología – FMVZ, Universidad Michoacana de San Nicolás de Hidalgo, Tarímbaro, Mich., CP. 58893, México.
E-mail: 1050629h@umich.mx

Breast cancer is a significant global health challenge, originating in the mammary gland and spreading via metastasis. The luminal A subtype shows an epithelial phenotype with lower aggressiveness and is characterized by ER- α overexpression and responsiveness to 17 β -estradiol (E2). This subtype constitutes the most prevalent breast cancer worldwide. Conversely, the triple-negative subtype lacks receptor overexpression, exhibiting heightened aggressiveness due to its mesenchymal phenotype. The epithelial characteristics of luminal cancer can be altered by cytokines from the microenvironment, such as IL-6, which induces epithelial-to-mesenchymal transition (EMT), thereby enhancing metastasis in breast cancer cells (1). Also, IL-6 has been implicated in tamoxifen (TMX) resistance by reducing ER- α levels; however, genes regulated by ER- α continue expressing, suggesting the involvement of an alternative receptor. GPR30/GPER is an alternative E2 receptor found in both luminal and aggressive triple-negative breast cancer, which is linked to early TMX resistance and cancer progression (2). The roles of GPR30 and IL-6 on metastasis have been individually established; however, their interplay remains unexplored (3). This study aims to elucidate GPR30's role in IL-6-induced metastatic properties in MCF-7 luminal breast cancer cells. Results indicate that GPR30 contributes to E2-induced proliferation because its inhibition with G15 diminished the proliferation rate of MCF-7 cells. Besides, GPR30 regulated EMT by modulating vimentin and E-cadherin levels in these cells. Interestingly, GPR30 also regulates the IL-6-induced migration, invasion, and TMX resistance in MCF-7 cells. This mechanism is calcium-dependent, as evidenced by inhibiting these properties with *Pertussis* toxin. Also, MDA-MB-231 triple-negative cells' basal and IL-6-induced metastatic properties are related to GPR30 activity. Moreover, Src homology 2-containing protein tyrosine phosphatase 2 (SHP2) appears implicated in IL-6-induced EMT and depends on GPR30 activity because receptor inhibition leads to decreased SHP2 protein activation. The above results suggest that GPR30 participates in IL-6-induced EMT by regulating SHP2. However, it is necessary to analyze the signaling pathway to provide therapeutic target options.

References

1. Y., Li, H., Zhang, Y., Merkher, L., Chen, N., Liu, S., Leonov, Y., Chen. (2022). Recent advances in therapeutic strategies for triple-negative breast cancer. *Journal of Hematology & Oncology*.15(1):121.
2. Z., Mo, M., Liu, F., Yang, H., H., Luo, Z., Li, G., Tu, G., Yang. (2013). *Breast Cancer Research*. 15(6):R114.
3. A., Abaurrea, A, M., Araujo, M, M., Caffarel. (2021). *International Journal of Molecular Sciences*.22(15):8334.

THROMBIN-INDUCED CYTOKINE SECRETION FROM RETINAL MÜLLER GLIA

Naomi Vargas Martínez, Irene Lee Rivera, Gabriela Rodríguez Rodríguez,
Ángel Alfonso Zaráin Herzberg, Ana María López Colomé

Instituto de Fisiología Celular, UNAM, Circuito Exterior, s/n, Ciudad Universitaria, México, CDMX, C.P. 04510, México.
Tel.: +525556225617; acolome@ifc.unam.mx

Müller cells are the main type of glia in the vertebrate retina. These cells are involved in the maintenance of tissue structure, homeostasis, damage detection, inflammation, and induce protection and repair responses. Müller cells are activated and induce gliosis. This process is characterized by morphological and physiological changes that involve the release of proinflammatory cytokines, among other factors, which, can promote retinal degeneration in the long term.

Similarly, upon the alteration of the blood-retina barrier, the retina comes in contact with blood serum components, including thrombin. It has been observed that thrombin, a pro inflammatory factor, induces gliosis in Müller cells. Moreover, in the pigment epithelium of the retina it has been observed that this protease induces cytokine secretion, that appears to depend on nuclear factor kappa B (NF- κ B). In addition, thrombin also triggers Mitogen-activated protein kinase (MAPK) signaling pathway, which might participate in the release of pro-inflammatory factors.

The aim of this study was to understand the mechanisms by which thrombin induces cytokine secretion in retinal Müller glia. We worked with MIO-M1 cell line, stimulated with thrombin for 24 and 48h. Cytokine expression was evaluated by qRT-PCR. Thrombin was shown to induce the expression of: CCL2, CXCL1, IL1B and IL1A at 24h of stimulation; whereas IL1RN was induced after 48h of stimulation. Thrombin specificity was assessed using a chelator: Hirudin, which was found to inhibit thrombin-induced cytokine expression. To evaluate the participation of the aforementioned signaling pathways in cytokine expression, their specific pharmacological inhibitors, U0126 for MAPK and BMS345541 for NF- κ B, were used.

In conclusion, thrombin induces the expression of CCL2, CXCL1, IL1B, IL1A and IL1RN in a specific manner, which is mediated by MAPK and NF- κ B signaling pathways. This could mediate the effect of thrombin in inducing gliosis.

This work was funded by PAPIIT-UNAM project IN201223.

THE PROTEIN TYROSINE PHOSPHATASE 1B (PTP1B) PROMOTES THE MIGRATION OF COLON CANCER CELLS THROUGH THE ACTIVATION OF A VAV1-RAC1 SIGNALING PATHWAY

Olga Villamar Cruz¹, Angélica Samantha Santos Reyes¹, Janisse Michelle Ramírez García¹, Héctor Iván Saldívar Cerón³, Marco Antonio Loza Mejía² and Luis Enrique Arias Romero¹

¹Unidad de Investigación en Biomedicina, Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México. Av. de los Barrios #1, Los Reyes Iztacala. Tlalnepantla, Estado de México, Mexico. C. P. 54090.
email: olga.villamar@unam.mx

²Facultad de Ciencias Químicas, Universidad La Salle Campus Ciudad de México, Benjamin Franklin 47, Cuauhtémoc, México 06140, México. ³Carrera de Medicina, Facultad de Estudios Superiores Iztacala, Universidad Nacional Autónoma de México. Av. de los Barrios #1, Los Reyes Iztacala. Tlalnepantla, Estado de México, México. C. P. 54090.

PTP1B is a non-transmembrane protein tyrosine phosphatase that plays a major role in insulin and leptin signaling. Recent evidence also indicates that PTP1B is overexpressed in different types of cancers, including colon, prostate, breast and ovarian tumors. In addition, experiments using cultured cells and mouse models of breast cancer have yielded conflicting results regarding the identity of the key substrates of PTP1B that mediate its oncogenic role in tumor development.

In this work, we identified VAV1, a tyrosine phosphorylated-regulated Rho guanosine nucleotide exchange factor (GEF) that participates in various cellular responses including actin cytoskeleton reorganization, gene transcription, and development and activation of immune cells, as a novel substrate of PTP1B by using a SILAC-based phosphoproteomic approach.

First, we performed molecular docking studies, which revealed stable interactions between the PTP1B catalytic domain and VAV1. In addition, *in vitro* phosphatase assays confirmed that a phosphopeptide corresponding to the residues 137-147 of VAV1 is dephosphorylated by PTP1B at Tyrosine residue 142. Next, co-immunoprecipitation, co-localization and Proximity Ligation Assays (PLA) showed that PTP1B interacts with VAV1 in a cellular context. Finally, we observed that the pharmacological inhibition of PTP1B impairs the migration of colon cancer cells and impedes the activation of Rac1, a downstream effector of VAV1, suggesting novel roles of this phosphatase in the regulation of cell motility through the activation of a VAV1-Rac1 signaling pathway.

ADRENOCEPTOR ALPHA1A (*ADRA1A*) INTEGRATES A SIGNALING SIGNATURE STATISTICALLY LINKED TO LONGER SURVIVAL OF LIVER HEPATOCELLULAR CARCINOMA PATIENTS

Yarely Mabell Beltrán Navarro¹, José Vázquez Prado², Guadalupe Reyes Cruz³, Jesús Adolfo García Sainz¹

¹Department of Cell Biology and Development, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Ciudad Universitaria, CP 04510, Mexico City, Mexico. Ap. Postal 70-248

Tel. +52 (55) 56225613 E-mail address: agarcia@ifc.unam.mx

²Department of Pharmacology and ³ Department of Cell Biology. CINVESTAV. Av. Instituto Politécnico Nacional 2508, Col. San Pedro Zacatenco, CP 07360, Mexico City, Mexico.

The alpha-1A adrenergic receptor is a G protein coupled receptor codified by the *ADRA1A* gene. This adrenoceptor is coupled to the heterotrimeric G_{q/11} proteins, leads to calcium mobilization and PKC activation and regulates multiple liver functions, like proliferation, regeneration and metabolism in response to adrenaline and noradrenaline. Although the hepatic functions of *ADRA1A* are well described, its role in hepatocellular carcinoma remains to be investigated. We hypothesized that *ADRA1A* expressed in liver cancer tissue is linked to a particular set of signaling partners potentially linked to patient outcome. We analyzed The Cancer Genome Atlas and found that expression of *ADRA1A* correlated with longer patient survival. Thus, we aimed to identify *ADRA1A*-signaling partners equally linked to longer patient survival. We employed several mining strategies: a signaling role for the genes, a positive correlation with *ADRA1A* when it's highly expressed, contrasting expression between normal and tumoral samples and similar correlation with patient survival. We identified 32 signaling transcripts preferentially correlated with *ADRA1A* in the high expression group, all of which exhibited a similar pattern of expression compared to normal tissue and survival curves. Further selection criteria led us to identify an *ADRA1A* transcriptional signature (p=0.0005, linked to longer survival) integrated by the adrenoceptor and five signaling companions including a phospholipid phosphatase: *PLPP6*, two RhoGEFs: *FGD4* and *FARP2*, a RabGAP: *TBC1D2B*, and a RTK adaptor: *BAIAP2*. Additionally, highly coexpressed GPCRs such as *DRD1*, *GPR17*, *SIPR1*, and *GPR182*, and RTKs like *KDR* and *TEK* similarly integrated transcriptional signatures with *ADRA1A* that correlated with longer survival (p=0.00021 and p=0.0041 for the GPCR and RTK signatures, respectively). The identified members of the transcriptional signatures were well coexpressed with hepatocyte markers, suggesting the integration of signaling networks likely beneficial, potentially contributing to a better prognosis of liver cancer patients.

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex biological molecule, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

SYSTEMS BIOLOGY & BIOINFORMATICS

EFFECT OF THE LYS62ALA MUTATION ON THE THERMAL STABILITY OF *BstHPr* PROTEIN BY MOLECULAR DYNAMICS

Salomón J. Alas-Guardado¹, Aranza C. Martínez-Zacarias¹, Edgar López-Pérez²

¹Departamento de Ciencias Naturales, Universidad Autónoma Metropolitana Unidad Cuajimalpa, Ciudad de México 05300, Mexico. ²Posgrado en Ciencias Naturales e Ingeniería, Universidad Autónoma Metropolitana Unidad Cuajimalpa, Ciudad de México 05300, Mexico
E-mail: salas@cua.uam.mx

We analyzed the thermal stability of the *BstHPr* protein through the site-directed point mutation Lys62 replaced by Ala residue using molecular dynamics simulations at five different temperatures: 298, 333, 362, 400, and 450 K, for periods of 1 μ s and in triplicate. The results from the mutant thermophilic *BstHPr*m protein were compared with those of the wild-type thermophilic *BstHPr* protein and the mesophilic *BsHPr* protein. Structural and molecular interaction analyses show that proteins lose stability as temperature increases. Mutant and wild-type proteins behave similarly up to 362 K. However, at 400 K the mutant protein shows greater structural instability, losing more buried hydrogen bonds and exposing more of its non-polar residues to the solvent. Therefore, in this study, we confirmed that the salt bridge network of the Glu3-Lys62-Glu36 triad, made up of the Glu3-Lys62 and Glu36-Lys62 ion pairs, provides thermal stability to the thermophilic *BstHPr* protein.

References

1. Razvi, A.; Scholtz, J.M. *Protein Sci.* 2006, 15, 1569–1578.
2. Razvi, A.; Scholtz, J.M. *Biochemistry* 2006, 45, 4084–4092.
3. Gómez-Flores, A.K.; López-Pérez, E.; Alas-Guardado, S.J. *Int. J. Mol. Sci.* 2023, 24, 9557.

DYNAMICS OF BACTERIAL COMMUNITIES IN RESPONSE TO DISTURBANCES

Roberto Álvarez-Martínez^{2,1}, Víctor Lázaro-Vidal^{1,2}

¹Facultad de Ciencias Naturales, Universidad Autónoma de Querétaro, Camino a Chichimequillas S/N, Querétaro.

²Laboratorio de Biología Cuantitativa, Facultad de Ciencias Naturales, Universidad Autónoma de Querétaro.

E-mail: roberto.alvarez@uaq.edu.mx

Microbial communities consist of diverse populations of bacteria, archaea, fungi, and viruses that interact with each other and their environment. These interactions regulate population dynamics, maintaining community diversity and functionality despite fluctuating abundances. As complex systems, microbial communities can absorb and buffer environmental disturbances, preserving their structure despite varying conditions. This stability is seen in the intestinal microbiota composition despite changes in diet, physical activity, and water consumption. However, strong disturbances can alter the community's composition.

We explored various databases to study microbial communities exposed to disturbances such as antibiotics, temperature changes, or infections. Using previously published studies, we evaluated common patterns and identified disturbed communities compared to their baseline state, quantifying diversity and dominance. We compared these values in disturbed, baseline, and post-disturbance states to analyze recovery capacity. We also conducted dysbiosis analyses and studied structural changes in the communities using multilayer co-abundance networks, applying two different inference algorithms.

Our research has shown that microbial communities, despite being significantly disturbed, have the potential for recovery. Computer simulations using two mathematical models, the generalized Lotka-Volterra (LVg) and the MacArthur consumer-resource model, demonstrated that following a disturbance, diversity decreases and dominance increases, favoring certain species over others. Some communities recovered, while others adopted different dynamics, highlighting the potential for recovery despite significant disturbances. Both dysbiosis and structural analyses supported these findings, indicating that disturbances alter community structure with potential recoveries or permanent changes. The simulations highlighted the importance of negative interactions in community stability and recovery, whereas positive interactions could adversely affect these metrics.

Microbial communities are susceptible to changes in their composition due to significant environmental disturbances, which can disrupt the system's preexisting dynamics. More intense disturbances hinder recovery and may promote alternative dynamics. Our research, which sheds light on how microbial communities respond to environmental changes, is crucial for predicting and managing the effects of infections or antibiotics on our microbiota and health. It also provides insights into the impact of climate change on soil microbial communities and the animals exposed to these changes.

EXPERIMENTAL AND COMPUTATIONAL STUDY OF BENZAZEPINTHIONES AS POTENTIAL INHIBITORS OF MYCOBACTERIUM TUBERCULOSIS HADAB DEHYDRATASE

Armando A. Ambrosio-Huerta¹, Rodrigo Aguayo-Ortiz¹, Roberto Martínez², Clara Espitia³,
Silvia J. Becerra-Anaya², Rosa María Chávez-Santos², Carol S. Martínez-Caballero²

¹Department of Pharmacy, School of Chemistry, UNAM, Mexico City 04510, Mexico. ²Institute of Chemistry, UNAM, Mexico City 04510, Mexico. ³Institute of Biomedical Research, UNAM, Mexico City 04510, Mexico.

E-mail: aambrosiohuerta@gmail.com

Tuberculosis (TB) remains one of the leading causes of death from infectious agents worldwide.¹ Among the main challenges in treating both drug-sensitive and drug-resistant TB are the length and adverse effects of current treatments.² Therefore, the search for novel compounds with antitubercular activity is essential for establishing new safe, and effective therapeutic alternatives. Proteins involved in the synthesis of the *M. tuberculosis* cell wall are attractive targets for the design of new active compounds.³ It has been reported that compounds containing thiourea or thiosemicarbazide groups (such as isoxyl and thioacetazone, respectively) are covalent inhibitors of *M. tuberculosis* HadAB dehydratase, a key enzyme involved in the synthesis of the mycolic acids external layer. This external structure is crucial for intrinsic resistance to many antibiotics and the evasion of the host immune response.³ Herein, we aim to characterize the inhibition mechanism of benzazepinethione-type compounds, whose antitubercular activity has been evaluated *in vitro*. It is hypothesized that, similar to isoxyl and thioacetazone, these compounds target HadAB. To evaluate this hypothesis, we plan to express the HadAB complex and verify the covalent binding of benzazepinethione-type compounds using mass spectrometry. Additionally, we aim to propose the specific inhibition mechanism of these compounds on HadAB using various computational tools. We have successfully conducted expression trials of HadAB, and we are currently working on the purification stage to proceed with the binding assays of the proposed compounds. Covalent docking with HadAB has been conducted to identify the binding mode of benzazepinethiones and their inhibition mechanism. We are working on molecular dynamics simulations and aim to evaluate the reactivity of the proposed compounds, as they are prodrugs that require enzymatic activation.⁴

References

1. World Health Organization. (2023). Global tuberculosis report 2023.
2. Vasiliu, A. et al. (2023). Clin. Microbiol. Infect. 29(6), 503-512.
3. Sreelatha, S. et al. (2023). Int. J. Biol. Macromol. 243, 1-16.
4. Laborde, J. et al. (2017) ChemMedChem 12, 1657-1676.

ANALYSIS OF GENE VARIATIONS IN TUMOUR AND HEALTHY TISSUES OF ACRAL LENTIGINOUS MELANOMA PATIENTS

Orlando Ángeles Martínez¹, Eduardo Carrillo Tapia², Javier Acuapan Henández¹,
L. Itzel. Quintas Granados², Norma E. Herrera Gonzalez

¹Universidad Mexiquense del Bicentenario, Ocoyoacac, Edo. Méx, ²Universidad Autónoma de la Ciudad de México, Colegio de Ciencias y Humanidades, Ciudad de México, México.
itzel.quintas@uacm.edu.mx

Acral lentiginous melanoma (ALM) is a type of melanocytic skin cancer, with abnormal cell cycle and growth. This tumor appears on the non-sun-exposed regions of the human body, such as the nail unit, palms of the hands and soles of the feet. ALM has a low incidence in the world population and diagnosis is difficult. Genetic factors have more influence in the development of ALM than environmental risk factors. Thus, some genes such as SLP2, have been associated with development of ALM. In this context, the SLP2 gene is located in chromosome 11 and encodes four protein variants such as SLP2-a, SLP2-b, SLP2-c, SLP2-d. In addition, SLP2 is a melanocyte membrane protein involved in transport mechanisms through its binding to Rab27A, resulting in the releasing of melanosome contents into the interstitial space. In the present work, single nucleotide polymorphisms (SNPs) in 5' untranslated regions of the SLP2-a gene were analysed in order to determine the potential association between gene variations and the function of this important protein in the development of ALM in Mexican patients. We found several gene variations in the 5' UTR of the SLP2-a gene, some of them were found in tumour and healthy areas. But other variations were specifically found in tumour areas of ALM patients, whereas in the healthy areas, this variation was not found, suggesting that SNPs in the 5' UTR of SLP2-a gene are possibly associated with the development of ALM.

References

1. Nakamura Y, Fujisawa Y, (2018). *Curr Treat Options Oncol*.

IDENTIFICATION AND FUNCTIONAL CHARACTERIZATION OF THE GENETIC CIRCUITS CONTROLLING CELL WALL METABOLISM IN JIPI PALM (*CARLUDOVICA* SPP.)

Jorge Luis Araujo Sánchez¹, Hugo Alejandro Méndez Hernández¹, Fulgencio Alatorre-Cobos¹

¹Centro de Investigación Científica de Yucatán A.C. 43th Street #130 x 32 and 34 Chuburná de Hidalgo Colony, Pinzón II, PC: 97205 Mérida, Yuc.
E-mail: fulgencio.alatorre@cicy.mx

Carludovica is a genus of flowering plants in the Cyclanthaceae family, widely distributed in Neotropical regions from Mexico and Guatemala to Ecuador and Bolivia. In our country, two species of this genus coexist: *Carludovica palmata* and *C. drudei*. These species are the essential raw material for producing of artisanal hats worldwide known as Panama hats. The manufacture of these hats requires high-quality fibers, making it crucial to understand the biological and physiological characteristics of these plants to ensure the sustainability of this resource. The leaf cell wall, composed of multiple biopolymers, represents one of the most complex structural networks in nature (Zhang *et al*; 2021). High-throughput multi-omics approaches have played an important role in elucidating growth, senescence, yield, and responses to biotic and abiotic stress in numerous crops (Yang *et al*; 2021). A transcriptomic approach was used in this study to unravel molecular genetics circuits controlling the physical and chemical properties of young leaves of *C. palmata* and *C. drudei*, where at a first instance it has been observed that the absence of certain gene expressions related with the photosystems apparatus in *C. drudei* directly impacts the quality of the plant tissue and, consequently, the fibers in specie. However, it is necessary to complement this study with a comprehensive omic approaches.

References

1. Zhang, B., Gao, Y., Zhang, L., & Zhou, Y. (2021). *Journal of Integrative Plant Biology*, 63(1), 251-272.
2. Yang, Y., Saand, M. A., Huang, L., Abdelaal, W. B., Zhang, J., Wu, Y., & Wang, F. (2021). *Frontiers in Plant Science*, 12, 563953.

COMPUTATIONAL SCREENING TO PREDICT MICRORNA TARGETS IN THE FLAVIVIRUS 3' UNTRANSLATED REGION FOR ANTIVIRAL DEVELOPMENT

Rodolfo Gamaliel Avila Bonilla¹, Juan Santiago Salas Benito²

¹Departamento de Genética y Biología Molecular, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Av. Instituto Politécnico Nacional #2508, Col. San Pedro Zacatenco, CDMX, CP 07360, ²Escuela Nacional de Medicina y Homeopatía, Instituto Politécnico Nacional, Guillermo Massieu Helguera #236, Frac. La Escalera-Ticomán, CDMX, CP 07320.
E-mail: rodolfo.avilab@cinvestav.mx

Flaviviruses are transmitted to humans by mosquitoes or ticks, and they represent an important public health problem worldwide. There are not specific antiviral treatments and the vaccines available are limited. They are single stranded RNA viruses with a single open reading frame flanked by two conserved untranslated regions (UTR) at the 5' and 3' end, important for viral replication and translational processes¹. MicroRNAs (miRNAs) are small non-coding RNAs that participate in posttranscriptional gene expression regulation. Since they have an important participation during viral infections, they have been proposed as potential antiviral agents².

In this work, we were retrieved the 3' UTR genomic sequences from several flaviviruses, including Dengue, Yellow fever, Zika, West Nile, Japanese encephalitis, Murray valley encephalitis, Saint Louis encephalitis, and Usutu viruses from the National Center for Biotechnology Information (NCBI) GenBank platform, and miRNA sequences from humans and mosquitoes from the miRBase database. Using RNAhybrid, Inta-RNA, miRanda, and StarMir algorithms, we selected those miRNAs that bind to 3'UTR from all flaviviruses tested. Considering the minimum free energy (MFE) for miRNA-target hybrids and thermodynamic principles, miR-6842-5p and miR-661 from humans; miR-9c, miR-2945-5p, miR-11924, and miR-282-5p from *Ae. aegypti*, and miR-79 from *Cu. quinquefasciatus* mosquitoes exhibited a greater degree of similarity in MFE predictions. The antiviral potential of these miRNAs shall be evaluated in vitro in future studies.

References

1. WC. Ng, R. Soto-Acosta, SS. Bradrick, et al (2017). *Viruses*, 9, 137.
2. LFR Gebert, IJ MacRae (2019). *Nature reviews*, 20, 21–37.

VIRAL METAGENOMICS IN HYPERSALINE ENVIROMENTS

LC. M. Guadalupe Ayala Rodríguez¹, PhD. Sonia Dávila Ramos², PhD. Hugo G. Castelán Sánchez³

¹Laboratorio de Metagenomica Viral y Bioinformatica, Universidad Autonoma del Estado de Morelos
E-mail: maria.ayalaro@uaem.edu.mx; Tel : 777 441 32 40 Av. Universidad 1001, Chamilpa, 62210 Cuernavaca, Mor.

Hypersaline environments have a higher concentration of chloride and sodium ions than the sea, and the presence of other ions such as Mg²⁺, Ca²⁺ and K is found, distributed throughout the world, There are organisms that have managed to adapt to these limiting physicochemical conditions (1). To better understand biodiversity in hypersaline environments, metagenomics has become the technique of choice. This is because it does not require the prior isolation and cultivation of organisms (2). In this work, a search was conducted for possible prokaryotic virus hosts using different bioinformatics strategies to identify CRISPR Cas sequences and the host-virus relationship that exists in these environments. From 6 metagenomic samples from salt mines located in different geographic regions: Spain, Peru and Antarctica; a taxonomic classification of viruses was carried out, which in order of abundance are Haloferacalesvirus, Haloviruses, Siphoviridae and Myoviridae. In addition, the phage-host relationship of Enterobacteria phage lambda, Burkholderia phage BcepIL02, Xanthomonas phage Xp15 and Salicola phage CGphi29, with Salinibacter ruber, Natribaculum longums, Natrinema sp. J7-2, Salarchaeum sp. JOR-1 and Haloquadratum walsbyi, these organisms are known for their ability of thrive in environments with high concentrations of salt ions.

References

1. Rothschild LJ, Mancinelli RL. Life in extreme environments. Nature. febrero de 2001;409(6823):1092-101.
2. Thomas T, Gilbert J, Meyer F. Metagenomics—a guide from sampling to data analysis. Microb Inform Exp. diciembre de 2012;2(1):3.

CLUSTERING BACTERIAL PROMOTER SEQUENCES USING SUPERVISED MACHINE LEARNING

P. Cambranis-Boldo^{1,2}, A. B. Farias³, E. Perez-Rueda² and G. Martinez⁴

¹FIQ-UADY, Yucatán México, ²IIMAS-UAIEY-UNAM, Yucatán México, ³LNCC, Brazil, ⁴Dalhousie University, Canada.
paulocambranis@gmail.com

The surge in next-gen sequencing has generated a vast amount of genomic data, impractical to classify manually. Machine Learning (ML) has become a crucial tool in bioinformatics for automating genomic sequence classification¹. Databases like CBDProm utilise algorithms leveraging DNA Duplex Stability (DDS) features to detect promoters from unannotated databases. However, clustering algorithms face challenges due to the high dimensionality of genomic data.

In this work, to circumvent the high-dimensionality problem associated with promoter sequences, a supervised clustering technique was employed. First, a XGBoost algorithm was trained on bacterial Sigma (σ) promoter sequences which belong to one of six well-known families in a One-vs-Rest manner, i.e., considering each σ family as a positive and the rest as negatives. This strategy ensures that the algorithm learns features that are specific to each family. Then, SHapley Additive exPlanation (SHAP) values were calculated from the model. SHAP values are commonly used to explain the decision-making process of ML models, highlighting the subtle differences amongst each class. Lastly, Uniform Manifold Approximation and Projection (UMAP), a dimensionality reduction algorithm, was used on the SHAP data for both visualization and simplification of cluster analysis. Using UMAP on the SHAP values is a novel strategy that can find new patterns in data².

While the XGBoost algorithm's performance could be improved, the process of reducing dimensionality with SHAP values produced graphs where the σ families can be clustered. This is the first time such a workflow has been applied to promoter sequences. Furthermore, an analysis of mean SHAP values revealed that the most relevant features for classification lie on the sequence motifs that are involved in σ factor recognition. Peaks in free-energy detected in previous analysis on the distinct DDS footprints of these sequences³ align with peaks in SHAP values detected in this work. This suggests that SHAP values not only enable sequence clustering but also highlight sequence fragments integral to promoter function.

References

1. Cassiano and Silva-Rocha. 2020. *mSystems*, 5, e00439-20
2. Cooper, et al. 2021. *Machine Learning and Principles and Practice of Knowledge Discovery in Databases*, 408–422.
3. Martinez, et al. 2021. *SN Applied Sciences*, 3, 740-750.

MICROALGAL PEPTIDES AGAINST THE MAIN PROTEASE (MPRO) OF SARS-COV-2: *IN SILICO* EVALUATION

David Mauricio Cañedo-Figueroa¹, Bianca Vianey Rivera-Serrano¹, Sandy Lucero Cabanillas-Salcido¹, Alberto Kousuke De la Herrán Arita¹, Josué Camberos-Barraza¹, Alejandro Camacho-Zamora¹, Carlos Daniel Cordero-Rivera², Rosa María del Ángel², José Manuel Reyes-Ruiz^{3,4}, Carlos Noe Farfan-Morales⁵, Alejandra Romero-Utrilla⁶, Carla Angulo-Rojo, Alma Marlene Guadrón-Llanos, Verónica Judith Picos-Cárdenas¹, Ángel Radamés Rábago-Monzón¹, Claudia Desiree Norzagaray-Valenzuela¹, Loranda Calderon Zamora¹, Marco Antonio Valdez-Flores¹, Luis Adrián De Jesús-González⁷, Juan Fidel Osuna-Ramos^{1*}

¹Maestría en Ciencias en Biomedicina Molecular, Facultad de Medicina, Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México. ²Departamento de Infectómica y Patogénesis Molecular, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Ciudad de México, México. ³Centro Médico Nacional "Adolfo Ruiz Cortines", Instituto Mexicano del Seguro Social, Veracruz, Mexico. ⁴Facultad de Medicina, Región Veracruz, Universidad Veracruzana, Veracruz, Mexico. ⁵Departamento de Ciencias Naturales, Universidad Autónoma Metropolitana, Unidad Cuajimalpa, Ciudad de México, Mexico. ⁶Departamento de Anatomía Patológica, Instituto Mexicano del Seguro Social, Culiacán, Sinaloa, Mexico. ⁷Unidad de Investigación Biomédica de Zacatecas, Instituto Mexicano del Seguro Social, Zacatecas, Zacatecas, Mexico.

*Mail: osunajuanfidel.fm@uas.edu.mx

COVID-19, caused by SARS-CoV-2, has impacted millions globally. Use of effective vaccination reduced hospitalization and mortality, vaccines do not fully address the virus's high mutability and infection rates. One of the most conserved structures among B-coronaviruses is the Main Protease (Mpro), essential for viral replication. This research aimed to identify peptides from microalgae, specifically *Phaeodactylum tricornutum*, and evaluate their interactions with Mpro using *in silico* tools. A molecular docking analysis was conducted with *Phaeodactylum tricornutum*-derived peptides against SARS-CoV-2 Mpro (PDB ID: 6LU7)¹. The synthetic peptide QTFSVLACY, effective against SARS-CoV-2 Mpro, was used as a reference for alignment with a public peptide database on UniProt. Three derivative sequences with high alignment scores were identified: A0A8J9SAR9 (Score: 9, Weight: 5.674 kDa), A0A8J9SAR9 (Score: 9, Weight: 5.674 kDa), and A0A8J9TDK2 (Score: 9, Weight: 4.791 kDa). Structural prediction was conducted using AlphaFold v2 and molecular docking with ClusPro, resulting in binding energies from -834.2 to -1214.2 kcal/mol, indicating favorable interactions. This approach demonstrates the potential of microalgae-derived peptides in antiviral drug development. Computational methods enable efficient initial screening and validation, identifying promising candidates for therapeutic development against COVID-19 and other viral diseases.

References

1. Johansen-Leete, J., Ullrich, S., Fry, S. E., Frkic, R. & col., (2022). Antiviral cyclic peptides targeting the main protease of SARS-CoV-2. *Chemical Science*, 13, 3826-3836.

EVOLUTIONARY HISTORY AND DISTRIBUTION ANALYSIS OF RHAMNOSYLTRANSFERASES IN THE FUNGAL KINGDOM

Joaquin Omar Chávez Santiago¹, Jorge Humberto Ramírez Prado², Bernardo Franco Bárcenas¹,
Gustavo Alexis Niño Vega¹ & Héctor Manuel Mora Montes¹

¹División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Noria Alta S/N; C.P. 36050; Guanajuato, Guanajuato.

²Centro de Investigación Científica de Yucatán, A. C., Chuburná de Hidalgo, Calle 43 No. 130 x 32 y 34; C.P. 97205; Mérida, Yucatán, México.
E-mail: hmora@ugto.mx

Rhamnose is a natural sugar that is part of glycoproteins and structural polysaccharides in plants, fungi, and bacteria. Once rhamnose is synthesized, enzymes called rhamnosyltransferases (RHTs) catalyze the transfer of rhamnose to specific glycoconjugates,

significantly impacting the structure and function of these glycoproteins. Despite advances in the knowledge of rhamnose synthesis in genera such as *Sporothrix* and *Botrytis*, there is a notable lack of information regarding the evolutionary history of RHTs in other fungal genera. Understanding how these enzymes evolved in different fungal lineages can provide valuable insights into the strategies and adaptations of these organisms.

This project aimed to analyze the evolutionary history and distribution of RHTs in the fungal kingdom. The results show a predominant distribution of putative *Rht1* and *Rht2* sequences in Ascomycota (99.76% and 99.35%, respectively), suggesting greater diversification and preservation of these enzymes in this phylum. The minority presence in Basidiomycota (0.24% and 0.65%, respectively) could indicate an early evolutionary divergence and a lower requirement for these types of RHTs in this phylum. Protein alignments were also performed to determine the structural similarity with the previously reported RHTs. Some preliminary protein alignments obtained RMSD values of 0.815 between *S. schenckii* and *Aspergillus niger* for *Rht1*, and 1.375 between *S. schenckii* and *Niveomyces insectorum* for *Rht2*. These results suggest significant structural similarities, and functional conservation throughout evolution, despite sequence variations. These results underscore the prevalence of RHTs in Ascomycota and offer key insights into their evolutionary paths and functional preservation, highlighting a structural resemblance among RHTs from various fungal species.

This work was supported by Consejo Nacional de Ciencia y Tecnología (Ciencia de Frontera 2019-6380), and Red Temática Glicociencia en Salud (CONACYT-México).

BIOINFORMATIC ANALYSIS OF WHOLE GENOMES OF STAPHYLOCOCCUS AUREUS ISOLATED FROM THE SKIN OF ATOPIC DERMATITIS PATIENTS AND HEALTHY SUBJECTS

Sergio Alland Colorado-Cortés^{1,2}, Ibáñez-Hernández Miguel Ángel Antonio², Pastelin-Palacios Rodolfo¹, Garza-Velasco Raúl¹, Moreno-Eutimio Mario Adán¹

¹Laboratorio de Inmunobiología. Facultad de Química, Universidad Nacional Autónoma de México (UNAM), Cd. Universitaria, 04510. Mexico City, Mexico. ²Laboratorio de Terapia Génica, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional (IPN) Casco de Santo Tomás, 11340. Mexico City, Mexico.

*Corresponding author: marioadan@quimica.unam.mx

Atopic Dermatitis (AD) is the primary chronic inflammatory skin disease, characterized by skin lesions, itching, scaling, redness, and sometimes crusting or bleeding due to scratching. These symptoms directly affect patients' quality of life, causing physical discomfort, sleep disturbances, aesthetic impact, and an increased risk of secondary infections. Previously, a genomic characterization of *Staphylococcus aureus* isolated from healthy subjects and AD patients was performed. We initially, conducted a bioinformatics analysis using whole genomes of *S. aureus* isolated from AD patients and healthy subjects from open access databases. The analysis began with constructing a phylogenetic tree through genomic alignments to compare the genetic proximity of *S. aureus* strains isolated from both groups. Our preliminary results, revealed that *S. aureus* strains isolated from AD patients cluster separately from those isolated from healthy subjects, indicating a possible genetic divergence that could have implications for the pathogenesis of AD. We then focused on protein-coding genes, specifically those encoding virulence factors. Using gene alignments, we compared the presence of these proteins in *S. aureus* strains from AD patients versus those from healthy subjects. Understanding the relationship between *S. aureus* virulence genes and the severity of AD could provide valuable insights for developing more effective therapeutic and preventive strategies. These findings could be instrumental to improve the quality of life for AD patients by enabling more precise management of *S. aureus* colonization, a factor contributing to the disease's severity.

Funding: Research and Technological Innovation Projects, UNAM. PAPIIT 2023 No. IA208423 and PAPIIT 2024 No. IN221024.

TRANSCRIPTOMIC ANALYSIS OF THE INFECTIVE PROCESS OF THE MISTLETOE *PSITTACANTHUS CALYCVLATUS* IN THE HOST TREE MESQUITE *PROSOPIS LAEVIQATA*

Marco Ramsés Cota-Pineda¹, Domancar Orona-Tamayo² and Miguel Ángel Hernández-Oñate

¹Centro de Investigación en Alimentación y Desarrollo, A.C. Coordinación de Tecnología de alimentos de Origen Vegetal, CP 83304 Hermosillo, Sonora, Mexico. ²CIATEC, A.C., Medio Ambiente y Biotecnología, CP 37545, León, Guanajuato, Mexico.

³CONAHCYT-Centro de Investigación en Alimentación y Desarrollo, A.C. Coordinación de Tecnología de Alimentos de Origen Vegetal, CP 83304 Hermosillo, Sonora, Mexico.

E-mail: miguel.hernandez@ciad.mx

The parasitic plants are organisms of the PLANTAE kingdom that are capable of making a connection between the vascular system of the host and himself to obtain the water and the nutrients that the parasitic plant needs to maintain its cycle life¹. The mistletoes are one of the parasitic plants with more prevalence in Mexico, more than 65% of the urban trees of León, Guanajuato are infested with mistletoes²; in the forest areas of Jalisco, around of 25% of the have mistletoe³; in Sonora, the elephant trees *Bursera michropylla* and *Bursera laxiflora* are infested in 27% and 6% respectively⁴. *Psittacanthus calyculatus* is one of the mistletoes with more presence in the central region of Mexico, and the mesquite *Prosopis laevigata* is one of their hosts of ecological importance⁵. The parasitic plants (including mistletoes) have a modified root called haustorium, this organ is the responsible of the interaction parasite-host, that is because this tissue makes the connection in the xylem of the host⁶. The haustorium initiation is mediated by haustorium initiation factors (HIFs), that are organic compounds generally exudated by the host, the host recognition by HIFs promotes the seed germination by promoting the accumulation of reactive oxygen species (ROS) in the root and the phytohormone metabolism^{7,8}. During the haustorium penetration through the host tissue, the haustorium secretes lignocellulolytic proteins that degrades the cell wall of the host cells, to reach the host xylem⁵. The specific biologic process of the mistletoe *P. calyculatus* during the infective stages in mesquite *P. laevigata* are still unknown. A transcriptomic analysis of the transcripts in the infective stages will make possible the understanding of the specific biologic process of the haustorium development and will make possible the development of novel strategies of population control of the mistletoes in the Mexican ecosystems.

References

1. Bouwmeester H, Sinha N, Scholes J. (2021). *Plant Physiology*. 185 1267-9.
2. Instituto Cultural de León. (2016). Muérdago: una amenaza para nuestros árboles.
3. Protección Ambiental, Asesoría y Proyectos, S. C. (PAAP), (2016) Diagnostico de las áreas forestales con afectación de muérdago en el ejido San Agustín, dentro del área de protección de flora y fauna la primavera, tomando como base el uso de imágenes de sensores remotos y validación en campo. 7-14.
4. Rodríguez-Pacheco, T. (2015). Estudio ecológico de las plantas parásitas en un matorral xerófilo de la costa de Sonora. Universidad de Sonora.
5. Aguilar-Venegas M, Quintana-Rodríguez E, Aguilar-Hernández V, López-García CM, Conejo-Dávila E, Brito-Argáez L, et al. (2023). *Plants*. 12, 464.
6. Kokla A, Melnyk CW. (2018). *Developmental Biology*. 442, 53-9.
7. Goyet V, Wada S, Cui S, Wakatake T, Shirasu K, Montiel G, et al. (2019). *Front Plant* 10, 1056.
8. Ibarra-Laclette E, Venancio-Rodríguez CA, Vásquez-Aguilar AA, Alonso-Sánchez AG, Pérez-Torres CA, Villafán E, et al. (2022) *Front Genet*. 13, 929490.

EARLY AND LATE TRANSCRIPTIONAL RESPONSES TO AUXINS AND CYTOKININS DURING THE INDUCTION OF SOMATIC EMBRYOGENESIS IN *COFFEA CANEPHORA*

Marcos D. Couoh-Cauich¹, Rosa María Galaz-Avalos¹, Enrique Ibarra-Laclette², Víctor M. Loyola-Vargas*

¹Centro de Investigación Científica de Yucatán, Unidad de Biología Integrativa, Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México, ²Red de Estudios Moleculares Avanzados, Instituto de Ecología A.C., Carretera Antigua a Coatepec 351, El Haya, C.P. 91070, Xalapa, Ver, Mexico.

*Correspondence: vmloyola@cicy.mx

In the model plant *Arabidopsis thaliana*, somatic embryogenesis (SE) is induced from immature embryos or apical meristems of seedling shoots, the latter being competent only for a short period¹. In contrast, SE in *C. canephora* is induced from leaf explants. Since the protocol was developed in 2006, there have been no significant changes in the number of embryos produced or development timelines. Given these characteristics, *C. canephora* represents a valuable model for exploring cellular differentiation, especially for woody perennial crops. In this work, we explored gene expression during 10 stages of the SE process in *C. canephora*: 14 and 9 days before induction, at the time of induction, 1h, 2h, 1d, 3d, 14d, 21d, and 28d post-induction. We analyzed genes involved in the biosynthesis, metabolism, signaling, transport and degradation of indole-3-acetic acid and cytokinins. Some genes from multigene families, such as *YUC*, *GH3*, *AUX/IAA*, *IPT* and *LOG*, did not exhibit the same expression pattern as the rest of their family members. Instead, they were coexpressed with genes from different regulatory modules, indicating that they play non-redundant roles in the SE process. Furthermore, we observed differential expression of genes responsive to different types of stress during the first hours of induction, while at 14 days, 21 days and 28 days we identified a co-expression network of the genes *WOX*, *BBM*, *ABI3*, *CUC*, and *ESR1*, as well as genes involved in cell cycle control, chromosome organization and DNA methylation. These time points coincided with the formation of embryogenic structures in the SE process studied. These findings suggest that the induction of somatic embryogenesis in *C. canephora* involves a complex gene regulatory network involving multiple signaling pathways and cellular processes, which could be harnessed for the improvement of woody perennial crops.

References

1. Mathew, M. M., & Prasad, K. (2021). *Development*, 148(6), dev195347.

STUDY OF PERIVASCULAR FIBER FORMATION IN AGAVE FOURCROYDES THROUGH A TRANSCRIPTOMIC APPROACH

Roberto Cruz-Balam¹, Marcos D. Couoh-Cauich¹, Enrique Ibarra-Laclette², Fulgencio Alatorre-Cobos¹

¹Centro de Investigación Científica de Yucatán, Unidad de Biología Integrativa, Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México, ²Red de Estudios Moleculares Avanzados, Instituto de Ecología A.C., Carretera Antigua a Coatepec 351, El Haya, C.P. 91070, Xalapa, Ver, Mexico.
E-mail: falatorreco@conahcyt.mx

Agave fourcroydes, commonly known as henequen, is an endemic plant species from the Yucatan Peninsula, Mexico. This plant has significant historical and cultural importance in the region, and it is a valuable resource for agave breeding for Brazil and Africa. Recently, there is a renewed interest on *A. fourcroydes* due to its heat tolerance and potential for commercial fiber production. However, unlike cotton or flax, the biological processes in agave fiber formation remains unclear.

The present research aims to analyze the genes involved in the lignification process of *A. fourcroydes* fibers. To achieve this, we sequenced 18 Illumina paired-end RNA-seq libraries, generating a total of 1.5 Gb of raw reads corresponding to whole leaves and fibers from three different stages of fiber development (15 cm, 40 cm, and 2 m). Quality filtering and assembly of the reads yielded a total of 603,560 contigs, of which only 264,962 could be translated into proteins. Among the analyzed genes, those involved in monolignol production were identified, such as PAL, CCR, and F5H. Additionally, genes responsible for the polymerization of these monolignols within the secondary cell wall were detected as differentially expressed genes, exhibiting significant expression levels when compared across the different ontogenetic stages evaluated. These genes encode enzymes such as peroxidases and laccases, which are essential for lignin polymerization and subsequent deposition. To complement the transcriptomic analysis, we detected in situ activities of LAC and POX enzymes. Our results suggest that autonomous and/or semi-autonomous mechanisms are operating during hard fiber formation. This study provides valuable insights into the molecular mechanisms underlying perivascular fiber formation in *A. fourcroydes* and contributes to a better understanding of the lignification process in this economically and culturally important plant species.

MICROBIAL COMMUNITY DYNAMICS THROUGH METAGENOMICS IN TWO EXPERIMENTAL BIODIGESTERS FOR RURAL COMMUNITY USE

Erika Cruz-Bonilla¹, Barbara B.Moguel², Augusto Franco³, Eliana Valencia-Lozano¹, Víctor Olalde Portugal¹ and Maribel Hernández-Rosales¹

¹Centro de Investigación y de Estudios Avanzados del I.P.N. Unidad Irapuato, Irapuato, Mexico,
²Centro de Geociencias, Universidad Nacional Autónoma de México, Juriquilla, Querétaro, Mexico,

³Universidad del Valle de Guatemala, Guatemala, Guatemala.

E-mail: erika.cruz@cinvestav.mx, maribel.hr@cinvestav.mx

Biodigesters are a small-scale sustainable alternative for gas production instead of fossil energies. Biodigesters main raw material for biogas production is biomass, which can be sourced from agricultural and crop waste, and animal manure. Gas production within biodigesters is a spontaneous process where the microbial communities within the biomass supplied are the responsible for its transformation, and their composition largely determines biogas production¹. However, the nature of these microbial communities is not fixed, there is a dynamic change in their composition throughout the gas production process, where different organisms take a leading role in the transformation of biomass. The identity of these lead organisms is also contingent upon the biodigester type and the composition of the primary materials used. Hence the importance of identifying the microbial communities present in the biodigesters in a personalized way and throughout the entire production process.

Here we present a characterization of the microbial consortia present in two biodigester models, along with a proposal for a multi-purpose bioinformatics pipeline for metagenome analysis. The biodigesters were supplied with animal manure from two rural communities in Irapuato, Guanajuato, where they are expected to alleviate wood and gas consumption for basic meal preparation. Biodigesters were operated and biological samples taken during a period of one month on five critical stages of gas production. Total DNA was extracted and sequenced by shotgun metagenomics. The pipeline developed here was used for processing the metagenomes, where quality of the reads, taxonomy assignment of sequences, assembly into MAGs and gene and functional annotation is performed automatically. From this analysis we identified in both biodigesters archaea from the genus *Methanosarcina* and *Methanothrix* as critical during the methanogenic phase², and the bacterium *Syntrophomonas wolfei* as a key player during acetogenic phase with the potential of bioaugmentation treatment³.

References

1. Weiland. (2010). Applied microbiology and biotechnology, 85, 849-860.
2. Zhang, Wang, Ma, Gao, Wang, Shi & Yang. (2019). Environment international, 126, 543-551.
3. Kushkevych, Kobzová, Vítězová, Vítěz, Dordević, & Bartoš. (2019). Biologia, 74, 1229-1236.

IN SILICO INHIBITORY POTENTIAL OF PEPTIDES HRA AND PK-2 AGAINST THE FUSION PROTEIN F OF RESPIRATORY SYNCYTIAL VIRUS

Deyanira Judith De Anda Alejandrez¹, Ángel Gabriel Alpuche Solís¹, Daniel Ernesto Noyola Cherpitel²

Instituto Potosino de Investigación Científica y Tecnológica, A.C.¹ Camino a la Presa de San José 2055, Lomas 4ta Secc, 78216 San Luis Potosí, S.L.P. Centro de Investigación en Ciencias de la Salud y Biomedicina. Universidad Autónoma de San Luis Potosí
² Av. Sierra Leona 550, Lomas de San Luis, San Luis Potosí, S.L.P.
deyanira.deanda@ipicyt.edu.mx

Respiratory diseases caused by viruses, such as Respiratory Syncytial Virus (RSV) and SARS-CoV-2, are the leading causes of mortality, particularly in children under three years of age and in adults with underlying health conditions. Therefore, there is an urgent need for effective treatments, then this study focuses on analyzing the replication inhibitory potential of two peptides, HRA and PK-2, against the fusion protein F of RSV through an *in silico* molecular docking assay.

Our working group demonstrated that the HRA peptide was not only effective in inhibiting the replication of the human metapneumovirus against which it was designed, but also against other viruses. Additionally, we designed the PK-2 peptide, to be targeted for the stem region of the SARS-CoV-2 spike protein. Given the promising nature of these peptides, we decided to evaluate their effectiveness in inhibiting the entry of RSV into HEP-2 cells.

To achieve this, we employed the Discovery Studio software to conduct molecular docking simulations. These simulations enabled us to identify the specific amino acids involved in the interactions between the peptides (HRA and PK-2) and the RSV fusion protein F. The docking results provided crucial insights into the binding affinities and the potential inhibitory effects of the peptides on the fusion protein, suggesting a strong inhibitory potential.

Furthermore, to ensure the safety of these peptides for potential therapeutic use, we conducted cytotoxicity assays using the MTT method on HEP-2 cell lines at various peptide concentrations. It is essential to demonstrate that the peptides are not cytotoxic before testing for inhibition of viral replication. The results from the MTT assays indicated that both HRA and PK-2 peptides had low cytotoxicity across 3.12 μ M and 100 μ M, underscoring their safety and potential as therapeutic agents.

In conclusion, the *in silico* analyses and cytotoxicity evaluations of HRA and PK-2 peptides strongly suggest that they could serve as effective inhibitors against RSV. *In vitro* inhibition assays are under development to demonstrate the results of the molecular docking. If we have positive results *in vitro*, we could in the future consider a trial in mice where the peptides are sprayed on the nasal mucosa and see if they stop virus replication. Our findings contribute significantly to the development of novel therapeutic strategies aimed at combating respiratory viral infections, particularly benefiting vulnerable populations such as young children and adults with comorbid conditions.

KEY PROTEINS FOR REGENERATION IN A. MEXICANUM: TRANSCRIPTOMIC INSIGHTS FROM AGED AND JUVENILE LIMBS

Aylin del Moral Morales^{1,2,3}, Cynthia Sámano Salazar¹, José Antonio Ocampo Cervantes⁴, Maya Topf^{2,5,6},
Jan Baumbach^{3,7}, Rodrigo González-Barrios⁸, Ernesto Soto-Reyes^{*}

¹Departamento de Ciencias Naturales, Universidad Autónoma Metropolitana-Cuajimalpa (UAM-C), Mexico City, Mexico.

²Centre for Structural Systems Biology (CSSB), Hamburg, Germany. ³Institute for Computational Systems Biology, University of Hamburg, Hamburg, Germany. ⁴Centro de Investigaciones Biológicas y Acuícolas de Cuemanco (CIBAC), Universidad Autónoma Metropolitana-Xochimilco (UAM-X), Mexico City, Mexico. ⁵Leibniz-Institut für Virologie (LIV), Hamburg, Germany. ⁶University Medical Center Hamburg-Eppendorf (UKE), Hamburg, Germany. ⁷Computational BioMedicine Lab., University of Southern Denmark, Odense, Denmark. ⁸Unidad de Investigación Biomédica en Cáncer, Instituto Nacional de Cancerología-Instituto de Investigaciones Biomédicas, UNAM, Mexico City, México.

Av. Vasco de Quiroga 4871, Cubículo 831, Col. Santa Fe, Delegación Cuajimalpa de Morelos, 05348, Mexico D.F.

*Correspondence: esotoreyes@cua.uam.mx

The axolotl, known for its remarkable regenerative abilities, is an excellent model for studying regenerative therapies. Nevertheless, the precise molecular mechanisms governing its regenerative potential remain uncertain. In this study, we collected samples from axolotls of different ages, including 8-year-old individuals and 8-month-old juveniles, from which we gathered their blastemas 10 days after amputation. Subsequently, we conducted a transcriptomic analysis comparing our samples to previously published experiments. Through transcriptomic analysis and co-expression networks, we unveiled a distinctive transcriptional response in the blastema, characterized by differential gene expression associated with processes such as bone and tissue remodeling, transcriptional regulation, angiogenesis, and intercellular communication. To gain deeper insights, we compared these findings with aged axolotls that showed no signs of regeneration 10 days after amputation. We identified four genes (*FSTL1*, *ADAMTS17*, *GPX7*, and *CTHRC1*) with higher expression in regenerating tissue compared to aged axolotls. Further scrutiny, including structural and homology analysis, revealed that these genes are conserved across vertebrate species. Our discoveries point to a group of proteins relevant to tissue regeneration, with their conservation in vertebrates suggesting critical roles in development. These findings also propose a novel gene set involved in axolotl regeneration, laying a promising foundation for future investigations across vertebrates.

HOW TO MODEL THE STRUCTURE-FUNCTION RELATIONSHIP OF PROTEINS?

Gabriel Del Río Guerra, Juan M. Hernández Castillo, Albros H. Poot

Instituto de Fisiología Celular, UNAM.

¹Department of Biochemistry and Structural Biology, Institute of Cellular Physiology at UNAM, Mexico City 04510, Mexico.

Email: gdelrio@ifc.unam.mx

The era of machine learning (ML) is meddling in different human activities, including science; of particular relevance to this work is the effectiveness of ML to predict fundamental aspects of proteins. For instance, AlphaFold, a deep-learning method developed by DeepMind, has been recognized as the best solution to the prediction of the three-dimensional structure of a protein from its sequence in the world CASP contest. The best protein function predictors at the community-wide world competition, CAFA, are all based on ML techniques. With the improving on computing power of current machines and the systematization of different high throughput molecular biology techniques, it is expected that ML will continue to surpass other more traditional models (e.g., classical physics) to achieve even more reliable predictions.

Despite these advances, different features are used to model protein structure and protein function, suggesting that these two observations (structure and function) are not related, however they are related, hence posing a paradox in the modeling of the structure-function relationship of proteins.

To solve this paradox derived from the increasing number of models currently developed to predict protein structure and/or function, it is important to keep in mind the Occam's razor criteria, that is, given two or more competing ideas to explain the same phenomenon, the simpler one is preferable. This is particularly important considering that ML models are usually far more complex than classical models, yet ML models are far better predictors. Two types of ML models are competing to achieve the best predictions about proteins; in the protein structure prediction, deep learning is the best model, while on the protein function prediction, shallow learning is the best. In general, deep-learning models are far more complex than shallow-learning models, hence shallow-learning models achieving similar prediction reliability than deep-learning ones, should be preferred. Furthermore, having models for the structure and function of proteins that are based on the same set of features is preferable considering that both phenomena are related. In the current work, we will show that a shallow-learning model with only 26 features is the best method in the task of protein structure classification, protein function prediction and protein-protein interaction prediction. The 26 features account for the densest regions of proteins, hence this finding relate the protein folding with the protein structure and protein function. We'll explain how this 26-features model lead us to foresee the protein folding problem as an irregular tessellation problem, which in turn may explain the occurrence of metastable proteins and intrinsically disordered regions of proteins or fully disordered proteins. Thus, this shallow-learning model provides a simpler and more reliable way to model proteins and provides a unified framework to study the structure-function relationship of proteins.

ANALYSING THE CO-EXPRESSION AND PROTEIN-PROTEIN INTERACTION NETWORKS RELATED TO THERMOTOLERANCE IN ARABIDOPSIS THALIANA

Ana Karen Diaz-Martínez¹*, Fernanda Lizeth Gutiérrez-Palomeque¹,
Humberto Herrera-Ubaldo², Cynthia Paola Rangel-Chávez¹

¹Departamento de Bioquímica. Instituto Tecnológico Superior de Irapuato, Silao-Irapuato km 12.5 El Copal, 36821 Irapuato, Gto.

²Department of Plant Sciences. University of Cambridge, Downing St., Cambridge CB2 3EA, United Kingdom.

*E-mail: diazanakaren33@gmail.com

Naturally, plants are subject to temperature changes. Sometimes these changes can exceed optimal conditions, causing plants to experience thermal stress. As a consequence, negative effects on growth may be observed, potentially affecting the productivity of various crops. On the other hand, plants have established thermotolerance mechanisms that allow them to withstand high temperatures; many of these mechanisms are mediated by protein-protein interactions (PPIs).

Additionally, previous studies have identified transcriptional regulators and other types of proteins involved in responses to high temperatures. The interactome of *A. thaliana* has also been analyzed and mapped to improve the understanding of the mechanisms by which proteins interact to perform cellular functions.

To achieve this objective, a literature review was conducted, considering all proteins involved in various processes of the high-temperature response, such as thermotolerance, thermomorphogenesis, transcriptional and post-transcriptional regulation, photosynthesis, circadian clock, and other mechanisms. From this information, a network with 187 proteins and 1712 interactions was obtained. Subsequently, we focused specifically on the subnetwork of proteins associated with the thermotolerance process, which consists of 169 proteins and 1643 interactions. In this subnetwork, we are currently conducting coexpression and PPI analyses to identify modules that work together in the thermotolerance process. This work will generate relevant information that can be leveraged to implement practices that help mitigate the effects of high temperatures on crops.

FUNCTIONAL AND STRUCTURAL STUDIES OF A NOVEL ENDOLYSIN FROM ICP1 PHAGE ACTIVE AGAINST MULTIDRUG-RESISTANT BACTERIA

Laura Espinosa-Barrera¹, Oscar Linares-Vergara¹, Beatriz Velazquez-Cruz¹, Cesar Cardona-Felix², Sara Centeno-Leija^{3*} and Hugo Serrano-Posada^{3*}

¹Laboratorio de Biología Sintética, Estructural y Molecular, Universidad de Colima, Carretera Los Limones-Loma de Juárez, 28627 Colima, Colima, México, ²CONAHCyT, Instituto Politécnico Nacional, Centro Interdisciplinario de Ciencias Marinas, 23096 La Paz, Baja California Sur, México, ³CONAHCYT, Laboratorio de Biología Sintética, Estructural y Molecular, Universidad de Colima, Carretera Los Limones-Loma de Juárez, 28627 Colima, Colima, México.

*Autores de correspondencia: scenteno0@ucol.mx, hserrano0@ucol.mx

Vibriophage ICP1-type are specific predators of *Vibrio cholerae*, the bacterium that causes cholera, with a recently described holin-endolysin-spanin lysis system tripartite finely modulated by an antiholin-mediated mechanism. Nonetheless, the structure, activity, or catalytic mechanism of the critical endolysins from ICP1 phages remains mysterious. In this work, a data mining of ICP1 phage genomes revealed an endolysin from *V. cholerae* phage JSF14 (OsLys). Biochemical characterization of OsLys endolysin showed optimal muralytic activity at pH 7.0, 37 °C supplementing with 10 mM NaCl or Ca²⁺. Moreover, OsLys exhibited lytic activity against multidrug-resistant G- bacteria: *V. cholerae* ATCC-25872, *V. parahaemolyticus* ATCC-17802, *Escherichia coli* BA196 y H2Bi strains, *Pseudomonas aeruginosa* PAO-01, and *Salmonella enterica*. Structure analysis revealed that OsLys is a modular endolysin with a conserved Peptidoglycan binding-1 domain (PG-1) followed by a cysteine-histidine-dependent amido-hydrolase/peptidase domain (CHAP). Phylogenetic analysis indicated that OsLys grouped with a conserved clade of endolysins modular of ICP1 phages, and the elements of the lytic system were identified in the genome of JSF14 Vibriophage. OsLys is the first endolysin characterized of ICP1 phage and was useful to preventing and controlling G- bacteria strains.

SCL and HSP acknowledge financial support from CONAHCyT grants CB-2018-A1-S-18011 and CF-2019-33-549477. CCF acknowledge financial support CF-2019-33-549477 from CONAHCyT. SCL and HSP also acknowledge financial support from CONAHCyT grant INFR-2021-17-316456. OLV and LEB thank for the master (No.800362) and posdoctoral (No. 218862) fellow-ships from CONAHCyT.

THEORETICAL STUDY OF THE NAMH HYDROXYLASE OF *MYCOBACTERIUM TUBERCULOSIS* AS A POTENTIAL TARGET IN THE DEVELOPMENT OF NEW ANTIBIOTICS

Roberto Sealtiel Farías Gaytán¹, Abraham Madariaga Mazón¹, Carol Siseth Martínez Caballero¹

¹Institute of Chemistry UNAM, Circuito exterior, C.U., 04510, Mexico City.

E-mail: seal1999@live.com.mx

Tuberculosis (TB) is an infectious disease caused by *Mycobacterium tuberculosis* that continues to be a significant threat to global public health. The appearance of strains resistant to multiple drugs has complicated their treatment, emphasizing the need to develop new antibiotics¹. A promising strategy is the study of the biosynthesis of peptidoglycan (PG), an essential component of the cell wall of *M. tuberculosis*. NamH hydroxylase is a cytoplasmic enzyme involved in this biosynthesis, and its inactivation generates sensitivity to β -lactam antibiotics and lysozyme, which makes it a potential target for new drugs².

The present work addresses an *in silico* perspective, in which we have used molecular modeling and virtual screening tools to identify potential NamH inhibitors. Using the three-dimensional NamH model predicted by AlphaFold and validated with tools such as SWISS-MODEL and Dogsite Scorer, we have identified two main active sites in the UlaG and Rieske domains of the protein. These sites were analyzed for their binding potential with ligands, using blind molecular coupling techniques with AutoDock Vina-GPU.

Virtual screening was carried out with a library of 12,123 compounds, including known anti-tuberculosis drugs and library molecules focused on anti-tuberculosis compounds. The results showed a diverse distribution of binding affinities, highlighting some candidates with a high probability of inhibiting NamH activity. These compounds presented greater binding affinities in the previously identified active sites, especially in the Rieske domain, suggesting their potential efficacy as NamH inhibitors.

These *in silico* findings provide a solid basis for the selection of compounds that can be validated experimentally in the future, an aspect that will contribute significantly to the development of new therapeutic approaches against tuberculosis, particularly for strains resistant to multiple drugs.

References

1. World Health Organization [WHO]. (2023). Global Tuberculosis Report 2023.
2. Raymond, J. B., Mahapatra, S., Crick, D. C. & Pavelka, M. S. (2005). *The Journal of Biological Chemistry*, 1(7), 326-333.

PHENOTYPIC AND GENOTYPIC EVALUATION OF ANTIBIOTIC RESISTANCE IN *PSEUDOMONAS AERUGINOSA* ISOLATED FROM HYDROTHERMAL WATER SAMPLES FROM CHIGNAHUAPAN, PUEBLA

Jennifer Vianey Fernández Dodero¹, Hugo G. Castelán Sánchez², Candelario Vázquez Cruz³, Patricia Sánchez Alonso³, Norma Elena Rojas Ruiz³

¹Licenciatura en Biotecnología. Facultad de Ciencias Biológicas. Benemérita Universidad Autónoma de Puebla.

²Department of Pathology and Laboratory Medicine, Western University, London, ON, Canada.

³Instituto de Ciencias. Benemérita Universidad Autónoma de Puebla. Instituto de Ciencias, BUAP. Edificio IC11, CU. Av. San Claudio y 24 Sur, Col. Jardines de San Manuel, CP 72570. Puebla, México. Tel. 2222295500 ext. 2536. candelario.vazquez@correo.buap.mx

Multiresistance can be defined as a bacterium's resistance to various antibiotics. *Pseudomonas aeruginosa* is one of the critical priority bacteria according to the WHO, belonging to the so-called ESKAPE group, which includes multidrug-resistant and potentially dangerous bacteria in the intrahospital setting. In this study, the resistance profile of *Pseudomonas aeruginosa* strains previously isolated from thermal water samples from Chignahuapan, Puebla, was determined.

The aim of this work is to compare the phenotype and genotype of antibiotic resistance in these thermal-origin strains. Additionally, whole-genome sequencing was performed to determine resistance genes in silico. The Tygs tool was used for taxonomic identification of the *Pseudomonas aeruginosa* genome and phylogenetic tree reconstruction. Subsequently, genome completeness was evaluated with MIGA, achieving 98.1% and 93.81% ANI. Genome visualization was performed using GCVIEW.

To determine antibiotic resistance, antibiograms were conducted under two nutritional conditions (P Agar and LB Agar), using commercial multidiscs for Gram-negative bacteria. The antibiograms indicated that *Pseudomonas aeruginosa* strain 39 exhibits resistance to a wide variety of antibiotics, with variations between the nutritional conditions used. In LB Agar, it showed resistance to 9 antibiotics (netilmicin, gentamicin, cefotaxime, carbenicillin, ampicillin, carbenicillin, cephalothin, cefotaxime, netilmicin, chloramphenicol, trimethoprim, and nitrofurantoin) and sensitivity to 3 (norfloxacin, amikacin, and ciprofloxacin). On the other hand, in P Agar, the bacterium was resistant to 10 antibiotics (netilmicin, gentamicin, cefotaxime, carbenicillin, ampicillin, carbenicillin, cephalothin, cefotaxime, netilmicin, chloramphenicol, trimethoprim, nitrofurantoin, and amikacin) and sensitive to 2 antibiotics (norfloxacin and ciprofloxacin).

The phenotypic resistance results in the laboratory were compared with the results of bioinformatic analysis using RGI (Resistance Gene Identifier) for the evaluation of antimicrobial resistance in silico, which utilizes the CARD database. The results show that *P. aeruginosa* is resistant to the antibiotics tested in vitro in the laboratory, as well as to some others that are not present in the commercial multidiscs used and their derivatives or composite presentations. Genomic analysis indicated that *P. aeruginosa* has resistance genes for 41 antimicrobials. However, many of these may not be expressed, indicating that under environmental pressures, they may be expressed, although this requires further investigation.

DEVELOPING A GENERATIVE AI ASSISTANT FOR REGULONDB: ENHANCING DATA ON *E. COLI* K-12 TRANSCRIPTIONAL REGULATION

Socorro Gama-Castro, Leonel Valencia-Estudillo, Gabriel Alarcón-Carranza, Heladia Salgado, Carlos F. Méndez-Cruz, Oscar Lithgow-Serrano, Ana Bonavides-Aguilar, Paloma Lara, Fanny Flores-Gallegos, César Bonavides-Martínez C., Julio Collado-Vides

Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México.
Avenida Universidad S/N, Cuernavaca, Mor. México.

RegulonDB is a comprehensive database that provides information about the regulation of gene expression in the bacterium *Escherichia coli* K-12. It encompasses a wide array of data, including details on transcription factors, regulatory networks, operons, promoters, terminators, and more. In this study, we explore the integration of ChatGPT, an advanced language model of Generative Artificial Intelligence (IA), as an assistant to improve usability of RegulonDB. Our main objective is to leverage ChatGPT to automate the curation process about genetic regulation within RegulonDB, and to develop a chatbot for efficient and user-friendly database queries.

By employing ChatGPT for automated curation, we aim to streamline database maintenance and update processes. In addition to automated curation, we are exploring the development of a ChatGPT-based chatbot designed to facilitate intuitive and precise queries to RegulonDB. Users will be able to pose questions in natural language and receive accurate, context-aware responses, improving the user experience. We envisage that this functionality will make the database more accessible to a broader audience, including researchers, educators, and students, by providing quick and straightforward access to its extensive information.

Moreover, integrating automated curation may be a crucial step towards the creation of a multi-organism RegulonDB. Expanding the scope of this database beyond *E. coli* will support a wider range of genomic studies and enable comparative analyses across different species. This multi-organism resource will offer valuable insights into the conservation and divergence of regulatory mechanisms, thereby enriching our understanding of transcriptional regulation in various bacterial species.

In conclusion, incorporating the ChatGPT capabilities into RegulonDB represents a significant advancement in database maintenance and user interaction. We consider that the automated curation and chatbot functionalities will not only improve the efficiency and accuracy of data curation but also promote broader and more effective use of RegulonDB. This innovative integration is poised to transform RegulonDB into a more dynamic, multi-organism resource, ultimately contributing to the advancement of genomic research.

EVALUATION OF TWO TAXONOMIC ASSIGNMENT METHODS FOR THE STUDY OF LIZARD GUT MICROBIOME USING METAGENOMIC DATA

Elizabeth Selene Gómez Acata, Nina Montoya Ciriaco, Luc Dendooven, Yendi Ebenezer Navarro Noya

Laboratorio de Interacciones Bióticas, Centro de Investigación en Ciencias Biológicas, Universidad Autónoma de Tlaxcala, Km 10.5 de la carretera San Martín Texmelucan S/N, San Felipe Ixtacuixtla, C.P. 90120, Ixtacuixtla de Mariano Matamoros, Tlaxcala.
E-mail: yendiebenezer.navarro.n@uatx.mx

Evaluating the microbial composition of the microbiome in an environment involves the taxonomic assignment of reads to a specific taxa using reference databases. Numerous bioinformatics tools have been designed to classify metagenomic data and estimate the abundance profiles of taxa. However, taxonomy assignment can vary depending on the method and databases used. In this study, we compared two methods for the taxonomic assignment of shotgun metagenomic reads. Samples were taken from the gut of the viviparous lizard *Sceloporus grammicus* Wiegmann, 1828, and metagenomic DNA was extracted and sequencing using the NovaSeq 6000 platform. After quality-filtered and adapter-removed reads, taxonomic assignment was done using the Kraken and the Qiagen CLC classifier. A total of 909 bacterial features were identified using the Qiagen CLC classifier with an abundance of 469,454 sequences, while 2475 features with an abundance of 4,267,212 sequences were obtained using the Kraken classifier. The analyses performed allow us to discuss the advantages and disadvantages of each approach, as well as their accuracy in identifying specific bacteria. Our findings provide valuable information for the selection of taxonomic tools in the study of the reptile microbiome.

COMPARATIVE TRANSCRIPTOMICS OF THE PHARMACOLOGICAL RESPONSE TO THE COMPOUND AZD5363 IN TWO HUMAN DUCTAL ADENOCARCINOMA CELL LINES

González Ortiz Rebeca, Sánchez Flores Fidel Alejandro
Rebeca.gonzalez@ibt.unam.mx; Alejandro.sanchez@ibt.unam.mx

Background. The importance in pharmacogenomics lies in specialized treatment, thus avoiding side effects (Roden et al., 2019). In the USA 3% to 7% of hospitalizations are due to adverse drug reactions; in Mexico they represent 9% per year (Checa Rojas, 2017). The effect of the compound AZD5363 on ductal adenocarcinoma at the transcriptional level in different cell lines (LNCaP and BX-CP3) can provide us with information about the mechanisms of cancer resistance, as well as valuable information for the design or combination of different drugs, based on the expression profile of the cells after receiving a treatment.

Objective. To characterize gene regulation by RNAseq and Bioinformatics, in ductal adenocarcinoma cells treated with the drug AZD5363 using transcriptomics.

Methods. Cell culture. The transcriptomic analysis started from the cancer lines LNCaP and BxPC-3 respectively. Both lines were cultured at 80% confluence. Cells were incubated for 16 h at 37 °C, 5 % CO₂, then control and target cells had their medium changed and target cells were supplemented with AZD5363 drug. RNAseq

Starting from the purified RNA, mRNA libraries were made. The first step of the computational analysis consisted of validating the quality of the reads using the FastQC tool. Differential expression was performed using the lima, edge2 and deseq methods.

Results and Conclusions. LNCaP and BX-CP3 cells with and without AZD5363, show a differential expresión profile, this implies a change in cell dynamics when there is a pharmacological treatment. Differential expression of 898 and 560 genes was found in the LNCaP and BX-CP3 lines, respectively. The under- and over-expressed genes are associated with macromolecule biosynthesis, as well as with the FoxO and TGF- β pathways respectively.

PIPELINE IMPLEMENTATIONS USING THE GALAXY PLATFORM FOR REFERENCE ASSEMBLY OF CHLOROPLAST GENOME FROM TRANSCRIPTOME DATA.

Ana Carolina González Trillo, Yolanda Herrera Arrieta¹, René Torres Ricario¹, Miguel Ángel Reyes-López²

¹Instituto, Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional, CIIDIR Instituto Politécnico Nacional, Unidad Durango. Sigma 119, 20 de Noviembre II, 34220 Durango, Dgo, ²Centro de Biotecnología Genómica del Instituto Politécnico Nacional. Reynosa, Tam.
Email: yherrera@ipn.mx

The use of transcriptome as a starting point in genomic studies offers a dynamic perspective on gene expression and allows precise genome reconstruction, revealing detailed sequence and structure information¹. While the conventional approach to genome assembly for comparison and phylogenetic analysis involves direct DNA sequencing^{2, 3}, there is growing interest in assembling genomes from transcriptome data⁴. Existing procedures require high-capacity processing equipment with a minimum of 6 cores, 12 threads, 32 GB RAM, and bioinformatics expertise^{5, 6}. This study aimed to implement and validate a previously established methodology⁶ for assembling chloroplast genome from transcriptome data via the Galaxy platform, an online data processing environment for users with limited computing power and bioinformatics experience⁷. The procedure was based on Senthilkumar et al. (2021) on assembling the chloroplast genome from the transcriptome of *Pterocarpus santalinus*, adapting it to Galaxy. The methodology follows five steps: (1) downloading Illumina reads with Faster Download and Extract Reads in FASTQ (Galaxy Version 3.0.10+galaxy0), (2) trimming low-quality reads with Trimmomatic (Galaxy Version 0.36.6), (3) mapping reads using Bowtie2 (Galaxy Version 2.5.3+galaxy0) against the chloroplast genome of *P. santalinus* MT249117.1⁸, (4) filtering matched reads with Samtools View (Galaxy Version 1.15.1+galaxy2), and (5) searching for the consensus sequence with IVAR (Galaxy Version 1.4.2+galaxy0). To validate the data, chloroplast genome annotation and comparative genomic analysis were performed. The results showed that using this methodology with *P. santalinus* transcriptome data, a chloroplast genome with a size of 158,967 bp can be assembled with 158 annotated genes, 8 rRNAs, and 36 tRNAs, matching Senthilkumar's protocol data (158,966 bp, 158 genes, 8 rRNAs and 36 tRNAs). Comparative alignment revealed high identity between the reference genome (MT249117.1) and the assembled transcriptome with no low identity regions, improving on Senthilkumar's method. Another highlight is the reduced processing time, IVAR in Galaxy takes 5-10 minutes to assemble the transcriptome, compared to about 30 minutes required by programs NOVOPlasty and GetOrganelle^{9, 10}. In conclusion, this modification of Senthilkumar's strategy for assembling chloroplast genomes from transcriptomes in Galaxy is more efficient, achieving higher identity with the reference genome in less time, making it valuable for projects with limited computing capacity and bioinformatics expertise.

References

1. Adams J (2008). *Nature Education*, **1**(1), 195.
2. Venket R, Kraft L, Mesny F, Rigerte L (2022). *Briefings in Bioinformatics*, **23**(2).
3. Twyford AD, Ness, RW (2016). *Molecular Ecology Resources*, **17**(5), 858–868.
4. Yu T, Mu Z, Fang Z, Liu X, Gao X, Liu J (2020). *Genome Research*.
5. Zhao D, Liu T, Yu T (2023). *Biology Methods and Protocols*, **8**(1).
6. Senthilkumar S, Ulaganathan K, Ghosh DM (2021). *Biotechnolgy*, **11**(8), 393.
7. Afga E, Baker D, Batut B, van den Beek M, Bouvier D, Cech M, Grüning B (2018). *Nucleic Acids Research*, **2**(46), W537–W5.
8. Hong Z, Wu Z, Zhao K, Yang Z, Zhang N, Guo J, Tembrock LR, Xu D (2020). *International Journal of Molecular Science*, **21**(11):3758.
9. Dierckxsens N, Mardulyn P, Smit G (2017). *Nucleic Acids Research*, **45**(4), e18.
10. Jin J, Yu WB, Yang JB, Song Y, dePamphilis C, Yi T, Li DZ. (2019). *BioRxiv*. 256479.

PLANT THERMOMORFOGENESIS: PROTEIN-PROTEIN INTERACTION AND GENE COEXPRESSION NETWORKS IN ARABIDOPSIS THALIANA

Fernanda Lizeth Gutiérrez-Palomeque^{1*}, Ana Karen Díaz-Martínez¹, Humberto Herrera-Ubaldo², Cinthya Paola Rangel-Chávez¹

¹Departamento de Bioquímica. Instituto Tecnológico Superior de Irapuato, Irapuato 36 821, Guanajuato México.

²Department of Plant Sciences, University of Cambridge, Cambridge CB2 3EA, United Kingdom.

Email: palomequelizeth92@gmail.com, humberto.herrera@cinvestav.mx, cynthia.rc@irapuato.tecnm.mx

Due to global climate change, plants may experience prolonged exposure to extreme temperatures, causing them to enter a state of thermal stress that generates changes in metabolism and morphology. Plants have developed response mechanisms to mitigate these effects, such as thermomorphogenesis, directional growth away from hot soils or surfaces or change in architecture to promote cooling. At the molecular level, these mechanisms are complex, varied, and mediated by protein-protein interactions (PPIs). In this sense, the construction and study of PPI networks are essential to understanding the role of proteins with known functions, as well as to finding new regulators involved in signalling processes and understanding the related biological mechanisms or processes.

Given the above, the present work aims to construct and analyze the global PPI network related to high-temperature responses and subsequently focus on a sub-network of the thermomorphogenesis process. The gene co-expression networks associated with this process are also being analyzed.

Information was collected on proteins involved in the heat stress response and their physical interactions. Then, an PPI network containing 187 proteins with 1712 interactions was constructed. Subsequently, we focused on those proteins related explicitly to the process of thermomorphogenesis and obtained a sub-network with 164 proteins and 1531 interactions. The networks showed that thermomorphogenesis is associated with thermotolerance, flowering, photosynthesis, adaptation, vernalization, freezing tolerance, circadian clock, and transcriptional and post-transcriptional regulation. Finally, with this information, we will perform analyses to characterize proteins with unknown functions to find new regulators, which, in the long term, will help understand thermomorphogenesis and, in the future, apply it for crop improvement.

SEARCHING FOR STRUCTURAL TRANSITIONS IN A PARALLEL PROTEIN NETWORK

Juan Martín Hernández Castillo¹ and Gabriel Del Río Guerra¹

¹Instituto de Fisiología Celular Universidad Nacional Autónoma de México,
Circuito Investigación Científica Ciudad Universitaria, Coyoacán.
E-mail: juanmartin@ifc.unam.mx

The 3D structure of a stable protein can be represented with a graph, where each node is an amino acid and each edge is a proximity relationship. By counting the different complete subgraphs (maximal cliques), 26 residue group classes (RCC)¹ can be identified, which defines a vector with enough information to predict the structural classification, function² and interactions between proteins³ using Machine Learning, outperforming more complex methods. Two proteins present different RCCs, so the angular aperture between their vectors allows to quantify their similarity. Two semi-parallel vectors are two proteins with nearly the same RCC counts, diverging less than 5°. It is to be expected that tertiary structures will tend to be parallel to similar tertiary structures, i.e., parallelism relationships are found between members of the same structural class (Mainly Alpha, Beta, Alpha Beta, Few Secondary Structures and Special, as on CATH database).

As my current progress shows, almost 100% of the protein domains registered in the PDB have at least one semi-parallel domain, allowing the construction of a network of parallelism that connects domains that are very different from each other, both in structure and origin. Even more surprising is the fact that there are parallel relationships between members of different structural classes and with this, structural similarities are found between homologous superfamilies which are different according to the CATH criterion. These relationships are structural transitions and do not occur at random, however the pattern that allows predicting specific transitions between different folds following parallelism relationships is yet to be discovered.

References

1. Ricardo Corral-Corral, et al. (2015) *Machine Learnable Fold Space Representation based on Residue Cluster Classes*. *Comp. Biol. and Chem.* **59** (2015) pp. 1-7.
2. F. Fontove, G. del Río (2020) *Residue Cluster Classes: A Unified Protein Representation for Efficient Structural and Functional Classification*. *Entropy* 2020 April, 20; **22** (4): 472
3. Albros H. Poot-Velez, et al. (2020) *Protein-Protein Interactions Efficiently Modeled by Residue Cluster Classes*. *Int. J. Mol. Sci.* 2020 Jul 6; **21** (13): 4787

IN SILICO STUDY OF GALANIN RECEPTORS AS POTENTIAL TARGETS FOR THE TREATMENT OF DEPRESSION

Beatriz Hernández-Estrada¹, Giovanny Aguilera Durán², Rubén Antonio Romo-Mancillas^{3*}

^{1,3}Facultad de Química, Universidad Autónoma de Querétaro, Centro Universitario, 76010, Querétaro, Querétaro, México.

²Laboratorio de Química Cuántica y Modelado Molecular, Unidad Académica de Ciencias Químicas, Universidad Autónoma de Zacatecas.

bhernandez71@hotmail.com¹, giovanny.aguilera@uaz.edu.mx², ruben.romo@uaq.mx³

Depression is a mental disorder characterized by loss of interest or pleasure in activities and cognitive, physical, emotional, and behavioral symptoms. It is estimated to affect 280 million people worldwide, of which 60% do not receive adequate care, of which one third of patients do not receive adequate treatment and 16% relapse. Depression is associated with lower life expectancy due to suicide rates.

Current research is focused on the pathways related with neurotransmission, being neuropeptides such as galanin of recent interest. Activation of GALR2 has antidepressant effects, whereas GALR1 and GALR3 have prodepressant effects. Antagonism of the latter is a potential strategy for the treatment of depression. Computational techniques, such as homology modeling and molecular dynamics, are essential to study these receptors and design peptides with potential antidepressant effect.

In the project, homology models of galanin receptors were constructed using the Robetta server using Rosetta-fold mode. Model templates were obtained for GALR1 (PDB: 7WQ3) and GALR2 (PDB: 7WQ4), and for GALR3 its UniProt sequence (ID: O60755) was used. The models were subjected to molecular dynamics simulations to evaluate their behavior. The simulations were performed in Gromacs 2023.4, using the CHARMM-GUI server to build the systems and the CHARMM36 force field. A three-point model (TIP3) was used for the water molecules, adding a CHL1, POPC, POPE, POPI, POPS membrane and the corresponding ions were added using NaCl at 0.15 M. Using an isothermal-isobaric assembly (NPT) at 1 atm pressure and 310.15 K. The systems were subjected to 5000 minimization steps followed by 6 equilibrium steps prior to production simulations. The obtained models were evaluated in MolProbity, showing high percentages of favored residues in the Ramachandran diagram. Finally, molecular docking was performed on the ClusPro public server with agonists (galanin and M617) and antagonists (m871 and M40), identifying relevant residues for receptor activity.

References

1. Rana, T., Behl, T., Sehgal, A., Singh, S., Sharma, N., Abdeen, A., & Bungau, S. (2022). Progress in Neuro-Psychopharmacology and Biological Psychiatry, 114, 110478. doi: 10.1016/j.pnpbp.2021.110478.
2. Demsie D., Altaye B., Weldekidan E., Gebremedhin H., Alema N., Tefera M., Bantie A. (2020) Biologics. Targets and therapy. 21;14:37-45. doi: 10.2147/BTT.S240715.

DETECTION OF TRANSCRIPTIONAL DETERMINANTS OF CELLULAR LONGEVITY IN HUMAN TISSUES USING A MACHINE LEARNING-BASED APPROACH

Jorge Adrián Islas^{1,2,*}, Selene Yunuén Mena¹ y Humberto Gutiérrez¹

¹Departamento de Genómica Computacional, Instituto Nacional de Medicina Genómica (INMEGEN), Periférico Sur 4809, Tlalpan, Ciudad de México, CP:14610, México.

²Posgrado de Ciencias Biológicas, Universidad Nacional Autónoma de México.

*joarian_96@outlook.com, +5255017324

In multicellular organisms, most postmitotic cell types have a short life and need to be continuously replaced. Neuronal cells, on the other hand have the longest lifespan as individual cells and need to survive for as long as the whole organism. These differences in cellular longevity correspond to differences in postmitotic cellular maintenance and, at present, the nature of the molecular machinery responsible for differences in postmitotic cell survival remains unclear. Using available expression data from primary human cells, for over 20 thousand genes, aligned with postmitotic cell longevity data for these same tissues and cell types, we identify, using jackknife correlations, 1616 genes whose levels of expression are robustly associated with postmitotic cellular longevity across various tissues, and 710 genes negatively correlated. Deploying a complementary strategy based on decision tree machine learning approaches (random forest regression models), we identify among the top 5% most highly ranked predicting transcripts a statistically significant overlap with positively correlated genes but a significant under representation of negatively correlated genes. Coexpression analysis shows that these cell longevity-associated genes display an overall level of coexpression in human brain tissue higher than expected by chance, suggesting a high level of functional coordination between these transcripts in long living nervous system tissue. Our results so far, identify a host of novel molecular determinants associated with long term neuronal survival.

Keywords: Longevity, Neuronal maintenance, ageing

KLF10: A CRUCIAL TRANSCRIPTION FACTOR IN MACROPHAGE DEFENSE AGAINST MYCOBACTERIUM TUBERCULOSIS

Hilda C. Leyva-Sánchez¹, Edgardo Madrid-Paulino¹, Tomás Villaseñor-Toledo¹, Verónica García Radilla¹, Rogelio Hernández Pando², Leonor Pérez Martínez¹, Martín Gustavo Pedraza-Alva¹

¹Department of Molecular Medicine and Bioprocesses, Institute of Biotechnology, National Autonomous University of Mexico, Cuernavaca, Morelos, Mexico. ²Department of Pathology, National Institute of Respiratory Diseases (INNSZ), Mexico City, Mexico.

Correspondence: Dr. Martín Gustavo Pedraza-Alva. Phone: (777) 3290 869. Email: gustavo.pedraza@ibt.unam.mx

Tuberculosis (TB) is a disease caused by the bacterium *Mycobacterium tuberculosis* (Mtb) and primarily affects the lungs, leading to high mortality and morbidity rates among the population. It is considered the leading cause of death due to antibiotic-resistant bacterial infections.

During infection, macrophages are the first responders of the immune system. Recognition of Mtb by macrophages leads to the digestion and phagocytosis of the bacteria in phagosomes, but Mtb can also exploit these cells for survival and replication within the human body. Additionally, Mtb regulates inflammation by inducing the expression of anti-inflammatory cytokines such as IL-10 and TGF β to maintain its survival. IL-6, TNF, and IFN- γ are crucial for activating macrophages' bactericidal functions and for granuloma maintenance.

Concurrently, the transcription factor Kruppel-like factor 10 (KLF10) emerges as a key player in the immune response to Mtb. Induced upon macrophage infection, KLF10 modulates the expression of both pro-inflammatory (e.g., IL-6, TNF) and anti-inflammatory (e.g., IL-10) cytokines, negatively regulating the former and positively regulating the latter. Bioinformatic analyses reveal putative KLF10 binding sites within these cytokine genes, suggesting a direct regulatory role in inflammation. To elucidate how Mtb modulates KLF10 transcriptional activity to favor an anti-inflammatory microenvironment, which is crucial in TB pathogenesis, we are performing ATAC-seq and RNA-seq analyses of Mtb-infected wild-type and KLF10 knockout macrophages. These findings will provide important insights into the molecular mechanisms underlying Mtb infection and disease, highlighting future therapeutic strategies and vaccine development against TB.

Work supported by CONACYT (Grants: CF/2019-40792). HCLS was supported by a CONACYT postdoctoral fellowship.

SEARCH FOR SELECTIVE INHIBITORS ON PTP1B OF PEPTIDE NATURE

Rodolfo Lizárraga, Martín González Andrade and Laura Álvarez

Facultad de Medicina, Escolar 411A, Copilco Universidad, Coyoacán, 04360 Ciudad de México, CDMX
E-mail: RALV117@hotmail.com

Type 2 Diabetes (T2DM): Represents approximately 90% of diabetes cases. It has a polygenic origin and can start with hyperinsulinemia but ultimately leads to insulin resistance ^[1]. Currently, there are various drugs for glycemic control with different molecular targets. The protein PTP1B has been highlighted as a potential target for treating this disease. PTP1B is a ubiquitous protein involved in insulin receptor desensitization, affecting glucose uptake in cells. It is a monomeric cytosolic protein encoded by the PTPN1 gene and consists of 435 residues distributed across three structural domains: a highly conserved N-terminal catalytic domain known as the PTP domain, an intrinsically disordered regulatory domain also called the proline-rich domain, and a hydrophobic C-terminal signaling domain ^[2].

However, PTP1B inhibitors have not yet passed clinical trials due to side effects, especially autoimmune disorders related to a homologous protein called TCPTP found in T lymphocytes. TCPTP, a phosphatase involved in T cell activation regulation, is abundantly expressed in hematopoietic cells. Despite high homology in the catalytic domain (74% amino acid sequence similarity). Selective inhibition of PTP1B over TCPTP remains one of the most challenging aspects of drug discovery due to this high homology ^[3]. In our quest for greater selectivity in PTP1B inhibition, we have explored a post-translational modification called sumoylation. This process involves the attachment of the Small Ubiquitin-like Modifier (SUMO) protein to specific lysine residues on PTP1B. To design peptides derived from complexes rationally, understanding this sumoylation process and having the complete protein, including its intrinsically disordered region (amino acids 279-400), are crucial for peptide design.

References

1. Authors of the American Association of Clinical Endocrinology Clinical Practice Guideline: Developing a Diabetes Mellitus Comprehensive Care Plan–2022 Update (Endocr Pract. 2022 Oct;28(10):923-1049.
2. Coronell-Tovar, A., Cortés-Benítez, F., & González-Andrade, M. (2023). The importance of including the C-terminal domain of PTP1B1-400 to identify potential antidiabetic inhibitors. Journal of enzyme inhibition and medicinal chemistry, 38(1)
3. Li, X., Wang, L., & Shi, D. (2016). The design strategy of selective PTP1B inhibitors over TCPTP. Bioorganic & medicinal chemistry, 24(16), 3343–3352

MOLECULAR DYNAMICS AND DOCKING ANALYSIS OF QUERCETIN AND PATULETIN FOR EVALUATING ANTITUMOR ACTIVITY

¹Aarón Saúl López Carrasco, ¹Gabriela Figueroa González, ²Laura Itzel Quintas Granados, ¹Edgar Yebrán Villegas Vázquez and ¹Octavio Daniel Reyes Hernández

¹Laboratorio de Farmacogenética, Unidad Multidisciplinaria de Investigación Experimental Zaragoza, Facultad de Estudios Superiores Zaragoza, Universidad Nacional Autónoma de México, Ciudad de México 09230, México. Tel: 5539005899.

²Universidad Autónoma de la Ciudad de México, Colegio de Ciencias y Humanidades, Doctor García Diego 168, Colonia Doctores, Alcaldía Cuauhtémoc, 06720.
E-mail: octavio.reyes@zaragoza.unam.mx

Flavonols, a class of flavonoids, have recently been shown to have anticarcinogenic effects. Quercetin and patuletin, which belong to this class of compounds, have historically been used therapeutically against various diseases and have recently been evaluated as anticancer agents due to their effects on the induction of apoptosis and inhibition of cell proliferation. However, there is little scientific evidence on the molecular mechanisms by which quercetin and patuletin exert their antitumor effects. In this context, it is of interest to analyze the possible interactions between quercetin and patuletin with biomolecules expressed during breast cancer development to increase the knowledge of the therapeutic effects of these compounds. These possible interactions were evaluated by molecular docking, a technique that allows the study of ligand-receptor interactions, to find therapeutic targets for the specific design of quercetin- and patuletin-based drugs. First, a search was performed in public databases such as PubChem, PDB, SIB, and GeneCards, using key terms such as “breast cancer,” “quercetin,” “patuletin,” “quercetin and docking,” “patuletin and docking,” “quercetin and cancer,” and “patuletin and cancer.” This search yielded information on proteins that have been associated with these flavonols and cancer. The identified proteins were analyzed using specialized software such as Protein Data Bank, AutoDockTools-1.5.6, and Chimera1.11rc. Our findings suggest that these flavonoids potentially interact with proteins involved in cell proliferation, migration, survival, and metabolism, such as phosphatidylinositol-3 kinase and aldo-keto reductase family 1 member B. In summary, our results present an overview of the potential of these flavonols with an anticancer effect in in vitro and in vivo models.

Funding. PAPIIT-IA206724 and PAPIIT-IN221824.

IDENTIFICATION OF MOLECULAR MARKERS, SNPS, ASSOCIATED WITH MORPHOLOGICAL CHARACTERISTICS OF *BIXA ORELLANA* L.

Ana Lucía López-Gurgua¹, Renata Rivera-Madrid¹, Jorge Humberto Ramírez Prado¹

¹Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, A.C.
Calle 43 No. 130, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México.
E-mail: renata@cicy.mx

Achiote (*Bixa orellana* L.) is a woody plant that produces a pigment called bixin, which accumulates mainly in the seeds. Bixin is important in the food, cosmetic, and pharmaceutical industries; however, no registered varieties of achiote exist to establish homogeneous plantings to ensure predictable seed production and pigment content. This study mainly aimed to identify SNPs in transcriptomes of three achiote variants for molecular markers and relate them to bixin content, flower color, and fruit dehiscence. SNPs were searched for in transcriptomes of three morphologically contrasting accessions of achiote (N4, N5, and P12). A total of 17,587 SNPs were identified using the GATK program. The variant rate was obtained with the help of the snpEff program, having 1 variant every 14,844 bases, a ratio $Ts/Tv=1.1497$. A random selection was made to relate them to the morphology of the accessions and subsequently use them as molecular markers to apply Marker Assisted Selection (MAS) in morphological traits of interest.

MODELING KREBS CYCLE FROM RAT LIVER, HEART, AND HEPATOMA MITOCHONDRIA

Álvaro Marín Hernández, Luz Hernández-Esquivel, Isis Del Mazo-Monsalvo, Silvia C. Pacheco-Velázquez, Rocío D. Feregrino-Mondragón, Diana X. Robledo-Cadena, Rosina Sánchez-Thomas, Ricardo Jasso-Chávez, Emma Saavedra

Departamento de Bioquímica, Instituto Nacional de Cardiología Ignacio Chávez,
Mexico City, 14080, Mexico
E-mail: alvaro.marin@cardiologia.org.mx

The Krebs cycle (KC) is essential for cell intermediary metabolism; it produces reducing equivalents (NADH and FADH₂) used for synthesize ATP and provides precursors for proliferation. In cancer cells, changes in enzyme activities suggest possible modifications in the steps that control the Krebs cycle flux compared to non-cancer cells. Therefore, we propose constructing a kinetic model of KC to establish the enzymes that may be inhibited to decrease the KC flux, specifically in cancer cells. The kinetic parameters (V_{max} and K_m) of Krebs cycle's enzymes were determined in isolated mitochondria for model construction. In general, the V_{max} values of all enzymes showed an increase in hepatoma mitochondria (HepM) compared to rat liver mitochondria (RLM). In contrast, the V_{max} values in rat heart mitochondria (RHM) were higher than in RLM and HepM. On the other hand, K_m values of the enzymes in the three types of mitochondria were similar. The levels of Krebs cycle metabolites showed variations in function of the oxidizable substrates used, in the following order: 2OG > pyruvate/malate > glutamine > succinate.

The kinetic models were constructed using the software COPASI. After an exhaustive refinement process of the kinetic models for each type of mitochondria, the simulations predicted the concentrations of all metabolites within the same order of magnitude determined experimentally or reported. The validated models indicated that NADH consumption (complex I) exerted a higher control on the Krebs cycle flux in HepM; meanwhile, it also exerted control in RHM but at a lesser extent. In contrast, the flux control in RLM was shared between 2OGDH and PDH. These results suggested that cancer cells may be more sensitive to the inhibition of complex I than heart and other non-cancer cells. Indeed, when cancer cells from cervix (HeLa), breast (MCF7), and prostate (PC3) were exposed to rotenone (complex I inhibitor), the cellular proliferation was more sensitive to rotenone than heart (H9c2) and non-cancer (MCF10A) cells. In contrast, the proliferation of all cells had a similar sensitivity to malonate, an inhibitor of succinate dehydrogenase, an enzyme that does not exert control on the Krebs cycle flux. Our results suggest that kinetic modeling and metabolic control analysis allow the identification of targets to design strategies to specifically inhibit the proliferation in cancer cells without toxic effects in normal cells, such as heart cells which are sensitive to standard chemotherapy.

PREDICTION OF THE STRUCTURAL EFFECT OF VARIANTS OF UNCERTAIN SIGNIFICANCE DETECTED BY NGS IN MEXICAN CHILDREN WITH B-CELL ACUTE LYMPHOBLASTIC LEUKEMIA

Daniel Martínez Anaya¹, Marian Valladares Coyotecatl¹, Ulises Juárez Figueroa¹, Rocío Juárez Velázquez¹ and Patricia Pérez Vera¹

¹National Pediatric Institute, Insurgentes Cuicuilco 3700-C 04530 Mexico City.
E-mail: pperezvera@yahoo.com Phone number:1084-0900 ext 1471.

Background. B-cell precursor Acute Lymphoblastic Leukemia (B-ALL) is the most common childhood malignancy among Mexican population. Over the last decade the next generation sequencing (NGS) analysis has revealed pathogenic variants (PV), clinically relevant for B-ALL management ¹. Nevertheless, a significant proportion of NGS variants are interpreted as uncertain significance (VUS) and a comprehensive analysis is necessary to clarify their role in B-ALL.

Objective. To predict the effect of VUS detected by NGS in children with B-ALL using bioinformatic algorithms and protein modeling analysis.

Methodology. DNA from bone marrow of 73 patients with B-ALL was sequenced using an NGS exome panel targeted to 205 cancer associated genes. The variants were classified using CancerVar platform. Variants were confirmed by Sanger sequencing. The pathogenicity was predicted using four different bioinformatic tools². For in silico mutagenesis analysis, we used the PDB crystal structures and PyMOL 4.6 software. To analyse the conservation of protein residues we used ClustalX2.1 program.

Results. We detected 1508 variants in 205 genes from 73 samples. The PV and VUS represent the 5% and 22% respectively; 21% of VUS were upgraded to potentially pathogenic (PP-VUS) by at least 3 pathogenicity predictors. The PP-VUS involve principally kinase signalling, epigenetic regulation, B-cell development and DNA repair genes. The structural analysis of JAK2p.L884P, JAK1p.P960L, FLT3p.E573G/p. Y589D and WHSC1p.E1099K revealed a steric hindrance, and loss of intramolecular interactions which possibly disturbs the stability of catalytic and regulatory domains. Additionally, the sequence alignment revealed a high grade of conservation (70-100%) among structurally related proteins of human and other species. Considering the relation between the 3D structure and biological function, we suggest that some of the PP-VUS should be scaled to in-vitro experimental analysis to obtain more insights about their functional consequences.

References

1. Martínez Anaya D et al (2022). Int J Mol Sci, (17), 9587.
2. Li MM et al. (2017). J Mol Diagn, (1), 4-23

VECTOR CAPACITY OF *Aedes aegypti* THROUGH THE HORIZONTAL INFECTION MECHANISM OF THE DENGUE LARVA-LARVA VIRUS

Mayra Mejía Sánchez¹, Rosalío Ramos-Payán², José Israel Torres Avendaño¹, Jorge Armando López Gutiérrez², Cuauhtémoc Villarreal Treviño³, Elsa Maribel Aguilar-Medina², Hipólito Castillo Ureta^{1*}

¹Faculty of Biology, Autonomous University of Sinaloa, Culiacán, Sinaloa, Mexico. ²Faculty of Chemical Biological Sciences, Autonomous University of Sinaloa, Culiacán, Sinaloa, Mexico. ³Regional Public Health Research Center, Tapachula de Córdova y Ordóñez, Chiapas, Mexico.

* Correspondence: hipolito.cu@uas.edu.mx

Introduction. Dengue is a disease classified as an emerging and re-emerging pathology, caused by the dengue virus (DENV), which is transmitted horizontally to humans through the bite of infected female mosquitoes. Additionally, there is evidence of vertical transmission of DENV from mother to child. Three modes of viral transmission among mosquitoes have been identified: sexual, transovarian and larva-larva viral excretion. These modes are thought to play a role in maintaining the virus in nature during periods of low viremia in humans. However, the implications of larva-larva transmission have a natural epidemiological model remain unclear, and its potential impact on public health has yet to be fully understood. **Objective.** To evaluate the mosquito-host transmission of the dengue virus through the horizontal larva-larva infection mechanism. **Materials and methods.** The virus was replicated in C3/36 cells, titration of the virus with the supernatant by plaque assays, exposure of feces with dengue viral particles for vector infection, completing the transmission cycle through Balb/c mice and confirmation of the infection using two different RT-PCR and indirect immunofluorescence tests. **Preliminary results.** When infecting the C6/36 cell line with DENV-2 viral replication supernatant, a change in the morphology of the infected cell was observed, demonstrating cytopathic damage at 72 hours post-infection, viral particles were observed in the cytoplasm of the cells, Likewise, its replication was demonstrated by seeing the replication curve and an expected 511 bp fragment in the agarose gel. **Conclusion.** C6/36 cells are highly permissive for dengue virus infection, so it was demonstrated that there is a highly infective viral stock for the continuation of the development of the research project.

UNVEILING THE PLASTIC-DEGRADING POTENTIAL OF EUKARYOTES: A COMPUTATIONAL APPROACH

Andrés Méndez-Zamora¹, Tania Díaz-Vidal¹, Marcela Robles-Machuca²,
Alejandro Pereira-Santana³, Leticia Casas-Godoy⁴

¹Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco (CIATEJ), Avenida Normalistas 800, Col. Colinas de la Normal, C.P. 44270 Guadalajara, Jalisco., ²Tecnología de alimentos, Secretaría de Investigación y Posgrado, Universidad Autónoma de Nayarit, Ciudad de la Cultura s/n, C.P. 63000, Tepic, Nayarit., ³CONAHCYT- CIATEJ, Parque Científico Tecnológico de Yucatán, 97302, Mérida, Yucatán., ⁴CONAHCYT- CIATEJ, Camino Arenero 1227, El Bajío, 45019, Zapopan, Jalisco.
E-mail: lcasas@ciatej.mx

Plastic pollution, particularly from polyethylene terephthalate (PET), poses a major environmental threat. Biodegradation of PET offers a promising solution, but the focus has primarily been on bacterial PETases [1,2]. This study explores the potential of eukaryotic PETases using *in silico* bioprospecting combining hidden Markov models (HMM), clustering techniques, molecular docking, and dynamic simulations, as well as experimental validation by HPLC.

We identified 424 putative PETase sequences from 219 eukaryotic organisms, across 13 classes, predominantly from the phylum Ascomycota. The diversity includes sequences from three kingdoms (Fungi, Metazoa, and Streptophyta), with Ascomycota representing the majority. The discovery of potential PETases in these kingdoms offers promise for enzyme diversity studies. Furthermore, these findings suggest untapped possibilities for plastic degradation within the Ascomycota phylum, hinting at unexplored sequences waiting to be discovered. On the other hand, within the 424 putative sequences, a 100% conserved motif was identified, including the putative catalytic serine residue, common in acyl hydrolases [3].

Further analysis highlighted 42 potential PETases with accessible binding sites, and molecular docking identified 6 promising candidates. The *Aspergillus luchuensis* sequence (*AluCut*) stood out with the lowest Gibbs free energy in docking and thermal stability in simulations. Experimental assays confirmed *Aspergillus luchuensis* PET-degrading activity. This integrated approach highlights the potential of eukaryotic organisms, particularly fungi, in PET degradation. This research expands the PETase universe and facilitates further exploration of eukaryotic microorganisms in bioremediation strategies. The findings underscore the importance of interdisciplinary approaches in enzyme discovery for sustainable plastic waste management.

References

1. Joo S, Cho IJ, Seo H, Son HF, Sagong HY, Shin TJ, (2018). Structural insight into molecular mechanism of poly(ethylene terephthalate) degradation. *Nature Communications*
2. Chow J, Pérez-García P, Dierkes RF, Zhang H, Streit WR (2023). The PET-Degrading Potential of Global Metagenomes: From In Silico Mining to Active Enzymes. In: Streit WR, Daniel R, editors. *Metagenomics: Methods and Protocols*. Springer US.
3. Ben Hlima H, Dammak M, Karray A, Drira M, Michaud P, Fendri I, (2021). Molecular and Structural Characterizations of Lipases from *Chlorella* by Functional Genomics. *Marine Drugs*.

ROLE OF THE PAS DOMAIN IN THE ACTIVITY OF DIGUANYLATE CYCLASE OR PHOSPHODIESTERASE OF THE CDGD PROTEIN IN *AZOSPIRILLUM BALDANIORUM* SP245

Dulce Andrea Montes Perez¹, María Luisa Xiqui Vázquez¹, Sandra Reyes Carmona¹, Beatriz Eugenia Baca¹ and Alberto Ramírez-Mata¹

¹Investigaciones en Ciencias Microbiológicas, Benemérita Universidad Autónoma de Puebla, Avenida San Claudio S/N, Puebla Pue, Mexico. C.P. 72570
E-mail: alberto.ramirez@correo.buap.mx Tel: 222 867 0044

Azospirillum baldaniorum Sp245, a plant growth-promoting rhizobacterium for its ability to fix nitrogen and produce phytohormones¹. A critical regulatory mechanism in *Azospirillum* spp. involves the second messenger cyclic di-GMP (c-di-GMP). This molecule plays a central role in diverse cellular processes, including biofilm formation, motility, and cell cycle regulation². Intracellular c-di-GMP levels are modulated by diguanylate cyclases (DGCs) and phosphodiesterases (PDEs). DGCs synthesize c-di-GMP, while PDEs degrade it. These enzymes are identified by the presence of characteristic motifs: GGDEF for DGCs and EAL for PDEs³. These domains enable bacteria to perceive environmental signals and transduce them into appropriate responses⁴. PAS (Per-ARNT-Sim) domains are particularly important sensory modules. They detect environmental stimuli such as light, redox states, and oxygen. Ligand binding induces conformational changes in PAS domains, which are then transmitted to other protein domains ultimately affecting protein function⁵. Structural studies reveal that PAS domains participate in the dimerization of DGCs and PDEs. A prime example is CdgD in *A. baldaniorum* Sp245. This protein possesses CHASE-PAS-DGC-EAL domains, suggesting a complex role in environmental signal perception and c-di-GMP metabolism⁶. Understanding signaling domains like PAS is crucial. These domains, upon receiving signals, modulate the catalytic activities of signaling cyclic nucleotide metabolizing enzymes (CMEs). While downstream c-di-GMP signaling components have been extensively studied, the upstream signals that CMEs respond to remain poorly understood. Relevant reviews are also scarce⁷. This research aims to elucidate the intricate regulatory networks within *A. baldaniorum* Sp245. Our approach involves structural modeling of PAS domain to identify amino acid residues involved in substrate binding. Based on this we will generate specific point mutations to precisely modify the protein. These studies will enhance our knowledge of *Azospirillum* signaling mechanisms and their implications for plant growth promotion⁸.

References

1. Mariotti, L., Scartazza, A., Curadi, M., Picciarelli, P., & Toffanin, A. (2021). *Plants*, 10(6), 1141.
2. Viruega-Góngora, V. I., & VIRUEGA GONGORA, V. I. (2023). CdgB es una proteína híbrida (GGDEF y EAL) localizada en la membrana citoplasmática de *Azospirillum baldaniorum* Sp245.
3. O'Neal, L., Ryu, M. H., Gomelsky, M., & Alexandre, G. (2017). *Journal of bacteriology*, 199(18), 10-1128.
4. Ramírez-Mata, A., López-Lara, L. I., Xiqui-Vázquez, M. L., Jijón-Moreno, S., Romero-Osorio, A., & Baca, B. E. (2016). *Research in microbiology*, 167(3), 190-201.
5. Stuffle, E. C., Johnson, M. S., & Watts, K. J. (2021). *Current opinion in microbiology*, 61, 8-15.
6. Mata, A. R., Pacheco, C. M., Cruz Pérez, J. F., Sáenz, M. M., & Baca, B. E. (2018). *BMC microbiology*, 18, 1-14.
7. Yu, Z., Zhang, W., Yang, H., Chou, S. H., Galperin, M. Y., & He, J. (2023). *FEMS Microbiology Reviews*, 47(4), fuad034.
8. Cruz-Pérez, J. F., Sáenz, M. M., & Baca, B. E. (2021). *BMC microbiology*, 18, 1-14

EXPLORING THE THERAPEUTIC POTENTIAL OF BERRY CACTUS IN RAT MICROBIOTA COMPOSITION WITH METABOLIC SYNDROME

Cesaré Ovando-Vázquez^{a,c}, Jorge Noé García-Chávez^a, Yadira Ramírez-Rodríguez^b, Victoria Ramírez^d, Claudia J. Bautista^e and Joyce Trujillo^{b,c}

^aCentro Nacional de Supercomputo, Instituto Potosino de Investigación Científica y Tecnológica (CNS-IPICYT), San Luis Potosí, 78216. México. ^bDivisión de Materiales Avanzados, Instituto Potosino de Investigación Científica y Tecnológica (DMA-IPICYT), San Luis Potosí, S.L.P. 78216. México. ^cConsejo Nacional de Humanidades, Ciencias y Tecnologías (CONAHCYT). ^dDepartamento de Cirugía Experimental. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán. Tlalpan, CDMX 14080. México.

^eDepartamento de Biología de la Reproducción. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán. Tlalpan, CDMX 14080. México. ^fDepartment of Biotechnology and Biochemistry, Cinvestav-Unidad Irapuato, Km. 9.6 Carr. Irapuato-León, 36824 Irapuato, Guanajuato, Mexico.

The complex condition known as metabolic syndrome (MetS) is impacted by both environmental and hereditary factors. Among other things, MetS is associated with obesity, dyslipidemia, hypertension, and hyperglycemia. The gut microbiota, a varied microbial population found in the gastrointestinal tract and linked to a number of metabolic illnesses, including MetS, has drawn a lot of interest lately. The berry cactus (*Myrtillocactus geometrizans*) is rich in betalains, sterols, pectins, and polyphenols. The metabolites of berry cacti exhibit anti-inflammatory, antiproliferative, hypolipemic, and hypoglycemic effects. This study used a rat model of MetS development fed with a high-fat diet to examine the effects of berry cactus fruit ingestion on gut microbial composition. Comprehensive metagenomic analyses of microbial 16S rRNA gene and metabolic indicators associated with MetS were obtained after 140 days of treatment.

There were established correlations between the metabolic parameters and the relevant microbial species, anticipated functions, and MetS-related pathways. According to our findings, the consumption of berry cactus juice reduced serum insulin, triglyceride, and glucose levels as well as the fat percentage of adipose, intestinal, and hepatic tissue. Moreover, therapy with berry cactus was linked to changes in the composition of the gut microbiota, favoring microbial phyla linked to metabolic health. In particular, a relationship with the *Parabacteroides* genus was found, indicating possible modes of action such as altering physiological pathways linked to the metabolism of fatty acids and increasing the availability of bioactive compounds from berry cacti.

These results shed light on the complex interactions between food, gut microbiota, and metabolic health, suggesting that berry cactus consumption may be a promising therapeutic strategy for people with MetS or at risk of developing it.

EVOLUTION OF HYPOXIA RESPONSE FACTORS IN THE PLANTAE KINGDOM

Janet Palacios-Martínez¹, Ana I. Chávez-Martínez¹, Leticia Vega-Alvarado¹ y Luis Cárdenas

¹Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, México. AP 510-3, Cuernavaca, Mor, 62250, México.
E-mail: luis.cardenas@ibt.unam.mx, janet.palacios@ibt.unam.mx

Plants are multicellular organisms that have colonized diverse environments. Among the most challenging habitats to colonize are those with limited oxygen availability. The relative oxygen concentration in multicellular organisms can integrate positional information, metabolic state and even environmental conditions. It has been described that oxygen is an essential molecule involved in cellular metabolism, which participates as a regulator of plant cell growth, differentiation and reproduction. In plants, for example, hypoxic niches exist with an essential role during development, but also in response to biotic and abiotic stress. Hypoxia is a condition, in which the availability of oxygen provides a protective environment, which facilitates quiescence and a reduced redox state that in turn promotes genomic stability. This is probably a constant in meristematic cell niches. However, when plants experience abiotic stress conditions such as flooding and waterlogging, a hypoxic condition is also imposed and this affects not only productivity but also crop quality. In this project we generated an *Open Big Data* repository of the transcriptional landscape of different tissues and organ stages with hypoxic niches such as the mycorrhizal nodule and arbuscule. We analyze the phylogenetic relationships of hypoxia response factors (HRF) in the *Plantae kingdom*. This analysis allows us to generate hypotheses about the evolution of the hypoxia response mechanism in model organisms. In our working group there is a great interest in understanding the cellular and molecular basis of both cell perception and reprogramming in response to hypoxia conditions in model plant organisms at the tissue and single cell level.

PREDICTION OF THE THREE-DIMENSIONAL STRUCTURE OF THE NRG1-RTG3 CHIMERIC COMPLEX

Dr. Edgar A. Ramírez González, Dr. Carlos Campero-Basaldua, Dra. Alicia Gonzalez-Manjarrez

Instituto de Fisiología Celular, UNAM.
Circuito Exterior s/n Ciudad Universitaria, Coyoacán, 04510 México D.F.
amanjarr@ifc.unam.mx

The regulatory role played by chimeric regulators has been described for Hap2-3-5-Gln3 and recently for Nrg1-Rtg3, this new type of regulation increases the repertoire of regulatory possibilities. Nrg1 acts by negatively regulating genes encoding enzymes involved in gluconeogenesis, those involved in the Krebs cycle, and those that metabolize carbon sources other than glucose. Rtg3 and Rtg1 form a transcription complex that activates genes of the retrograde signaling pathway between the mitochondria and the nucleus in response to mitochondrial dysfunction, resulting in the induction of antioxidant defenses and stress resistance. In previous work, the existence of a new chimeric regulator formed by Nrg1 and Rtg3 was demonstrated. Also, it was observed that increasing the concentration of alanine in the culture medium promoted the formation of the Nrg1-Rtg3 complex, indicating that alanine could act as a co-regulator. In this work, we predicted the tertiary structure of Nrg1, Rtg3, the promoter region recognized by Rtg3, and analyzed whether alanine interacted directly or indirectly in the structure. First, the I-TASSER server was used to create structural models of the Nrg1 and Rtg3 regulators, as well as the DNA region containing the promoter sequence recognized by Rtg3 in the ALT2 gene. These models were then validated with equilibrium molecular dynamics using the NAMD3 program. The stability of the complex was tested by RMSD which was measured throughout the 100 ns simulation. When the molecules were in equilibrium, a timestep of the Nrg1 and Rtg3 dynamics was selected for analysis with COFACTOR and COACH server to find possible ligand binding sites and predict possible functions of the regulators. Subsequently, a molecular docking between Nrg1, Rtg3, and the promoter was performed with these structures to analyze the stability of the chimeric complex, as well as the total energy of the molecule and the binding energy.

References

1. Hernández H, Aranda C, López G, Riego L, González A. Hap2-3-5-Gln3 determine transcriptional activation of GDH1 and ASN1 under repressive nitrogen conditions in the yeast *Saccharomyces cerevisiae*. *Microbiology*. 2011;157(Pt 3):879-889.
2. Campero-Basaldua C et al. 2023 Neo-functionalization in *Saccharomyces cerevisiae*: a novel Nrg1-Rtg3 chimeric transcriptional modulator is essential to maintain mitochondrial DNA integrity. *R. Soc. Open Sci.* 10:231209.

TITLE: PHAZ AS A POSSIBLE PET DEGRADER?

Graciela Jesed Reyes-Trujillo, Yovani Aguilar-Carrillo, Lucía Soto-Urzúa, Lino Javier Martínez-Soto and Luis Javier Martínez-Morales*

Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias.
Benemérita Universidad Autónoma de Puebla, Puebla, México.

C.P. 72592. Phone number: 222 185 51 06. *Corresponding author: luis.martinez@correo.buap.mx

Due to the imminent plastic pollution worldwide, alternatives offer promising solutions, such as bioremediation to accelerate the degradation of plastics, the generation of biodegradable polymers, and/or the use of enzymes that degrade plastics. Enzymes have been discovered capable of cleaving the ester bonds of the linear PET polymer, thus increasing plastic degradation and improving plastic recycling processes. Another way to reduce waste is to use the byproducts generated after enzymatic catalysis, giving rise to a circular economy in which these byproducts become raw materials. On the other hand, PHB is a linear biopolymer produced by some organisms and microorganisms, such as *Azospirillum baldaniorum* Sp245. The PHB polymer is degraded by the enzyme PHB-depolymerase (PhaZ), which cleaves the ester bonds. The sequences and 3D structures of PhaZs and some hydrolases that have been proposed and/or proven to have the capacity to degrade PET were obtained, and multiple alignments were generated in the Molecular Evolutionary Genetics Analysis (MEGA) and Clustal Omega software, where superfamilies were found. of Alpha Beta hydrolase, from the alignments, phylogenetic trees were constructed in the MEGA and MABL software, in which some of the PHB depolymerase can be seen in the same clade of the PET enzymes that have been verified. Molecular docking of these with the PHB and PET substrates were obtained. Through *in silico* analysis, a model is presented in which the enzyme could cleave the ester bond, and PhaZs are proposed as remediators at an industrial level.

Keywords: Enzymes, PhaZs, remediation.

MACHINE LEARNING PREDICTION OF BACTERIAL-PHAGE SPECIFICITY USING GENOMIC FINGERPRINTS OBTAINED BY VIRTUAL HYBRIDIZATION

Juan-Antonio Rodríguez de la Cruz¹, Hueman Jaimes Díaz¹, Alfonso Méndez Tenorio¹

¹Laboratory of Biotechnology and Genomic Bioinformatics, Department of Biochemistry, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, México City, Mexico
E-mail: juanantonio_2604@hotmail.es

The use of bacteriophages has been shown as a therapeutic alternative to face the crisis of antibiotic resistance. This type of virus shows a high specificity towards the bacteria it attacks, so an effective treatment must be carried out by a phage adapted to the infection strain. Characterizing the specificity of the phage in the laboratory is usually time-consuming, so a computational model that carries out this activity and thereby filters therapeutic candidates could allow not only earlier treatment but a higher success rate. The approach used in this work to develop this model is the use of Machine Learning (ML), a branch of Artificial Intelligence, which consists of a group of algorithms focused on tasks such as prediction and classification. However, the nature of the task involves the use of the genomes of the phages, which despite being small, they continue to be disorganized and unfiltered data that will make it difficult to train ML algorithms. As an option to overcome this problem, the frequency of virtual probes known as Virtual Genomic Fingerprints from the VAMPhyRE¹ program was used, which could be helpful as a method of dimensionality reduction, since in addition to exponentially reducing the size of the data to be analysed, produces tabular data ideal for the development of ML systems. 100 genomes of different bacteriophages specific to each of the following bacteria were used: *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*. Five different ML models were compared. As results, scores close to perfect performance in the ROC-AUC metric were obtained in all models, indicating outstanding performance in the detection of phage specificity. These scores were obtained even using only 10% of the data, which shows that virtual probes in conjunction with ML models allowed a great distillation of information for this task.

Reference

1. Tenorio, A. M., Cortés, P. F., Trejo, A. G., Díaz, H. J., Rosales, E. R., Rodríguez, A. M., ... & Kenneth, L. B. (2006). In silico evaluation of a novel DNA chip based fingerprinting technology for viral identification. *Revista latinoamericana de microbiología*, 48(2), 56-65.

ANALYSIS OF THE INTERACTION BETWEEN TMPRSS2, ACE2, AND FURIN AGAINST SPIKE OF SARS-COV-2

Víctor Rodríguez-Escobar¹, Carlos A. Matias Cervantes¹, María T. Hernández-Huerta¹,
Eduardo L. Pérez-Campos², Carlos Romero-Díaz²

¹CONAHCYT – Faculty of Medicine and Surgery, Autonomous University “Benito Juárez” of Oaxaca. Oaxaca, Mexico.

²Division of Graduate Studies and Research, National Technology of Mexico/IT Oaxaca. Oaxaca, Mexico.

E-mail: rodesc.med@gmail.com.

Introduction. The mechanism of infection of the coronavirus type 2 causing severe acute respiratory syndrome (SARS-CoV-2) is based on the binding of the receptor-binding domain (RBD) of the Spike glycoprotein to the angiotensin-converting enzyme 2 (ACE2) receptor¹. To trigger the fusion of the viral and cellular membranes, Spike requires proteolytic activation by the transmembrane serine protease type 2 (TMPRSS2) and furin (FUR)¹. The interactions between TMPRSS2, ACE2, and FUR with the Spike protein have been described by molecular docking². However, there are no specific reports of interactions between RBD and TMPRSS2. The objective of this work was to determine probable interaction sites in the TMPRSS2-RBD complex that suggest biological activity.

Methodology. The crystallized models of TMPRSS2, FUR, and Spike RBD were obtained from the Protein Data Bank (PDB) database. The complete Spike model was obtained by molecular modeling on the CHARMM-GULORG platform. The membrane-associated ACE2 model was modeled using AlphaFold, subjected to 20 nanoseconds of molecular dynamics with AMBER20 for its stabilization. Energy was deputed and minimized using UCFC Chimera 1.16. The spike-ACE2, Spike-TMPRSS2, Spike-furin, ACE2-TMPRSS2, and TMPRSS2-RBD complexes were docked using Hex 8.0.0+CUDA. The interactions between amino acids in the complexes were identified using UCFC Chimera 1.16 and compared with previously reported interactions^{2,3,4,5}.

Results and Discussion. Intermolecular interactions were obtained in all the complexes subjected to docking, mainly on the S1/S2 and S2 cleavage sites and RBD of Spike and the catalytic sites of the proteases. Similarities were identified with the reported interactions^{4,5} and the differences are attributed to the methods used to obtain the structural models and the software used for the analysis of the complexes.

Conclusion. The TMPRSS2-RBD complex showed intermolecular interactions mainly composed of positively charged amino acids in the receptor chain, giving it the ability to form ionic bonds and hydrogen bonds, which suggests biological activity over the RBD domain. The interaction between TMPRSS2-RBD in the SARS-CoV-2 infection mechanism has not yet been explored.

References

1. Takeda, M. (2022). Proteolytic activation of SARS-CoV-2 spike protein. *Microbiology and Immunology*, 66(1), 15–23.
2. Lan, J. Et al (2020a). Structure of the SARS-CoV-2 spike receptor-binding domain bound to the ACE2 receptor. *Nature*, 581.
3. Senapati, S., Et al. (2021). Contributions of human ACE2 and TMPRSS2 in determining host-pathogen interaction of COVID-19. *Journal of Genetics*, 100(1).
4. Vankadari, N. (2020). Structure of Furin Protease Binding to SARS-CoV-2 Spike Glycoprotein and Implications for Potential Targets and Virulence. *The Journal of Physical Chemistry Letters*, 11(16), 6655–6663.
5. Stevens, B. R. (2020). TMPRSS2 and ADAM17 interactions with ACE2 complexed with SARS-CoV-2 and BOAT1 putatively in intestine, cardiomyocytes, and kidney. *BioRxiv*, 2020.10.31.363473.

UNRAVELING THE DISTINCT BIASES OF THE GENOMIC LANDSCAPE OF LUNG ADENOCARCINOMA FROM MEXICAN PATIENTS

Bertha Rueda Zarazúa^{1,4}, Humberto Gutiérrez¹, Humberto García Ortiz¹, Lorena Orozco¹, Joaquín Zúñiga², Federico Ávila Moreno³, Jorge Meléndez Zajgla¹

¹Instituto Nacional de Medicina Genómica. ²Instituto Nacional de Enfermedades Respiratorias.

³FES-Iztacala, UNAM. ⁴Posgrado en Ciencias Biológicas, UNAM.

Periférico Sur 4809, Arenal Tepepan, Tlalpan, 14610, CDMX.

(55) 5350 1900 ext. 1901 y 1902

jmelendez@inmegen.gob.mx

Lung cancer continues to exhibit the highest mortality rates worldwide. Recent studies suggest that there are differences in carcinogenic processes between populations, which may be associated to specific risk factors, socioeconomic aspects or genetics. Nevertheless, the majority of studies are based in populations with European ancestry and there is a need for further research on under-represented populations. This study analyzed whole exome sequencing data from 25 Mexican lung adenocarcinoma patients using a cohort mostly of European ancestry (TCGA-Pancancer) as a reference cohort. We propose a new strategy for limited cohort studies using a gene population-based approach to extract relevant information from genomic data.

36% of the patients carry a Pathogenic or Likely-Pathogenic germline variant with the SERPINA1 gene being the most frequently affected gene (12%). The mutational signatures: SBS32, SBS85, SBS12, SBS19 were present at significantly higher proportions in our cohort compared to the European cohort (Bonferroni-adj $P < 0.0001$). Interestingly, the smoking-associated signature SBS4 was absent in smoking Mexican patients ($p < 0.01656$). Among the genes not shared with the reference cohort, and with the highest potential for positive selection, there is a significant enrichment of protein-protein interactions, suggesting an effective functional association among these genes. Frequencies of somatic variants in actionable genes like EGFR (24%) and KRAS (16%) were comparable to other Mexican studies. However, SLC36A4 (20%, $p=0$), AP1S1 (8%, $p=0$) and TP53 (16%, $p=0.00005$) showed significant differences with the reference cohort. Genes associated with the necroptosis pathway were overrepresented among potential positive selection targets in our cohort. This study highlights distinct genomic biases in the genomic landscape of lung adenocarcinoma in this Mexican cohort when compared with a cohort of European ancestry, suggesting the need of targeted precision medicine strategies in Latin American populations.

STRUCTURAL ANALYSIS OF SARS-COV-2 MPRO PROTEASE VARIANTS FOR THE DISCOVERY OF BROAD-SPECTRUM INHIBITORY DRUGS USING *IN SILICO* TOOLS

Carlos Alfonso Sánchez-López¹, Varinia López-Ramírez¹, Divanery Rodríguez-Gómez¹
& Juan Vázquez-Martínez^{2*}

¹Departamento de Ingeniería Bioquímica, Tecnológico Nacional de México / ITS Irapuato, Carretera Irapuato Silao Km. 12.5, C.P.:36821 Irapuato, Guanajuato, México. ²Departamento de Ingeniería Química, Tecnológico Nacional de México / ITS Irapuato, Carretera Irapuato Silao Km. 12.5, C.P.:36821 Irapuato, Guanajuato, México. Tel. (+52) 462-152-22-73

*E-mail: juan.v.m@irapuato.tecnm.mx

Since the WHO declared the COVID-19 pandemic caused by the SARS-CoV-2 virus on March 11, 2020, more than 77 million cases have been recorded worldwide, with over 6 million resulting in deaths. SARS-CoV-2 has constantly mutated, giving rise to new variants with distinct characteristics such as partial or total resistance to certain medications, and increased transmissibility. The main protease (Mpro), which cleaves polyproteins into functional proteins for the formation of new viral particles, has not been exempt from mutations in the new variants. Therefore, the objective of this work was to generate a consensus sequence from the Mpro variants to identify broad-spectrum drugs that inhibit the development of new Coronavirus variants. The work was divided into three stages: 1) the development of the consensus sequence using EMBOSS and the subsequent prediction of the tertiary structure with AlphaFold2, 2) molecular docking experiments using the AutoDock Vina software, and 3) molecular dynamics experiments using the GROMACS software. The results showed that the Mpro active site is conserved in the consensus sequence obtained from 150 SARS-CoV-2 variants with mutations in the ORF1a gene according to the PANGO lineage. Additionally, the results of molecular docking and molecular dynamics experiments allowed to identify four potential broad-spectrum inhibitors of Mpro, against current and future coronavirus variants.

References

1. Gutierrez-Villagomez, J. M., Campos-García, T., Molina Torres, J., López, M. G., & Vázquez-Martínez, J. (2020). Alkamides and Piperamides as Potential Antivirals Against the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). *The Journal of Physical Chemistry Letters*. doi:10.1021/acs.jpcclett.0c01685
2. Hu, Y.; Lewandowski, E. M.; Tan, H.; Zhang, X.; Morgan, R. T.; Zhang, X.; Jacobs, L. M. C.; Butler, S. G.; Gongora, M. V.; Choy, J.; Deng, X.; Chen, Y.; Wang, J. (2023). Naturally Occurring Mutations of SARS-CoV-2 Main Protease Confer Drug Resistance to Nirmatrelvir. *ACS Cent. Sci.* 9 (8), 1658–1669, DOI: 10.1021/acscentsci.3c00538

AI REVOLUTIONIZING WATER QUALITY: HARNESSING MICROALGAE AND BACTERIA FOR SUSTAINABLE REMEDIATION

Mishael Sánchez-Pérez^{1,2,3}, Jose Juan Torres Martinez^{3,4}, Cesaré Ovando-Vázquez¹,
Ruth Elena Soria Guerra³, J.L. Morán-López^{1,2}

¹Grupo de Ciencia e Ingeniería Computacionales, Centro Nacional de Supercómputo. ²División de Materiales Avanzados, IPICYT Camino a la Presa San José 2055. Col. Lomas 4 sección CP. 78216. San Luis Potosí S.L.P. ³Laboratorio de Biotecnología de Células Vegetales y Bioinformática, Facultad de Ciencias Químicas, Universidad Autónoma de San Luis Potosí, Av. Dr. Manuel Nava 6, 78210 San Luis Potosí, Mexico. ⁴Laboratorio de Biotecnología y Bioenergía, Facultad de Ciencias Biológicas, Universidad Autónoma de Coahuila.
E-mail: Mishael.sanchez@ipicyt.edu.mx

Microalgae are unicellular organisms found in mostly all aquatic environments that show and interesting behavior on cleaning up water pollution. They can absorb carbon dioxide (CO₂) and inorganic nutrients from water. Some types of microalgae species like *Chlorella* and *Scenedesmus* also have the function of accumulating heavy metals and organic pollutants.

Bacteria also play a critical role in cleaning water pollution by breaking down organic compounds and reducing inorganic contaminants.

The interaction of microalgae and bacteria present a combined strength to sustainable water management and environmental restoration, generating cleaner and healthier water resources in the future.

The use of next-generation sequencing technologies helps to capture the metabolic functions of certain biological phenomena, but the treatment of massive data poses a challenge. Metatranscriptomics datasets consist of a vast amount of transcript expression patterns that provide clues to biological behavior.

The use of machine learning algorithms in biological data has been instrumental in predicting gene functions, identifying regulatory elements, and detecting patterns in DNA, RNA, and protein sequences. The conjunction of all these techniques helps elucidate the mechanisms that could boost the reduction of contaminants and reduce the cost of water bioremediation.

IN SILICO ANALYSIS OF THE BINDING OF THE OPPA PROTEIN OF *YERSINIA PSEUDOTUBERCULOSIS* WITH DIFFERENT LIPASES

Andrea Sánchez-Ríos, Graciela Jesed Reyes-Trujillo, Lucía Soto-Urzúa,
Lino Javier Martínez-Soto and Luis Javier Martínez-Morales*

Centro de Investigaciones en Ciencias Microbiológicas, Instituto de Ciencias.
Benemérita Universidad Autónoma de Puebla, Puebla, México. C.P. 72570.
Phone number: 222 185 51 06. *Corresponding author: luis.martinez@correo.buap.mx

There are 17 species of the genus *Yersinia*, of which only three are pathogenic for humans and animals (*Y. pestis*, *Y. enterocolitica*, and *Y. pseudotuberculosis*); they are Gram-negative zoonotic bacteria that cause gastrointestinal infections in animals and humans, except for *Y. pestis*. They present the type III secretion system encoded in the pYV plasmid. This system is a key component for pathogenicity in the *Yersinia* genus because it facilitates the injection of proteins, increasing their virulence in host cells and causing inhibition of the immune response.

The periplasmic oligopeptide binding protein (OppA) of *Y. pseudotuberculosis* is part of the oligopeptide transport system (Opp). However, it has been shown to have chaperone-type activity, favoring the folding of denatured proteins. This indicates that there are proteins with a double function. This multifunctional capacity of OppA could have implications for the survival of *Yersinia* under different conditions.

Protein sequence analyses were carried out in the InterPro database of lipases from organisms and microorganisms; these lipases shared the Alpha/Beta hydrolase superfamilies. Subsequently, phylogenetic analyses were carried out with the different lipases and the OppA protein in the Molecular Evolutionary Genetics Analysis (MEGA) and Méthodes with algorithms pour la Bio-informatique (MABL) software, where *Y. pseudotuberculosis* was found in the same clade of some fungus. Finally, molecular couplings were obtained, where the interaction between some lipases and the OppA protein was observed. These analyses contribute to the knowledge of virulence and pathogenicity between host cells and *Y. pseudotuberculosis* as well as in similar pathogens, in addition to providing information to develop prevention strategies in gastrointestinal diseases or even be used to develop therapeutic approaches.

Keywords: *Yersinia pseudotuberculosis*, OppA, lipases, docking.

DIFFERENTIAL MOLECULAR INTERACTIONS BETWEEN IBERIOTOXIN AND HUMAN SLO3 AND SLO1 POTASSIUM CHANNELS

Jorge Arturo Torres Juárez¹, Ana Gabriela Hernández Puga¹ and Ana Alicia Sánchez Tusie¹

¹Faculty of Medicine, Autonomous University of Querétaro, Clavel 200 Querétaro, Qro.

E-mail: ana.sanchez@uaq.mx

The steady increase in human population and the accompanying high unintended pregnancy rates pose a pressing threat to public health. As such, novel male contraceptive methods are urgently needed. To satisfy this, there are validated molecular targets such as the SLO3 channel and computational methods that can facilitate structural insights for drug development. Nonetheless, with the ubiquitous expression of SLO1, paralogue of SLO3, potential inhibitors must have a higher preference to SLO3 if they are to reach market. Here, we evaluated the differential molecular interactions between the human SLO3 and SLO1 channels and iberiotoxin, a toxin that selectively blocks SLO channels. For this, molecular docking and dynamics were implemented on the channel-toxin complexes to help elucidate atomistic details of their interaction and binding energy. We saw that iberiotoxin has a similar binding energy to both channels but interacts in a distinct manner with both channels. Particularly, Ser8 and Arg25 of iberiotoxin diverges in their interaction with the residues Val283 and Asn260 of SLO3 and the corresponding residues Tyr359 and Ala336 of SLO1. Together, our results suggests that SLO1 possess additional supports for iberiotoxin coupling. Knowledge of the key residues in the molecular interface of iberiotoxin blockage identified can help guide and hasten non-hormonal contraceptive development.

MICROBIAL DIVERSITY ANALYSIS VIA METAGENOMICS IN 'EL IZTETE' PUBLIC LANDFILL, TEPIC NAYARIT

Jesús Urbar Ulloa, María Marcela Robles Machuca, Alejandro Pereira-Santana,
Pedro Javier Escobar-Turriza, Leticia Casas-Godoy*

Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco. Camino Arenero 1227, El Bajío, 45019,
*lcasas@ciatej.mx, +52 (33) 334-55200 Ext. 2017.

The sanitary landfill known as 'El Iztete,' located in Tepic, Nayarit, was originally intended for the disposal of various types of waste compacted in layers and covered with soil to protect the environment. However, over time, it has evolved into an open dump, accumulating more than 135,000 tons of waste annually. This situation has resulted in the exposure of natural resources and local populations to significant pollution.

As is the case in other ecosystems, microorganisms within the landfill exploit all available carbon sources to survive. This process is not an exception in 'El Iztete,' leading to the generation of a unique microbial diversity which represents an untapped resource that could be used for innovations in biotechnological fields.

This research work details the processes of soil sample collection, genomic DNA extraction, sequencing, genome assembly and reconstruction, and data analysis of microbial genomes found in the 'El Iztete' sanitary landfill. These findings are valuable for analyzing microbial diversity and identifying microorganisms relevant for bioremediation and other biotechnological applications.

UNLOCKING NITROGEN'S ROLE DURING SOMATIC EMBRYOGENESIS INDUCTION OF *C. CANEPHORA*: A TRANSCRIPTOMIC VIEW

Brigitte V. Vargas-Morales¹, Rosa María Galaz-Avalos², Hugo Alejandro Méndez Hernández³, Ana O. Quintana-Escobar⁴ and Víctor M. Loyola-Vargas^{5*}

¹⁻⁵Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, Calle 43, 130, Colonia Chuburná de Hidalgo, Mérida, Yucatán, C.P. 97200.

*E-mail: vmloyola@cicy.mx; Phone: (52) 999 942 83 30, Ext. 243

Somatic embryogenesis (SE) is a process in which embryos are formed from somatic cells, bypassing the need for gamete fusion to regenerate plants. This process can be obtained *in vitro* from vegetative tissue explants, serving as a tool for massive plant propagation and a model to study embryo formation and development in *Coffea canephora*. Acquiring cell embryogenic capacity involves a complex signaling network and reprogramming gene expression patterns usually triggered by external stimuli, ranging from stress conditions to plant growth regulation factors¹. Inorganic nitrogen (N) and its concentration in the culture medium affect the response of explants to the induction of SE, a process in which auxin (AUX) and cytokinin (CK) are critical to enable somatic embryo formation in several species^{2,3}. Some studies have suggested a crosstalk between plant growth regulators and N in plants⁴. Therefore, understanding the biochemical and molecular mechanisms by which the nitrogen source participates in SE induction is crucial for manipulating and improving the process. This work employs RNA-seq, a powerful tool commonly used to identify global gene expression changes at specific times and conditions to understand how these changes participate in development⁵. We will harness the utility of RNA-seq to study changes in the transcriptome profile of SE induction in *C. canephora*, particularly when the ratio of nitrate and ammonium in the medium is modified.

References

1. Méndez-Hernández, H. A., Ledezma-Rodríguez, M., Avilez-Montalvo, R. N., Juárez-Gómez, Y. L., Skeete, A., Avilez-Montalvo, J., & Loyola-Vargas, V. M. (2019). *Frontiers in Plant Science*, 10, 77.
2. Duarte-Aké, F., Márquez-López, R. E., Monroy-González, Z., Borbolla-Pérez, V., & Loyola-Vargas, V. M. (2022). *Planta*, 256(6), 113.
3. Avilez-Montalvo, J. R., Quintana-Escobar, A. O., Méndez-Hernández, H. A., Aguilar-Hernández, V., Brito-Argáez, L., Galaz-Avalos, R. M., ... & Loyola-Vargas, V. M. (2022). *Plants*, 11(15), 2013.
4. Xing, J., Cao, X., Zhang, M., Wei, X., Zhang, J., & Wan, X. (2023). *Plant Biotechnology Journal*, 21(7), 1320-1342.
5. Quintana-Escobar, A. O., Nic-Can, G. I., Avalos, R. M. G., Loyola-Vargas, V. M., & Gongora-Castillo, E. (2019). *PeerJ*, 7, e7752.

COMPARATIVE GENOMIC ANALYSIS OF CUTICULAR BIOSYNTHESIS IN FLESHY FRUITS

José A. Valenzuela-Avilés^{1*}, Carmen A. Contreras-Vergara¹; Abraham Cruz-Mendivil²;
Carlos Calderón-Vázquez³, Martín E. Tiznado-Hernández¹, Miguel Á. Hernández-Oñate^{1**}

¹Centro de Investigación en Alimentación y Desarrollo, A.C. Carretera Gustavo Enrique Astiazarán Rosas No. 46, Hermosillo, Son. CP. 83304. ²Instituto Politécnico Nacional, Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional. Bulevar Juan de Dios Bátiz Paredes #250, Col. San Joachin, Guasave, Sin. CP. 81049.
E-mail: *jvalenzuela222_79@estudiantes.ciad.mx, **miguel.hernandez@ciad.mx

The cuticle is a lipidic layer synthesized by epidermal cells that cover aerial organs in land plants. It performs essential functions including transpiration regulation, gaseous exchange, protection against UV radiation and mechanical damage¹. In fleshy fruits it is involved during ontogeny and postharvest life, in the latter being involved in softening and pathogen resistance. Cuticle biosynthesis is conserved in plants; however, its composition and structure vary between species and organs, and are influenced by environmental conditions², suggesting a specific and complex regulation for each species. In this sense, information about it in fleshy fruits remains incomplete. Therefore, this study aims to identify the homologous genes involved in the cuticular biosynthesis in several fleshy fruits and model plants to identify changes in gene family evolution, gene structure and regulatory elements.

A wide database search was performed to gather genomic information, we assessed its quality using BUSCO and QAST. The orthologous genes were identified with Orthofinder and InterPro was used to establish the conserved domains per orthogroup. We generated a phylogenetic tree using MAFFT and FastTree to assess the evolution of the cuticular gene families using CAFE5. We identified the orthologues related to cuticular biosynthesis and its conserved domains in the genome of 40 species of fleshy fruits. We also found that certain orthogroups involved in cuticular biosynthesis have evolved at a faster rate, suggesting these genes could give these fruits an evolutionary advantage to response to biotic and abiotic stress. These candidate genes could serve as a guide for enhancing fleshy fruit traits and developing varieties. Furthermore, they could improve our understanding of plant epidermis formation mechanisms and offer fresh perspectives on research into plant cuticle formation.

References

1. Liu L, Wang X, Chang C. Toward a smart skin: Harnessing cuticle biosynthesis for crop adaptation to drought, salinity, temperature, and ultraviolet stress. *Frontiers in Plant Science: Frontiers Media S.A.*; 2022.
2. Kong L, Liu Y, Zhi P, Wang X, Xu B, Gong Z, Chang C. Origins and evolution of cuticle biosynthetic machinery in land plants. *Plant Physiology*. 2020;184(4):1998-2010.

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex of atoms, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

TOXICOLOGY & PHARMACOLOGY

VIAN-C4551 IN EYE DROPS REACHES THE RETINA IN SUPRATHERAPEUTIC CONCENTRATIONS AND REVERSES DIABETES-INDUCED EXCESSIVE VASOPERMEABILITY IN THE RETINA OF RODENTS

Elva Adán-Castro¹, Daniela Granados Carrasco¹, Juan Pablo Robles^{1,2}, Magdalena Zamora¹, Lourdes Siqueiros-Marquez¹, Jose Fernando García-Rodrigo¹, Gonzalo Martínez de la Escalera¹, Carmen Clapp*

¹Instituto de Neurobiología, Universidad Nacional Autónoma de México, Querétaro, Querétaro, México.

²VIAN Therapeutics, CA, United States.

*clapp@unam.mx, Campus UNAM 3001, 76230 Juriquilla, Qro., (442) 238 10 28

Introduction. Topical ophthalmic instillation for the treatment of diabetic macular edema (DME) and diabetic retinopathy (DR) would provide a non-invasive alternative to the current antiangiogenic therapy involving physician administered intravitreal injections. However, reaching the retina after eye drops is challenging due to numerous ocular barriers and there is no eye drop formulation approved for the treatment of DME and DR. VIAN-c4551 is a highly potent and stable antiangiogenic cyclic peptide. We aimed to investigate the access of VIANc-4551 in eye drops to the retina and vitreous and its effect against diabetes-induced retinal vascular leakage.

Methods. Ocular pharmacokinetics (PK) of VIAN-c4551 in eye drops was measured in rats and rabbits; and the effect of eye drops containing VIAN-c4551 was tested in the rodent DR model induced by streptozotocin.

Results. After topical ocular instillation, VIAN-c4551 reached μM and nM concentrations in the retina-choroid and vitreous respectively, substantially exceeding its pM potency to inhibit endothelial cell permeability. The concentration of VIAN-c4551 peaked at 6 hours and was maintained for 24 hours. The PK profile was confirmed using VIAN-c4551-FITC in eye drops. A single eye drop containing 0.5% VIAN-c4551, delivered daily for 5 days, reversed the increase in retinal vascular leakage due to diabetes in rats and mice.

Conclusions. VIAN-c4551 in eye drops reaches the back of the eye at supratherapeutic concentrations providing a non-invasive, once-a-day potential intervention for reversing retinal vascular leakage in DME, DR, and other vascular retinopathies.

Supported by Secretaría de Educación, Ciencia, Tecnología e Innovación de la Ciudad de México (SECTEI-061-2023).

ANTI-*HELICOBACTER PYLORI* ACTIVITY, CYTOTOXICITY AND *IN VIVO* GASTROPROTECTIVE EFFECT OF DIACETYLCURCUMIN AND ITS METAL DERIVATIVES

Almanelly Agabo-Martínez¹, Erika Gomez-Chang¹, William Meza-Morales², Marco A. Obregón-Mendoza², Yair Álvarez-Ricardo², Raúl G. Enríquez², Irma Romero¹

¹Depto. de Bioquímica, Facultad de Medicina, ²Instituto de Química, Universidad Nacional Autónoma de México, Cd. Universitaria, 04510 Coyoacán, CDMX, México
E-mail: irma@bq.unam.mx

Introduction. *Helicobacter pylori* bacterium is the main risk factor for the development of gastritis, peptic ulcer, adenocarcinoma, and MALT lymphoma. Eradication treatments combine the administration of antibiotics with an acid secretor inhibitor; however, these therapies fail mainly due to antibiotic resistance. In the search for new drugs to control *H. pylori* infection, compounds have been isolated from medicinal plants. Curcumin, obtained from *Curcuma longa*, showed anti-*H. pylori* and gastroprotective activities^{1,2}. However, its therapeutic use is limited due to its low bioavailability. To improve curcumin pharmacokinetic profile, diacetylcurcumin (DAC) and four metal complexes were synthesized from it³. In this work the anti-*H. pylori* activity, the cytotoxic effect on gastric adenocarcinoma cells (AGS) and the gastroprotective effect of DAC and its 4 metal derivatives were investigated.

Methods. Antibacterial activity was determined by the broth dilution method according to CLSI. Cytotoxicity was evaluated by MTT cell viability assay. The gastroprotective effect of the compounds was assessed by an acute ethanol induced ulcer model in mice.

Results. *Antibacterial activity:* DAC₂-Cu and DAC₂-Mn were the compounds that best inhibited the growth of *H. pylori* (MIC= ~15 µM), followed by DAC₂-Mg, DAC₂-Zn and DAC. *Cytotoxicity:* All compounds have cytotoxic effects on AGS cells, DAC₂-Zn and DAC₂-Mg had the lowest IC₅₀ (15 µM at 48 h). *Gastroprotection:* The best antiulcer effect was obtained with DAC₂-Cu, and DAC₂-Zn at a dose of 200 mg/kg (~70% gastroprotection).

Conclusion. These results will contribute to the knowledge of the biological activities of these compounds and their possible use in *H. pylori* eradication therapies.

Acknowledgements. To DGAPA PAPIIT, UNAM-IN207622, CONAHCyT, FOINS-PRONACES 307152. AAM is a PhD student of the PDCB, UNAM and is a fellow from CONAHCyT-1246957

References

1. Morsy, M. A., y El-Moselhy, M. A. (2013). *Pharmacology*, 91, 5-6.
2. Gupta, S. C., et al. (2013). *The AAPS journal*, 15, 195–218.
3. Meza-Morales, W., et al. (2019). *Molecules*. 24, 1598

EFFECT OF PIOGLITAZONE ON KIDNEY DAMAGE AFTER EXPOSURE TO CADMIUM IN WISTAR RAT

Daniel Isaac Alcántara Jara¹, Víctor Enrique Sarmiento-Ortega¹, Samuel Treviño Mora¹

Institute of Physiology. Meritorious Autonomous University of Puebla.

Prol. de la 14 Sur 6301, Cd Universitaria, Cdad. Universitaria, 72592 Heroica Puebla de Zaragoza, Pue.

Correo: Samuel_trevino@hotmail.com; Telephone number: 222 229 5500

Cadmium is a toxic agent with a minimal presence in the human body, but with a great predominance in our environment, because we are exposed to the metal in various daily activities. The kidney is one of the main organs affected by chronic exposure to cadmium. Cadmium nephrotoxicity has been reported after chronic ingestion of the metal and this may be associated with metal-induced metabolic dysfunction. Many animal studies have shown that the use of Pioglitazone, a PPAR γ agonist, could potentially reduce the risk of diabetes-induced nephropathy. Therefore, the objective of this work was to evaluate the effect of pioglitazone on the improvement of kidney damage due to cadmium exposure. Wistar rats were used exposed to cadmium at a concentration of 15 ppm for 3 and 5 months, to which a dose of pioglitazone of 2 mg/kg body weight per day was administered for 30 days. After treatment, weight and height and blood and urine samples were evaluated to evaluate markers of kidney damage (Albumin, Urea, Creatinine, Creatinine clearance, and electrolytes). The kidneys were removed for histological evaluation of renal morphology using H&E, and renal fibrosis by red 80 staining. Finally, the concentration of IL-18, IL-6, IL1 β , and TFG- β was evaluated, both in serum and urine by Elisa techniques. The results showed that exposure to cadmium generates kidney damage with elevation of urea, and creatinine, decreases creatinine clearance, and increases serum sodium retention, widening of the glomerulus occurs, and the architecture of the tubules is lost, coupled with an increase in collagen deposition. For its part, the use of pioglitazone improves markers of renal damage in serum and urine, increases creatinine clearance and improves renal cytoarchitecture and reduces renal fibrosis. Finally, the concentration of IL-18 decreases. With these results, we show that treatment with pioglitazone presents good results in treating nephropathy generated by exposure to cadmium.

ANTITUMOR ACTIVITY OF INCOMPTINE A IN A MURINE BREAST CANCER MODEL BY DOWNREGULATION OF HEXOKINASE II

Angel Giovanni Arietta García^{1,2}, Fernando Calzada Bermejo³, Francisco Javier Alarcón Aguilar⁴,
Elihú Bautista⁵, Rosa María Ordoñez Razo¹

¹Posgrado en Biol. Experimental, DCBS, Universidad Autónoma Metropolitana, Iztapalapa (UAMI). México CDMX, México, ²Unidad de Investigación Médica (UIM) en Genética Humana, UMAE Hospital de Pediatría 2° Piso, Centro Médico Nacional Siglo XXI (CMN-SXXI), CDMX, México, ³UIM en Farmacología, UMAE Hospital de Especialidades, 2° Piso CORSE, CMN-SXXI, CDMX México, ⁴Lab. de Farmacología, Dpto. Ciencias de la Salud, DCBS, UAM-I, CDMX, México, ⁵CONAHCYT—Consortio de Investigación, Innovación y Desarrollo para las Zonas Áridas, Instituto Potosino de Investigación Científica y Tecnológica A.C., San Luis Potosí CP 78216, México.
E-mail: angelo_arietta@hotmail.com

Background. Breast cancer (BC) is the most common and lethal cancer in women worldwide.¹ In breast cancer there are various adaptations that allow its proliferation and survival. One of them is the overexpression of enzymes, as the hexokinase II (HKII) which, in addition to participating in metabolism, it can inhibit apoptosis.² Incomptine A (IA) is a sesquiterpene lactone isolated of *Decachaeta incompta*, which has recently demonstrated cytotoxic activity in different breast cancer cell lines due HKII downregulation.³ However, the antitumor activity of IA has not been tested in an *in vivo* model. Therefore, the aim of this study was to evaluate the antitumor activity of IA in a murine model of breast cancer and propose a mechanism of action that involves HKII. **Methods.** 4T1 murine breast cancer cells were inoculated subcutaneously into the abdominal mammary gland area at a concentration of 1×10^5 cells/mouse in 100 μ L. After 7 days of inoculation, animals with palpable tumor were selected and the antitumor activity of IA at different doses was evaluated. Tumors from treated mice were compared with untreated mice. To evaluate the effect of IA on HKII, western blot studies were performed. **Results.** We demonstrated that IA-treatment decreases the tumor growth in female BALB/c mice inoculated with 4T1 breast cancer cells. Additionally, the expression of HKII was decreased in IA-treated tumors.

Conclusion. These results suggest that IA possesses antitumor activity against breast cancer cells by downregulation of HKII and could potentially be used as a new anticancer agent for the treatment of breast cancer.

References

1. Sung, H.; Ferlay, J.; Siegel, R. L.; Laversanne, M.; Soerjomataram, I.; Jemal, A.; Bray, F. (2020) Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. *CA Cancer J Clin*, 71.
2. Schoeniger, A.; Wolf, P.; Edlich, F. (2022). How Do Hexokinases Inhibit Receptor-Mediated Apoptosis? *Biology (Basel)*, 11 (3).
3. Arietta-Garcia, A. G.; Calzada, F.; Ramirez-Sanchez, I.; Bautista, E.; Garcia-Hernandez, N.; Ordonez-Razo, R. M. (2023). Advances in the Properties of Incomptine A: Cytotoxic Activity and Downregulation of Hexokinase II in Breast Cancer Cell Lines. *Int J Mol Sci*, 24 (15).

THE COMBINED THERAPY OF DICLOFENAC-ITRACONAZOLE REDUCES THE SIZE OF *MADURELLA MYCETOMATIS* GRAINS AND STIMULATES CYTOKINES OF THE TH1 AND TH17 PROFILE

Iván Alejandro Banda-Flores¹, José David Torres-Tirado¹, Gabriela Pérez-Flores¹,
Mónica Jasso-Romo¹, Sarai C. Rodríguez-Reyes², Luis Antonio Pérez-García^{1*}

¹Facultad de Estudios Profesionales Zona Huasteca, Universidad Autónoma de San Luis Potosí, Romualdo del Campo #501, Fracc. Rafael Curiel, C.P. 79060, Cd. Valles, S.L.P. ²Instituto de Nutrigenética y Nutrigenómica Traslacional, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara.

*E-mail: luisantonio.perez@uaslp.mx

Eumycetoma is a granulomatous disease caused by fungi, characterized by the formation of subcutaneous grains surrounded by inflammatory infiltrate which generally appears in extremities after traumatic implantation of etiological agents, mainly *Madurella mycetomatis*. Treatment commonly includes the use of azoles and surgery to remove grains, although this scheme has proven to be ineffective, lengthy and with high rate of neglect from patients. It has been suggested that inflammation plays a key role on therapy effectiveness, acting as a barrier preventing antifungals to reach the fungal grains. In this work, we employed an eumycetoma murine model to evaluate the effect of combined diclofenac-itraconazole therapy on the size of grains and secreted cytokines. Four groups of immunocompetent BALB/c mice infected with *M. mycetomatis* were treated for 28 days as follows: 1) saline solution as control, 2) diclofenac, 3) itraconazole and 4) itraconazole-diclofenac. After completion of treatment, infected organs and blood plasma were collected. Organs were histologically treated. On the other hand, IL-1 β , IL-6, IL-17A, IL-17F, IL-22, IL-25, IL-33, sCD40L, IL-4, IL-21, IL-23, IL-31, IFN- γ , and TNF- α levels were measured in plasma. Our results showed that after 4-week period of treatment, size of grains and inflammation were reduced in mice treated with the itraconazole-diclofenac combination. Th1 and Th17 cytokine profile predominated in this group along with diclofenac-treated group, whilst a Th2 profile predominated in itraconazole monotherapy group. These results suggest that azole-NSAID therapy may be a potential alternative for the treatment of eumycetoma caused by *M. mycetomatis*.

This work was funded by Consejo Potosino de Ciencia y Tecnología, Project 2380 (Agreement FME/2023/SE-08/06).

EFFECT OF CHRONIC OZONE POLLUTION ON IL-6 AND CD4 LEVELS IN THE HIPPOCAMPUS OF RATS

Pamela Barragán-Reséndiz, Selva Rivas Arancibia, Alfredo Miranda-Martínez,
Marlen Valdez Fuentes, Erika Rodríguez-Martínez

Departamento de Fisiología, Facultad de Medicina, Universidad Nacional Autónoma de México.
Av. Universidad No. 3000. C.P 04510. Col. Universidad Nacional Autónoma de México, Coyoacán. CDMX.
Tel. (52) 55 56232500
barraganresendizpamela@gmail.com

Introduction. Environmental pollution has become a serious public health problem. One of the main environmental pollutants is ozone (O_3). Chronic exposure to O_3 plays a very important role in the development of various diseases since it induces the loss of redox balance, which is accompanied by alterations in the inflammatory response. During inflammatory processes, microglia cells and adaptive immune system cells, such as CD4 T cells, become activated. IL-6 plays a crucial role in differentiating CD4 T cells and activating the immune response, contributing to inflammation.

Objective: determine the effect of chronic exposure to low ozone doses on CD4 and IL-6 levels in the hippocampus of rats. **Methodology.** for this study, 72 Wistar rats were used, which were randomly divided into six groups (n=12): 1) Control exposed only to ozone-free air, 2) 7 days, 3) 15 days, 4) 30 days, 5) 60 days and 6) 90 days of exposure to ozone for 4 hours daily at 0.25 ppm. Once the groups had completed their ozone exposure time, the rats were deeply anesthetized and sacrificed. The hippocampus was extracted and processed using Western blot and immunohistochemistry techniques, using the following antibodies: IL-6 and CD4. **Results.** The data indicates a significant increase in IL-6 levels after 15 and 60 days of exposure to O_3 ($p < 0.05$). There is a significant increase in CD4 levels observed only after 60 days of exposure to O_3 ($p < 0.05$). **Discussion.** CD4 lymphocytes are activated after 60 days of exposure to O_3 in response to increased IL-6 levels, suggesting a potential differentiation towards the TH17 response. **Conclusions:** In the hippocampus, chronic exposure to low doses of O_3 leads to changes in the inflammatory response. This includes increased levels of interleukin IL-6, which activates the differentiation of the Th17 response mediated by CD4 lymphocytes.

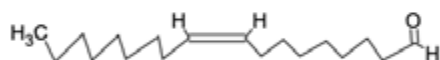
Thanks to the PAPIIT-IN204324 project awarded to S.R-A. Co-responsible E. R-M

OLEIC ACID INDUCE NEUROPROTECTION IN BRAIN REGIONS OF RATS TREATED WITH DEXRAZOXANE

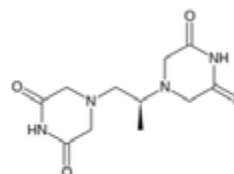
David Calderón Guzmán¹, Norma Osnaya Brizuela¹, Maribel Ortiz Herrera², Armando Valenzuela Peraza¹, Gerardo Barragán Mejía², Ernestina Hernández García³, Francisca Trujillo Jimenez³

Laboratorios Neurociencias¹, Bacteriología Experimental², Farmacología³. Torre de Investigación Dr. Joaquín Cravioto. Subdirección de Medicina Experimental. Instituto Nacional de Pediatría. Ciudad de México. C.P. 04530. México. Email: solodavid2001@yahoo.com.mx

Background. Oleic acid (OA) is a monounsaturated compound with antioxidant property. Worldwide increase in obesity prevalence across age groups has remained unabated in different countries. In Mexico, this tendency has continued to rise until date, affecting both young and old. Metabolic syndrome (MS) is a group of conditions characterized high abdominal fat accumulation, high systolic and diastolic blood pressure, high density lipid cholesterol, and high blood glucose and triglycerides. Irrational consumption of fatty foods and meats has been linked with oxidative stress with its concomitant adverse health effects. Oxidative stress due to nitric oxide is the main cause of alterations in the mitochondrial ultrastructure and DNA damage. It has been reported that exogenous polyunsaturated fatty acids such as oleic and linoleic acids can temper the cytotoxic activity of drugs, and dexrazoxane has been used to protect against toxic side effects of this drugs. The protection properties of OA have been demonstrated. In fact, OA can interact with proteins and increase their potentials to spread to other tissues. This work aims to analyze the effect of oleic acid and dexrazoxane on the levels of dopamine in rat brain. **Methods.** Wistar rats – gender male; age 6 weeks and average weight 150g–were recruited for the study and equally divided into groups. The treatments given to the animals consist of drugs in combination with OA. Group A (control), received NaCl 0.9%; group B, received only oleic acid (OA) (1.5ml/rat); and group C was treated with Dexrazoxane (50mg/rat) + OA. Every treatment was by intraperitoneal route and the administration was every 24 h for 5 days. At the end of treatment were sacrificed and their brains were extracted and sectioned into cortex, striatum, and cerebellum/medulla oblongata (CMO). It was employed in the homogenization of every section of the brain and used to assay the Dopamine levels with fluorescence methods using FL Win Lab version 4.00.02 software. **Results.** The concentration of Dopamine increased several folds in all regions of the groups treated with drug combinations CDX + OA, respect control groups. Dexrazoxane increased dopamine levels in cortex, striatum and CMO in the animals, and in the group treated with OA, the others biomarkers evaluated in this study did not witness any changes. **Conclusion.** This results may be explained either by alterations in the metabolic processes of OA and dysfunction of the mitochondria in dopaminergic neurons.



Oleic Acid



Dexrazoxane

EVALUATION OF ECOTOXIC EFFECTS OF STIBNITE (Sb₂S₃) IN AQUATIC PLANT MODEL (*ELODEA CANADENSIS*)

Luis Adolfo Ceniceros-Morales¹, Ana Paola Rivera Torres¹, Maria Guadalupe Nieto-Pescador¹,
Rene Homero Lara-Castro¹, Patricia Ponce-Peña¹, Francisco Javier Rios-Fránquez¹,
Gerardo Alfonso Anguiano-Vega^{1*}

¹Facultad de Ciencias Químicas Durango, Universidad Juarez del Estado de Durango Av. Veterinaria s/n Circuito Universitario
Col. Valle del sur C.P. 34120. Durango, Dgo, México.

*E-mail: gerardo.anguiano@ujed.mx

Antimony (Sb) is a metal with diverse applications, notably as in the preparation of various alloys. In previous works, it has been described that Sb generates health toxicity problems in humans and wild organisms, when found in the environment, due to the exploitation of mining areas; However, concentration studies place it as a rare and low-concentration contaminant. Sb has been reported as responsible for alterations in growth, development, peroxidation and oxidative stress, among others¹; However, there are not many reports of its capacity as a genotoxic agent².

In this work, the effect of genotoxic damage was evaluated in an aquatic plant model (*Elodea canadensis*), through bioassays of exposure to various concentrations of Sb from 1 to 2000 ppm for 7 and 15 days of exposure; using single cell gel electrophoresis or “comet assay”. In a 7-day trial, obvious damage was observed in the leaf tissue: darkening of the ends of the leaf and stem. Slides were prepared for the evaluation of the comet assay and the corresponding electrophoresis was performed. When observing the nuclei under a fluorescence microscope, apparent differences were found between the unexposed and exposed groups.

This work is a pioneer in the genotoxic study of Sb using comet assay. Sb is currently recognized as a plant toxicant capable of altering the cellular condition of the leaf and stem, where the comet assay is a sensitive and appropriate technique to evaluate genotoxicity in the plant model, *Elodea canadensis*.

References

1. Tang, H., Meng, G., Xiang, J., Mahmood, A., Xiang, G., SanaUllah, N., Liu, Y., & Huang, G. (2022). Toxic effects of antimony in plants: Reasons and remediation possibilities—A review and future prospects. *Frontiers In Plant Science*, 13
2. Obiakor, M. O., Tighe, M., Pereg, L., Maher, W., Taylor, A. M., & Wilson, S. C. (2021). A pilot in vivo evaluation of Sb(III) and Sb(V) genotoxicity using comet assay and micronucleus test on the freshwater fish, silver perch *Bidyanus bidyanus* (Mitchell, 1838).

IDENTIFICATION OF POTENTIAL AGONISTS FROM NATURAL SOURCES FOR PPAR- γ IN THE ERA OF MACHINE LEARNING

José Luis Velasco-Bolom¹, Sergio Hernández-Herrera¹, Ana C. Murrieta²,
Ramón Rodríguez-Villanueva² and Flavio F. Contreras-Torres^{1,2,*}

¹Unidad de Medicina Experimental, The Institute for Obesity Research, Tecnológico de Monterrey, Monterrey, NL, 64849, Mexico. ²Laboratorio de Nano y Microestructuras. Escuela de Ingeniería y Ciencias. Tecnológico de Monterrey, Monterrey, NL, 64849, Mexico
*e-mail: contreras.flavio@tec.mx

Peroxisome proliferator-activated receptor gamma (PPAR- γ) is a nuclear receptor that plays a significant role in regulating adipogenesis and glucose metabolism. PPAR- γ is considered an attractive drug target for addressing metabolic disorders, including obesity and inflammation diseases. Synthetic PPAR- γ agonists such as thiazolidinediones (TZDs) can modulate the orthosteric site; however, TZDs compounds (including rosiglitazone, RGZ) can show undesirable side effects, such as weight gain, fluid retention, and cardiovascular dysfunction. The discovery of novel modulators is crucial to understanding the associated pathologies and developing effective therapeutic strategies.

The present study explores the potential of plant-derived natural compounds to act as PPAR- γ ligands. The COCONUT database was screened using machine learning (ML) approaches (support vector machine algorithm, SVM), leading to 47 potential ligands from over 400,000 entries. Furthermore, we analyzed and compared the strength of interactions with different ligands. Molecular docking with these 47 molecules yields nine molecules in the range of -8.2 and -9.1 kcal/mol. Additionally, the stability of docked complexes was evaluated, and the energetics in protein-ligand structures were estimated through molecular dynamics (MD) simulations (AMBER 22, 500 ns). We predict three potential compounds with the best energetic values and interactions. Natural compounds from catechin, proanthocyanidin, and capsaicin can be allocated into the orthosteric site of PPAR- γ acting as modulators or potential agonists. In particular, the capsaicin compound showed similar interactions at residue positions reported from other ligands of PPAR- γ . The compound also exhibited the most favorable energetic interaction compared with the reported agonist RGZ.

In conclusion, this study aimed to characterize the chemical space of natural compounds and identify novel potential ligands for the PPAR- γ receptor. The era of ML is revolutionizing drug discovery processes, enabling the identification of effective and safe therapeutic candidates more efficiently with the capability to screen thousands of molecules in very low periods. However, the complex interplay of multiple targets will require the incorporation of additional levels of information, and in vitro studies will be needed to confirm our findings.

DIFFERENTIAL EXPRESSION OF PIEZO1 IN PROSTATE CANCER CELL LINES

Jesús Emiliano Covarrubias-Lobatón^{1,2}, Sara Luz Morales-Lázaro^{3,4}, Ruth Rincón-Heredia⁵,
Luisa Durán-Pasten¹ and Enoch Luis^{1,6}

¹Laboratorio Nacional de Canalopatías, Instituto de Fisiología Celular, UNAM, Cto. Exterior s/n, C.U., 04510 Ciudad de México
²Posgrado en Ciencias Bioquímicas, UNAM, ³División de Neurociencias, Instituto de Fisiología Celular, UNAM Cto. Exterior s/n,
C.U., 04510 Ciudad de México, ⁴Centro de Investigación sobre el Envejecimiento, CINVESTAV Sur, Calz. de los Tenorios 235,
Coapa, Rinconada de las Hadas, Tlalpan, 14330 Ciudad de México, CDMX ⁵Unidad de Imagenología del Instituto de Fisiología
Celular, UNAM, Cto. Exterior s/n, C.U., 04510 Ciudad de México, ⁶Investigador CONAHCYT – Instituto de Fisiología Celular, UNAM,
Cto. Exterior s/n, C.U., 04510 Ciudad de México.
Teléfono: 55 5616 2282 Ext. 81449. E-mail: enoch@ifc.unam.mx

Piezo1, a mechanically activated ion channel, described for the first time in 2010 by Coste and collaborators¹, plays an important role in many physiological functions such as breathing, vasoconstriction, heart beating, intestinal motility, mechanotransduction in chondrocytes², and also cancer progression. However, the Piezo1 expression levels in various kinds of cancer, including Prostate Cancer (PCa), remain unknown.

The aid of this work is to study, by western blot (WB), immunofluorescence (IF), and calcium imaging, if there is a differential expression of Piezo1 in normal and cancer cell lines from the prostate: (1) RWPE-1, which serves as a healthy prostate control, (2) LNCaP, a cell line that is capable of expressing the Androgen Receptor and considered less aggressive than others, and (3) PC-3, a level IV metastatic cell line, as the most aggressive.

We determined the total levels of protein by WB assays, finding a trend to decrease the Piezo1 protein levels as cancer appears and increases its aggressiveness; in this way, we observed higher Piezo1 levels in RWPE-1, followed by LNCaP, and an apparent minor level in PC-3 cells. To corroborate WB results and determine differences in functional expression, we made Fluo4-calcium imaging experiments. Cells were stimulated with the Piezo1 agonist, Yoda1 (30 µM). All the cell lines responded to Yoda1; however, the RWPE-1 cell line showed the most robust response with an amplitude signal of 3.75 ± 0.03 (n = 1827), followed by LNCaP with a response mean of 2.15 ± 0.05 (n = 307; P<0.001), and finally, the PC-3 having the lowest response 1.78 ± 0.04 (n = 1680; P<0.001). These functional responses could be explained as the consequence of Piezo1 localization on the cell. By IF, we found that in PC-3 cells, Piezo1 is localized at the cytosol; in contrast, LNCaP and RWPE-1 expressed Piezo1 mainly at the cell membrane. Finally, this differential expression of Piezo1 plays a role in cell viability, where the incubation of the Piezo1 agonist (Yoda1) and antagonist (Dooku1) has a different impact on cell viability in these three cell lines.

In summary, Piezo1 has a differential expression in prostatic tissue, RWPE-1>LNCaP>PC-3. These findings could lead to understanding some of the cancer hallmarks and pathophysiology to eventually modify with therapeutic purposes, nevertheless further studies must be realized.

References

1. Coste, B., Mathur, J., Schmidt, M., Earley, T. J., Ranade, S., Petrus, M. J., Dubin, A. E., & Patapoutian, A. (2010). *Science*, 330, 55-60
2. Qin, L., He, T., Chen, S., Yang, D., Yi, W., Cao, H. & Xiao, G (2021). *Bone Res*, 9, 44.

MOLECULAR COUPLING OF DIELS-ALDER ADDUCT DERIVED FROM ANTHRANILIC ACID ON COX 1 AND 2

Eric Suriel Cruz Ruiz¹, Guevara Salazar Juan Alberto¹, Jessica Rubí Morán Díaz¹,
José Guadalupe Trujillo Ferrara, Mónica Griselda Arellano Mendoza²

¹Department of Pharmacology, Higher School of Medicine-IPN, Plan of San Luis and Salvador Díaz Mirón S/N, Col. Santo Tomás, CP11340, Mexico City. ²Laboratory of Chronic-Degenerative Diseases, Higher School of Medicine-IPN, Plan of San Luis and Salvador Díaz Mirón S/N, Col. Santo Tomás, CP11340, Mexico City.
Email: eric60368@gmail.com, jguevaas@ipn.mx, emi_monica@yahoo.com

Current anti-inflammatory drugs have significant adverse effects, or may not be effective for some patients, highlighting the importance of having diverse therapeutic options. Therefore, the development of new anti-inflammatory drugs is crucial to improve the quality of life of patients.

The work team designed new chemical entities, selecting the lead compound through pharmacophore -based design, starting from salicylic acid, whose bioisostere is anthranilic acid. After completing the design, two series of compounds were generated: series 5, which consists of *N*-phenylmaleamic acids monosubstituted in the *ortho*, *meta* and *para* positions with carboxylic acid; while series 6, consisting of *N*-phenylmaleimides monosubstituted in the same positions with carboxylic acid. It was proposed to evaluate these compounds through a molecular docking study directed at the catalytic site of the COX-1 and COX-2 enzymes to predict the binding of Diels-Alder adducts on these enzymes. The stereoisomers of each adduct were evaluated, and NSAIDs such as acetylsalicylic acid, mefenamic acid, indomethacin and celecoxib were used as positive controls.

The validation of the study showed RMSD values of 0.00 Å, for both enzymes. In the case of the evaluation on COX-1, series 5 showed ΔG values in a range of -9.22 to -10.92 kcal/mol, the most similar being the *ortho* substituted compound; While series 6 presented Gibbs free energy values between -8.41 and -9.61 kcal/mol, the compound with meta-substitution presented the best affinity. With reference to the COX-2 enzyme, the *N*-phenylmaleamic acids showed values of -8.26 to -9.71 kcal/mol, and the *N*-phenylmaleimides presented values of -8.45 to -9.06 kcal/mol, whose results showed for both cases the more promising compounds than those substituted in the *meta* position. These values are comparable to those of the positive controls, indicating a good ligand-enzyme interaction, which means that they could probably be potentially anti-inflammatory and/or analgesic substances.

CHANGES IN LIVER FUNCTION IN RATS CHRONICALLY EXPOSED TO FLUORIDE PRENATALLY AND POSTNATALLY

Fernanda Marlen Enríquez Sánchez¹, Miguel Ángel López Vázquez², María Esther Olvera Cortés²,
Liliana Valdez Jiménez¹, Paola Trinidad Villalobos Gutiérrez¹,
María de la Luz Miranda Beltrán¹, María Isabel Pérez Vega¹

¹Centro Universitario de los Lagos (CULagos), Universidad de Guadalajara, Lagos de Moreno, Jalisco, México,

²Centro de Investigación Biomédica de Michoacán, IMSS, Morelia, Michoacán, México.

E-mail: misabel.perez@academicos.udg.mx

The effect of prolonged exposure to fluoride (100 ppm F⁻) prenatally and postnatally on the serum concentration of AST, ALT, total protein (TP), albumin (ALB), and total bilirubin (TB) of male *Wistar* rats exposed up to 130 days postnatally was evaluated. The exposed animals exhibited a reduction in food consumption and body weight gain, accompanied by increased serum fluoride concentration and alterations in the incisors (dental fluorosis). Similarly, the group exposed from a postnatal stage exhibited an increase in the concentration of AST and TB, as well as a decrease in the concentration of TP and ALB, compared to the control group. In contrast, the animals that began exposure from gestation exhibited a significant decrease in the concentration of ALB in comparison to the control group. Additionally, differences were observed in the serum concentration of AST, ALT, and TP according to the beginning of exposure to F⁻.

The findings demonstrate that prolonged fluoride exposure results in alterations in the parameters studied in hepatic functionality. However, these alterations manifest differently according to the stage at which the fluoride exposure begins.

EFFICACY OF THE CYTOTOXIC EFFECT OF 6-PENTADECYL SALICYLIC ANACARDIC ACID IN OLIVE OIL NANODROPS IN DIFFERENT CELL LINES

Elizabet Estrada-Muñiz¹, Christyna Rosales-González¹, Carlos Ruiz-Suárez²,
Alejandra. G. Valdéz-Lara² and Libia Vega^{1*}

¹Center for Research and Advanced Studies of the National Polytechnic Institute, Toxicology, Mexico City, Mexico;

²Cinvestav, Monterrey Unit, Nuevo Leon, Mexico. *E-mail: lvega@cinvestav.mx

Cancer is the second most common cause of death in the world. The treatment of choice against it is chemotherapy which produces deleterious side effects such as myelosuppression and leukopenia. The 6-pentadecyl salicylic anacardic acid (6SA) has demonstrated cytotoxic activity against several transformed cell lines, and antineoplastic effects *in vivo*. However, 6SA has poor solubility in water, therefore, searching for delivery vehicles is important. Oil-based nanodrops (NDROP) offer solubilization of highly lipophilic drugs and increases in their bioavailability. Thus, this work aimed to evaluate the efficacy of the cytotoxic activity of 6SA in NDROPs formulation compared to 6SA solubilized in DMSO by Neutral Red (NR) and tetrazolium salts (MTT) assays using untransformed fibroblasts and several cancer cell lines.

To produce 6SA-charged NDROPs, 6SA (20 mg) was dissolved in 1 ml of olive oil and 99 ml of distilled water and high-pressure homogenization was applied. Cytotoxic effect of NDROPs was evaluated in human cancer cell lines HCT116, RKO and Lovo (colon), K562 (lymphoblast) and NK-92 MI (natural killer cell), 4T1 mouse breast cancer and untransformed human fibroblasts (GM03440, and CRL1-459) grown in 96-well plates, exposed for 48 h to 6SA (0, 12.5, 25, 50, 100, 200 μ M), or 6SA-NDROPs (3.6, 7.2, 14.4, 28.8, 57.6 and 115 μ M). MTT assay showed that 6SA is cytotoxic in all cell lines evaluated, with inhibitory concentrations of 50% of viability values (IC_{50}) ranging from 62.9 to 88.9 μ M. The NR uptake assay showed IC_{50} values from 39.11 to 110.4 μ M. When 6SA was formulated in NDROPs the IC_{50} values, using the MTT assay, ranged between 63.6 and 100.2 μ M and between 11 and 124.6 using the NR assay.

We concluded that 6SA-NDROPs have similar efficacy than 6SA solubilized in DMSO because the olive oil nanoemulsion did not modify the cytotoxic activity of 6SA. Improved efficacy was observed in some colon cancer cell lines treated with the 6SA-NDROPs, suggesting that the NDROP formulation could be a potential formulation to administer 6SA as an anticancer treatment in humans.

Funding was provided by CONACHyT, grant 21067.

EVALUATION OF THE ADMINISTRATION OF A PHYTOCHEMICAL TO A MURINE MODEL OF ACUTE GASTRITIS

Verónica Edith Gallegos-Hernández¹, Ehekatzin García-Valdés¹, Patricia Vergara-Aragón²,
Mónica Rosalía Jaime Fonseca¹, Rosalba Desiré Romero-Palma², Vannia Daniela Merced-Canales²,
Mónica Michele Hernández-Cruz¹, Maximiliano Loredo-Rosales¹, Nazarett Tabita Mendoza-Centeno¹,
Frida Azucena Valentín García¹

¹Universidad Nacional Autónoma de México, Facultad de Medicina, Fisiología, México.

²Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada, Legaria, IPN, México.

E-mail: vegh9401@gmail.com

The purpose of this work is to evaluate the effect of the administration of different doses of a phytodrug in a murine model of acute gastritis. Materials and methods: The extraction and characterization of the GAHV extract was carried out. Gastritis was induced in the animals (n=4) and the GAHV extract was administered at 5.5, 11, 16.5 and 22 mg/kg b. w., two control groups were developed: positive control and negative control. After 21 days of intragastric administration of the extract, the gastritis was induced through the intragastric administration of 100µL of ethanol at a 5 mL/kg dose. The animals were sacrificed 1 hour after the acute gastritis induction, the stomachs retrieved, and the histological effects of the administration were analyzed. Results: The GAHV extract has an IC50 of DPPH of 0.4349±0.0713 mg/mL, meanwhile in the ABTS test, the extract shows 0.2242±0.0147 TEAC. The extract shows gastroprotective effects and these are dose dependent, when the phytochemical doses were higher the gastroprotective effects were bigger. Conclusion: The administration of different doses of extract (5.5, 11, 16.5 and 22 mg/kg) does not cause greater development of gastric injury when compared to the damage caused by the administration of the vehicle (p=0.389). At low doses (5.5 and 11 mg/kg) the GAHV extract protects the gastric mucosa from the erosive effect of the oral administration of the vehicle composed of PEG+DMSO+CMC 0.5%.

References

1. Azer, S. y Akhondi, H. (2022). Gastritis. PMID: 31334970.
2. Hernández-Rodríguez, S., Quiroz-Reyes, C. N., Ramírez-Ortiz, M. E., Ronquillo-de Jesús, E., & Aguilar-Méndez, M. Á. (2020). Optimización del Proceso de extracción Asistida por ultrasonido de compuestos Fenólicos de Justicia Spicigera Schltdl. Mediante la Metodología de Superficie de Respuesta. TIP Revista Especializada En Ciencias Químico-Biológicas, 23. doi:10.22201/fesz.23958723e.2020.0.246

IMPACT OF LITHIUM CHLORIDE AND LITHIUM CARBONATE ON DNA REPAIR AND APOPTOSIS IN CERVICAL CANCER CELLS

Juan Carlos García Acosta, Axel Fernando Ramírez Rubio, Laura Itzel Quintas Granados, Edgar Yebrán Villegas Vázquez, Octavio Daniel Reyes Hernández, Gabriela Figueroa González

Laboratorio de Farmacogenética, Unidad Multidisciplinaria de Investigación Experimental Zaragoza, Facultad de Estudios Superiores Zaragoza, Universidad Nacional Autónoma de México, Ciudad de México 09230, México.
Tel: 5539005899, e-mail: gabriela.figueroa@zaragoza.unam.mx

Cervical cancer (CC) is the fourth most common cancer among women, with more than 500,000 new cases per year and about 250,000 deaths worldwide. CC is a serious public health problem, and efforts are directed towards the development of new therapeutic approaches. In this context, lithium salts have shown enormous potential because of their impact on different signaling pathways related to the hallmarks of cancer, such as apoptosis, replication, cell death, and genomic instability, among others(1). However, the effects of different lithium salts on tumor cell hallmarks are still not fully understood. In this investigation, we evaluated the effect of lithium chloride (LiCl) and lithium carbonate (Li_2CO_3) on DNA fragmentation and apoptosis induction in CC (HeLa and SiHa) and HaCaT (non-tumor cell line) cells. First, IC_{50} values were determined for each of these salts, and then the Comet and TUNEL assays were used to evaluate the effect of lithium on DNA fragmentation. Additionally, the relationship between lithium salts and the induction of apoptosis was established by determining the expression of apoptosis and DNA repair biomarkers such as CAS3, PARP-1, pChk1S317, and RAD51 by Western blot. According to the results, lithium salts induce DNA fragmentation in both tumor cells and the non-tumor cell line, with LiCl producing higher fragmentation than Li_2CO_3 . We also observed that both lithium salts participate in the induction of apoptosis in the different cell lines. In HeLa cells, we observed 16% and 19% apoptosis in the presence of LiCl and Li_2CO_3 , respectively. In SiHa cells, we observed 63% and 38% induction of apoptosis in the presence of LiCl and Li_2CO_3 , respectively. In HaCaT cells, 24% and 8% induction of apoptosis was observed in the presence of LiCl and Li_2CO_3 , respectively. Our preliminary data indicate that lithium salts induce DNA fragmentation and have a regulatory effect on the apoptosis process.

Funding: The financial and academic support of the project PAPIIT-IA208422, PAPIIT-IA206724, PAPIIT-IN222321, and PAPIIT-IN221824 to Dirección General de Asuntos del Personal Académico (DGAPA).

Reference

1. Villegas-Vázquez EY, Quintas-Granados LI, Cortés H, González-Del Carmen M, Leyva-Gómez G, Rodríguez-Morales M, et al. Lithium: A Promising Anticancer Agent. *Life*. 2023 Feb 15;13(2):537.

EVALUATION OF THE ANTIVIRAL ACTIVITY AGAINST CHIKUNGUNYA OF THE DICHLOROMETHANE EXTRACT OF THE STEM OF *OCIMUM BASILICUM* IN VITRO

José Carlos González Arce¹, Araceli Posadas Mondragón¹, José Angel Santiago Cruz^{1,2}, Angélica Pérez Juárez¹, Norma Estela Herrera González¹, Jessamyn Reyna Crespo Sandoval^{1,2}, Rocío Jiménez Jiménez^{1,2}, José Leopoldo Aguilar Faisal¹

¹Escuela Superior de Medicina-IPN, Plan de San Luis y Díaz Mirón s/n, Col. Casco de Santo Tomas, Alcaldía Miguel Hidalgo, C.P. 11340, Ciudad de México. ²Escuela Nacional de Ciencias Biológicas-IPN, Prolongación de Carpio y Plan de Ayala s/n, Col. Santo Tomás C.P. 11340 Alcaldía Miguel Hidalgo CDMX. México.
E-mail: aposadasm@ipn.mx

Introduction. *Ocimum basilicum* is widely used in traditional medicine for its antimicrobial, antiparasitic, anti-inflammatory, antioxidant and antiviral effects¹. Regarding antiviral activity, extracts and purified compounds have been evaluated against hepatitis B, adenovirus, coxsackievirus, herpes virus and enterovirus^{2,3}. The antiviral activity of the lipophilic hexane extract of *Ocimum basilicum* has been tested against Dengue virus (DENV) and Chikungunya virus (CHIKV), in which a significant reduction in the DENV titer was obtained after pre- and post-treatment, but no anti-CHIKV activity was observed⁴. In this study, the antiviral activity of the dichloromethane extract of the stem of *Ocimum basilicum* against the CHIKV was evaluated. **Methods.** The stem of the *Ocimum basilicum* plant was macerated, and processed with dichloromethane; the solvent was evaporated under reduced pressure. Vero and BJ cells were cultured, the cytotoxicity of the extract was evaluated at concentrations: 100, 50, 25, 12.5 and 6.25 µg/mL using the MTT assay (3-(4,5-dimethylthiazol-2-yl). Antiviral effect assay (co-treatment): Vero and BJ cells were seeded in 24-well plates, and subsequently the mixture of 25 PFUs (plaque-forming units) of CHIKV and the *Ocimum basilicum* extract (100, 50, 25, 12.5 and 6.25 µg/mL) was performed; it was left incubating for 1 h, subsequently plate reduction assay was performed. **Results.** No cytotoxic effect was observed in the BJ cells, only an 85% reduction in viability at 100 µg/mL was observed in the Vero cells. Pre-treatment with the extract reduced CHIKV PFUs by 100% at all concentrations evaluated in Vero cells. A dose-dependent reduction of CHIKV PFUs was observed in both Vero and BJ cells in co-treatments with the extract. **Conclusion.** The dichloromethane extract of the stem of *Ocimum basilicum* showed a reduction of CHIKV PFUs up to 50% in Vero and BJ cells.

References

1. Kuldeep Dhama a, Khan Sharun et al.(2023). A Comprehensive Review on Chemical Profile and Pharmacological. Vol. 39, No. 1, 119–147 <https://doi.org/10.1080/87559129.2021.1900230>.
2. Chiang, L. C.; Ng, L. T.; Cheng, P. W.; Chiang, W.; Lin, C. C. 2005. Antiviral Activities of Extracts and Selected Pure Constituents of *Ocimum Basilicum*. Clin. Exp. Pharmacol. Physiol, 32(10), 811–816.
3. Chattopadhyay, D.; Naik, T. N. 2007. Antivirals of Ethnomedicinal Origin: Structure-activity Relationship and Scope. Mini. Rev. Med. Chem. 7(3), 275–301.
4. Joshi RK, Agarwal S, Patil P, Alagarasu K, Panda K, Cherian S, Parashar D, Roy S. 2023. Anti-Dengue Activity of Lipophilic Fraction of *Ocimum basilicum* L. Stem. Molecules. 2;28(3):1446. doi: 10.3390/molecules28031446.

BACTERICIDAL AND BACTERIOSTATIC ACTIVITY OF SECONDARY METABOLITES FROM *MAGNOLIA VOVIDESII* AGAINST ORAL BACTERIA

Alma Rosa González Pérez¹, Ruth Reyes Cortes², Suria Gisela Vásquez Morales², Juana Elizabeth Reyes Martínez²

¹Doctorado en Ciencias (Biología), Universidad de Guanajuato, ²Departamento de Biología, Universidad de Guanajuato, Noria Alta S/N; C.P. 36050; Guanajuato, Gto.
Tel: (+52) 473-7320006 Ext: 8158
E-mail: juana.reyes@ugto.mx

The Human oral microbiome comprises commensal, symbiotic and pathogenic microorganisms.¹ Oral pathogens form biofilms on tooth surfaces causing caries, periodontal diseases and in some cases systemic diseases. Caries and periodontal diseases are among the most prevalent major infectious diseases worldwide.²

On the other hand, various species of the Magnoliaceae family synthesize a wide variety of biologically active compounds, including alkaloids, flavonoids, neolignans and terpenoids. Many of these compounds are of medicinal importance and have been reported to exhibit insecticidal, antifungal and antimicrobial activity.³ *Magnolia vovidesii* is endemic from Veracruz, Mexico and is an endangered species due the human agriculture activity.

The main objective of this work was to evaluate the antimicrobial activity of *Magnolia vovidesii* botanical extracts on *Enterococcus faecalis*, *Streptococcus mutans* and *Streptococcus sanguinis*, bacteria usually identified in oral cavity, as well as to determine the effect of these extracts on the formation of biofilms. In addition, Magnolia bioactive compounds as honokiol, magnolol and estragoles were tested on its bactericidal effect and the inhibition of biofilm formation.

The bactericidal and bacteriostatic activity of *M. vovidesii* leaf, seed and sarcotesta extracts and chemical compounds (honokiol, magnolol and estragole) were evaluated throughout disc diffusion test and microdilution method. The effect on biofilm formation of botanical extracts and standards were determined by crystal violet staining assay.

Extracts from leaf, seed, and sarcotesta [100 mg/mL] inhibit the growth of *E. faecalis*, *S. mutans*, and *S. sanguinis* on the planktonic state. Honokiol, magnolol and estragole inhibited the growth and biofilm formation of oral bacteria with different efficiencies. Botanical extracts and secondary metabolites of *M. vovidesii* such as honokiol, magnolol, and estragole proved to be potential candidates to prevent or treat of oral diseases caused by pathogenic organisms.⁴

References

1. Arweiler, N. B., & Netuschil, L. (2016). J Adv Exp Med Biol, 902, 45-60. https://doi.org/10.1007/978-3-319-31248-4_4
2. Li, J., Helmerhorst, E. J., Leone, C. W., Troxler, R. F., Yaskell, T., Haffajee, A. D. & Oppenheim, F. G. (2004). J Appl Microbiol, 97(6), 1311-1318. <https://doi.org/10.1111/j.1365>
3. Hernandez-Rocha, J.V., Vásquez-Morales, S.G. 2023. Molecules 28, no. 12: 4681. <https://doi.org/10.3390/molecules28124681>
4. González-Pérez AR. 2022. Actividad bactericida y bacteriostica de metabolitos secundarios de *Magnolia vovidesii* contra bacterias bucales. Tesis de Maestría, Universidad de Guanajuato.

VIAN-C4551 IN EYE DROPS INHIBITS THE RETINAL VASOPERMEABILITY INDUCED BY THE INTRAVITREAL INJECTION OF VEGF IN RATS AND MICE

Daniela Granados Carrasco¹, Elva Adán-Castro¹, Juan Pablo Robles^{1,2}, Magdalena Zamora¹, Lourdes Siqueiros-Marquez¹, Jose Fernando García-Rodrigo¹, Gonzalo Martínez de la Escalera¹, Carmen Clapp^{*}

¹Instituto de Neurobiología, Universidad Nacional Autónoma de México, Queretaro, Queretaro, Mexico.

²VIAN Therapeutics, CA, United States.

* clapp@unam.mx, Campus UNAM 3001, 76230 Juriquilla, Qro., (442) 238 10 28

Introduction. Overproduction of vascular endothelial growth factor (VEGF) contributes to the excessive retinal vasopermeability leading to visual loss in diabetic macular edema. Intravitreal injections of inhibitors of VEGF are first-line therapy, but the invasiveness of frequent intravitreal injections and suboptimal responders demand better treatments. Topical ocular instillation provides a non-invasive alternative. Vasoinhibin is an endogenous protein that blocks VEGF-induced vasopermeability in the retina. The vasoinhibin analog, VIAN-c4551, is a highly potent and stable cyclic heptapeptide. Here, we tested whether VIAN-c4551 in eye drops inhibits VEGF-induced retinal vasopermeability in rodents.

Methods. Wistar rats and CD1 mice were treated with a single eye drop containing different concentrations of VIAN-c4551 at different times before the intravitreal injection of VEGF. The extravasation of Evans blue-stained albumin evaluated retinal vasopermeability.

Results. VIAN-c4551 in eye drops inhibited VEGF-induced vascular leakage in a dose dependent manner. VIAN-c4551's potency was high (0.005% minimum effective dose), significant at 3 hours, maximal at 12 hours, and lasted 24 hours.

Conclusions. VIAN-c4551 in eye drops inhibits excessive retinal vasopermeability with high potency for 24 hours. These findings support VIAN-c4551 as non-invasive, once-a-day potential intervention for preventing the progression of retinal vascular leakage in diabetic macular edema.

Supported by Secretaría de Educación, Ciencia, Tecnología e Innovación de la Ciudad de México (SECTEI-061-2023).

ANACARDIC 6-PENTADECYL SALICYLIC ACID MODIFIES THE EXPRESSION OF CELL CYCLE-RELATED PROTEINS IN JURKAT AND PERIPHERAL BLOOD MONONUCLEAR CELLS

Gabriela Guerrero-Palomo, Sitali Olguín-Reyes, Laura López-Bañuelos, Elizabet Estrada-Muñiz, Libia Vega

Department of Toxicology, Center for Research and Advanced Studies of the National Polytechnic Institute. Ave. IPN 2508, Zacatenco, 07360, Mexico City, Mexico
e-mail: lvega@cinvestav.mx

Anacardic 6-pentadecyl salicylic acid (6SA) is one of the active components of *Amphipterygium adstringens*. This plant is used in traditional medicine to treat symptoms of malaria, vascular and gastric diseases. It has been reported that 6SA has antitumoral activity in vitro and in animal models of breast cancer, antibacterial effects and is a possible modulator of the immune system. Because some reports indicated that 6SA can inhibit the activity of some acetyltransferases, we hypothesize that it can produce changes in regulatory acetylation patterns on proteins that control cell cycle, DNA repair and proliferation, such as E2F1, NF- κ B and Rb, that modulate stability of other cell cycle regulators, such as Cyclin A, or the sensing of DNA damage as Rad50. In this study, we used the transformed cell line of lymphocytes Jurkat and peripheral blood mononuclear cells (PBMCs) obtained from apparently healthy male individuals exposed to 6SA (25 μ M) before, after, and at the same time as cisplatin (Cpt, 10 μ M used as a DNA damage inducer) at different times (6-72 h) to evaluate the expression of NF- κ B, E2F1, Cyclin A, Rad50, Rb and pRb by Western blot. Treatment with 25 μ M of 6SA was not cytotoxic in Jurkat cells as measured by neutral red incorporation (NRU) assay. The exposure of PBMCs and Jurkat cells to 6SA alone did not cause cell cycle arrest, but when Jurkat cells were treated with 6SA and CisPt, we observed an increase in the G₁ phase, and a reduction in the S and G₂ phases. Treatment with 6SA did alter the expression of E2F1 and Cyclin A at 24 h, while the expression of NF- κ B, Rad50, Rb, and pRb changed from the first hours of exposure in Jurkat cells. The data suggest that 6SA induces differential changes at the level of protein expression in Jurkat cells respect to PBMCs.

This work was supported by the National Council of Humanities, Science and Technology of Mexico (CONAHCyT) number 21067 and postdoctoral fellowship CVU 371305.

REPOSITIONING IMIPRAMINE FOR *IN VITRO* ANTIPARASITIC EFFECTS IN *GIARDIA LAMBLIA*

Xareni Zinereth Herrera Valero¹, Angela Daniela Saldaña Alvarado¹, Sendar Daniel Nery Flores¹, Lizeth Guadalupe Campos Múzquiz¹, Martha Monzerrath Orozco Sifuentes¹, Raúl Rodríguez Herrera¹, Filiberto Gutiérrez Gutiérrez², Lissethe Palomo Ligas^{1*}

¹Facultad de Ciencias Químicas, Unidad Saltillo, Universidad Autónoma de Coahuila. Saltillo, Coahuila, 25280, México.

²Departamento de Farmacobiología, Centro Universitario de Ciencias Exactas e Ingenierías, Universidad de Guadalajara, Guadalajara, Jalisco, 44430, México. Tel: 8442118749

*email: lissethe_palomo@uadec.edu.mx

Drug repositioning involves identifying novel therapeutic applications for existing or investigational drugs beyond their original indications¹. Imipramine, a tricyclic antidepressant approved by the FDA and EMA, is primarily used for treating depression, anxiety disorders, and chronic pain². Recent studies have investigated its potential efficacy in colorectal cancer and its antiviral properties, suggesting broader therapeutic applications^{2,3}.

Among gastrointestinal infections caused by protozoans, giardiasis is considered a silent and neglected disease in urgent need of new therapeutic options⁴. Although preliminary studies have suggested the anti-giardial potential of imipramine, comprehensive characterization of its antiprotozoal effects has not been reported. This investigation documents the effects of imipramine on the growth and adherence of *Giardia lamblia* trophozoites. Additionally, the potential induction of cell death and alterations in the immunolocalization of *Giardia* cytoskeletal proteins were evaluated using fluorescence microscopy.

The results of *in vitro* experiments demonstrated that imipramine inhibits the growth and adherence of the parasite. Significant alterations in the distribution of *Giardia lamblia* cytoskeletal proteins were observed, along with evidence of apoptosis-like or necrosis-like cell death. This research highlights new potential antiparasitic effects of an antidepressant medication on *Giardia lamblia*, accelerating the development of new treatments through drug repositioning.

References

1. Pushpakom, et al. (2018). Drug repurposing: Progress, challenges and recommendations. *Nat Rev Drug Discov*, 18(1), 41–58.
2. Asensi-Cantó, et al. (2023). Effects of imipramine on cancer patients over-expressing Fascin1; description of the HITCLIF clinical trial. *Front Oncol*, 13.
3. Kutkat, O., et al. (2022). Robust antiviral activity of commonly prescribed antidepressants against emerging coronaviruses: *in vitro* and *in silico* drug repurposing studies. *Sci Rep*, 12(1), 12920.
4. Argüello-García, et al. (2020). Drug resistance in *Giardia*: Mechanisms and alternative treatments for Giardiasis. *Adv Parasitol*, 107, 201–282.

ELECTROPHYSIOLOGICAL ANALYSIS OF *CENTRUROIDES EXILICAUDA* VENOM ON ION CHANNEL ACTIVITY IN MOUSE DORSAL ROOT GANGLION NEURONS

Darién Huerta-González¹, E. Luis^{2,3}, María Zambrano-Astorga¹, Sergio A. Román-González^{3,4}, N. Caram-Salas^{1,3,*}

¹Departamento de Innovación Biomédica, Centro de Investigación Científica y de Educación de Ensenada, Ensenada BC, México; ²Laboratorio Nacional de Canalopatías, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Ciudad de México, México. ³Investigador(a) por México, CONAHCYT, Ciudad de México. ⁴Instituto Nacional de Medicina Genómica.

E-mail: * ncaram@cicese.mx

Scorpion venoms can induce various physiological responses, including anaphylactic shock, inflammation, arrhythmia, and acute pain, through neurotoxins targeting ion channels (Nav, Cav, Kv). These neurotoxins have potential as tools for developing treatments for neuropathic pain, epilepsy, and other nervous system diseases. This study focuses on the venom of *Centruroides exilicauda*, an endemic species from Baja California, known for causing intense pain and prolonged numbness. We evaluated *C. exilicauda* venom to identify neurotoxins affecting dorsal root ganglion neurons (DRG) ion channels and the human Nav1.7 channel.

C. exilicauda scorpions were captured in Ensenada, México, and venom was extracted via electrostimulation. The venom was tested on DRGs of C57 male mice (7-10 days postpartum, n = 4) using calcium imaging recordings and patch-clamp electrophysiology. The calcium imaging experiment showed that the total venom increases the intracellular calcium concentration dose-dependently. The venom-responding neurons also responded to capsaicin, suggesting a role as nociceptors. Macroscopic ion currents were generated by 0 mV rectangular pulses (100 ms duration) applied every 5 seconds from a holding voltage of -70 mV. Patch-clamp preliminary results indicate that *C. exilicauda* venom reduces inward ionic currents by 35.6% ($p=0.0124$, $n=8$) and outward ionic currents by 39% ($p=0.0328$, $n=8$). Additionally, patch-clamp experiments on HEK293 cells expressing the human Nav1.7 channel showed that *C. exilicauda* venom reduces inward current by 65% ($p=0.0008$, $n=7$).

These findings suggest the potential to prevent and generate neuron depolarization and action potential generation differently. Inhibition could be significant for treating pain. Ongoing work involves detailed electrophysiological analysis of fractionated venom using High-Performance Liquid Chromatography (HPLC) to identify components with therapeutic potential for pain treatment.

EFFECT OF CHRONIC β -CAROTENE TREATMENT ON STEM MARKERS OF BREAST CANCER CELL LINES

Juan Carlos Juárez-Cruz¹, Josefina Atrisco-Morales², Miguel Ángel Mendoza-Catalán³,
Mónica Ramírez⁴, Eduardo Castañeda-Saucedo¹ and Napoleón Navarro Tito¹

¹Laboratorio de Biología Celular del Cáncer, ²Laboratorio de Investigación Clínica, ³Laboratorio de Biomedicina Molecular, ⁴Laboratorio de Proteómica y Genómica Funcional, FCQB-UAGro. Av. Lázaro Cárdenas S/N Ciudad Universitaria. C.P. 39087. Chilpancingo de los Bravo, Gro. Méx.
E-mail: nnavarro@uagro.mx

Background. Breast cancer presents recurrence, chemoresistance, and metastasis associated with cancer stem cells, which are subpopulations of tumor cells with the capacity for self-renewal and giving rise to heterogeneous lineages of cancer cells¹. β -carotene (BC) is an antioxidant phytochemical with anti-tumor capacity in various cancer models². BC regulates various stem markers in models of colon cancer and neuroblastoma; however, the effect of BC on stem markers in breast models has been poorly explored³. **Objective.** Determine in breast cancer cell lines the effect of chronic BC treatment on clonal capacity, spheroid formation capacity, anoikis, and β -catenin expression levels. **Methodology:** MCF7, T47D, and MDA-MB-231 cells were treated for 14 days with 0, 10, 25, and 50 μ M of BC. After treatment with BC, cell viability was determined by counting using trypan blue. Additionally, the clonal capacity was determined by clonogenicity assay, and the sphere formation capacity was determined by the mammosphere formation assay (MFA). To determine the effect of the treatment on anoikis, the MFA cells were reseeded in adherent culture conditions, grown for 12 days, and finally counted with trypan blue. The relative expression levels of β -catenin were determined by Western blot. **Results.** We found that the total number of viable cells decreases in a dose-dependent manner in MCF7 and MDA-MB-231 cells, but no changes in T47D cells were observed. Clonogenicity assays showed a significant dose-dependent decrease in the size and number of colonies of MDA-MB-231 cells a slight decrease in the size and number of colonies of MCF7 cells, but no changes in T47D cells were observed. MFA showed a significant dose-dependent decrease in the mammosphere number of MCF7 and MDA-MB-231 cells but no change in T47D cells. The results of reseeded cells showed a significant dose-dependent decrease in the viable cell number of MCF7 and MDA-MB-231 cells. Treatment with BC in T47D and MDA-MB-231 cells decreases β -catenin levels in a dose-dependent manner. In contrast, in MCF7 cells, viability increases significantly at 10 and 25 μ M, but decreases below basal levels at 50 μ M. **Conclusions.** The effect of chronic treatment with BC displayed differential effects on stem markers in breast cancer models but showed a marked effect on stem markers in triple-negative breast cancer cells, MDA-MB-231.

References

1. Zheng, Q., Zhang, M., Zhou, F., Zhang, L., & Meng, X. (2021). *Frontiers in Pharmacology*, 11, 599965.
2. Niranjana, R., Gayathri, R., Mol, S. N., Sugawara, T., Hirata, T., Miyashita, K., & Ganesan, P. (2015). *Journal of Functional Foods*, 18, 968-985.
3. Lee, K. E., Kwon, M., Kim, Y. S., Kim, Y., Chung, M. G., Heo, S. C., & Kim, Y. (2022). *Nutrition Research and Practice*, 16, 161-172.

DISCOVERING POTENTIAL ANALGESICS FROM BAJA CALIFORNIA BARK SCORPION VENOM (*CENTRUROIDES EXILICAUDA*) USING A METABOLOMIC APPROACH

Eugenio Leyva-Figueroa¹, Rommel A. Carballo-Castañeda¹, Alan Hernández-Melgar¹,
Aldo Moreno-Ulloa¹, Nadia Caram-Salas^{1,2}

¹Biomedical Innovation Department, CICESE, Carretera Ensenada-Tijuana No. 3918, Zona Playitas, Ensenada, B.C. México.

²CONAHCYT, Avenida Insurgentes Sur 1582, Crédito Constructor, Benito Juárez, Ciudad de México, 03940, México.

E-mail: ncaram@cicese.mx Tel. 646 175-05-00 ext. 27272

Scorpion venom, known for its neurotoxic effects in mammals, holds promise for its potential analgesic properties, particularly in addressing healthcare challenges, such as chronic pain. Despite the existing body of research supporting the potentially beneficial effects of scorpion venom in pain management, these venoms, especially one of the endemic Baja California scorpions, *Centruroides exilicauda* (*Ce*), still need to be fully characterized. In this study, we investigated the metabolome of the venomous glands (VG) of this scorpion to uncover metabolites potentially associated with its analgesic properties. Untargeted mass spectrometry-based metabolomics was used to analyze the VG of *Ce* scorpions, employing a wide range of chemoinformatics tools for data analysis. This included GNPS for putative annotation; *in silico* prediction tools such as SIRIUS, MolDiscovery, and DEREPLICATOR+ to discover potential metabolites; ClassyFire and CANOPUS for classification; and DataWarrior for data visualization and analysis. Metabolite profiling uncovered 384 unique metabolites in the VG of *Ce* scorpions, including peptidomimetics, amino acid derivatives, and lipids. Specific metabolites belonged to chemical classes associated with analgesic drugs, such as piperidines, indoles, phenanthrenes, and pyrrolidines. We then compared the molecular structures of the identified metabolites with those of FDA-approved drugs in the ChEMBL database using t-distributed stochastic neighbor embedding (t-SNE) to gain insight into potential analgesic metabolites. These projections allowed us to identify potential metabolites that cluster near FDA-approved drugs for neurological conditions, suggesting a solid link to the analgesic properties of scorpion venom. Our study uncovered a wide variety of metabolites with an extensive range of potential analgesic properties in the venom glands of *Ce* scorpions, highlighting their potential as a source for biomedical innovation and promising prospects for the development of new analgesic treatments.

STUDY OF THE EFFECT OF GLUTATHIONE PEROXIDASE IN ORGANOTYPICAL CULTURES OF RAT CARTILAGE EXPOSED TO NANOPARTICLES WITH GLUTATHIONE

Laura Denise López Barrera¹, Karen E. Álvarez Castillo¹, Roberto Díaz Torres¹ y Patricia Ramírez Noguera¹

¹Laboratorio 9 "Toxicología Celular" UIM, Facultad de Estudios Superiores Cuautitlán, Universidad Nacional Autónoma de México. Km 2.5 carretera Cuautitlán-Teoloyucan, San Sebastián Xhala, Cuautitlán Izcalli, Estado de México, México, C.P. 54714.

Corresponding autor: laura.lobar@cuautitlan.unam.mx

Introduction. Osteoarthritis (OA) is a degenerative pathology of the joints that occur when the cartilage that cushions the ends of bones in joints breaks down progressively deteriorate¹. Cartilage is a firm, slippery tissue that allows movement joint without practically friction. Eventually, if the cartilage wears away completely, the bone it will rub against the bone. Nanoparticles are complex systems, an escalation nanometric, made up of at least two components, one of which is the active ingredient or biologically active molecule and, the second, is the system itself that allows a special function related to the diagnosis, treatment or prevention of a disease^{1,2}. **Objective.** Evaluate the cell viability, as well as the effect on GSH-dependent antioxidant enzymes by exposure of antioxidant nanosystems in an organotypic culture of rat cartilage. **Method.** The sacrifice of a 5-week-old Wistar rat to obtain the ribs, the treatment was performed necessary to carry out the organotypic culture. Nanoparticles were prepared by the method of ionic gelation according to the proposed methodologies with some modifications, these were characterized through Malvern's Zetasizer . To evaluate the antioxidant response, exposed systems to NP-Q and NP-Q/GSH at different concentrations, as well as some were exposes a H₂O₂. The enzymatic activity of GSH, GPx and cell viability (LDH) were evaluated by spectrophotometric methods. **Results.** Evaluation of the activity of different enzymes show the modification of the redox state of the cells after exposure with GSH-loaded nanoparticles. Likewise, exposure to NP-Q/GSH increased the cellular functionality of the systems, in response to their exposure to an extra contribution of cellular antioxidant (GSH). **Conclusions.** NP-Q/GSH at concentrations used help modulate the effect of H₂O₂, so the enzymatic activity of GPx, and LDH, in systems exposed to these NPs decreased as an adverse effect of the increase in antioxidant capacity of the cell induced by the extra contribution of defense against oxidative damage exerted by GSH encapsulated in NPs.

This work was financed by the project Programa interno de catedras de investigación 2024, con el Proyecto "Estudio de la actividad toxicológica y terapéutica de agentes xenobióticos" con clave CI2476.

References

1. Dycus, D, Au. A., Grzanna M., Wardlaw J. y Frondoza C. Modulation of inflammation and oxidative stress in canine chondrocytes. *AJVR* , JULY 2013, 74 (7): 983-989.
2. Mobasheri, A. Konrad, H. Shakibaei, M. and Henrstin Y. Antioxidants and Osteoarthritis. *Systems biology of free radicals and antioxidants* MAY 2014, 2997-3026.

KIDNEY AND LIVER DAMAGE FROM A COMBINED NSAID-ANTIFUNGAL THERAPY FOR TREATMENT OF EUMYCETOMA

Manuel Martínez-Hernández¹, Luis Antonio Pérez-García¹, José David Torres-Tirado¹, Mónica Jasso-Romo¹, Sarai C. Rodríguez-Reyes², Gabriela Pérez-Flores^{1*}

¹Facultad de Estudios Profesionales Zona Huasteca, Universidad Autónoma de San Luis Potosí, Romualdo del Campo #501, Fracc. Rafael Curiel, C.P. 79060, Cd. Valles, S.L.P. ²Instituto de Nutrigenética y Nutrigenómica Traslacional, Centro Universitario de Ciencias de la Salud, Universidad de Guadalajara.

*E-mail: gabriela.perez@uaslp.mx

Eumycetoma is a chronic granulomatous disease that forms grains in the subcutaneous tissue accompanied by pus and pain. The main etiological agent is *Madurella mycetomatis*¹. Conventional treatment (administration of itraconazole and surgery) has proven to be inefficient and has toxic effects. The objective of this work was to determine the magnitude of liver and kidney damage in mice that received a combined NSAID-antifungal pharmacotherapy, which have been studied in our work team as a treatment for eumycetoma. The methodology consisted in administering the following drugs: itraconazole (ITZ), terbinafine (TBN), liposomal amphotericin B (AmB lip) and amphotericin B deoxycholate (AmB deoxy) either in monotherapy or in combination with diclofenac as NSAID to different groups of BALB/c mice, which were sacrificed after 28 days of treatment to collect their liver and kidneys. The samples were analyzed by traditional histological techniques to assess histopathological modifications. In microscopic evaluation, different criteria were used to assess the liver (steatosis, blood congestion, necrosis, and hepatocellular repair) and the kidneys (permeability, blood congestion, necrosis, and tubular edema). The results showed that the ITZ-DC group presented the highest evidence of hepatocellular repair, with a significant difference over all groups. This might be due to the toxicity generated by the combination of the two drugs. Moreover, ITZ-DC group presented the highest tubular edema in the kidneys, with a significant difference against the control and AmB deoxy. Using PAS staining we found that the matter congesting the tubules was of glycosidic nature. These results suggest that the combination of ITZ-DC causes more damage to the kidneys and liver than that caused by the other combinations of NSAID-antifungal or the monotherapy.

This work was funded by Consejo Potosino de Ciencia y Tecnología, Project 2380 (Agreement FME/2023/SE-08/06).

EVALUATION OF ANTINEOPLASTIC COMPOUNDS IN BREAST CANCER CELL LINES

Natalia Mata de los Rios¹, Carmen Carolina Alvarado González¹, Iván Arenas Sosa², Gerardo Corzo Burguete²,
Roxana Olguín Alor³, Gerardo Pável Espino Solís¹

¹Laboratorio de Investigación Traslacional, Facultad de Medicina y Ciencias Biomédicas, UACH, Nuevo Campus Universitario, CP 31125, Chihuahua, Chihuahua, México. ²Instituto de Biotecnología, UNAM, Av. Universidad 2001, Chamilpa, 62210 Cuernavaca, Morelos, México. ³Laboratorio Nacional de Citometría de Flujo, Instituto de Investigaciones Biomédicas, UNAM, Escolar 33, C.U., Coyoacán, 04510 Ciudad de México, México. E-mail: gespinos@uach.mx

Breast cancer encompasses a group of diseases with various subtypes, molecular profiles, presentations, and behaviors ⁽¹⁾. Treatment modalities vary depending on the molecular profile. The standard treatment is chemotherapy, which lacks specificity, is associated with significant adverse effects, and has a propensity to develop resistance, enforcing the discovery of new therapies ⁽²⁾. Based on this, the study of various components with the intention of discovering chemotherapeutic agents was initiated; from medicinal herbs to animal venoms, such as scorpions ⁽³⁾. This has shown promise; it is known that scorpion venom is a mixture of neurotoxins and other biologically active peptides that display antibacterial, immunomodulatory, and anticancer activities, among others. Several peptides derived from these venoms have demonstrated activity in neoplastic cells, both in vitro and in vivo ⁽⁴⁾. Therefore, the objective of this work was to study protein fractions isolated from the venom of the scorpions *Chihuahuanus cohuailae* and *Chihuahuanus crassimanus*. Their effect on cell proliferation was evaluated through assessing the expression of the Ki-67 marker and on the induction of apoptosis using Annexin V and 7AAD by flow cytometry approaches. It was found that no fraction significantly reduced cell viability or increased apoptosis on the other hand, that fraction 43 reduced Ki-67 expression to a greater extent at a lower concentration.

References

1. Kashyap D, Pal D, Sharma R, Garg VK, Goel N, Koundal D, et al. (2022). *Biomed Res Int*, **18**, 1-16.
2. Yin L, Duan JJ, Bian XW, Yu S cang (2020). *Breast Cancer Research*, **22**, 61.
3. Yu C, Li Y, Chen G, Wu C, Wang X, Zhang Y (2022). *Journal of Zhejiang University-SCIENCE B*. **23**, 547-63.
4. Ortiz E, Gurrola GB, Schwartz EF, Possani LD (2015). *Toxicon*, **93**, 125-35.

EVALUATION OF ARTICHOKE (*CYNARA SCOLYMUS*) EXTRACT AND WEIGHT CONTROL IN A MODEL OF INDUCED MENOPAUSE

Nazarett Tabita Mendoza-Centeno¹, Patricia Vergara-Aragón¹, Fabiola Mallon Mercado², Vannia Daniela Merced-Canales¹, Rosalba Desiré Romero-Palma¹, Mónica Michele Hernández-Cruz¹, Maximiliano Loredo-Rosales¹, Joshua Imanol Cervantes Andraca¹, Verónica Edith Gallegos-Hernández², Ehekatzin García-Valdés²

¹Universidad Nacional Autónoma de México, Facultad de Medicina,

²Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada Unidad Legaria, IPN.

E-mail: mc22mecn7350@facmed.unam.mx

This experiment studied the effects of the administration of an ethanolic extract of artichoke (*Cynara scolymus*), in order to treat symptoms caused by hormonal dyscontrol, in an animal model of menopause induced by oophorectomy. The extraction of phenolic compounds and flavonoids from artichoke was performed through maceration, soxhlet method, among others. Once the extracts were characterized, two experiments were carried out with the murine model: E1: dose-response curve: 48 male Wistar rats with a body weight of 220-260 g were used, 3 groups were formed to establish different types of administration and the extract was diluted in different substances using different doses to observe its effect: intraperitoneal route extract diluted in ethanol: (0.2; 0.1; 0.05; 0.025) (g Artichoke/ kg bw); intraperitoneal route extract diluted in water : (0.2; 0.1; 0.05; 0.025) (g Artichoke/ kg bw) and oral route, extract diluted with saline solution: (0.05; 0.025) (g Artichoke/ kg bw). 30 minutes after the administration of the treatments, the animals were sacrificed with an overdose of sodium pentobarbital. E2: Effect on a menopause model: Wistar strain female rats weighing 200-250 g oophorectomized were used. Three groups were established: SHAM, OVX (oophorectomized rats) and OVX+ALC (oophorectomized rats+artichoke extract). At the end of the experiment, they were sacrificed with an overdose of 4% paraformaldehyde perfusion anesthesia. Organs from both experiments were recovered and analyzed histologically with H&E staining. The ED50 for the ethanolic artichoke extract was 0.025 g alc/kg b.w., After administration of the extract in a menopause model, the body weights of the animals decreased compared to the control (p<0.05). Artichoke extract did not damage the cytoarchitecture of the rat small intestine. The observed effects may be due to the high content of antioxidant compounds in the extract.

References

1. Rezazadeh, K.; Rahmati-Yamchi, M.; Mohammadnejad, L.; Ebrahimi-Mameghani, M.; Delazar, A. Effects of Artichoke Leaf Extract Supplementation on Metabolic Parameters in Women with Metabolic Syndrome: Influence of TCF7L2-Rs7903146 and FTO-Rs9939609 Polymorphisms. *Phytotherapy Research* 2018, 32 (1), 84–93. <https://doi.org/10.1002/ptr.5951>.

BEHAVIORAL AND GABAERGIC ALTERATIONS AFTER CHRONIC ATRAZINE EXPOSURE IN THE FEMALE ALBINO RAT

M.S. Mendoza-Trejo¹, J.J. Sanchez-Yepe¹, T. Acevedo-Huergo¹, R. Corona², G.R. Sevilla-Parra¹,
V. Pastor-Martínez Ulloa¹, and V. M. Rodríguez¹.

¹Departamento de Neurobiología Conductual y Cognitiva. Instituto de Neurobiología, Universidad Nacional Autónoma de México. ²Departamento de Neurobiología Celular y Molecular. Instituto de Neurobiología, Universidad Nacional Autónoma de México. Blvd. Juriquilla 3001. C.P. 76230. Querétaro, Qro. México. Tel. (442) 2381061 ext. 34061.
E- mail: vmrodriguez@comunidad.unam.mx

Studies performed mainly in male rodents have shown that exposure to the widely used herbicide atrazine (ATR) produces disruptions in motor behavior and memory, and alters dopamine, GABA, and glutamate markers in basal ganglia. However, in female rats, studies that evaluate the neurotoxic effects of chronic atrazine exposure are sparse. The present study aimed to evaluate the alterations in motor behavior, anxiety, memory, olfaction, and GABA levels in brain regions of female rats chronically treated with 0, 1, or 10 mg ATR/kg body weight daily for 14 months. Female rats treated with 1 or 10 mg ATR showed alterations in locomotor activity along the treatment. Impairments in olfactory function were also present in both groups exposed to 1 or 10 mg ATR, although no alterations in anxiety, memory, or motor coordination tasks were observed. Increased tissue GABA levels were found in the ventral midbrain of 1 and 10 mg ATR/kg groups, but no alterations were found in the striatum or nucleus accumbens. These results show that, in females, chronic ATR exposure transiently alters locomotor activity, disrupts olfaction, and increases the GABA content in the ventral midbrain.

Acknowledgments: Dra. Alejandra Castilla, Ing. Luis Antonio Prado Loeza, María Eugenia Rosas Alatorre, and Ing. Omar González Hernández.

Supported by UNAM-DGAPA-PAPIIT IN 212522.

CURCUMIN AND METFORMIN ENHANCES CYTOTOXICITY AND CELL DEATH ON CERVICAL CANCER CELL LINES

Marisol Mir García, Jonathan Gerardo Sánchez Peralta, Margarita Bahena Román,
Vicente Madrid Marina, Carla O. Contreras Ochoa*

Centro de Investigación Sobre Enfermedades Infecciosas, Instituto Nacional de Salud Pública
*Av. Universidad no. 655, Col. Santa María Ahuacatlán, Cerrada los Pinos y Caminera, Cuernavaca Morelos, México, CP 62100,
Teléfono 777 3293000 extensión 2204, Email: ccontreras@insp.mx

Cervical cancer is a major public health problem worldwide, chemotherapy with antineoplastic drugs is the current treatment, but secondary effects, non-specificity and chemoresistance reduce the result. Therefore, it is relevant to find new alternatives of treatment. Metformin (Met) is a synthetic compound used for type 2 diabetes treatment, while Curcumin (Cur) is a polyphenol isolated from a plant and has antioxidant, anti-inflammatory and immunomodulatory properties. Met and Cur have anti-tumoral effect, reduce cell proliferation and induce cell death. **Objective.** Evaluate the cytotoxic effect of Met in combination with Cur on cervical cancer cell lines and identify cellular and molecular targets activated by these compounds. **Material and methods.** HeLa (HPV18⁺) and SiHa (HPV16⁺) cells were treated with several doses of Met and Cur during 48 hours, cell viability was measured with MTS reagent. Expression of genes involved in cell proliferation and survival (*Cyclin D1*, *p21*, *NFKB*, *STAT3*), apoptosis cell death (*Bax*, *Bcl-2*), autophagy (*LC3B*), and carcinogenesis (*E6*, *E7*) were evaluated by real time RT-qPCR; additionally, apoptosis induction was measured by Caspases-3/7 activity by luminescence. **Results.** Met and Cur induce a dose-dependent cytotoxic effect, Met IC₅₀ was 80 mM and 50 mM for SiHa and HeLa respectively, while Cur IC₅₀ was 150 μM in both cell lines. The combined treatment of Met+Cur significantly enhanced cytotoxicity between 32% and 82%. Met and Cur induced a differential expression of the genes evaluated, the overexpressed genes were mainly *NFKB* and *LC3B*, followed by *Bcl-2* and *Cyclin D1*; whereas *STAT-3* decreased. In contrast, *P21* decreases in SiHa and increased in HeLa, and *Bax* remained unchanged. Importantly, Cur decreased the expression of the E6 and E7 oncogenes in both cell lines. Met or Cur individually induced apoptosis through caspase 3/7 activation, whereas the combined treatment Met+Cur decreased this activity. **Conclusions.** Curcumin enhanced the cytotoxicity of Metformin in HPV cells; this cytotoxic and antiproliferative effect was associated with apoptosis and autophagy cell death induction. The differential expression of genes was dependent of individual or combination treatment, viral genotype (18 or 16), or cell type/tumor. The inhibition of HPV oncogenes E6 and E7 by Curcumin could prevent cellular transformation. Curcumin and/or Metformin activate or inhibit several signaling pathways, which allows them to proposed as potential candidates for adjuvant chemotherapy for cervical cancer treatment.

ANTIMICROBIAL ACTIVITY OF SYNTHETIC PEPTIDES DERIVED FROM SCORPION *SUPERSTITIONIA DONENSIS*

Rodolfo Miranda-Espino¹, Georgina Gurrola-Briones¹, Jimena Cid-Uribe¹, Fernando Zamudio¹,
Lourival Domingos Possani Postay¹

¹Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Universidad 2001, Cuernavaca Morelos 62210, México.

Recent years have brought with them a considerable increase of antibiotic resistant pathogens, which have turned into a top global public health and development threat. Consequently the development of novel antibiotics has become crucial. Venoms, especially scorpion venoms, have proven to be a rich source of diverse antibiotic molecules. Of the components of the venom, non-disulphide bridge peptides (NDBPs) are of particular interest, and the venom gland transcriptome of *Superstitionia donensis* suggest a greater than normal diversity in this component. Here five peptides derived from the venom gland transcriptome of the scorpion *Superstitionia donensis* were selected, synthesized, and purified after which, their haemolytic and antimicrobial activities were screened and measured. Antimicrobial activity against Gram-negative and Gram-positive bacteria *in vitro* together with low haemolytic activity in the selected peptides SDO_NDBP4_5 and SDO_NDBP4_3 suggests it is possible to select for desirable characteristics in antimicrobial peptides derived from scorpion venom, whose clinical application has been traditionally hindered by their high toxicity.

References

1. Rodríguez de la Vega RC, Schwartz EF, Possani LD. Mining on scorpion venom biodiversity. *Toxicon* [Internet]. 2010 Dec 15 [cited 2023 Oct 14];56(7):1155–61. Available from: <https://www.sciencedirect.com/science/article/pii/S0041010109005285>
2. Almaaytah A, Albalas Q. Scorpion venom peptides with no disulfide bridges: A review. *Peptides* [Internet]. 2014 Jan [cited 2023 Oct 14];51:35–45. Available from: <https://linkinghub.elsevier.com/retrieve/pii/S0196978113003537>
3. Jiménez-Vargas JM, Ramírez-Carretero S, Corzo G, Possani LD, Becerril B, Ortiz E. Structural and functional characterization of NDBP-4 family antimicrobial peptides from the scorpion *Mesomexovis variegatus*. *Peptides* [Internet]. 2021 Jul 1 [cited 2023 Aug 8];141:170553. Available from: <https://www.sciencedirect.com/science/article/pii/S0196978121000619>
4. Santibáñez-López CE, Cid-Uribe JI, Batista CVF, Ortiz E, Possani LD. Venom Gland Transcriptomic and Proteomic Analyses of the Enigmatic Scorpion *Superstitionia donensis* (Scorpiones: Superstitioniidae), with Insights on the Evolution of Its Venom Components. *Toxins* [Internet]. 2016 Dec 9 [cited 2023 Aug 8];8(12):367. Available from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5198561/>

ANTIFUNGAL MICONAZOLE INHIBITS KV10.1 AND NAV1.7 ION CHANNELS

Enikar Patlán^{1,2}, María Luisa Durán-Pastén², Sara Luz Morales-Lázaro³, Ricardo González-Ramírez⁴, Ruth Rincón-Heredía⁵, Enoch Luis^{2,6}

¹Posgrado en Ciencias Bioquímicas, UNAM, CDMX; ²Laboratorio Nacional de Canalopatías, Instituto de Fisiología Celular, UNAM, CDMX; ³Instituto de Fisiología Celular, UNAM, CDMX. ⁴Departamento de Biología Molecular e Histocompatibilidad, Hospital General Dr. Manuel Gea González, Secretaría de Salud, CDMX; ⁵Unidad de Imagenología, Instituto de Fisiología Celular, CDMX; ⁶Investigador por México CONAHCYT, CDMX.

E-mail: enikar@ciencias.unam.mx

Miconazole belongs to the imidazole family and is recognized as an antifungal agent. Its primary targets include fungal infections of the epidermis, oral cavity, and vagina. In recent years, miconazole has shown potential as an inhibitor of various ion channels, such as Kv11.1. This inhibitory effect is particularly intriguing as it may modulate other ion channels implicated in pathological conditions like cancer. Consequently, we sought to investigate the inhibitory effects of miconazole on voltage-dependent Kv10.1 and Nav1.7 channels. Kv10.1 is predominantly expressed in the central nervous system, particularly in the olfactory bulb, while Nav1.7 is found in sensory neurons innervating the skin. Both channels, however, are overexpressed in a variety of tumors, including melanoma. In our study, using patch-clamp electrophysiology, we assessed the impact of miconazole on HEK cells that expressed the human Kv10.1 and Nav1.7 channels. We observed a dose-dependent inhibition of Kv10.1 activity by miconazole, with an IC₅₀ value of $23.6 \pm 5.0 \mu\text{M}$. At $100 \mu\text{M}$, miconazole completely abolished Kv10.1 currents (at +50 mV), reducing them from $12.6 \pm 1.4 \text{ nA}$ in control conditions to $0.7 \pm 0.2 \text{ nA}$ in the presence of miconazole ($n = 10$; $P < 0.001$, paired t-test). Molecular docking simulations made us hypothesize that miconazole exerts its inhibitory effect through pore blockage at the S6 segment, specifically targeting phenylalanine residue 495. In contrast, miconazole did not show a dose-dependent effect on HEK-Nav1.7 channels, showing only a 41% inhibitory effect at $100 \mu\text{M}$ concentration, reducing Nav1.7 currents from $-652.9 \pm 80.5 \text{ pA}$ in control conditions to $-190.8 \pm 18.2 \text{ nA}$ in the presence of miconazole ($n = 7$; $P = 0.0164$, paired t-test). Additionally, we observed that miconazole ($30 \mu\text{M}$) reduced the viability of wild-type HEK cells by 31%, HEK-Nav1.7 by 26%, and HEK-Kv10.1 by 6%; it suggests a protective role of Kv10.1 against cytotoxicity by miconazole. Furthermore, using the annexin V assay, we demonstrated that miconazole induces cellular apoptosis at low concentrations in HEK-Kv10.1. It is worth noting that miconazole received FDA approval in 1974 and has successfully passed all safety tests. Therefore, considering its safety profile, repurposing miconazole as a potential treatment for tumors overexpressing Kv10.1 and Nav1.7 ion channels appears promising.

References

1. Kikuchi, K., Nagatomo, T., Abe, H., Kawakami, K., Duff, H. J., Makielski, J. C., January, C. T. & Nakashima, Y. (2005). Blockade of HERG cardiac K⁺ current by antifungal drug miconazole. *British Journal of Pharmacology*, 144(6), 840-848. <https://doi.org/10.1038/sj.bjp.0706095>
2. Luis, E., Anaya-Hernández, A., León-Sánchez, P. & Durán-Pastén, M. L. (2022). The Kv10.1 Channel: A Promising Target in Cancer. *International Journal of Molecular Sciences*, 23(15), 8458. <https://doi.org/10.3390/ijms23158458>

MACHINE LEARNING INNOVATES DRUG DISCOVERY PROCESSES: POTENTIAL LIGANDS FOR CANNABINOIDS

Ana C. Murrieta¹, Ramón Rodríguez-Villanueva¹, José Luis Velasco-Bolom²,
Gerardo García-Rivas², Flavio F. Contreras-Torres^{1,2,*}

¹Laboratorio de Nano y Microestructuras. Escuela de Ingeniería y Ciencias. Tecnológico de Monterrey, Monterrey, NL, 64849, Mexico. ²Unidad de Medicina Experimental, The Institute for Obesity Research, Tecnológico de Monterrey, Monterrey, NL, 64849, Mexico.
*e-mail: contreras.flavio@tec.mx

The endocannabinoid system, including CB1 and CB2 receptors and ligands anandamide and 2-arachidonoylglycerol, is linked to inflammation, pain, stress, cardiovascular health, and obesity¹. CB1 activation increases appetite and food intake, whereas CB2 modulation regulates inflammatory responses². While approved pharmacotherapies targeting CB1 have existed for appetite regulation³, the quest for safe and effective therapies persists.

This study used machine learning (ML) to explore the chemical space of potential ligands (CB1-antagonists and CB2-agonists) with criteria of functional similarity in over 400,000 molecules from the COCONUT database. Support Vector Machine (SVM), Random Forest (RF), and Deep Neural Networks (DNN) evaluated train/test splits, scoring metrics, and time performance for binary classification. Predictions were validated with different input formats (PubChem, Morgan, MACCSKeys, and Daylight). RF led to 21 CB1 and 52 CB2-ligands from 6 chemical families, with accuracy of 0.94 (CB1) and 0.88 (CB2). Predicted molecules were structurally optimized (Open Babel v3.0.1). CB1 (PDB 5TGZ) and CB2 (PDB 8GUR) crystals were prepared for modeling (Modeller v10.4). Ten independent docking experiments (Qvina2.1) per crystal were averaged, comparing the best interactions with endogenous and reported ligands. Molecular dynamics simulations (AMBER22) were conducted for top-ranked molecules. We report favorable interactions in 2-arylbenzofuran flavonoids with CB1 (-11.5 kcal/mol) that are more stable compared to anandamide (-8.3 kcal/mol) and rimonabant (-10.5 kcal/mol). Moreover, indoles and dibenzazepines appeared to show enhanced affinity with CB2 (-12.9 kcal/mol) compared to anandamide (-7.9 kcal/mol) and 9GF (-9.7 kcal/mol).

This study highlights the potential of ML in exploring chemical space and identifying structures with success probabilities for CB receptors. Our method efficiently screens molecules, thorough evaluates binding, and assesses conformation stability, highlighting potential for cost-effective therapeutic strategies with reduced health risks.

References

1. U. Pagotto, et al. (2005). *Annals of Medicine*, 37, 270-275.
2. F. Rossi, et al. (2018). *International Journal of Molecular Sciences*, 19, 2690.
3. C. Fu et al. (2016). *Journal of Agricultural and Food Chemistry*, 64, 9571-9585.

PROTECTIVE EFFECT OF AN AQUEOUS EXTRACT OF MAQUI BERRY IN AN ACUTE MODEL OF CARBON TETRACHLORIDE-INDUCED LIVER INJURY IN WISTAR RATS

Carlos Enrique Orozco Barrios¹, Mario Gil Moreno Pérez², David Reyes Corona²,
Vicente Sandoval Herrera², Erika Bueno González Bosc³

¹Unidad de Investigaciones Médicas en Enfermedades Neurológicas Centro Médico Nacional Siglo XXI, IMSS. Av. Cuauhtémoc 330, Doctores, Cuauhtémoc, Ciudad de México, CDMX. Teléfono: 5591933329. E-mail: crls2878@gmail.com

²Departamento de Farmacología, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional. Av Instituto Politécnico Nacional 2508, San Pedro Zacatenco, Gustavo A. Madero, 07360 Ciudad de México, CDMX.

³Benemérita Universidad Autónoma de Puebla. 4 Sur #104 Col. Centro C.P. 72000, Puebla de Zaragoza, Puebla, México

Introduction. Acute liver injury and acute liver failure are syndromes characterized by a rapid loss of functional hepatocytes in a patient with no evidence of pre-existing liver disease. Alcohol consumption, infections, neoplasms and exposure to hepatotoxic compounds are the most frequent causes of acute liver damage. Maqui Berry (*Aristotelia chilensis*, MQ) is a species native to Chile which has been recognized for its multiple benefits that are attributed to its high content of polyphenols, as well as its wide variety of anthocyanins and flavonoids that confer effects against oxidative stress and inflammation. **Objective.** In this study, we propose that aqueous extract of MQ is able to protect from carbon tetrachloride (CCL4)-induced acute liver damage due to its strong antioxidant and anti-inflammatory capacity. **Methods.** 32 healthy male rats of Wistar strain between 200 and 250 g were divided into 4 groups: Control group (Vehicle CCl4 (Veh CCl4) and Vehicle MQ (Veh MQ)), Damage group ((CCL4) + Veh MQ), Experimental group (CCl4 + MQ) and Maqui group (Veh CCl4 + MQ). CCl4 was administered orally at a dose of 2 g/kg body weight and Veh MQ or MQ extract were administered by the same route 2 hours after CCl4 or Veh CCl4 administration. Rats were sacrificed 24 hours later and whole blood was extracted for biochemical determination of liver function (alanine aminotransferase, ALT and gamma-glutamyl transpeptidase, GGT), total liver extracts for determination of oxidative stress and metabolic damage (Malondialdehyde, MDA and glycogen), and liver tissue for histology (hematoxylin and eosin and Pas staining). **Results.** Aqueous extract of MQ is able to decrease acute liver damage by decreasing serum ALT but not GGT concentration, decreases oxidative stress marker MDA and increases hepatic glycogen concentration. Histologically, MQ is able to maintain the histoarchitecture of the hepatic lobule and recover the glycogen present in it. **CONCLUSIONS:** The single dose of aqueous extract of MQ is able to protect from acute liver damage produced by CCl4 by decreasing oxidative stress, metabolic damage and maintaining the normal structure of the hepatic lobule.

CHARACTERIZATION OF THE VENOM OF THE SCORPION *NULLIBROTHEAS ALLENII*

Nancy Sarai Ortiz Ventura¹, Fernando Zamudio Zuñiga³, Jimena Isaias Cid Uribe⁴,
Lourival Domingos Possani Postay²

Instituto de biotecnología UNAM, Av. Universidad 2001, Chamilpa, 62210 Cuernavaca, Mor.
E-mail: nancy.ortiz@ibt.unam.mx

Scorpions are cosmopolitan animals that have conquered many ecological environments, except for the poles. There are approximately 2,762 species worldwide, with around 281 found in Mexico. Scorpion venoms contain over 200 compounds, most of which are proteins or peptides, while a smaller amount consists of non-protein compounds whose function is still unknown. The study of non-protein compounds has led to the identification of neurotransmitters, lipids, alkaloids, and benzoquinones in the venom of several scorpion families, which show biotechnological applications. Mexico has only one representative of the Chactidae family, *Nullibrotheas allenii*, a scorpion endemic to Baja California Sur. There are only a few reports about the venom composition of Chactidae scorpions, none of which included non-protein compounds. The venom was obtained by electrical stimulation, solubilized and extracted. We used organic solvents to extract 25 non-protein compounds from *N. allenii* venom. The non-protein compounds were submitted to mass spectrometry determination. These components have a molecular weight composition that ranges between 200-900 Da. They have a color (yellowish), which is a unique characteristic of this venom species. The possible physiological function of these components is now being studied.

References

1. Al-Asmari, A. K., Kunnathodi, F., Al Saadon, K., & Idris, M. M. (2016). Elemental analysis of scorpion venoms. *Journal of venom research*, 7, 16–20.
2. Santibáñez-López, C. E., Francke, O. F., Ureta, C., & Possani, L. D. (2015). Scorpions from Mexico: From Species Diversity to Venom Complexity. *Toxins*, 8(1), 2.

***p,p'*-DDE ALTERS MONOCYTE-TO-MACROPHAGE DIFFERENTIATION MARKERS OF HUMAN PERIPHERAL BLOOD MONONUCLEAR CELLS**

José R. Palacios-Valladares, Christian D. Ortiz-Robles, Omar B. Rivera-Maya, Rosa M. García-Hernández, Mariano E. Cebrian, Emma S. Calderón-Aranda

Centro de Investigación y de Estudios Avanzados (Cinvestav) del Instituto Politécnico Nacional (IPN),
Departamento de Toxicología, Ciudad de México, México. Av. Instituto Politécnico Nacional 2508,
San Pedro Zacatenco. Cd. México, Mexico. CP 07360
scalder@cinvestav.mx

Dichlorodiphenyltrichloroethane (DDT) is an organochlorine pesticide used to combat disease-carrying vectors. Due to the adverse effects on animals and humans, the use of DDT was prohibited decades ago. Even though it is forbidden, detectable levels of DDT and its more persistent metabolite DDE are still present in different matrices. *In vitro* and *in vivo* studies showed that *p,p'*-DDT and *p,p'*-DDE affect immune cell function, such as the macrophages, decrease inflammatory markers production, and resistance to infections. However, no studies have been reported on the effects of these compounds on monocyte-to-macrophage differentiation markers. The objective of the present study was to determine if *p,p'*-DDE affects the differentiation markers of monocytes to macrophages. Human peripheral blood mononuclear cells (PBMCs) were obtained from 30 healthy male volunteers. residents of Mexico City, without occupational exposure history to pesticides. The basal levels of *p,p'*-DDE, and other organochlorine pesticides were quantified in donors' serum and *ex vivo* effects of *p,p'*-DDE were evaluated. Twenty-four organochlorine pesticides, including *p,p'*-DDT, and *p,p'*-DDE, were quantified by gas chromatography with a microelectrode capture detector. The monocytes were stimulated and induced to macrophages with 5 ng/ml of GMC-SF for seven days, in the presence or absence of *p,p'*-DDE (25, 250, 1250, and 2500 ng/ml). The markers CD14, CD16, CD68, and HLA-DR were evaluated at the began and the end of time by flow cytometry. Results. Only levels of *o,p'*-DDT, *p,p'*-DDE, and *o,p'*-DDD were detected in the order of ng/mL. The *p,p'*-DDE at none of the concentrations caused cytotoxicity nor affected the expression of the CD14 and CD16 markers. However, *p,p'*-DDE decreased, inverse to the concentration, the expression of the CD68 and HLA-DR. Data support the idea that *p,p'*-DDE is still present in environmentally exposed humans. Our findings suggest that exposure to *p,p'*-DDE decreases the differentiation of monocytes into macrophages, which leads us to imagine an inadequate response of these cells to an insult and an increase in susceptibility to infections.

This study was funded by the Mexican Council for Science and Technology (grant A1-S-32913).

TRANSCRIPTIONAL CHARACTERIZATION OF THE ANTITUMOR ACTIVITY OF LAHERRADURIN ON AN *IN VITRO* MODEL OF COLORECTAL CANCER

Eduardo Perez Arteaga^{1,2}, Nadia Judith Jacobo Herrera²,
Carlos Guadalupe Pérez Plasencia³, Sergio Juárez-Méndez⁴

¹Facultad de Medicina, Universidad Nacional Autónoma de México, Escolar 411A, Copilco Universidad, Coyoacán, 04360 Ciudad de México. ² Instituto Nacional de Ciencias Médicas y de la Nutrición Salvador Zubirán, Vasco de Quiroga 15, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México. ³ Instituto Nacional de Cancerología, Av. San Fernando 22, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México. ⁴ Instituto Nacional de Pediatría, Av. Insurgentes Sur 3700-Letra C, Insurgentes Cuicuilco, Coyoacán, 04530 Ciudad de México.

Corresponding Author:

Nadia Judith Jacobo Herrera Ph D.

Vasco de Quiroga 15, Belisario Domínguez Secc 16, Tlalpan, 14080 Ciudad de México.
nadia.jacobo@gmail.com Tel.: +52 5516500646

Cancer is the second leading cause of death worldwide. Among the types of cancers with the highest incidence, colorectal cancer (CRC) stands out, ranking 3rd in incidence and 2nd in mortality for both sexes. Treatment of CRC is according to the clinical stage at which it is diagnosed. In more advanced stages, surgery is performed followed by adjuvant chemotherapy. However, recurrence remains a common challenge, with a 5-year survival rate of only 50-60%. So, the search and the continual development of newer and more effective therapies are necessary. In this scenario, natural products could be a rich source of secondary metabolites with anticancer potential. Due to their biological activity, natural products have been extensively investigated for the identification of novel drug candidates. Laherradurin, an acetogenin extracted from different species of *Annoneceae* seeds, exemplifies this approach. It has been reported that laherradurin has growth inhibition activity *in vitro* and *in vivo* CRC models. This activity has been attributed to the laherradurin capacity of binding the electron transport chain complex 1 and blocking its function (Jacobo-Herrera et al., 2019). Also, it has been demonstrated that other acetogenins can inhibit HIF-1 α activity, altering angiogenesis and metabolic processes in breast cancer cell lines (Coothankandaswamy et al., 2010). However, the molecular mechanisms underlying these effects have not been defined yet. In this project, we evaluated the effect of laherradurin on the transcriptome of the CRC cell lines hct116 and sw620 by the Affymetrix Clariom S human whole transcriptome microarray technology. So far, our findings indicate that laherradurin induces differential gene expression in CRC cell lines. We identified 152 downregulated and 244 upregulated genes in HCT116 cell line, while in SW620 cell line, we found 1855 downregulated and 722 upregulated genes. Interestingly, the downregulated genes in both cell lines, are significantly enriched in molecular pathways involved in tumour progression, such as Wnt/B catenin, PI3K/AKT and MAPK signalling pathways.

References

1. Coothankandaswamy, V., Liu, Y., Mao, S.C., Morgan, B., et al. 2010. The Alternative Medicine Pawpaw and Its Acetogenin Constituents Suppress Tumor Angiogenesis via the HIF-1/VEGF Pathway. *J. Nat. Prod.* 73:956–961.
2. Jacobo-Herrera N., Pérez-Plasencia, C., et al., 2019. A Selective Acetogenins and Their Potential as Anticancer Agents. *Front. Pharmacol.* 10:783.

ANTIMICROBIAL AND ANTIFUNGAL PROPERTIES OF METHANOLIC EXTRACTS FROM THREE SPECIES OF MAGNOLIOPSIDA

Angélica Eulalia Pérez Cid¹, Dennis Adrián Infante Rodríguez²,
Mario J. Aguilar-Méndez^{1*}, Suria Gisela Vásquez Morales²

¹Unidad Profesional Interdisciplinaria de Ingeniería Campus Guanajuato Instituto Politécnico Nacional. Silao de la Victoria Gto. México. ²Departamento de Biología, División de Ciencias Naturales y Exactas, Universidad de Guanajuato, Noria Alta SN, Noria Alta, AP 36050, Guanajuato, Guanajuato., México.

*E-mail:mjaguilarm@ipn.mx

Over an extensive period, microorganisms have been a constant presence in our daily lives, some of which are pathogens for humans. Antibiotic therapy has increased the microbial resistance, so we propose the interaction plant components with bacteriostatic/bactericidal activities as an alternative solution. We intend to address this problem with two approaches: Identify new plant species that can provide bioactive compounds to counteract microbial resistance to drugs and to add value to these plants, especially to three species that are endangered according to the The Red List of Magnoliaceae. The study is carried out through the antibiograms test using the Kirby-Bauer method. For this purpose, 13 extracts of Magnolias obtained from three different plant tissues were evaluated: seeds, polyfollicles, and leaves. Our microbial models were *Escherichia coli* (ATCC 25922), *Staphylococcus aureus* and *Aspergillus niger*. Our aim is to evaluate the antifungal and antibacterial activity of methanolic extracts from Magnoliopsida. Furthermore, we will establish the minimum inhibition concentration of the extracts on our microbial models. Positive results were obtained from plant extracts, mainly from seed and polyfollicle extracts against *S. aureus* and *E. coli*, but none against *A. niger*. This suggests broad-spectrum antibacterial activity and no antifungal activity. Inhibition was observed at concentrations of 2400 mg/mL for *Magnolia perezferrarae*, 800 mg/mL for *Magnolia pugana*, and 700 mg/mL for *Magnolia vovidesii* from seed extracts. To ensure the antimicrobial sensitivity of *S. aureus* and *A. niger*, an antibiogram was performed with commercial antibiotics. These results will provide insights to alternatives to antimicrobial therapy, using a variety of bioactive compounds. This represents a more comprehensive and sustainable approach to addressing this important public health issue, while also promoting responsible use of natural resources. Furthermore, this study seeks to contribute to the knowledge of the biodiversity and conservation of Magnoliopsida plant species. By highlighting the importance of these organisms in pharmacological research, their potential as sources of bioactive compounds is emphasized.

URINARY 1-HYDROXYPYRENE AS A BIOMARKER OF EXPOSURE TO POLYCYCLIC AROMATIC HYDROCARBONS IN PREGNANT WOMEN IN THE PUEBLA CITY

Jesús Azahel Piña López, Lucio Antonio Ramos Chávez, Anabella Handal Silva, José Luis Morán Perales, María Eugenia Gensebatt Bonaparte, Wendy Argelia García Suastegui

Benemérita Universidad Autónoma de Puebla
72000 Puebla, Pue.
2222295500-5649
wendy.garcias@correo.buap.mx

Exposure to environmental pollutants during pregnancy can have adverse effects on maternal and fetal health. In this context, 1-hydroxypyrene (1-OHP) is a biomarker of exposure to polycyclic aromatic hydrocarbons (PAHs), which are present in the particulate matter (PM) in the air we breathe. The ability to metabolize these compounds varies among individuals due to genetic polymorphisms in metabolic enzymes such as cytochrome P450 (CYP1A1, CYP2E1) and Glutathione S-Transferase (GSTT1, GSTM1). The aim of this study is to identify if there is a correlation between urinary 1-OHP levels and PM10 and PM2.5 concentrations in the resident area and how genetic polymorphisms in metabolic enzymes influence urinary 1-OHP levels. A sample of 84 pregnant women attended at the Hospital de la Mujer in Agua Santa, Puebla, was included in this study. Urine and blood samples were collected for genetic and 1-OHP analysis, and a questionnaire was applied to obtain information about habits, education level, monthly economic income, and age. Daily PM10 and PM2.5 data were also collected from four air quality monitoring stations (Agua Santa, Las Ninfas, BINE, and UTP) during the gestation period, while urine and blood samples were collected between August and November 2018. Polymorphisms in CYP1A1 (*2A, *2C, *4), CYP2E1 (RsaI), GSTT1 (null variant), and GSTM1 (null variant) were investigated. Data obtained from the questionnaires provided basic demographic information. In the environmental analysis, daily averages of PM2.5 showed that the BINE station reported several daily overtake the standard (Mexican Official Standard NOM-SSA1-025-2021), and the annual averages of PM2.5 at all stations overtake the maxim limits of reference. We observed a positive correlation between urinary 1-OHP concentrations and PM2.5 and PM10 reported at the air monitoring station closest to their residence ($r=0.36$, $P=0.0096$; $r=0.22$, $P=0.046$, respectively). Although genetic polymorphisms are still being analyzed, the hypothesis is that the concentration of 1-OHP in the urine of pregnant women is influenced by polymorphic variants in cytochrome P450 and Glutathione S-Transferase.

Keywords. Genetic polymorphisms, 1-hydroxypyrene (1-OHP), particulate matter (PM10, PM2.5), pregnant women, cytochrome P450, Glutathione S-Transferase.

IMMUNE CHARACTERIZATION EFFECT OF SCORPION VENOM FROM MUNICIPALITY OF CORONADO, CHIHUAHUA

Adrian Rivas Muñoz¹, Carmen Carolina Alvarado González¹,
Iván Arenas Sosa², Gerardo Pável Espino Solís¹

¹Laboratorio de Investigación Traslacional, Facultad de Medicina y Ciencias Biomédicas, UACH, Nuevo Campus Universitario, CP 31125, Chihuahua, Chihuahua, México. ²Instituto de Biotecnología, UNAM, Av. Universidad 2001, Chamilpa, 62210 Cuernavaca, Morelos, México.
E-mail: gespinos@uach.mx

Scorpions are common throughout Mexico, although they are not considered a public health problem everywhere¹. In the state of Chihuahua, while scorpion sting incidents are not as numerous as in central states, there is still a great variety of species and subspecies with significant medical value. Therefore, this study aims to characterize a species of scorpion collected near the municipality of Coronado, Chihuahua. Despite the medical importance of arachnids, there is a general lack of knowledge about them. Although they are often considered “dangerous for humans,” only a few species of spiders and scorpions pose a real threat². Proper identification is crucial. The collected scorpions, identified as “very dangerous” based on their morphology and identification keys, were confirmed to be of great medical importance. This research aims to identify these specimens through DNA to determine their species and the effect of this venoms over different immune cells.

References

1. Ponce Saavedra, J. (2003). “El alacranismo en México: un problema de salud pública”. En *Acta Zoológica Mexicana*, 20(2), 1-10
2. Coddington J A, Levi H W. 1991. Systematics And Evolution Of Spiders (Araneae). *Annual Review of Ecology and Systematics* 22: 565-592.

PREPARATION AND CHARACTERIZATION OF MATRIX MICROSPHERES OF ACETYLSALICYLIC ACID AND ACETAMINOPHEN, USING SODIUM ALGINATE BY THE IONIC GELATION TECHNIQUE

Edith C. Rivera¹, Adrián J. Rivera², José A. Hernández³

Instituto Politécnico Nacional. Unidad Profesional Interdisciplinaria de Ingeniería, Campus Guanajuato.
E-mail: jahernandezma@ipn.mx; Tel. 477-115-6320. CP. 37293

The present project, consisted of producing microcapsules through external ionic gelation of two active ingredients: acetaminophen and acetylsalicylic acid, as a modified release pharmaceutical form. The microcapsules were prepared using sodium alginate, calcium chloride and the active ingredients. Parameters such as morphology and particle size, active ingredient content, encapsulation efficiency, moisture content, among others, were determined. The drug was incorporated into preformed alginate beads (impregnation) or simultaneously incorporated during the gelation phase (encapsulation). Particle size and bead morphology were evaluated by standard optical microscopy measurement for bead production, averaging 1.7 and 1.8 mm for acetaminophen-encapsulated and impregnated beads respectively, while for encapsulated AAS beads and impregnated, an average of 2.4 and 2.2 is maintained, respectively. Obtaining a correlation factor of 99%, which is why they are considered reliable. The active ingredient content and encapsulation efficiency were obtained through quantification in a spectrophotometer and calibration curves, observing that, in encapsulation, drug entrapment was greater compared to impregnation. Thus, in 0.5 g of beads with acetaminophen of each type, an average concentration of 387.1 mg/L is found for the encapsulated beads, and 189.6 mg/L for the impregnated beads. Furthermore, it was determined that in 0.3 g of beads with acetylsalicylic acid of each type, there is an average concentration of 13.61 mg/L for the encapsulated beads, and 11.885 mg/L for the impregnated beads.

SYNERGICAL EFFECT OF SPC13-DIC WITH COMMONLY USED ANTIBIOTICS IN GRAM NEGATIVE, POSITIVE AND MULTI-RESISTANT BACTERIA

Carmen I Rodríguez A.¹, Ma. Del Carmen Gutiérrez Villafuerte¹, Lourival Domingos Possani Postay², Georgina Gurrola-Briones², Fernando Zamudio², Jesús Garza³, Lucero Valladares Cisneros¹ and Luz María Rico²

¹Laboratorio de Neurofarmacología, Centro de Investigación en Biotecnología, Universidad Autónoma del Estado de Morelos, Av. Universidad #1001, Cuernavaca, Morelos, México. ²Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Universidad #1001, Cuernavaca, Morelos, México.

³Instituto Nacional de Salud Pública, CISEI Av. Universidad 655, Cuernavaca, Morelos 62508, México.

E-mail: cira_1607@yahoo.com.mx

In recent years, the practice of combined therapy has shown to have several advantages, which is why the synergy between an antibiotic and an antimicrobial peptide (AMP) becomes a promising alternative in the era of resistance. SPC13, present in the venom of *S. polymorpha*, is a 13kDa AMP with bactericidal against *S. aureus* (MIC of 128µg/ml) and bacteriostatic against *E. coli* (128µg/ml) activities as well as 1.7% of hemolytic activity¹. Therefore, in this work, the fractional inhibitory concentration of SPC13-DIC, a synthetic anionic AMP of 8 amino acids tryptic derivative of SPC13, was explored as a synergist of antibiotics. SPC13-DIC was synthesized on solid phase and purified by HPLC-RP. Once purified, it was tested by the checkerboard method in 4 combinations, among SPC13-DIC and antibiotics used in *S. aureus* infections(ATCC29213): Gentamicin, Amikacin, Cefotaxime, and Vancomycin. Our results showed that 3 of them could have synergy having one indifferent effect (Vancomycin/PAM). Concerning *E.coli* (ATCC25922), the combinations of the 4 antibiotics used (Gentamicin, Amikacin, Cefotaxime, and Nitrofurantoin), only Cefotaxime/AMP showed a synergistic effect while the others, an antagonistic effect. The effect on *K. pneumoniae* was also tested with 2 antibiotics (Cefotaxime and Imipenem); however, no synergistic effect was observed in any combination. The hemolytic activity in human erythrocytes with the testing combinations showed percentages < 3.4%. SPC13-DIC could be considered a synergist of commonly used antibiotics in *S. aureus*, being more susceptible to the presence of the peptide and decreasing the MIC of currently used antibiotics.

Reference

1. Rodríguez-Alejandro C.I and MC Gutiérrez (2019) Antimicrobial Activity of SPC13, New Antimicrobial Peptide Purified from *S. polymorpha* Venom, *Anti-Infective Agents*, 233-238

ESTABLISHMENT OF A DOXORUBICIN-RESISTANT SW620 CELL LINE FOR MULTIDRUG RESISTANCE STUDIES

Rodríguez López Yanet Eunice¹, Octavio Daniel Reyes Hernández¹, Laura Itzel Quintas Granados², Edgar Yebrán Villegas Vázquez¹, Gabriela Figueroa González¹

¹Laboratorio de Farmacogenética, Unidad Multidisciplinaria de Investigación Experimental Zaragoza Universidad Nacional Autónoma de México – Facultad de Estudios Superiores Zaragoza. ²Universidad Autónoma de la Ciudad de México, Colegio de Ciencias y Humanidades, Doctor García Diego 168, Colonia Doctores, Alcaldía Cuauhtémoc, 06720. Tel: 5539005899 , E-mail: gabriela.figueroa@zaragoza.unam.mx

Doxorubicin (Dox) is a well-known chemotherapeutic agent capable of suppressing proliferation in cancer cells, triggering apoptosis, and restraining mitosis and cell cycle progression. However, frequent application of Dox results in the emergence of resistance in cancer cells, leading to treatment failure. Multidrug resistance (MDR) in cancer cells to various anticancer drugs remains an obstacle to successful chemotherapy (1). A major mechanism driving the development of MDR is the overexpression of P-glycoprotein (Pgp) in the MDR cell phenotype. Pgp is an ATP-binding cassette (ABC) membrane transporter that mediates the efflux of cytotoxic drugs in cancer cells (2). Leading to MDR and a lower success rate of chemotherapy. Since Dox is the chemotherapeutic agent used in colorectal cancer therapies, studying Dox resistance might enhance our understanding of this phenomenon. This research focused on the development of a Dox-resistant cell line derived from the colon cancer cell line SW620. First, the parental cell line was gradually exposed to increasing concentrations of Dox (0.0046µM-0.98µM). The inhibitory concentration (IC₅₀) was determined through a Sulforhodamine B colorimetric assay to assess the cytotoxicity of Dox in both parental and resistant colon cancer cells (SW620/Dox). The relative expression level of Pgp was determined by flow cytometry and validated by Western Blot. We successfully obtained a Dox resistant colon cancer cell line (SW620/Dox). We documented the morphological changes observed in SW620/Dox cells, which included round shapes, irregularities, and star-shaped forms. The recovery time was approximately two weeks to acquire a morphology similar to the parental cells. It was determined that the SW620/Dox cell line, having a resistant phenotype, can be used as a research model in studies focused on MDR evasion.

Funding: PAPIIT-IA208422, PAPIIT-IA206724, PAPIIT-IN222321, and PAPIIT-IN221824 to Dirección General de Asuntos del Personal Académico (DGAPA).

References

1. Bukowski K, Kciuk M, Kontek R. Mechanisms of multidrug resistance in cancer chemotherapy. Vol. 21, International Journal of Molecular Sciences. MDPI AG; 2020.
2. Engle K, Kumar G. Cancer multidrug-resistance reversal by ABCB1 inhibition: A recent update. Vol. 239, European Journal of Medicinal Chemistry. Elsevier Masson s.r.l.; 2022.

BIOPHYSICAL AND PHYSICOCHEMICAL CHARACTERIZATION OF NOVEL POTASSIUM CHANNEL BLOCKER 3-FLUORO-5-METHYLPYRIDIN-4-AMINE

Yang Sun^{1*}, Sofía Rodríguez-Rangel^{2*}, Lauren L. Zhang¹, Jorge E. Sánchez Rodríguez² & Pedro Brugarolas¹

¹Department of Radiology, Massachusetts General Hospital and Harvard Medical School, Boston, MA 02114, USA.

²Departamento de Física, Universidad de Guadalajara, Guadalajara, Jalisco 44430, México.

*These authors contributed equally

E-mail: jorge.srodriguez@academicos.udg.mx

4-aminopyridine (4AP) is a specific voltage-gated potassium (K^+) ion channel (K_v) blocker clinically indicated to improve walking ability in people with multiple sclerosis. 4AP binds to exposed K_v channels ($K_v1.1$ and $K_v1.2$, mainly) in demyelinated axons, reducing the leakage of intracellular K^+ and enhancing nerve impulse. It has been shown that addition of fluorine (F) or methyl (CH_3) in position 3 of 4AP produced the 4AP derivatives 3-fluoro-4-aminopyridine (3F4AP) and 3-methyl-4AP (3Me4AP) capable to block the Shaker K_v channel with greater lipophilicity and blockage potency than 4AP, respectively^{1,2}. Based on this fact, and the latent demand to find novel molecules with suitable physicochemical, biophysical and pharmacological properties as well as binding affinity that can potentially be radiolabelled and used as PET radiotracers³, in this work we introduce the 3-fluoro-5-methylpyridin-4-amine (5Me3F4AP), a novel 4AP derivative, with ability to block K_v channels⁴. We measured the ability of 5Me3F4AP to block K_v channels in terms of the half-maximal inhibitory concentration (IC_{50}) evaluated in the Shaker K_v ion channel expressed in *Xenopus* oocytes under voltage-clamp. We characterized the biophysical (IC_{50} and dependence) and physicochemical properties ($logP$, and permeability to an artificial membrane). Our results demonstrate that 5Me3F4AP has a similar efficiency of blockage than 4AP and 3F4AP (IC_{50} (in μM) = 301 vs 293 and 244, respectively). We analyzed the curve by the equation used by Hermann⁴ to determine the electric fraction (f_e), which represents the fraction of the electric field that the compound must cross through the channel pore to reach its binding site. A value of 0.4 was obtained for 5Me3F4AP and 4AP indicating that binds the same site on the Shaker K_v pore. Compared to 3F4AP, 5Me3F4AP exhibits comparable basicity (pK_b = 7.46 ± 0.01 vs. 7.37 ± 0.07), greater lipophilicity ($logP$ = 0.664 ± 0.005 vs. 0.414 ± 0.002) and higher permeability to an artificial brain membrane (P = 88.1 ± 18.3 vs. 31.1 ± 2.9 nm/s). The results suggest that 5Me3F4AP is a good candidate for PET radiotracer development. This work was recently published⁵.

References

1. Brugarolas, P., Sánchez-Rodríguez, J. E., Tsai, H. M., Basuli, F., Cheng, S. H., Zhang, X., ... & Popko, B. (2018). Scientific reports, 8(1), 607.
2. Rodríguez-Rangel, S., Bravin, A. D., Ramos-Torres, K. M., Brugarolas, P., & Sánchez-Rodríguez, J. E. (2020). Scientific reports, 10(1), 52.
3. Brugarolas, P., Wilks, M. Q., Noel, J., Kaiser, J. A., Vesper, D. R., Ramos-Torres, K. M., ... & El Fakhri, G. (2023). European journal of nuclear medicine and molecular imaging, 50(2), 344-351.
4. Hermann, A., & Gorman, A. L. (1981). The Journal of general physiology, 78(1), 63-86.
5. Sun, Y., Rodríguez-Rangel, S., Zhang, L. L., Sánchez-Rodríguez, J. E., & Brugarolas, P. (2024). Scientific Reports, 14(1), 11105.

PHARMACOLOGICAL POTENTIAL OF THE SAP OF *SEDUM RUBROINCTUM* R.T. CLAUSEN FOR THE TREATMENT OF EYE INFECTIONS

Anuar Salazar Gómez¹, Fernando Uriel Rojas Rojas¹, Luis Fernando Hernández Zimbrón¹

¹Escuela Nacional de Estudios Superiores Unidad León, Universidad Nacional Autónoma de México (ENES-León UNAM), Blvd. UNAM 2011, León, Guanajuato 3768, México.
E-mail: asalazarg@enes.unam.mx

Eye infections are among the main reasons for consultation in eye care services¹. In many cases, the initial approach to maintaining good eye health often involves self-treatment using medicinal plants. Ethnobotanical surveys have documented that the leaf juice of certain species of the *Sedum* genus is extracted and used in traditional Mexican medicine to treat eye, skin, and mouth infections^{2,3}. However, limited studies have been conducted to identify the biological activities of these plants. This study aimed to investigate the antimicrobial activity of the leaf juice of *Sedum rubroinctum* R.T. Clausen against three microorganisms associated with eye infections and to evaluate its effect on the viability of human gingival fibroblast. Two consecutive methods were used to obtain the leaf juice. The first method involved compressing the green and red leaves of *S. rubroinctum*, following their traditional medicinal use. The second method consisted of triturating the remaining plant material to obtain a homogenate. The remaining plant material was used to prepare a methanol extract. Total phenolic and flavonoid contents were determined using the Folin-Ciocalteu and aluminum chloride methods. The antimicrobial tests were conducted against the microorganisms *S. epidermidis* ATCC 12228, *S. aureus* ATCC 2592, and *C. albicans* ATCC 10231 using agar diffusion assays and the assessment of anti-biofilm potential using crystal violet. The homogenized red leaves of *S. rubroinctum* exhibited antimicrobial activity against *S. epidermidis* and showed a higher total phenolic content compared to the other samples. Importantly, the leaf juice of *S. rubroinctum* demonstrated no cytotoxicity toward gingival fibroblasts at concentrations up to 10% (v/v) after a 24-h exposure. The methanol extract of red leaves also proved effective, inhibiting the formation of biofilms by *S. aureus* (76%). These findings suggest that the leaf juice of *S. rubroinctum* holds promise as an alternative antibacterial and anti-biofilm agent for the treatment of eye infections. However, further pharmacological studies are necessary to confirm their safety and effectiveness.

References

1. World report on vision. Geneva: World Health Organization;2019. Licence: CC BY-NC-SA 3.0 IGO.
2. García, V. N., Gonzalez, A., Fuentes, M., Aviles, M., Ríos, M. Y., Zepeda, G., Rojas, M. G. (2003). Journal of ethnopharmacology, 87(1), 85-88.
3. Salazar-Gómez, A., Velo-Silvestre, A. A., Alonso-Castro, A. J., Hernández-Zimbrón, L. F. (2023). Pharmaceuticals, 16(10), 1432.

ANTICANCER EFFECTS OF MORIN HYDRATE TARGETING CYTOCHROMES P450 IN PEDIATRIC RHABDOMYOSARCOMA CELLS

Rebeca Santes-Palacios and Lizeth Enid López-Rodríguez

Laboratorio de Toxicología Genética, Instituto Nacional de Pediatría, Insurgentes Sur 3700-C, Insurgentes Cuicuilco, Coyoacán, Ciudad de México, México, 04530.
E-mail: rsantesp@pediatria.gob.mx

In Mexico, pediatric rhabdomyosarcoma (RMS) is the fourth leading cause of cancer-associated death with a range of response to antineoplastic drugs of 20-30%, which decreases the efficacy of chemotherapy. In addition, conventional antineoplastic drugs generate toxicity in most patients. Morin hydrate is a naturally occurring polyphenolic compound that possesses anticancer activity through various mechanisms such as antioxidant activity, modulation of liver metabolism enzymes (cytochromes P450) or inhibition of cell proliferation, so it could be a novel candidate to treat RMS.

Cytochrome P450 enzymes are members of a superfamily of heme proteins crucial in the metabolism of procarcinogens into carcinogens, as well as a variety of anticancer drugs. They play a significant role in tumor evolution and the tumor's response to treatment. Therefore, the modulation of their expression and activity has been considered a mechanism in the search for new anticancer agents.

To determine if morin hydrate modulates the gene expression of some cytochromes P450 involved in RMS pathogenesis and the response to chemotherapy, *in vitro* experiments were performed in a pediatric RMS cell line (ATCC CRL-2061) treated with morin hydrate for 48 h. Specifically, qRT-PCR experiments revealed that after 48 hours of treatment with morin hydrate (150 μ M), the gene expression of CYP3A4, CYP1B1, and CYP2E1 increased by 4-fold, 34-fold, and 16-fold, respectively, compared to control cells. This modulation was further characterized for CYP1B1 regulation through the aryl hydrocarbon pathway, showing a decrease in *AHR* and *ARNT* levels by 1.5-fold and 1.9-fold, respectively, and an upregulation of *AHRR* by 1.3-fold.

Molecular docking suggests that π - π stackings between morin hydrate and phenylalanine of the cytochrome P450 catalytic site are important for protein-ligand interaction.

These findings collectively suggest that morin hydrate influences key enzymes involved in RMS pathogenesis and treatment response. Thus, morin hydrate holds promise as a potential therapeutic agent for RMS, warranting further investigation into its mechanisms of action and therapeutic efficacy.

IN VITRO GENE THERAPY MODEL TO ASSESS THE ANTICANCER ACTIVITY OF THE PROAPOPTOTIC PEPTIDE CTMP-4

Ma. Teresa Silva-Guzmán, Elizabeth Ochoa-Muñoz, Karla Ramírez Estrada, Isaías Balderas-Rentería, Aldo O. González-Cruz, Eder Arredondo-Espinoza

Universidad Autónoma de Nuevo León, UANL, Facultad de Ciencias Químicas, Laboratorio de Farmacología Molecular y Modelos Biológicos. Av. Guerrero S/n, Treviño, 64570 Monterrey, N. L., México.
eder.arredondosp@uanl.edu.mx

Currently, among one million publications related to cancer treatment, over 500 mention pro-apoptotic peptides as agents of interest. At this point, the interest in CTMP-4 arises, which has been identified as a derivative of the C-terminal modulator protein (CTMP) therefore, is capable of inducing apoptosis by inhibiting the Akt pathway. Recognising the need to evaluate the intracellular activity of the pro-apoptotic peptide CTMP-4, we propose a tetracycline-induced stable expression system using a cellular model that employs MCF7 and A549 cell lines to assess its cytotoxic effect, crucial property for future cancer therapies.

To accomplish this, the coding sequence of CTMP-4 was retrieved from Simon et al. (2009) and synthesised into pUC57 by GenScript. Cloning was performed using the restriction sites KpnI and BamHI in the pTRE3G-ZsGreen1 expression vector, was confirmed by PCR. Using a tetracycline-induced stable system, we achieved cellular expression and determined anticancer effects of the pro-apoptotic peptide in MCF7 and A549 cell lines. This provides a framework for studying cancer-targeted cytotoxic activity in a stable cell line model for future assays.

References

1. Wang L, Wang N, Zhang W, Cheng X, Yan Z, Shao G, et al. Therapeutic peptides: current applications and future directions. *Signal Transduct Target Ther.* 2022 Dec 14;7(1):48.
2. Li. CM, Haratipour P, Lingeman RG, Perry JJP, Gu L, Hickey RJ, et al. Novel Peptide Therapeutic Approaches for Cancer Treatment. *Cells.* 2021 Oct 27;10(11):2908.
3. Simon PO, McDunn JE, Kashiwagi H, Chang K, Goedegebuure PS, Hotchkiss RS, et al. Targeting AKT with the proapoptotic peptide, TAT-CTMP: A novel strategy for the treatment of human pancreatic adenocarcinoma. *Int J Cancer.* 2009 Aug 15;125(4):942–51

DOES SPC13, AN ANTIMICROBIAL ACTIVITY FOUND IN THE VENOM OF THE *SCOLOPENDRA POLYMORPHA*, ALSO HAVE HISTONE H3 PROPERTIES?

¹Lucero Valladares Cisneros, ¹Ma. Del Carmen Gutiérrez Villafuerte

Universidad Autónoma del Estado de Morelos. ¹Centro de Investigación en Biotecnología. Av. Universidad No. 1001. Col Chamilpa. Cuernavaca, Morelos. C. P. 62209. (777)3297057 Ext. 3522, email: carmengu@uaem.mx

Antimicrobial peptides are crucial elements of innate immunity for the protection of organisms against pathogens, these peptides have been found in plants, animals, and other organisms.

Furthermore, histones with antimicrobial properties have been also proposed as components of the innate defense system, acting against pathogenic microorganisms in vertebrates and invertebrates (1, 2).

Recently, our group reported an antimicrobial peptide (SPC13), extracted from the venom of *Scolopendra polymorpha*, with a molecular weight of 13 kDa (3). Its partial sequence showed a sequence identity of 98% concerning histone H3 of *Scolopendra viridis* (GenkBank: DQ222181.1). Therefore, in this study, we were interested in determining if SPC13 has characteristics of a functional histone too.

The extraction of the venom from *S. polymorpha* was performed by mechanical stimulation of the forcipules. Thereafter, the SPC13 peptide was purified by 16% SDS-PAGE and electroelution, and its antimicrobial activity was assayed against *S. aureus* by the agar diffusion method. DNA and histones were obtained from the whole body of *S. polymorpha* according to Pecina (4) and Sidoli (5) respectively. SPC13 and histones recognition assay was performed by western blot with anti-histone H3; subsequently, the sequences of histone H3 were aligned with the partial sequences of SPC13. Finally, a DNA-Protein binding assay was performed on a 1% agarose gel in which the histone binding was incubated with either DNA or SPC13 peptide to observe its interaction. Results showed that, although the anti-histone H3 antibody did not recognize SPC13, the DNA-Protein binding test demonstrated that SPC13 can interact with DNA.

References

1. Kawasaki, et al. (2003). *Biochemical and Biophysical Research Communications*, 312:1082–1086.
2. Lee, D., et al. (2009). *Journal of Investigative Dermatology*, 129: 2489–2496.
3. Rodríguez-Alejandro, C. and Gutiérrez, M.C. (2018) enviado
4. Pecina, Q. V., (2008) *Protocolo para la extracción de adn de insectos*.
5. Sidoli, S., et al. (2016). *J. Vis. Exp.* (111), e54112. Pag. 1-11. Kawasaki, et al. (2003). *Biochemical and Biophysical Research Communications*, 312:1082–1086.

IDENTIFICATION OF ANTI-CD36 ANTIBODIES AND CHARACTERIZATION OF THEIR BIOLOGICAL ACTIVITY

Marco Velasco-Velázquez¹, Cecilia Mata-Cruz¹, Keila M. Gómez-Castellanos²,
Juan Carlos Almagro², Mayra Pérez-Tapia²

¹Department of Pharmacology, School of Medicine, UNAM. 2ENCB, IPN
Ciudad Universitaria, Coyoacán, CDMX CP 04510. Tel. 55 56232282
marcovelasco@unam.mx

CD36 is a membrane protein that is expressed in multiple cell types such as adipocytes, macrophages, platelets, endothelial cells, cardiomyocytes, dendritic cells, epithelial cells, erythrocytes, and muscle cells. CD36 is a receptor for ox-LDL and fatty acids that is overexpressed in different types of cancer promoting tumor development, metastasis, drug resistance, stemness, and modulation of the anti-tumor immune response. Therefore, CD36 is a potential therapeutic target in cancer. Herein, we aimed to obtain a CD36-blocking antibody fragment.

Identification of anti-CD36 was carried out by solid-phase panning of a semisynthetic library of human scFv displayed in filamentous phages. Positive clones were produced in *E. coli* bacteria and the supernatants were purified by protein L affinity chromatography. The binding of the purified scFv to CD36 on cells (HepG2 and THP-1-derived macrophages) was evaluated by flow cytometry. The effect of selected clones on CD36 function was assessed by incubating cells with fluorescent CD36 ligands (palmitic acid or oxLDL) and quantifying their intake by flow cytometry. The phenotypic effect of one function-blocking scFv clone was analyzed in sphere formation assays with HepG2 cells and in lipid-droplet accumulation assays in THP-1-derived macrophages.

Clone D11 obtained from our panning bound to both recombinant and membranal CD36. In competition assays we demonstrated that D11 epitope overlaps with that of the blocking antibody JC63.1. D11 reduced the cell intake of palmitic acid and oxLDL in cellular assays and impaired tumorsphere-formation efficiency in HepG2 cells and lipid-droplet accumulation in macrophage-like cells.

Our results demonstrated that the anti-CD36 scFv clone D11 effectively blocks CD36 functions. This scFv will be further developed in order to obtain an antibody with therapeutic potential.

Supported by CONAHCYT A1-S-18285 and PAPIIT UNAM IN206622.

LITHIUM-INDUCED REGULATION OF CELLULAR PROCESSES IN COLORECTAL CANCER: AN *IN VITRO* ANALYSIS

Edgar Yebrán Villegas Vázquez¹, Ximena Paola Becerril Vigueras¹, Samantha Andrea Porras Vázquez¹, Jorge Manuel Almanza Torres¹, Laura Itzel Quintas Granados², Octavio Daniel Reyes Hernández¹, Gabriela Figueroa González¹

¹Laboratorio de Farmacogenética, Unidad Multidisciplinaria de Investigación Experimental Zaragoza, Facultad de Estudios Superiores Zaragoza, Universidad Nacional Autónoma de México. ²Universidad Autónoma de la Ciudad de México, Colegio de Ciencias y Humanidades, Doctor García Diego 168, Colonia Doctores, Alcaldía Cuauhtémoc, 06720. E-mail:gabriela.figueroa@zaragoza.unam.mx

Cancer is a process involving the growth and multiplication of abnormal cells, resulting in the development of a tumor. All over the world, cancer remains one of the leading causes of death. Colorectal cancer (CRC), which includes cancer of the colon and rectum, is caused by the aberrant proliferation of glandular epithelial cells in the colon¹. Worldwide, CRC is the four most diagnosed type of cancer and the fifth-leading cause of cancer-related mortality, representing a significant public health problem². The treatment of CRC mainly includes surgical procedures, chemotherapy, and radiotherapy. The development of additional chemotherapeutic agents is still necessary³. In this work, we evaluated the cytotoxic effect of lithium carbonate (Li_2CO_3) on CRC tumor lines to position this salt as a potential alternative therapy for this type of cancer. The cytotoxic effect of Li_2CO_3 was evaluated at both cellular and molecular levels. First, we determined the half inhibitory concentration (IC_{50}) of Li_2CO_3 in CRC cell lines (HCT-116 and SW-620) and in non-tumoral cells (CRL1790). Further, the antiproliferative activity of lithium was assessed using the Sulforhodamine B (SRB) assay, and the apoptotic activity was evaluated using the TUNEL assay. At the molecular level, we evaluated the expression of genes involved in proliferation, apoptosis, and autophagy, such as *gsk3 β* , *cyclin d*, *lc3*, *cas3*, and *bcl-2*, by semiquantitative PCR. Finally, the effect of lithium on autophagic biomarkers (p62, LC3) at the post-translational level was determined through Western Blot assay using β -actin as the loading control. The IC_{50} values for Li_2CO_3 were 8.14 mM for HCT-116 cells, 16.5 mM for SW-620 cells, and 13.7mM for CRL1790 cells. The evidence indicates that the lithium salt induces DNA fragmentation in HCT-116, SW-620 and CRL1790 cells. Moreover, Li_2CO_3 trigger apoptosis and increase the expression of genes *bcl-2* and *cas3* associated with apoptotic cell death. Interestingly, we observed a decrease in the expression of genes *gsk3 β* , *cyclin d* (associated with proliferation) and increase the expression of *lc3* (associated with autophagy). Finally, the expression of p62 and LC3- β proteins increase in the presence of Li_2CO_3 . In summary, Li_2CO_3 induce apoptosis and upregulate the expression of *cas-3* and *bcl-2*, genes associated with apoptotic cell death, inhibits cell proliferation, and promotes autophagy in CRC cell lines.

Funding. PAPIIT-IA206724 and PAPIIT-IN221824. E.Y.V.-V. is grateful to Dirección General de Asuntos del Personal Académico for a postdoctoral scholarship

References

1. Hanahan, D., Hallmarks of cancer: new dimensions. *Cancer discovery*, 2022. 12(1): p. 31-46.
2. Ferlay, J., et al., Global cancer observatory: cancer today. International Agency for Research on Cancer. Lyon, France, 2020..

CHLOROPHYLL QUANTIFICATION IN AQUATIC PLANT (*ELODEA CANADENSIS*) EXPOSED TO HEAVY METAL STIBNITE (Sb_2S_3) IN BIOASSAY

Ana Paola Rivera Torres¹, Luis Adolfo Ceniceros-Morales¹, Maria Guadalupe Nieto-Pescador¹,
Rene Homero Lara-Castro¹, Patricia Ponce-Peña¹, Francisco Javier Rios-Fránquez¹,
Gerardo Alfonso Anguiano-Vega^{1*}

¹Facultad de Ciencias Químicas, Durango, Universidad Juárez del Estado de Durango Av. Veterinaria s/n Circuito Universitario Col. Valle del sur C.P. 34120. Durango, Dgo, México.

*E-mail: gerardo.anguiano@ujed.mx

Chlorophyll is a green pigment in plants that absorbs light energy and transforms it into chemical energy through photosynthesis. Heavy metals can inhibit chlorophyll production and reduce plants' ability to produce energy and growth. Antimony (Sb) is reported as a heavy metal of high toxicity for animal species and humans mainly; however, the possible physiological effects that it may generate in plant species are unknown in detail. The main source of Sb contamination is through the dissolution of the mineral Stibnite (Sb_2S_3). The effect of Sb_2S_3 on the chlorophyll content in the aquatic plant model of the plant *Elodea canadensis* was examined to understand the impact of metal accumulation on the chlorophyll content of the aquatic plant, for this purpose, was determined by spectrophotometry the concentration of chlorophyll a, chlorophyll b, and total chlorophyll of thirty *E. canadensis* plants isolated and under experimental conditions, exposed in to 10 different bioassay to concentrations of 1 to 2000 parts per million of Stibnite (Sb_2S_3). A significant inhibitory effect was found on chlorophyll-a, chlorophyll b, and total chlorophyll in the plant. And a significant decrease in the overall chlorophyll content in *E. canadiensis* after accumulation of Sb_2S_3 during 7 days of exposure.

References

1. Shakya K., Chettri M.K., Sawidis T. (2008). Arch Environ Contam Toxicol, 54, 412-421.
2. J Val, L. Heras, E. Monge (1985). Nuevas ecuaciones para la determinación de pigmentos fotosintéticos en acetona, 231-238.
3. Haiying Tang, Guiyuan Meng, Junqing Xiang, Athar Mahmud (2022). Toxic effects of antimony in plants: Reasons and remediation possibilities—A review and future prospects, 01-05.

GENOTOXIC EFFECT IN ONION BULBS (*ALLIUM CEPA*) DUE TO EXPOSURE TO SPHALERITE (ZNS)

Gladis Anelsi Fernández Lozano¹, Luis Adolfo Cenicerros-Morales¹, Angel Díaz Duarte¹,
María Guadalupe Nieto-Pescador¹, Rene Homero Lara-Castro¹, Patricia Ponce-Peña¹,
Francisco Javier Rios-Fránquez¹, Gerardo Alfonso Anguiano-Vega^{1*}

¹Facultad de Ciencias Químicas, Durango, Universidad Juárez del Estado de Durango Av. Veterinaria s/n
Circuito Universitario Col. Valle del sur C.P. 34120. Durango, Dgo, México.

*E-mail: gerardo.anguiano@ujed.mx

Heavy metals are chemical elements called colloquially; chemically they can be recognized as highly electron-dense metallic elements and generally of high toxicity. The detailed characterization of the toxic effects of heavy metals in biological systems, including those generated at a genotoxic level, is a topic of great interest for environmental and public health in contaminated sites. The comet assay is a test of genotoxicity. short term; used to detect a wide range of DNA damaging. It is currently used in studies to monitor environmental and occupational exposure in the human or wild population. Zinc (Zn) is a metal that can be considered essential for biological systems, but it must be found in low concentrations; high concentrations of Zn generate an imbalance in the oxidation/reduction system in cells, which can generate severe physiological alterations. Sphalerite (ZnS) is a zinc sulfide mineral, endemic to certain regions of northern Mexico where it can occur with calcareous soils rich in carbonate compounds. In this work, bioassays of exposure to 12,000 parts per million of sphalerite were developed in onion bulbs (*Allium cepa*) for 15 and 30 days, with the intention of evaluating the effect that the presence of carbonates the soils were irrigated with two solutions: one rich in carbonate (CaO_3^{-2} 5mM, SO_4^{-2} 5mM) and another rich in sulfur (SO_4^{-2} 5 mM). Genotoxicity was evaluated by comet assay; the longer the exposure time, the more genotoxic damage occurs in the onion bulbs. Exposure for 30 days to sphalerite is significantly greater than exposure for 15 days. both for the bioassay on carbonates and sulfates. Genotoxic damage in the groups exposed to sphalerite with sulfates for 15 and 30 days was significantly higher than in the groups exposed to sphalerite with carbonates. We can recognize that the presence of carbonate in soils generates a protective effect by reducing genotoxic damage, probably due to the formation of zinc carbonate complexes that reduce the availability of the heavy metal.

EXPLORING LITHIUM SALTS FOR AUTOPHAGY INDUCTION AND CELL VIABILITY: NOVEL THERAPEUTIC STRATEGIES FOR CERVICAL CANCER

Alejandro Israel Castillo Montoya¹, Gareth Omar Rostro Alonso¹, Fernando Daniel García López¹, Edgar Yebrán Villegas Vázquez¹, Octavio Daniel Reyes Hernández¹ and Gabriela Figueroa González¹

Universidad Nacional Autónoma de México-Facultad de Estudios Superiores Zaragoza.
Batalla 5 de Mayo SN, Ejército de Oriente Zona Peñón, Iztapalapa, 09230, Ciudad de México, CDMX.
gabriela.figueroa@zaragoza.unam.mx

Cervical cancer (CC) is the second leading cause of cancer death in women in Mexico and the third leading cause worldwide, with 348,874 deaths reported in 2022¹. Current treatments for CC include surgery, radiotherapy, chemotherapy, and immunotherapy². However, using these procedures depends on the stage of cancer development, with lower survival rates in the most advanced stages. Therefore, exploring alternative treatments is crucial. This study investigates the potential of repositioning lithium salts (LiCl and Li₂CO₃) as an alternative treatment due to their ability to regulate various signaling pathways, such as autophagy and apoptosis³. The potential of lithium salts in autophagy induction was evaluated by analyzing the relative expression of the autophagic flux markers such as LC3BII and p62, as well as the detection of autophagosomal structures by confocal microscopy. Data were validated by flow cytometry, and cell viability was analyzed by the tetrazolium salt (MTT) assay in tumor (HeLa) and non-tumor (HaCat) cells to contrast the effects of this potential treatment between the two *in vitro* models. Finally, our findings contribute to the repositioning of lithium salts for cancer treatment, as they offer a higher success rate and fewer adverse effects compared to current anticancer treatments.

Funding. PAPIIT-IA208422, PAPIIT-IA206724, PAPIIT-IN222321, and PAPIIT-IN221824 to Dirección General de Asuntos del Personal Académico (DGAPA).

References

1. Ferlay J., et.al. (2024). Global Cancer Observatory: Cancer Today. Lyon, France: International Agency for Research on Cancer.
2. Wang, J.-J., et.al. (2018). Tumor microenvironment: recent advances in various cancer treatments. *Eur Rev Med Pharmacol Sci* (vol. 22, 3855-3864).
3. Villegas-Vázquez, E. Y., et.al. (2023). Lithium: A Promising Anticancer Agent. *Life* (Vol. 13, Número 2).

The background of the entire page is a composite image. On the left, there is a close-up of a green leaf with prominent veins. On the right, there is a glowing orange and yellow molecular structure, possibly representing a protein or a complex of atoms, with various spheres and connecting lines. The overall color palette is warm, dominated by oranges, yellows, and greens.

POSTER SESSION

XXXIV NATIONAL MEETING OF BIOCHEMISTRY

OTHERS

MESENCHYMAL-AMOEBOID TRANSITION IN TRIPLE-NEGATIVE BREAST CANCER LINE, MDA-MB-231

Arturo Aguilar-Rojas^{1*}, Daniela Rodríguez-Cruz^{1,2}, Mauricio Flores-Fortis^{3,4}, Eunice López-Muñoz¹, Miguel Ángel López Valle⁵, Elena Aréchaga Ocampo⁴

¹Mexican Social Security Institute, Medical Research Unit in Reproductive Medicine, High Specialty Medical Unit in Gynecology and Obstetrics No. 4, Mexico City, Mexico. ²Center for Research and Advanced Studies of the National Polytechnic Institute, Department of Molecular Biomedicine, Mexico City, Mexico. ³Metropolitan Autonomous University, Cuajimalpa Unit, Engineering and Natural Science Doctoral Program, Mexico City, Mexico. ⁴Metropolitan Autonomous University, Cuajimalpa Unit, Department of Natural Science, Mexico City, Mexico. ⁵Mexican Social Security Institute, Breast Tumor Service, High Specialty Medical Unit in Gynecology and Obstetrics No. 4, Mexico City, Mexico.

*a_aguilar@unam.mx

Breast cancer (BC) is the leading cause of cancer-related death among women. The high mortality rate is mainly due to metastasis. During tumor progression, the extracellular matrix (ECM) changes and produces more type-I collagen, which increases tumor stiffness. This alteration impacts cell migration, invasion, and metastasis. Specifically, cancer cells change their gene expression to promote the epithelial-to-mesenchymal transition (EMT). Then, a transition from mesenchymal to amoeboid (MAT) migration is observed. With these changes, malignant cells can migrate more easily through the stiffer microenvironment. Despite the importance of MAT, many aspects of it remains unexplored due to the difficulty to replicate in *in vitro* systems the conditions observed *in vivo* samples.

To address this limitation, we developed a three-dimensional (3D) growth system that replicates the different levels of stiffness observed during breast tumor progression. We used this model to study the migration and invasion of the Triple-Negative BC (TNBC) cell line MDA-MB-231, which is able to promote metastasis. Our results indicate that an increase in ECM stiffness induces a transition of the cells to a rounder morphology with bleb-like protrusions. We quantified how this transition is associated with more persistent migration, enhanced invasion capacity, and reduced secretion of matrix metalloproteinases. Our findings suggest that the proposed 3D growth conditions, especially those with high collagen concentrations, mimic key features of MATs, providing a new platform to study the physiology of migratory transitions and their role in BC progression.

This work was supported by the “Coordinación de Investigación en Salud-IMSS”, Project No: R-2018-785-126. D. R-C. is postgraduate students from the “Programa de Maestría en Biomedicina Molecular, Departamento de Biomedicina Molecular, CINVESTAV, México”. M. F-F is postgraduate student from the “Posgrado en Ciencias Natural e Ingeniería, Universidad Autónoma Metropolitana, Unidad Cuajimalpa, México”. They respectively received scholarships No. 1102304 and No. 472137 from “Consejo Nacional de Humanidades, Ciencia y Tecnología (CONAHCYT-México)”.

TOWARDS BIG DATA ANALYSIS OF A SUPERBUG'S MOLECULAR EPIDEMIOLOGY

Alejandro Aguilar-Vera, Valeria Mateo-Estrada, Alfredo Hernández, Santiago Castillo-Ramírez

Programa de Genómica Evolutiva, Centro de Ciencias Genómicas UNAM.
Av. Universidad s/n Col. Chamilpa, Cuernavaca Morelos México. C.P. 62210.
E-mail: aaguilar@ccg.unam.mx

Acinetobacter baumannii is a major public health problem due to its resistance to last-resort antibiotics, such as carbapenems. The World Health Organization classifies it as a priority 1 critical bacteria and new antibiotics are urgently needed to contain it. Most genomic epidemiology studies are focused on hospitals in different regions of the world. However, if we want to understand the dissemination of this organism under the One Health approach, global studies considering multiple hosts (human, animal, or environmental isolates) are needed. The rapid increase in publicly available genomes allows the implementation of big data approaches to better understand the transmission of *A. baumannii*. The implementation of a massive *A. baumannii* database was designed implementing innovative methodologies that facilitate the obtention of information and the better execution of bioinformatics programs, seeking the optimization of computing resources. We started with 20,188 genomes acquired from public databases that included all levels of assembly. A quality analysis allowed the correct species assignment, based on Average Nucleotide Identity (ANI), and generated a subset of 14,273 genomes, which were manually curated to obtain the greatest number of metadata, classified according to the host. Thus, 87.2% of the genomes belonged to human-related sources, 2.98% corresponded to environmental isolates, 2.78% came from animals and 7.02% could not be assigned to a class. The use of ANI-based intraspecies diversity units was a fundamental part of the implementation as it added an unprecedented level of granularity to the database, which in turn provides the ability to analyze the data to a higher level of resolution. This database will allow a much finer molecular epidemiology of this bacterial species and help with the identification of possible pandemic and novel clones. In addition, it will allow studies on the biological characteristics of the variants that infect non-human hosts to investigate whether they can be a potential source of infection or protection for humans. Importantly, this database will be also very helpful to better characterize the microevolution and spread of the genetic basis of virulence and resistance of this bacterium.

STUDY OF THE STABILITY OF THE RB PROTEIN AND ITS MODULATION BY MDM2

Gabriel Alonso Pérez, Jesús Hernández Monge, Vanesa Olivares Illana

Laboratorio de Interacciones Biomoleculares y Cáncer, Instituto de Física, Universidad Autónoma de San Luis Potosí, Privadas del Pedregal #1570 Av. Chapultepec, San Luis Potosí, San Luis Potosí.
Phone: +524442164132 E-mail: gabrielalonsoperez06@gmail.com

The Rb protein was the first tumoral suppressor discovered, as well as the first related to a malignancy, particularly to cancer of the retina thereby its name “Rb” for Retinoblastoma. Alongside p53, Rb and other factors are related to cell cycle regulation, cell survival, mitosis, and other several stress signals. Therefore, malfunctioning of these proteins can trigger the loss of cell cycle control thus promoting overgrowth of tumors, which could cause the apparition of cancer.

The Rb protein has a lot of functions in the cell, one of the most important is the regulation of the cell cycle by inhibiting the E2F's family factors which are essential to foster the cell cycle from G1 to S phase.³ To achieve this, the Rb protein undergoes posttranslational modifications such as phosphorylation. Rb is a protein with fourteen phosphorylatable residues, each can potentially cause conformational changes that affect the binding properties with other proteins.³

The Rb regulation is carried out by MDM2, an E3 ubiquitin ligase that is involved in its degradation via proteasome either dependent or independent of ubiquitin modification.⁴ However, under genotoxic stress the MDM2 protein switches into a translation factor that enhances the synthesis of the Rb protein.¹ Still, our group determined that in the M phase, Rb is subjected to degradation by MDM2.

Because Rb is a protein with versatile conformations, in this work we aimed to see how the punctual phosphorylation of Rb may affect the stability of this protein in the cell, either in the presence or not of MDM2.

References

1. Hernandez-Monge J, Martínez-Sánchez M, Rousset-Roman A, Medina-Medina I, Olivares-Illana V. (2020) MDM2 regulates Rb levels during genotoxic stress. *EMBO Rep.* 2021 Jan 7;22(1). Epub 2020 Nov 13.
2. Polager, S., & Ginsberg, D. (2009). p53 and E2f: partners in life and death. *Nature Reviews Cancer*, 9(10), 738–748. doi:10.1038/nrc2718
3. Rubin, S. M. (2013). *Deciphering the retinoblastoma protein phosphorylation code. Trends in Biochemical Sciences*, 38(1), 12–19. doi:10.1016/j.tibs.2012.10.007
4. Uchida, C., Miwa, S., Kitagawa, K., Hattori, T., Isobe, T., Otani, S., ... Kitagawa, M. (2004). *Enhanced Mdm2 activity inhibits pRb function via ubiquitin-dependent degradation. The EMBO Journal*, 24(1), 160–169. doi:10.1038/sj.emboj.7600486

MANIFESTACIONES BUCALES PRESENTES EN LOS PACIENTES CON ENFERMEDAD DE PARKINSON DE LA CIUDAD DE DURANGO Y SU RELACIÓN CON LA PROGRESIÓN DE ESTA ENFERMEDAD

Leslie Mariana Apodaca Ayala¹, Sergio Manuel Salas Pacheco¹, Omar Tremillo Maldonado¹, Rogelio González González², Francisco Xavier Castellanos-Juárez², Osmel La Llave León²

¹Facultad de Odontología, Universidad Juárez del Estado de Durango
Email: lesliem_003@hotmail.com

Palabras clave: Enfermedad de Parkinson, manifestaciones bucales, cavidad bucal.

Introducción. Los pacientes con enfermedad de Parkinson (EP) presentan manifestaciones bucales causadas por la dificultad para realizar una correcta higiene bucal, efectos secundarios de fármacos y la disminución del flujo salival. Entre estas manifestaciones encontramos enfermedad periodontal causada principalmente por la deficiente higiene bucal de los pacientes debido a la dificultad de realizar esta actividad de manera eficiente ocasionada por los síntomas motores o movimientos involuntarios; de igual manera se presentan úlceras bucales, malestar de boca ardosa y xerostomía. Estas manifestaciones presentan una serie de malestares que el paciente percibe a lo largo de su enfermedad, las cuales no se toman en cuenta como fenómenos relacionados a la EP.

Objetivo. Identificar las manifestaciones bucales en pacientes con enfermedad de Parkinson y analizar su relación con la progresión de la enfermedad.

Materiales y métodos. Se realizaron diagnósticos de xerostomía utilizando la técnica de flujo salival no estimulado según Navazesh (FSNE), de úlceras bucales mediante un examen clínico complementario y una inspección sistematizada de la mucosa bucal, al igual que malestar de boca ardosa en la que se utilizó la Clasificación clínica del SBA Lamey y Lewis y, por último, se utilizó el Índice de Necesidades de Tratamiento Periodontal de la Comunidad (CPITN) para la enfermedad periodontal, de igual manera para evaluar la progresión de enfermedad de Parkinson se realizó el examen de la MDS-UPRDS en 3 pacientes mayores de 70 años.

Resultados. Se obtuvo una media en el UPDRS de 69.6 puntos. Referente a manifestaciones bucales, el 100% dio positivo al diagnóstico de xerostomía, con una media de .22 ml/min, al realizar una correlación estadística se obtuvo una $P= 0.376$, lo cual indica que no importa el nivel de progresión de EP, la secreción salival siempre será baja. El 20% presentó una clasificación tipo 3 de SBA, por otro lado los pacientes que presentaron úlceras tienen una media de UPDRS de 37, mientras los que no presentaron tienen una media de 101.66, al comparar los dos grupos se obtuvo una $P= 0.08$, esta tendencia parece indicar que a menor UPDRS o menos progresión de la EP, mayor la probabilidad de padecer úlceras. En relación a enfermedad periodontal el 80% es candidato a tratamiento de la misma con una media de UPDRS de 50, por otro lado el 20% que corresponde al puntaje más alto del UPDRS de 148, esta condición seguramente está ligada a la enfermedad Periodontal, indicando que esta podría estar relacionada a un mayor puntaje en el UPDRS.

Discusión y conclusiones. Se identificó la presencia de xerostomía, enfermedad periodontal, úlceras bucales y SBA, este último con una baja prevalencia. La frecuencia de estas manifestaciones se encontraron en pacientes con EP avanzada como en pacientes con EP leve.

Referencias

1. Harris Ricardo, J., Fortich Mesa, N., & Díaz Caballero, A. (2013). Fisiopatología y manifestaciones bucales de la enfermedad de Parkinson: Una revisión actualizada. *Avances en Odontoestomatología*, 29(3), 151-157.
2. Silvestre, F. J., Silvestre-Rangil, J., & López-Jornet,
3. P. (2015). Burning mouth syndrome: a review and update. *Rev Neurol*, 60(10), 457-463.

MOLECULAR INSIGHTS INTO FOLATE DYNAMIC METABOLISM IN COMMON BEAN (*PHASEOLUS VULGARIS*) SEEDS DURING STORAGE

Itzel Astrid Aviña-Ávalos^{1,2}, Sara M. Garza-Aguilar², Arturo Tlelo-Reyes², Liliana Elizabeth García-Valencia², Perla Azucena Ramos-Parra², Rocío Isabel Díaz de la Garza^{1,2*}

¹Instituto Tecnológico y de Estudios Superiores de Monterrey, Institute for Obesity Research; ²Instituto Tecnológico y de Estudios Superiores de Monterrey, School of Engineering and Science. Eugenio Garza Sada Sur 2501, col. Tecnológico, Monterrey, Nuevo León. C.P. 64700..

E mail: rociodiaz@tec.mx

Micronutrient deficiencies affect mainly pregnant and lactating woman and young children in developing countries, leading to severe clinical conditions. The common bean (*Phaseolus vulgaris*), a widely consumed grain in Latin America, Africa and Asia, is a high nutritional value crop, being an important source of folate (providing 24.75% of dietary reference value, cooked), also called vitamin B9. The latter is relevant due to their role in amino acid and purine synthesis, crucial for regions where deficiencies are prevalent. However, seed storage, an inevitable process in the supply chain, can alter common bean properties, although little is known about how it affects micronutrient content. This study evaluated the effects of different storage conditions (temperature, relative humidity and time) on the folate contents and profile in three commercial bean varieties in Mexico (Mayocoba, MY; Negro Jamapa, NJ; and Pinto Saltillo, PS). Folate content was quantified by HPLC-electrochemical detector in bean grains during a 180-day storage time at different temperature and relative humidities. Mild temperature and humidity caused an increase in folate levels of 1.4-1.6 at 180-days storage in the three varieties. Transcriptomic analyses showed that this result correlated with changes in gene expression patterns of genes involved in folate biosynthesis and one-carbon (1C) metabolism (p-adj <0.05). The transcription of folate biosynthesis genes showed positive regulation, particularly the expression of the genes *ADCS* (*aminodeoxychorismate synthase*) and *GCHI* (*GTP cyclohydrolase I*), which code for the two committed steps of folate synthesis in the three varieties. The enzyme folylpolyglutamate synthase (FPGS), responsible for folate accumulation and stability, increased in transcripts of 2.53, 5.61 and 2.88 times in the MY, NJ, and PS respectively, this expression pattern correlated with increases in folate polyglutamylation during storage. For the first time, we report molecular activities that lead to micronutrient accumulation in stored seeds after six months. Our results identified optimal storage conditions to keep and improve seed nutritional value; also, they provide a framework for further research on the changes of seed metabolism during storage, an overlooked research area.

COMMON BEAN TRANSCRIPTION FACTORS FROM THE AGL/MADS AND SPL FAMILIES DRIVE THE EXPRESSION OF THE MIR172C GENE, A KEY REGULATOR OF COMMON BEAN N-FIXING SYMBIOSIS

Litzzy Ayra Pardo, Gladys E. Jiménez-Nopala, Carlos de la Rosa Ureña, Sara I. Fuentes Membreño, Mario Ramírez Yáñez, Alfonso Leija Salas, Jesús Montiel González and Georgina Hernández Delgado*

Centro de Ciencias Genómicas, UNAM, Campus Morelos Av. Universidad S/N, Cuernavaca, Mor. 62210.

*777/3290864 gina@ccg.unam.mx

The N-fixing symbiosis (SNF) between legume plant and soil bacteria commonly known as rhizobia, relevant for sustainable agriculture, is a complex process that is tightly regulated in both symbionts. Our group has demonstrated the participation of the microRNA172c (miR172c) – APETALA2-1 (AP2-1) node as a key regulator of the common bean (*Phaseolus vulgaris*) SNF with *Rhizobium etli* (Nova Franco et al., 2015). Similar function of this node was reported for soybean SNF (Wang et al., 2014). In this work we assessed the transcription regulation of the Mir172c gene. Our bioinformatic analysis of the miR17c promoter sequence (5 kb upstream of the ATG start codon) revealed statistically over-represented binding sites for AGL (AGAMOUS-LIKE PROTEIN) (Íñiguez et al., 2015) as well as for SPL (SQUAMOSA PROMOTER BINDING PROTEIN-LIKE) TFs. Yun et al. (2022) reported that in soybean, the SPL9d TF directly binds to the miR172c promoter and activates its expression. We reported the relevance of common bean AGL transcription factors as positive regulators of various processes of the SNF (Ayra et al., 2021). On this basis, we analysed if AGL/(PvFUL) and PvSPL9 TFs regulate the expression of common bean MiR172c gene. To achieve this, we conducted an effector/reporter assay in *Nicotiana benthamiana* leaves transfected with vectors bearing reporter genes coding for fluorescent protein. Our results confirmed that both, AGL/(PvFUL) and PvSPL9 TFs, drive the expression of the Mir172c gene. In addition, we used a reverse genetic approach evaluating the transcript level of MiR172c in common bean transgenic roots/nodules with overexpression or silencing of AGL/(PvFUL) or PvSPL9 TFs. As expected, the amount of miR172c varied depending on the level of expression of each TF in transgenic root/nodules. In this study, we demonstrate that AGL and SPL9 transcription factors regulate MiR172c gene in common bean nodulation, thus playing an important role in the control of SNF.

References

1. Ayra, et al (2021). Front. Plant Sci, 12, 1-17.
2. Iñiguez, et al (2015). Plant Signaling & Behavior, 10, 1-4.
3. Nova-Franco, et al (2015). Plant Physiology, 168, 273-291.
4. Wang, et al (2014). Plant Cell, 26, 4782-4801.
5. Yun, et al (2021). New Phytologist, 233, 1881-1899.

RAPID IDENTIFICATION OF BIOACTIVE COMPOUNDS FROM AGAVE BY MS-DART AS POTENTIAL BIOMARKERS FOR ARQUEOLOGICAL STUDIES

Emanuel Bojórquez-Quintal^{1,7}, Antonio Basurto-Muñoz^{2,3}, Eugenio Mancera², Rafael Montiel³, Hilda Ramos-Aboites³, Fernando González-Zozaya⁴, Carlos Lazcano-Arce⁵, Luis Rojas-Abarca¹, Carlos Cruz-Cárdenas⁶

¹Laboratorio de Análisis y Diagnóstico del Patrimonio, El Colegio de Michoacán, Cerro de Nahuatzen 85, Jardines del Cerro Grande, 59370, La Piedad, Michoacán, México. ²Departamento de Ingeniería Genética, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Unidad Irapuato, Irapuato, Mexico. ³Laboratorio Nacional de Genómica para la Biodiversidad, Unidad de Genómica Avanzada, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Km 9.6 Libramiento Norte Carretera Irapuato-León, Irapuato, Guanajuato, México. ⁴Centro INAH Colima, Instituto Nacional de Antropología e Historia, Colima, México. ⁵Instituto de Investigaciones Antropológicas, UNAM, Ciudad Universitaria, Ciudad de México, México. ⁶Centro Nacional de Recursos Genéticos-Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias, Tepatitlán de Morelos, Jalisco, México. ⁷CONAHCYT.
E-mail: emanuel@colmich.edu.mx; jedabojoquezqu@conacyt.mx

Agaves (*Agave sp.*) and their byproducts have been used since pre-Hispanic times¹ in medicine, food, traditional cuisine, and the production of fermented beverages². However, biomarkers and the metabolic profile of fermented and distilled agave products have remained underexplored for the study of these products^{3,4}. This work presents the rapid identification of bioactive compounds present in fermented agave juice and their potential use for principal component analysis. Contemporary fermented beverages of different geographical origin, production practices and agave species were analyzed. The resulting profiles were used as references in the identification of organic residues of agaves in ceramic vessels of pre-Hispanic origin. The analysis was carried out by Direct Analysis Mass Spectrometry in Real Time (MS-DART) at 300°C in positive mode using directly the fermented agave must, identifying molecular ions and adducts (m/z) of molecules belonging to sugars, phenolic compounds, organic acids, fermentation products and saponins. Of these latter molecules, diosgenin (415 m/z), gentrogenin (429 m/z), hecogenin (431 m/z), chlorogenin (433 m/z), and kammogenin (445 m/z) were identified. Hecogenin and kammogenin were of interest as biomarkers for studies of organic residues in pre-Hispanic vessels associated with agaves.

References

1. Correa-Ascencio, M., et al. (2014). PNAS, 111, 14223-14228.
2. Pérez-Zavala, M.L., et al. (2020). Science of food and Agriculture, 10, 5324-5333.
3. Bermúdez-Bazán, M., et al., (2021). Molecules, 26, 6789.
4. Ojeda-Linares, C., et al. (2021). Foods, 10, 2390

STUDYING NUCLEOPHAGY IN THE LONGEST-LIVED RODENT, THE NAKED MOLE-RAT

Luis Antonio Buendía Sánchez, Laura Cano, Berenice Franco, Susana Castro*

Instituto de Fisiología Celular, UNAM.
04510, +52 (55) 5622-5676, scastro@ifc.unam.mx

Background. Aging is the accumulation of molecular and cellular changes over time that lead to a progressive decline in physical and mental capacity. These changes can cause the accumulation of senescent cells, genome damage, and decreased autophagic capacity. Autophagy is a degradation pathway for intracellular components that diminishes with age, contributing to an increase in senescent neurons (neurosenescence) and the development of degenerative diseases such as cancer, diabetes, Alzheimer's, and Parkinson's. Stimulation of autophagy increases the lifespan of organisms. DNA damage can be physiologically removed by micronuclei, which are eliminated via the autophagic pathway (nucleophagy); their persistence generates senescence, inflammation, and/or genomic instability. The naked mole-rat, *Heterocephalus glaber*, can live up to 40 years without showing signs of aging. Most of their lives, they remain healthy and active, without presenting cancer, neurodegeneration, accumulation of senescent cells, or decreased autophagy. **Objectives.** To analyze whether there are micronuclei in naked mole-rat fibroblasts (NSF) basally and in response to DNA damage, as well as whether they colocalize with components of the autophagic machinery. To evaluate whether the inhibition of autophagy in NSF increases micronuclei and cellular senescence. **Methods.** We will evaluate the response to DNA damage with etoposide in NSF through immunofluorescence against the histone γ H2Ax and Lamin B1 to quantify the presence of micronuclei. LC3B, Beclin1, and LysoTracker will be used as autophagy markers to analyze their dynamics with an autophagy inhibitor: Chloroquine. **Results:** Using higher doses of etoposide in NSF does not represent any advantage over what is already established in mice. Analyzing the dynamics of the damage response by γ H2Ax in naked mole-rat fibroblasts, we can infer that one hour of repair is sufficient for NSF to repair most of the damage caused by etoposide. **Conclusions.** It is still necessary to quantify whether there are micronuclei and whether they are processed through nucleophagy, as well as to use an autophagy inhibitor to compare its dynamics in NSF, in relation to mouse fibroblasts. The immunofluorescences already performed will be used to analyze the dynamics of micronuclei. **Keywords.** Aging, Naked Mole-Rats, Autophagy

IDENTIFICATION OF AIM24 AS A YEAST COMPLEX II MEMBRANE DOMAIN ASSEMBLY FACTOR

Yolanda Camacho-Villasana¹, Ulrik Pedroza-Dávila¹, Sven Dennerlein², Peter Rehling²
and Xochitl Pérez-Martínez^{1*}

¹Departamento de Genética Molecular, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Mexico City, México. ²Institute for Cellular Biochemistry, University of Goettingen, Goettingen, Germany.
³E-mail: xperez@ifc.unam.mx

Unlike the rest of the electron transport chain, all four complex II subunits are encoded in the nucleus¹. Subunits Sdh1 and Sdh2 form the hydrophilic domain in the mitochondrial matrix. On the other hand, subunits Sdh3 and Sdh4 form the hydrophobic domain anchoring the complex to the inner mitochondrial membrane.

Four complex II assembly factors are known (Sdh5-8) which are conserved between yeast and animals². All four proteins are involved in the assembly, stability, or acquisition of prosthetic groups of subunits Sdh1 or Sdh2³. In contrast, it is unknown which proteins promote assembly of the membrane subunits Sdh3 and Sdh4 as well as heme group binding. In order to identify possible complex II chaperone candidates for assembly of the membrane portion, we tagged subunits Sdh3 and Sdh4 with a hemagglutinin epitope in yeast. Afterwards, immunoprecipitation of both proteins was performed, and the binding proteins were identified by LC-MS. Among the eluted proteins, Aim24 showed an interaction with both subunits Sdh3 and Sdh4. Immunoblot analysis confirmed that Aim24 interacts exclusively with the membrane domain subunits and not with the entire complex II. The $\Delta aim24$ null mutant was characterized by growth conditions, complex II subunit levels and in-gel activity in native conditions. In conclusion, Aim24 is an assembly factor of complex II membrane domain.

References

1. Bezawork-Geleta, A., Rohlena, J., Dong, L., Pacak, K., & Neuzil, J. (2017). Trends in biochemical sciences, 42(4), 312-325.
2. Van Vranken, J. G., Na, U., Winge, D. R., & Rutter, J. (2015). Critical reviews in biochemistry and molecular biology, 50(2), 168-180.
3. Sharma, P., Maklashina, E., Cecchini, G., & Iverson, T. M. (2020). The roles of SDHAF2 and dicarboxylate in covalent flavinylation of SDHA, the human complex II flavoprotein. Proceedings of the National Academy of Sciences, 117(38), 23548-23556.

LACCASE PROTON RELAY MECHANISM FROM *THERMUS THERMOPHILUS* HB27

María Cristina Cardona Echavarría¹, Enrique Rudiño Piñera¹, Gloria Saab-Rincón² and Ricardo Miranda Blancas¹

¹Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología, UNAM, Av. Universidad 2001, Cuernavaca, Morelos 62210, México. ²Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, UNAM, Av. Universidad 2001, Cuernavaca, Morelos 62210, México.

E-mail: maria.cardona@ibt.unam.mx, ricardo.miranda@ibt.unam.mx.

Laccases are oxidoreductase enzymes that depend of four electrons and four protons for each catalytic cycle, reducing one molecule of oxygen to two water molecules^{1,2}. However, the proton relay mechanism remains unclear for most laccases, including *T. thermophilus* laccase (Tth-MCO)³. This work describes eight new crystallographic structures of Tth-MCO with varying radiation doses. These structures reveal a potential proton relay mechanism from the protein surface to the trinuclear catalytic copper binding site, showing proton transport between residues E451 and D452. An alignment of the amino acid sequences of bacterial and fungal laccases reveals a high conservation of the amino acids E451, D452, and H139, a residue near the surface and close to D452. These conserved amino acids are present in bacteria but not in fungi. Additionally, mutants of the implicated amino acids (E451L, H139S and E451L-H139S) were generated, and their enzymatic activity was measured to determine their kinetic parameters. A significant decrease in oxidoreductase activity was observed, particularly in the E451L mutant, indicating the importance of these residues in the proton relay of this laccase. The correct folding of the mutants was assessed using circular dichroism and molecular dynamics, with artificial intelligence algorithms employed to predict the structures of the mutants. The analyzed structural data provided insights into the proton relay mechanism and were used to elucidate the enzymatic behavior of Tth-MCO. Despite the lack of complete structural information, the multi-step proton relay mechanism appears to be applicable to most bacterial laccases.

References

1. H. Claus, (2004). *Micron*, 35, 93-96.
2. K. Miyazaki (2005). *Extremophiles*, 9, 415-425.
3. H. Serrano-Posada, S. Centeno-Leija, S.P. Rojas-Trejo, C. Rodríguez-Almazán, V. Stojanoff, E. Rudiño-Piñera (2012). *Sect. D Biol. Crystallogr.*, D71, 2396-2411.

RESISTIN AND ADIPONECTIN MODULATE ABCA1 AND ABCG2 TRANSPORTERS IN AN *IN VITRO* PLACENTA MODEL

Bexidiu Castillo-Escoto^{1,2}, Aurora Espejel-Nuñez¹, Sandra Parra-Hernández¹, Arturo Flores-Pliego¹, Araceli Rojas-Bernabé^{2,3}, América Vanoye-Carlo⁴, Sonia Nava-Salazar^{1*}

¹Departamento de Inmunobioquímica, Instituto Nacional de Perinatología, Montes Urales 800, Col. Lomas Virreyes, Del. Miguel Hidalgo, C.P. 11000, Ciudad de México. ²Sección de Estudios de Posgrado e Investigación, Escuela Superior de Medicina, Instituto Politécnico Nacional, Salvador Díaz Mirón, Miguel Hidalgo, Santo Tomás, 11340, Ciudad de México, México.

³Hospital de la Mujer, Secretaría de Salud, Salvador Díaz Mirón 374, Santo Tomás, Miguel Hidalgo, 11340 Ciudad de México, México. ⁴Laboratorio de Neurociencias. Instituto Nacional de Pediatría, Av. Insurgentes Sur 3700-Letra C, Insurgentes Cuicuilco, Coyoacán, 04530 Ciudad de México, México.

*E-mail: s_nava_s@yahoo.com

Pregnant women with obesity generate an imbalance of different inflammatory markers and adipokines, including adiponectin and resistin. Several studies in liver cell and macrophage models suggest that these adipokines regulate some cholesterol transporters, including ABCA1 and ABCG2, two important members expressed in the human placenta, where they regulate cholesterol transport and drug transport. Their regulation in the placenta has been less explored, as well as the impact on the transport of cholesterol from the mother to the fetus and its effect on fetal development. Therefore, the present study aimed to evaluate the effect of resistin and adiponectin on ABCA1 and ABCG2 transporters in an *in vitro* placental model.

The project was approved by the IRB (2023-1-28). For placental explant culture, women with healthy full-term pregnancy, normal pregestational BMI (18.5 -24.9 Kg/m²), and cesarean delivery were included. JEG-3 and BeWo trophoblast cell lines were used, and a HepG2 liver carcinoma cell line was used as a positive control. The effect of adiponectin was evaluated at concentrations of 10 and 20 ng/μL and for resistin at 50 and 100 ng/μL. Expression of ABCA1 and ABCG2 and the transcription factors PPARα, PPARγ, and LXRα was analyzed with TaqMan real-time PCR assays and at the protein level by immunohistochemistry. Descriptive statistics (mean ± SD), one-way ANOVAs and Tukey's multiple comparison tests were performed according to the treatment performed (GraphPad Prism V.8.4.0).

In vitro placental models, a positive correlation was found between resistin concentration and ABCA1 and ABCG2 expression, whereas for adiponectin treatments, it had opposite effects on both genes. ABCA1 and ABCG2 are regulated by adiponectin and resistin, probably by PPARα, PPARγ, and LXRα-dependent pathways. These studies suggest that adiponectin and resistin during pregnancy may influence ABCA1 and ABCG2 levels; these molecules regulate maternal-fetal cholesterol transport, so alterations in this pathway could be involved in fetal development and growth.

RAPID CHARACTERIZATION OF COMMERCIAL ESSENTIAL OILS MAJOR COMPONENTS BY DIRECT METHODS (ATR-IR AND MS-DART)

Ana V. Coria-Téllez¹, Luis Rojas-Abarca¹, Emanuel Bojórquez-Quintal^{1,2}

¹Laboratorio de Análisis y Diagnóstico del Patrimonio, El Colegio de Michoacán, A.C., La Piedad, Michoacán, México. ²CONAHCYT
E-mail: anac@colmich.edu.mx

Essential oils (EOs) are liquid preparations obtained from aromatic plants, herbs and spices, which contain a complex mixture of low molecular weight compounds in different concentrations and wide variety of biological activities. EOs are currently widely marketed as therapeutic auxiliary due to their various health applications¹; however, the quality of commercial EOs can vary due to different factors. This work explores the application of direct DART^{2,3} mass spectrometry and ATR-IR⁴ spectroscopy methods to determine the chemical profile of commercial EOs of thyme, lemon, mint, cypress, oregano, lemongrass, frankincense, lavender, cedar and tea tree. In the DART-MS spectra, it was possible to identify main components, terpenes such as pinene, terpinene, limonene and aromatic compounds of the EOs. In ATR-IR, the specific chemical pattern for each EO is distinguished. Chemical profile, botanical family, traditional use and therapeutic properties contrasted between commercial EOs. The results indicate that MS-DART and ATR-IR can be methods that allow the reliable, rapid and direct identification of molecules present in EOs.

References

1. Predy, V.R. (2016). Essential oil in food preservation, flavor and safety. 3-9.
2. Chacón, C., et al. (2021). *Agronomy*, 11, 1-13.
3. Huber, S., et al. (2022). *Analytical Methods*, 14, 3875
4. Agatonovic-Kustrin, S., et al., (2020). *Applied sciences*, 10, 1-12.

AGRONOMIC AND MOLECULAR EVALUATION OF POLYPLOID TOMATO

Fátima Clarita Cota Ruiz¹, Leandro Alberto Núñez Muñoz¹, Rosa Irma Obando Gonzalez¹, Roberto Ruiz Medrano^{1,2}, América Padilla Viveros², Beatriz Xoconostle Cázares^{1,2}

¹Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Av. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, 07360 Ciudad de México, CDMX.
E-mail: bxoconos@cinvestav.mx

Tomato, a solanaceous plant which produces a colourful fruit with yellow to deep red hue, is one of the most important horticultural crops in the world. Mexico is the largest the producer worldwide with 1,663,316 tons exported in 2019. Due to its importance, and its susceptibility to several biotic and abiotic factors, different strategies to obtain more resistant plants have been developed to generate novel varieties with agronomic characteristics that provide resistance to pests and diseases, as well as tolerance to abiotic stress¹. One way to increase genetic diversity is through the induction of polyploidy; this technique allows the breeder to modify a plant by increasing the number of chromosomes, and thus the proportion of allelic genes. This in principle contributes to the emergence of specific traits, and improves phenotypic variability, plant vigour, resistance to abiotic stress and adaptability to new habitats. To this end antimitotic agents such as colchicine and oryzalin² are generally used. A cherry variety with black fruit was subjected to colchicine treatment, obtaining plants with different degrees of mixoploidy³. Their agronomic performance was also evaluated, and their transcriptome profile was also obtained and compared to untreated plants. The genetic variation obtained in the new varieties and their agronomic characteristics will be presented.

References

1. SAGARPA (2017). Planeación agrícola nacional 2017-2030. www.sader.gob.mx
2. Zhou, K., Fleet, P., Nevo, E., Zhang, X., & Sun, G. (2017). *Scientific Reports*, 7(1), 8503.
3. Obando Gonzalez, R. I. (2022). Tesis de maestría del Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional.

SERCA PUMPS COUNTERACT TETANIC CONTRACTIONS WHILE MAINTAINING THE FORCE AMPLITUDE OF SINGLE TWITCH CONTRACTIONS

Adán Dagnino Acosta; Daniel Perea Ruiz & Kevin E. Villa-Vuelvas

Centro Universitario de Investigaciones Biomédicas de la Facultad de Medicina, Universidad de Colima, Avenida 25 de Julio #965, Colonia Villas de San Sebastián, Colima, Col. México.
E-mail (ADA): dagninoa@ucol.mx

The dynamic interplay between calcium and the critical components that regulate the contractile mechanism of skeletal muscle has been investigated for the last few decades. It is now clear that calcium released by type 1 Ryanodine Receptors (RyR1), activated by an electrical stimulus sensed by DHPR channels physically coupled to RyR1, bind to troponin C, which in turn activates contraction by unblocking the interaction of myosin with actin.

Interestingly in fast-twitch skeletal muscle a strong calcium buffering is exerted by parvalbumin, limiting the transient increases of “cytosolic” calcium to 10-20 ms. In this relative fast process of calcium buffering, it is unclear the role of SERCA pumps (rate of calcium pumping = $2.4 \text{ Ca}^{2+} \text{ pump}^{-1}\text{s}^{-1}$)¹ in the kinetic behavior of contraction and persistence of contractions (fatigue). The goal of this study was to establish the temporal effect of SERCA pump inhibition on tetanic and single twitch contractions². For this purpose, we employed EDL and FDB muscles isolated from C57BL6 mice to measure the impact of SERCA pump inhibition with CPA on both, the kinetic of force generation and free calcium increases when muscle is electrically stimulated.

The obtained results show that inhibition of SERCA pumps with CPA modify the relaxation kinetic of tetanic contractions. However, this behavior is not dramatically affecting the decay half-times of fitted exponential functions. The effect of SERCA pumps inhibition on the calcium kinetic measured in single fibers isolated from FDB muscle was a conditional slowing of the recovery phase of transient calcium increases (induced by electrical stimulation).

In conclusion, SERCA pumps conditionally counteract the cytoplasmic calcium increases even in the presence of parvalbumin, suggesting an effect of reduced interaction time of calcium with troponin C during the relaxation phase of contraction.

References

1. Barclay, C. J., Woledge, R. C., & Curtin, N. A. (2007). *J. Mus Res Cell Mot*, 28(4–5), 259–274.
2. Lee, C. S., Dagnino-Acosta, A., et. al. (2015). *Skeletal Muscle*, 5(1).

PURIFICATION AND CHARACTERIZATION OF THE ESTERASE TA0887 FROM *THERMOPLASMA ACIDOPHILUM*

Alejandro Delgado Rey¹, M. Leticia Santos Martínez¹ and Samuel Lara-González¹

¹Instituto Potosino de Investigación Científica y Tecnológica A.C., División de Biología Molecular, San Luis Potosí, 78216
E-mail: samuel.lara@ipicyt.edu.mx

Microbial esterases have a broad range of biotechnological applications due to their high activity, specificity, and stability in various environments¹. However, few archaeal esterases have been characterized, despite archaea being a primary source of extremophilic enzymes². In this study, a novel carboxyl esterase, Ta0887, from the thermoacidophilic archaeon *Thermoplasma acidophilum* was successfully cloned and overexpressed in *Escherichia coli* BL21 Star as a His-tagged protein for its purification and subsequent structural and biochemical characterization. Dynamic light scattering assays revealed that Ta0887 is a monomer in solution. The aggregation temperature of Ta0887 was approximately 70 °C. The crystal structure of Ta0887, determined at a resolution of 1.93 Å, shows a canonical α/β hydrolase fold with an α -helical cap domain and a catalytic triad consisting of Ser95, Asp187, and His215. Enzyme assays using p-nitrophenyl (pNP) esters with different chain lengths as substrates confirmed its esterase activity, yielding the highest activity with pNP butyrate (C4). Interestingly, Ta0887 exhibited substrate inhibition at high pNP butyrate concentrations. The K_m and V_{max} values, calculated using the Luong substrate inhibition model, were 0.79 mM and 0.009 $\mu\text{mol}/\text{min}$, respectively. Moreover, the optimum reaction temperature of the esterase was 70 °C, and it retained over 60% residual activity within the range of 50-75 °C, demonstrating a broad range of reaction temperatures. Remarkably, Ta0887 displayed excellent thermostability, retaining 100% and 63% residual activity after incubation at 75 °C and 80 °C, respectively, for 2 h. To our knowledge, this is the first esterase characterized from *T. acidophilum*. The structural and biochemical insights gained from this study could provide a foundation for future engineering of Ta0887 to enhance its potential for industrial applications.

References

1. Casas-Godoy L, Gasteazoro F, Duquesne S, Bordes F, Marty A, Sandoval G (2012). *Methods in Molecular Biology*, **861**, 3-38.
2. Meghwanshi G. K, Verma S, Srivastava V, Kumar R (2022). *Biotechnology Advances*, **61**.

CHARACTERIZATION OF THE INHIBITORY EFFECTS OF COAGULASE-NEGATIVE STAPHYLOCOCCI FROM HEALTHY SUBJECTS ON THE GROWTH OF STAPHYLOCOCCUS AUREUS ISOLATED FROM ATOPIC DERMATITIS PATIENTS

Díaz-Ramos Leticia Itzel^{†‡}, García-Bautista Gabriel Alejandro^{†‡}, Velazquez-Sámano Guillermo², Vanegas-Flores Claudia Leticia¹, Rangel-Castañeda Itzia Azucena¹, Garza-Velasco Raúl¹, Pastelin-Palacios Rodolfo¹, Moreno-Eutimio Mario Adán^{1*}

¹Departamento de Biología. Facultad de Química, Universidad Nacional Autónoma de México (UNAM). Cd. Universitaria, 04510. Mexico City, Mexico. ² Servicio de Alergia e Inmunología, Hospital General de México. "Dr. Eduardo Liceaga", México City, Mexico.

*Corresponding author: marioadan@quimica.unam.mx

†These authors contributed equally to this work.

Atopic dermatitis (AD) is one of the most common chronic inflammatory skin diseases, especially in children. It is characterized by episodes of intense pruritus, erythema, and eczematous lesions. A common feature in individuals with AD is the high prevalence of skin colonization by *Staphylococcus aureus*, present in up to 90% of cases, compared to less than 30% in healthy individuals. This raises the question of why *S. aureus* is less likely to be isolated from individuals without AD. The aim of this study was to isolate *Staphylococcus* strains from the skin of healthy subjects and determine if these strains can inhibit the growth of *S. aureus*, subsequently characterizing the mechanism of inhibition. Subjects without atopic dermatitis who consented to participate in the study were sampled. Microbiological samples were taken from the forearm and neck using a 16 cm² nitrocellulose membrane, which was then placed on MSA medium. After 48 hours, colony growth was analyzed and identified using Biotyper MALDI-TOF. An inhibition assay was conducted on 38 *S. aureus* strains previously isolated from AD patients of varying severity. Finally, the mechanisms of *S. aureus* inhibition by the isolated strains were characterized. After sampling 35 subjects without AD, 117 strains were obtained, of which 0.5 % were mannitol-positive and coagulase-negative. Eleven strains showed inhibition of the majority of *S. aureus* strains isolated from AD patients. The inhibition was due to products secreted by these bacteria, which were subsequently characterized. In conclusion, the skin of individuals without AD have *Staphylococcus* bacteria capable of inhibiting the growth of *S. aureus* isolated from AD patients. Funding: Research and Technological Innovation Projects, UNAM. PAPIIT 2023 No. IA208423 and PAPIIT 2024 No. IN221024.

p53 MUTANTS INDUCE AN AGGRESSIVE PHENOTYPE THROUGH OVEREXPRESSION OF MIR-27B-5P IN CANCER

José Edwin Dolores-García^{1,2}, Guadalupe Domínguez-Gómez³, Nancy Patricia Rodríguez-Flores², Maximiliano Ramírez Vela², Héctor Serrano⁴ and José Díaz-Chávez²

¹Doctorado en Biología Experimental, DCBS, Universidad Autónoma Metropolitana Iztapalapa, Mexico City 09340, ²Unidad de Investigación Biomédica en Cáncer, Instituto de Investigaciones Biomédicas, UNAM/Instituto Nacional de Cancerología, Mexico City, ⁴Departamento de Ciencias de la Salud, Universidad Autónoma Metropolitana Iztapalapa, Mexico City 09340. edwindolores1995@gmail.com

p53 tumor suppressor is considered “the guardian of the genome”, because it plays an essential role in maintaining genomic stability. In normal cells, p53 is activated by DNA damage induced by various types of cellular stress. However, p53 is mutated in more than half of all human cancers. In our laboratory, global profiling of miRNAs was performed in cells stably transfected with the three more frequent p53 mutants (p53-R248Q, p53-R273C, and p53-R175H); where we found two putative miRNAs that these three p53 mutants could regulate: miR-182-5p and miR-27b-5p. In this study, we propose to evaluate the effect of miR-27b-5p overexpression induced by p53 mutations on the ability to induce cell proliferation, migration, and invasion in cancer cell lines. We performed stable transfections in the Saos-2 cell line of the three more frequent p53 mutants in cancer. p53 protein levels were measured by western blot and miR-27b-5p expression was analyzed by qRT-PCR, as well as possible miRNA target genes; likewise, the proliferation was measured by CCK8, and migration assays were performed with transwell inserts. Our results show that stable transfections of the three p53 mutants in Saos-2 resulted in the overexpression of miR-27b-5p, as well as the downregulation of the target gene MXI1 (negative regulator of MYC) in our model. Then, by inhibiting miR-27b-5p, we observed increased MXI1 gene expression and decreased proliferation. On the other hand, we also observed a decreased in cell migration when miR-27b-5p was inhibited, compared to the control. In summary, we have found that the three more frequent p53 mutants induce overexpression of miR-27b-5p, which could be a new type gain-of-function of p53 mutants to induce an aggressive phenotype in cancer cells.

MOLECULAR DOCKING OF GLYCEROL KINASE DRIVEN MONOPHOSPHORYLATION OF SMALL ALCOHOLS

Wendy Escobedo-Hinojosa^{1*}, Nahuel Matias Ko¹, Bernhard Hauer², Julian L. Wissner¹

¹Laboratorio de Productos Naturales Marinos. Unidad de Química en Sisal, Yucatán. Facultad de Química de la Universidad Nacional Autónoma de México. ²Department of Technical Biochemistry, Institute of Biochemistry and Technical Biochemistry, University of Stuttgart, Stuttgart, Germany.

*Corresponding author: Wendy Escobedo-Hinojosa. Unidad de Química en Sisal, Yucatán. Puerto de Abrigo S/N, UNAM, 97355 Sisal, Yucatán. Tel. +527771594447.

E-mail: wendyitzeleh@quimica.unam.mx

Glycerol kinase (GlpK) from *Escherichia coli* catalyzes the ATP dependent glycerol monophosphorylation^[1,2]. Strikingly, contrary to the expected specific substrate scope, this exceptional biocatalyst also enables the direct introduction of a non-protected phosphate group to the hydroxy group of a wide range of small alcohol substrates^[3]. Previously, we investigated the biotechnological potential of GlpK to produce small monophosphorylated alcohols^[4]. Such results demonstrated that GlpK displays advantageous catalytic rates suitable for up-scaling approaches towards the tested substrates, supporting the feasibility of using GlpK as biocatalyst. However, to understand how such variety of substrates are fitting in the active site, molecular docking studies are needed. To identify the amino acid residues involved in the biocatalytic conversion we develop a computational pipeline. The established workflow allowed to perform 1) the generation and refinement of the three-dimensional model, 2) the ligand optimization, employing quantum chemical calculations to explore different conformations and energetically favorable structures of the ligands, 3) the molecular docking simulations, where the optimized ligands are virtually docked into the binding site of the modeled protein after adjusting the conditions, predicting their interactions and orientations. The protocol allowed to visualize the binding affinities, towards the five substrates successfully converted by GlpK: glycerol, propane-1,3-diol, (*R*)-(-)-1,2-propanediol, ethane-1,2-diol, and 2-methylpropan-1-ol. Our findings are crucial to understand the structural aspects of ligand-protein interactions of the mentioned substrate panel with GlpK.

Reference

1. SI Hayashi, EC Lin, (1967). Purification and properties of glycerol kinase from *Escherichia coli*, *J Biol Chem*, 242, 1030-1035
2. JW Thorner, H Paulus, (1973). Catalytic and allosteric properties of glycerol kinase from *Escherichia coli*, *J Biol Chem*, 248, 3922-3932
3. MJ Novotny, WL Frederickson, EB Waygood, MH Saier, Allosteric regulation of glycerol kinase by enzyme IIIglc of the phosphotransferase system in *Escherichia coli* and *Salmonella typhimurium*, *J Bacteriol*, 162 (1985) 810-816, doi: 10.1128/JB.162.2.810-816.1985.
4. Escobedo-Hinojosa, W., Wissner, J.L., Hauer, B, (2021). A real-time ³¹P-NMR-based approach for the assessment of glycerol kinase catalyzed monophosphorylations, *MethodsX*, 8: 101285

GENERATION OF HUMAN ANTIBODIES IN NSG MICE AGAINST SPECIFIC TARGETS OF ALZHEIMER'S DISEASE

Jeniffer Espinoza-Hernández¹, Omar Rojas-Gutiérrez¹, Karina López-Olvera¹, Luis A. Valencia-Flores¹
Sonia Mayra Pérez-Tapia^{1,2}, Martha Pedraza-Escalona^{*1,2,3}

¹Unidad de Desarrollo e Investigación en Bioterapéuticos, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, CDMX, México. ²Laboratorio Nacional para Servicios Especializados de Investigación, Desarrollo e Innovación (I+D+i) para Farmoquímicos y Biotecnológicos, LANSEIDI-FarBiotec-CONAHCyT, CDMX, México. ³CONAHCyT-Unidad de Desarrollo e Investigación en Bioterapéuticos, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, CDMX, México.

*E-mail: martha.pedraza@udibi.com.mx

Alzheimer's Disease (AD) is a neurodegenerative pathology characterized by the accumulation of beta-amyloid plaques, which leads to the loss of synapses, atrophy and neuronal death. The generation of human antibodies specific for the molecular targets of AD can be offer alternative solutions¹. In this study, we develop a platform using NSG (NOD scid gamma) humanized mice with human hematopoietic cells to produce specific human antibodies against two AD targets²: 1) the Amyloid Beta Protein (A β), which is involved in the formation of amyloid plaques that interfere with communication between brain cells, affecting the hippocampus and causing neuronal dysfunction³; and 2) TREM2 (Receptor Triggering Expressed on Myeloid Cells-2), a key microglia protein, that detects pathological changes in the brain and triggers an adaptive defensive response⁴.

Mice were humanized by preconditioning with busulfan, a bifunctional alkylating agent with a selective immunosuppressive effect on the bone marrow. This induces an effective reconstitution of human immune cells in NSG mice⁵. Subsequently, human CD34⁺ cells, with the capacity to generate human lymphoid and myeloid cells, are injected into the NSG mouse⁶. The proliferation of human cells in mice was measured by flow cytometry using antibodies against CD19, CD20 and CD45, used as human cellular markers. Fourteen weeks after humanization, the mice were immunized with the antigens of interest (TREM2 and A β monomers), the immunization was performed three times intramuscularly and a fourth by direct immunization in the spleen. The levels of human immunoglobulins, specific hIgG, were measured by indirect enzyme-linked immunosorbent assay (indirect ELISA). The established platform is not only relevant for AD, but it can also be adapted to generate antibodies against other therapeutic targets in various diseases⁷.

References

1. Carvajal, C. (2016). *Medicina Legal de Costa Rica*, 33(2), 104–122.
2. Blümich S, Münz C, Kípar A, Pellegrini G. (2021). *Veterinary pathology*, 58(1), 161–180.
3. Kulkarni, B., Kumar, D., Cruz-Martins, N. et al. (2021). *Mol Neurobiol*, 58, 5239–5252.
4. Chen, Y., & Colonna, M. (2021). *Nature Neuroscience*, 25(1), 3–4
5. Brehm, M. A., Racki, W. J., Leif, J., & Greiner, D. L. (2012). *Blood*, 119(12), 2778–2788
6. Jara-Segura, E., Jensen-Gamboa, E., Clínic, L., & Suárez, J. (2018). *Rev. Colegio de Microb. Quim. Clin. De Costa Rica*, 24(2), 2215–3713.
7. Choi, B., Chun, E., Kim, M., Kim, S.-T., Yoon, K., Lee, K.-Y., & Sung Joo Kim. (2010). *Journal of Clinical Immunology*, 31(2), 253–264

STUDIES OF THE EFFECT OF THE INTERACTION OF THE E1B- 55K PROTEIN WITH RNA ON TRANSCRIPTION AND SPLICING OF ADENOVIRUS LATE VIRAL MRNA

Diana Laura Flores García and Ramón A. González

Centro de Investigación en Dinámica Celular, Universidad Autónoma del Estado de Morelos.
Av. Universidad 1001, Cuernavaca, Morelos, 62209
Tel: 777 3297020. E-mail: dlaura.fgarcia@gmail.com; rgonzalez@uaem.mx

Adenoviruses (AdVs) are double-stranded DNA viruses without an envelope that possess an icosahedral capsid structure. Belonging to the *Adenoviridae* family, they are ubiquitous agents frequently acquired in the early years of life and mainly cause acute respiratory infections, gastroenteritis, cystitis, conjunctivitis, among others. Immunosuppressed patients are especially susceptible to more severe complications. AdV infection induces the reorganization of nuclear components in the infected cell; such reorganization of the nucleus leads to the recruitment of cellular proteins necessary for the expression and replication of the viral genome to defined nuclear sites known as viral replication compartments (RCs). Viral DNA is localized in the RCs, and cellular and viral proteins that mediate DNA replication, transcription and post-transcriptional processing of viral mRNAs are recruited to RCs, one of these viral proteins being the E1B 55K (E1B). The AdV E1B protein is a multifunctional protein that participates in various processes necessary for the efficient formation of RCs, as well as for efficient replication and transcription of the viral genome, and inhibition of the host cell's antiviral response, however, the molecular mechanisms implicated in these processes have not been identified. The E1B protein interacts with viral RNA during infection through an RNA-binding motif that shares similarity with those found in some cellular RNA-binding proteins (RNPs). Previously, our group demonstrated that mutations in the RNP motif of E1B alter the production of viral progeny, the timing and accumulation level of viral DNA, as well as the expression level of viral genes and viral proteins. Moreover, measurement of viral pre-mRNA and mature mRNA levels suggested E1B-RNA binding may be required for mRNA splicing. To attempt to distinguish whether E1B RNA binding is implicated in viral DNA synthesis, mRNA production or splicing, here we have used an experimental strategy developed in the group, in which adenovirus RCs are isolated from AdV-infected cells. The isolated RCs maintain *in vitro* the molecular activities that they display in the cell nucleus. Therefore, we have used isolated RCs from cells infected with AdV WT or virus mutants with amino acid substitutions in E1B's RNP motif to measure *de novo* synthesis of viral DNA and mRNA, and viral pre-mRNA processing by splicing. Unexpectedly our results indicate that E1B-RNA binding is required for efficient viral DNA synthesis.

References

1. Fields, B. N., D. M. Knipe, et al. (2007). *Fields Virology*, Wolters Kluwer Health/Lippincott Williams & Wilkins
2. Horridge J., Leppard K., (1998) *Journal of virology*. vol 71 p. 9374–9379 “RNA-Binding Activity of the E1B 55-Kilodalton Protein from Human Adenovirus Type 5”
3. Hidalgo, P, Gonzalez R., (2019). *FEBS Letters* 593 p. 3504–3517 “The biology of the Adenovirus E1B 55K protein”.
4. Tejera B, López RE, Hidalgo P, Cárdenas R, Ballesteros G, Rivillas L, et al. (2019) *PLoS ONE* 14(4): e0214882. “The human adenovirus type 5 E1B 55kDa protein interacts with RNA promoting timely DNA replication and viral late mRNA metabolism”.

GENDER LEADERSHIP IN BACTERIOLOGY AND ARCHAEA RESEARCH IN LATIN AMERICA

Luis Fuentes¹, Rocío Bustillos¹, Yagul Pedraza Pérez¹, Liliana López¹, Beatriz Laug¹, Yuriria Santoyo¹,
Vianey Marín¹, Jesús López², Miguel Castañeda Lucio¹ and Ricardo Carreño¹

¹Centro de Inv. Microbiológicas, Instituto de Ciencias, Benemérita Universidad Autónoma de Puebla, Ciudad Universitaria, Puebla, Pue. ²Jardín Botánico Universitario, Benemérita Universidad Autónoma de Puebla, Ciudad Universitaria, Puebla, Pue.
E-mail: luis.fuentes@correo.buap.mx

Scholar production worldwide is growing at an exponential rate, presenting very particular features. Among them, it shows great gender inequality in different scientific branches. At a global level, different studies show that female researchers receive less funding for their scientific proposals compared to male researchers, reflected in a lower number of publications. Likewise, a lower number of citations have been reported for articles with women in the leadership role compared to articles with men in the same position. Fortunately, some geographical regions of knowledge show less imbalance in production and leadership between genders. This work presents bibliometric results of women's participation in the production of scientific articles indexed in the Web of Science database (Clarivate) in Latin American countries in the field of bacteriology and research in the taxonomic domain Archaea. This area of knowledge shows a tendency towards gender parity and inclusive dominance of women as scientific leaders in our geographical area. Global production was analyzed with the participation of authors assigned to institutions in Latin American countries over a period of 20 years, which includes more than 14,000 experimental and reviewed articles published in more than 1,400 journals. Particularly, we present a diagnosis of the publication of articles, their impact, and the trend over time in the Latin American countries with the greatest participation in scientific production. Brazil and Argentina show gender parity in scientific leadership. Mexico and Chile show a trend to parity and with adequate policies could achieve it soon.

Acknowledgments. This work was partially funded by the Vicerrectoría de Investigación y Estudios de Posgrado, BUAP.

POLY(VINYL FORMAL) MEMBRANES IN CATHETER MANUFACTURING: A STUDY OF CROSSLINKING EFFECTS AND AI-ENHANCED DRUG RELEASE PREDICTIONS

Igor Garcia-Atutxa¹, Laura A. Palomares² and Francisca Villanueva-Flores^{3*}

¹Universidad Católica de Murcia (UCAM), Av. de los Jerónimos, 135, 30107, Murcia, Spain. ²Departamento de Medicina Molecular y Bioprocesos. Instituto de Biotecnología. Universidad Nacional Autónoma de México, Ave. Universidad 2001. Col. Chamilpa. 62210. Cuernavaca, Morelos, México. ³Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada, Instituto Politécnico Nacional, Boulevard de la Tecnología, 1036 Z-1, P 2/2, 62790 Atlacholoaya, Morelos. *fvillanuevaf@ipn.mx

The development of medical devices for controlled drug delivery requires high precision and rigorous quality controls to ensure their safety and efficacy. This study showcases the development of innovative poly(vinyl formal) (PVFM) membranes embedded with heparin, designed with varying cross-linking degrees. The anti-thrombogenic properties of these membranes make them promising candidates for catheter manufacturing. We employed the Korsmeyer-Peppas model to analyze the drug release kinetics from these membranes. Our investigations revealed that the membranes exhibit an anomalous release mechanism under low cross-linking conditions. In contrast, at higher crosslinking levels, they display behavior akin to Super Case II, suggesting a breakdown in the polymeric network. These insights are pivotal for advancing our understanding of enhancing the durability and functionality of medical catheters. Additionally, the non-Fickian release behavior observed offers an excellent opportunity to evaluate the effectiveness of traditional pseudo-empirical models, such as zero-order, first-order, Higuchi, and hyperbolic tangent (tanh) models, in comparison to cutting-edge artificial intelligence-based models like the Generalized Regression Neural Network (GRNN). Our comparative analysis demonstrated that although traditional models can provide an adequate fit, the GRNN model exhibited superior accuracy across all tested conditions. This model's capacity to replicate system behavior and learn from experimental data enables it to make extremely precise predictions about drug release under new and untested conditions. This capability significantly enhances the design and optimization of new drug delivery systems and bolsters quality control measures in large-scale production environments. This study represents a significant advancement in pharmacology and has practical implications. We have laid a robust foundation for future research and development in drug delivery technologies by integrating advanced machine-learning techniques. This underscores the potential of neural networks to transform the pharmacodynamic modeling landscape, promising substantial improvements in therapeutic outcomes and manufacturing efficiencies, thereby benefiting the healthcare industry as a whole.

HGF MODULATES THE MAPK/ERK 1/2 PATHWAY TO EXERT ITS HEPATOPROTECTIVE EFFECT IN A MURINE MODEL OF EXPERIMENTAL CHOLESTASIS

Maricruz García-Barrera^{1,2,3}, Genaro J. Rosales-Muñoz^{1,2,3}, Natalia Nuño-Lámbarri⁴
Luis E. Gómez-Quiroz^{2,3}, Leticia Bucio-Ortiz^{2,3}

¹Posgrado en Biología Experimental, DCBS. Universidad Autónoma Metropolitana Unidad-Iztapalapa. Mexico, City.

²Área de Medicina Experimental y Traslacional, Departamento de Ciencias de la Salud, Universidad Autónoma Metropolitana-Iztapalapa., Mexico City 09340, Mexico. ³Laboratorio de Medicina Experimental, Unidad de Medicina Traslacional, IIB/UNAM, Instituto Nacional de Cardiología Ignacio Chávez, Mexico City 14080, Mexico. ⁴Translational Research Unit, Medica Sur Clinic & Foundation, Mexico City, Mexico. ⁵ Department of Surgery, Faculty of Medicine, Universidad Nacional Autónoma de México, Mexico City, Mexico.

Av. San Rafael Atlixco No. 186, Col. Vicentina, Iztapalapa México, D.F., A. P. 55-534, CP. 09340 Tel/Fax.: 5804-4730.

E-mail: lebo@xanum.uam.mx

Introduction and Objectives. Intrahepatic cholestasis, defined as partial or total obstruction of bile flow, causes chronic inflammation and excessive production of reactive oxygen species (ROS). In vivo and in vitro studies indicate that HGF generates hepatoprotective effects within 24 hours of administration. We characterize the systemic protection mechanisms in experimental cholestasis.

Methods. Male CD1 mice aged 8-10 weeks were randomly divided into 4 experimental groups: 1) untreated control group (NT), 2) ANIT-treated group via intragastric administration at a dose of 60 mg/kg, 3) ANIT+HGF-treated group, where HGF will be administered at a dose of 10 µg/kg intravenously 24 hours after ANIT administration, and 4) control group treated only with HGF. Mice were sacrificed at 30 h, 36 h, and 48 h post-treatment initiation for liver tissue and serum collection. The collected samples were used for biochemical assays and Western Blot.

Results. Our proteomic results suggest that HGF/c-MET can reverse cholestatic damage through adaptive mechanisms by MAPK/ERK 1/2 signaling, which was corroborated when we analyzed the protein content of PP2A phosphatase 6h after HGF administration. At 36h, activation of the ERK 1/2/PKCδ pathway is observed in mice treated only with the cholestatic agent, possibly as a last resort to avoid damage. The interconnection between ERK 1/2 and cell proliferation was analyzed by key cell cycle proteins. Increased P27 and P21 were observed in the ANIT+HGF group at 30h and increased Cyclin A and Aurora A, suggesting increased S- and M-phase cells to compensate for damage. The HGF/c-MET pathway not only induces proliferation but also affects the regulation of genes involved in redox homeostasis. Temporal analysis of antioxidant proteins showed an initial increase of GSTM, GSTM2, and γ-GCS at 6h and a specific increase of GPx4 at 36h, with a subsequent decrease of γ-GCS at 48h.

Conclusions. In summary, our study demonstrates that HGF modulates a hepatoprotective response through MAPK/ERK 1/2 signaling, promoting cell cycle activation and antioxidant response through modulation of the glutathione system. These findings significantly contribute to our understanding of the potential hepatoprotective mechanisms of HGF in experimental cholestasis, furthering our knowledge of liver pathophysiology. Conahcvt:#1320

THE INHIBITORY EFFECT OF RTBL-1 ON COLORECTAL CANCERS IN VITRO IS RELATED TO THE PRESENCE OF THE EPIDERMAL GROWTH FACTOR RECEPTOR

Francisco Javier Luján-Méndez¹, Guadalupe García-Alcocer¹, Patricia García-López²,
Laura Cristina Berumen Segura¹, Roberto Ferriz-Martínez³, Anette Ramírez-Carrera⁴, Teresa García-Gasca³

¹Laboratorio de Genética y Experimentación Biológica, Posgrado en Ciencias Químico Biológicas, Facultad de Química, UAQ.

²Laboratorio de Farmacología, Subdirección de Investigación, Instituto Nacional de Cancerología.

³Laboratorio de Biología Celular, Facultad de Ciencias de Naturales, UAQ. ⁴Licenciatura en Químico Farmacéutico Biólogo, Universidad Autónoma de Nayarit.

tggasca@uaq.edu.mx

rTBL-1 is a recombinant lectin from Tepary bean (*Phaseolus acutifolius*), whose effects on colon cancer cell lines have been studied by our group¹, as well as its antitumor activity in colorectal cancer (CRC) in vivo². rTBL-1 retains the proapoptotic activity of its native homolog in CRC cells in vitro³. The objective of this work was to evaluate whether the inhibitory bioactivity of rTBL-1 on CRC is dependent on the presence of EGFR. The concentration-response effects were explored in a range of 0 to 100 µg of rTBL-1/mL using two murine CRC cell types, EGFR+ (MC-38 cells) and EGFR- (CT-26). At 8 h of exposure, MC-38 cells recorded the highest responsiveness, with statistically significant drops in survival from only 20 µg of rTBL-1/mL ($p = 0.0127$). While CT-26 cells were significantly affected from >30 µg/mL ($p < 0.0001$). Lethal concentrations 50 (LC50) of 23.50 and 30.01 µg rTBL-1/mL for MC-38 cells CT-26 cells, respectively, were obtained. By flow cytometry it was observed that LC50 values produces apoptotic responses in MC-38 and CT-26 cells, with 2.36- and 3.5-fold increases in total apoptosis, respectively respect to control cells. MC-38 cells underwent apoptosis more gradually than CT-26 cells, whose reached the largest cell subpopulation in late apoptosis of each trial ($p < 0.0001$). Finally, series of western blots revealed significant increases in the proteolytic activation of caspase-3, as well as in the inactivation by cleavage of the substrate protein PARP1, in cells treated with rTBL-1, confirming the completion of apoptotic processes in both types. These results indicate that although the ability of rTBL-1 to induce apoptosis in CRC cells is not strictly dependent on EGFR, its expression could be associated with deeper responses. Therefore, preclinical evaluation of rTBL-1 in CRC is relevant, regardless of the level of EGFR expression in the tumor. Detailed comparisons in cytotoxic kinetics indicate that despite presenting a 21.69% greater tolerance, EGFR- cells experience a faster resolution of their apoptosis, compared to MC-38 cells. This asynchrony could indicate the involvement of unexplored mechanisms independent of the EGFR pathway in the antitumor activity of rTBL-1.

Keywords. Apoptosis, colorectal cancer, EGFR, lectins, *Phaseolus acutifolius*, Tepary bean

References

1. Moreno-Celis U, López-Martínez FJ, Cervantes-Jiménez R, Ferriz-Martínez RA, Blanco-Labra A, García-Gasca T. 2020. *Molecules*. 25 1021; doi:10.3390/molecules25051021.
2. Moreno-Celis U, López-Martínez J, Blanco-Labra A, Cervantes-Jiménez R, ... García-Gasca T. 2017. *Molecules*. 22, 1670; doi:10.3390/molecules22101670.
3. Dena-Beltrán, J. L., Nava-Domínguez, P., Palmerín-Carreño, D., Martínez-Alarcón, D. ,... García-Gasca, T. 2023. *Pharmaceuticals*. 16(2) <https://doi.org/10.3390/ph16020290>

RELATIONSHIP BETWEEN EXTERNAL PH, Ca^{2+} OSCILLATIONS AND INTRACELLULAR PH IN HUMAN SPERMATOZOIDS

Angela Raquel García Martínez, Claudia Lydia Treviño Santa Cruz and Julio César Chávez Zamora

The Biotechnology Institute
62170, 7774932556, angela.garcia@ibt.unam.mx

Sexual reproduction is an important process that ensures the prevalence of a species over time and involves the fusion of male and female gametes. In this project, we will focus on the male gamete, that is, the sperm. The sperm is a cell consisting of a head where the acrosome and nucleus are located, and a flagellum that is divided into three parts: the midpiece (where the mitochondria are contained), the principal piece, and the end piece. In the sperm membrane, there are some important ionic channels, such as Catsper, a Ca^{2+} channel, Slo3, a K^+ channel, and Hv1, a proton channel. These channels have important functions in carrying out fertilization. Recently, Ca^{2+} oscillations have been observed in sperm; when experiments were performed to measure intracellular Ca^{2+} at different pH levels, we observed that the basal Ca^{2+} tends to increase when the medium is alkalinized, while when the medium is acidified, the basal Ca^{2+} tends to decrease. Likewise, Ca^{2+} oscillations tend to disappear at alkaline pH, while at acidic pH they are more constant, as previously reported. We also found a relationship between membrane potential and pH, as by measuring membrane potential at different times and different pH levels, we determined that membrane potential increases as pH increases. Finally, membrane potential was related to lifestyle, as donors who smoke, drink, are constantly stressed, and stay up late have an altered membrane potential, while donors with healthy habits and an excellent quality of life have good semen quality and their E_m is not altered. It was concluded that pH influences both membrane potential and Ca^{2+} and Ca^{2+} oscillations, also that Catsper may have a relationship with Ca^{2+} receptors, and that the reason for the cessation of spontaneous Ca^{2+} oscillations at pH 8 is due to an inhibition in the receptors of internal Ca^{2+} uptake pools due to alkalinization, since it has been recorded that they are sensitive to pH. In addition to the fact that unhealthy habits generate the production of more free radicals, which causes a change in membrane potential.

CYCLIC NUCLEOTIDE-GATED ION CHANNELS: A POTENTIAL NEW PLAYER IN PLANT ANTIVIRAL DEFENSE AGAINST BEGOMOVIRUS INFECTION

Ana María García Montelongo^{1,2}, Edgar Bonales-Alatorre¹, Yair Cárdenas-Conejo²

¹Centro universitario de investigaciones biomédicas (CUIB), Universidad de Colima¹, ²Laboratorio de Biología biosintética y estructural (LaBioSEM), Tecnoparque CLQ, Universidad de Colima
ebonales0@uacol.mx, yair.cardenas@gmail.com

Geminiviral infections in plants pose a continuous threat, significantly impacting crop production worldwide. Within the *Geminiviridae* family, the *Begomovirus* genus is the most diverse and widespread, encompassing over 445 species. Begomoviruses (BGVs) evade the plant defense system, regulated by RNA interference (RNAi), by utilizing RNA silencing suppressor proteins encoded in their genomes. Recent reports indicate that the calcium signaling pathway activates the RNAi mechanism in response to viral intrusion in plants¹. One of the primary calcium channels that respond to pathogen infections are the cyclic nucleotide-gated ion channels (CNGCs)². However, the role of CNGCs in the antiviral response against BGVs remains unclear. This study aimed to identify and characterize CNGCs in the transcriptomes of plants responding to begomovirus infection. To achieve this, we searched public transcriptomes of plants infected by begomoviruses to identify deregulated CNGC genes (Log2 change ≤ -1 , ≥ 1 , p-value ≤ 0.05). We identified 17 differentially expressed CNGC genes (DEGs) from solanaceous, cucurbitaceous, fabaceous, and malvaceous plants infected with BGVs. Three CNGC genes from *Solanum lycopersicum* were the most common DEGs. From these three *S. lycopersicum* genes, we selected five homologous genes in *Nicotiana benthamiana* for further molecular and functional characterization. The molecular characterization of these five proteins confirmed that they are true CNGCs. To verify that these genes respond to the presence of BGVs, we will challenge *N. benthamiana* plants with the begomovirus TYLCV and measure the expression levels of these CNGCs. Subsequently, we will silence CNGCs that show changes in expression to determine their role in the antiviral response against BGVs. These findings will provide new insights into the role of CNGCs in the plant antiviral response and could lead to the development of new strategies for crop protection against begomoviruses.

References

1. Wang Y, Gong Q, Wu Y, Huang F, Ismayil A, Zhang D, Li H, Gu H, Ludman M, Fátýol K, Qi Y, Yoshioka K, Hanley-Bowdoin L, Hong Y, Liu Y. A calmodulin-binding transcription factor links calcium signaling to antiviral RNAi defense in plants. *Cell Host Microbe*. 2021 Sep 8;29(9):1393-1406.e7. Epub 2021 Aug 4. PMID: 34352216.
2. Lee S, Lee HY, Kang HJ, Seo YE, Lee JH, Choi D. Oomycete effector AVRblb2 targets cyclic nucleotide-gated channels through calcium sensors to suppress pattern-triggered immunity. *New Phytol*. 2024 Feb;241(3):1277-1291. Epub 2023 Nov 28. PMID: 38013595.

STUDY OF CALCIUM OSCILLATIONS INDUCED BY MEMBRANE DEPOLARIZING PULSES IN HUMAN SPERM

Mariam Gasga Tehuintle¹, Claudia Lydia Treviño Santa Cruz^{1*}, Paulina Torres Rodríguez^{1*}

¹Instituto de Biotecnología, Universidad Nacional Autónoma de México, Av. Universidad 2001, Col. Chamilpa, C.P.62210 Cuernavaca, Morelos, México

*E-mail: paulina.torres@ibt.unam.mx, claudia.trevino@ibt.unam.mx

In sperm Ca^{2+} oscillations are important for sperm motility changes and the regulation of the acrosome reaction¹. These oscillations can occur spontaneously or upon addition of different compounds called inducers. Among them, KCl addition to sperm, produces a depolarization of the still unknown the molecular identity of the channel responsible for these Ca^{2+} oscillations and whether or not they are dependent on the capacitation status of the sperm². Capacitation is a series of biochemical changes that sperm undergo inside the female tract and are required for successful fertilization. Therefore, is important to characterized them, since these oscillations may be related to the sperm fertilizing capacity³.

In the present work, the response in $[\text{Ca}^{2+}]_i$ to depolarizing pulses with KCl was evaluated using different Ca^{2+} and K^+ channels inhibitors, in both capacitated (CAP) and non capacitated (NC) conditions. Semen sample that fulfill the parameters established by the WHO (2010) for normozoospermia and from donors that signed the informed consent approved by the IBT bioethics committee, were used. Motile sperm were separated using the swim-up technique and subsequently incubated in the presence or absence of the following CatSper channel inhibitors (NNC 055-0396 and mibefradil), Slo1 inhibitors (penitrem, and slotoxin), Slo3 inhibitor (clofilium) under CAP and NC conditions. The Ca^{2+} indicator Fluo-3 AM was used to monitor Ca^{2+} changes using a spectrofluorometer. To elicit plasma membrane depolarizations a particular protocol was used that included the addition of valinomycin and increasing concentrations of KCl.

Our results show that the inhibitors mibefradil, NNC and clofilium reduce the depolarization induced- Ca^{2+} transients. There is a significant (ANOVA with Post Hoc Tukey, $p < 0.05$) decrease in the amplitude of the transients compared to the control. These results suggest that CatSper and Slo3 are the channels involved in this response. We also observed that the Ca^{2+} transients capacitation are only present in sperm in CAP conditions. This may be due to the molecular changes that the sperm undergoes in during capacitation that modifies factors such as pH and voltage that directly influence the activity of the channels involved in these oscillations.

References

1. Mata-Martínez E, Sánchez-Cárdenas C, Chávez JC, et al (2021). *Biosystems*, 209.
2. Ferreira Juan J., Lybaert Pascale, Puga-Molina Lis C., Santi Celia M (2021). *Frontiers in Cell and Developmental Biology*, 9.
3. Mark C Kelly, Sean G Brown, Sarah M Costello, et al (2018). *Human Reproduction*, 33, 1023-1033.

SEQUENCE SPACE ANALYSIS OF HOMOLOGOUS PROTEINS WITH (β/α)8 FOLD

Estefani Gaytan-Nuñez¹, Lorenzo Segovia¹

¹Instituto de Biotecnología, Universidad Nacional Autónoma de México, 62110 Cuernavaca Morelos, Mexico. +52(777)3291862
E-mail: lorenzo.segovia@ibt.unam.mx

This project focuses on the analysis of the TIM-barrel domains, the (β/α)8 fold, a common protein fold with a wide functional diversity. We studied the relationship between the potential amino acid sequence variability and the folding and function of TIM-barrels. TIM-barrel domains are abundant and catalytically diverse, catalyzing six of the seven classes of enzymatic reactions defined by the Enzyme Commission (EC). Their structural and functional flexibility makes them ideal models for protein redesign.

The sequence space represents protein variability, including every possible amino acid change in sequence. It is defined as a high-dimensional array, where each residue is represented by a dimension with the 20 possible amino acids. The evolution of proteins can be described as a walk in the sequence space. Proteins explore this space through mutations and selective pressure systems. Deep scanning mutagenesis shows the tolerance to mutations and the function of residues in a labor intensive way. On the other hand, computational methods facilitate rapid exploration of the sequence space.

Our strategy proposes that the comparison of the sequence space of TIM-barrels, generated by the ProteinMPNN neural network, will identify variability of residues. This approach uses probability matrices (position x amino acid) analysis based on structural alignments between domains. The overall objective of the project is to determine the effectiveness of ProteinMPNN in capturing TIM-barrels variability through probability matrix analysis. The methodology starts with the collection of representative TIM-barrel domains among databases. As a second step, the probability matrices of residue substitutions that could satisfy a TIM-barrel fold are inferred. Finally, the sequence space samples must be compared based on structural alignments.

This project will provide a deeper understanding of the sequence-structure-function paradigm in TIM-barrel domains, which is crucial for biotechnology and the design of new proteins with specific functions. The identification of residue positions that tolerate variability along TIM-barrel domain diversity, will be a step forward in engineering more efficient and versatile proteins.

PHYLOGEOGRAPHY OF *ZAMIA LODDIGESII* MIQ IN THE GULF OF MEXICO

Marcel Arturo Gómez García¹, Francisco Limón Salvador¹, Jonathan García-Román²

¹Facultad de Ciencias Biológicas y Agropecuarias-Región Poza Rica – Tuxpan, Universidad Veracruzana, carretera Tuxpan Tampico Kilómetro 7.5 Universitaria 92870 Tuxpam de Rodríguez Cano, Ver. México. ²Facultad de Medicina-Región Poza Rica-Tuxpan, Universidad Veracruzana, Lázaro Cárdenas no. 801, Col. Morelos 93340 Poza Rica, Ver. Postal Code: 93182 E-mail: marcelarturog@gmail.com Phone Number: +52 294 770 5341

Introduction: Cycads are plants of the order Cycadales with a long evolutionary history. In Mexico, the family Zamiaceae includes *Zamia loddigesii*, present in several states along the Gulf of Mexico¹. Phylogeography studies the distribution of genealogical lineages and their relationship with ecological and geological factors². **Background:** Cycads originated 280 million years ago; however, the current groups diversified less than 20 million years ago³. Mexico is the second country with the larger number of cycad species; *Zamia loddigesii* is found in several regions and types of vegetation, which has driven studies explaining the wide distribution of cycads and the causes of their dispersion⁴. **Aims:** This study aims to establish the phylogeographic relationships of *Zamia loddigesii* in the Gulf of Mexico region by analyzing genetic relationships understand the causes of its distribution. **Methods:** Genomic DNA will be extracted from 10-12 individuals throughout their natural distribution in the states of Tamaulipas and Veracruz. Amplification and sequencing of two specific genes in cycads will be performed, followed by sequence analysis using MUSCLE in MEGA for alignment and genetic variation analysis. The study will be complemented with the use of ArcGIS to analyze environmental conditions and relate them to the obtained results. Additionally, a SAMOVA analysis will be conducted to identify population groups and genetic barriers. Finally, dendrograms will be created using UPGMA and Neighbor-Joining methods to explore the evolutionary relationships of *Zamia loddigesii* populations.

References

1. Nicolalde-Morejón, F., González-Astorga, J., Vergara-Silva, F., & Vovides, A. P. (2011). Perspectivas sistemáticas de *Zamia* (Zamiaceae) en Megaméxico: de la taxonomía alfa a los códigos de barras genéticos. *Revista mexicana de biodiversidad*, 82(2), 341-355.
2. Domínguez-Domínguez, O.; Vázquez-Domínguez, E. «Filogeografía: aplicaciones en taxonomía y conservación». *Animal Biodiversity and Conservation*, 2009, Vol. 32, Núm. 1, p. 59-70, <https://raco.cat/index.php/ABC/article/view/132225>.
3. Calonje, M., Meerow, A. W., Griffith, M. P., Salas-Leiva, D., Vovides, A. P., Coiro, M., & Francisco-Ortega, J. (2019). A time-calibrated species tree phylogeny of the New World cycad genus *Zamia*. (Zamiaceae, Cycadales). *International Journal of Plant Sciences*, 180(4), 286-314.
4. Nicolalde-Morejón, F., Martínez-Domínguez, L., González-Aguilar, M. A., & Stevenson, D. W. (2024). Reestablishment of *Zamia lawsoniana* (Zamiaceae, Cycadales), an endemic species of Mexico, with first description of the ovulate strobilus. *Acta Botanica Mexicana*, (131)

THE ROLE OF THE UBIQUITINATION IN THE REPLICATION OF THE NEUROTROPIC ASTROVIRUS VAI

Jaqueline Gómez, Tomás López, Susana López, Carlos F. Arias

Departamento de Genética y Fisiología Celular, Instituto de Biotecnología, UNAM

Human astroviruses (HAstV) have been clearly associated with gastroenteritis in children. However, fatal encephalitis caused by the astrovirus VAI has also been reported.

The Ubiquitin-Proteasome System has been shown to be necessary in the replication cycle of the HAstV8 and VAI. It was reported that HAstV8 requires the proteasome activity in the early stage of the replicative cycle and depends on ubiquitination to produce viral progeny. In VAI, it was reported that proteasome inhibition caused viral progeny to decrease, but the role of ubiquitination on the replication of this virus is unknown.

In this work, we investigated the importance of the ubiquitination in the VAI replication cycle, using the ubiquitination inhibitor Pyr-41. We have found that the production of viral progeny and the synthesis of viral protein were reduced in the presence of Pyr-41 in a dose-dependent manner suggesting that ubiquitination is necessary for the replication.

In addition, the infectivity and viral yield were lower when Pyr-41 was added during the absorption of the virus to the cells. These results suggest that ubiquitination plays an important role in the early stages of VAI replication.

ROLE OF SERCA PUMPS IN THE SHAPE OF THE CONTRACTION-RELAXATION KINETICS OF SOLEUS MUSCLE AND ITS EFFECT ON FORCE PRODUCTION AND FATIGUE

Ana Marcela González Bedoy, Campos Ornelas José Miguel, D. en C. Adán Dagnino Acosta

Centro Universitario de Investigaciones Biomédicas de la Facultad de Medicina, Universidad de Colima, Avenida 25 de Julio #965, Colonia Villas de San Sebastián, Colima, Col. México.
E-mail: agonzalez74@uacol.mx

Skeletal muscle is a fundamental part of the human locomotor system, contributing significantly to mobility, posture, and several other physiological functions. The soleus muscle is noted for its high density of slow-oxidative fibers (type I), which gives it unique metabolic and contractile properties, with cellular adaptations for prolonged activities. A unique feature of the soleus muscle is the predominant expression of the SERCA1a isoform located in sarcoplasmic reticulum. SERCA pumps regulate intracellular calcium signals, crucial for muscle kinetics, which uses energy derived from ATP hydrolysis, to transport two Ca^{2+} ions into the sarcoplasmic reticulum (Periasamy, 2017). Inhibition of SERCA, with compounds such as Cyclopiazonic Acid (CPA) modify the muscle function, however, not enough data is available for a proper understanding of the effect of SERCA on muscle contraction of slow muscle. We employed CPA for partial inhibition of SERCA pumps and monitored the temporal effect of this drug in sustained contractions induced with trains at supramaximal sustained electrical stimulation. Additionally, a force-frequency study was conducted to understand the effect of this drug in the maximal force generation. Finally, the shape of contraction in single twitches or tetanic contraction was analyzed at several incubation times with CPA.

We conclude that SERCA pumps inhibition modify the contraction amplitude of single twitches and left shift the frequency dependence of tetanic contractions. Contractions induced with trains of middle to high intensity are affected differentially in correlation with CPA incubation time, generating a double decay relaxation at 5 minutes incubation which is completely converted to a single slow decay after 15-20 minutes. Finally, we suggest that SERCA pump is a relevant element that modulates the twitch generation of force and fusion by slowing the relaxation kinetics of slow-twitch skeletal muscle.

References

1. Periasamy, M., et. al (2017). *Comprehensive Physiology*, 7(3), 879–890

SURFACE ENGINEERING OF THE ENCAPSULIN NANOCOMPARTMENT OF MYXOCOCCUS XANTHUS FOR THE GENERATION OF PROTEIN DELIVERY VEHICLES

Sac Nicté Gómez-Barrera, Ismael Bustos Jaimes

Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México, Av. Universidad 3000.

E-mail: ismaelb@unam.mx

Encapsulin nanocompartments (ENCs) are a novel type of protein cages found in bacteria that selectively encapsulate proteins having a specific cargo-loading peptide (CLP). Naturally occurring protein cargoes are involved in resistance to oxidative stress and other metabolic tasks. However, heterologous proteins fused to the CLP have also been encapsulated. In this study, the surface of the ENC of *Myxococcus xanthus* (MxENC) was analyzed and modified to carry a biorthogonal conjugation peptide (SpyTag) to decorate the MxENCs with any targeting protein previously fused to the SpyTag orthogonal pair, the SpyCatcher domain. Two regions on the MxENC surface were selected: the 157-loop and the C-terminus. The engineered encapsulins kept the competence for self-assembly into ENCs. The PreS1 hepatocyte targeting peptide was successfully conjugated to both engineered ENCs. The modified nanocompartments underwent comprehensive characterization for stability, cargo loading, and cellular uptake in HepG2 cells, demonstrating their potential as targeting delivery vehicles. These results provide valuable insights into the design and customization of nanocompartments, opening up possibilities for improved drug delivery applications in biotechnology and nanomedicine.

DESCRIPTION OF SYCP3 ACCUMULATIONS IN PRIMARY SPERMATOCYTES OF THE FIRST SPERMATOGENIC WAVE OF PREPUBERTAL MICE

Luis Pablo Guzmán Vargas¹, Sebastián Pacheco Gutiérrez¹, Sandra Nicole Sánchez Mejía¹,
Olga Margarita Echeverría Martínez², Nayeli Torres-Ramírez², Rosario Ortiz²

^{1,2}Laboratorio de Microscopía Electrónica, Facultad de Ciencias, Universidad Nacional Autónoma de México,
México DF 04510, México.

E-mail: luispablogmg@ciencias.unam.mx

Meiosis is the cell division in reproductive cells that rise to eggs or sperm and to the next generation in sexually reproducing organisms. During meiotic prophase I, a tripartite protein structure called the synaptonemal complex (SC) is formed, which mediates homologous chromosome pairing, synapsis and recombination between non-sister chromatids¹. One of the proteins of the SC is Synaptonemal Complex Protein 3 (SYCP3), which constitutes a fundamental part of the lateral elements, being basic to correctly carry out the meiotic process. In order to identify this protein we perform immunodetection of surface spreads of spermatocytes and paraffin-embedded testis sections. We have been found that in the first spermatogenic wave of the rat (*Rattus norvegicus*), SYCP3 forms clusters in primary spermatocytes during meiotic prophase I, although the reason for this phenomenon has not been found yet. For the purpose to determinate this phenomenon is not unique to the rat, a mouse model (*Mus musculus*) was used to study the dynamics of SYCP3 during the first spermatogenic wave. Immunodetections were performed in surface spreads against constitutive SC proteins such as: SYCP3, Synaptonemal Complex Protein 1 (SYCP1), Structural maintenance of chromosomes 3 (SMC3); as well as a protein related to SUMOylation, Small ubiquitin like modifier 2/3/4 (SUMO-2/3/4) in prepubertal mice aged 10, 12, 14, 16, 18, 20, 22, 24 and 27 days, and 3-month-old adult mice were used as controls. ELISA essay was also performed to measure the concentration of testosterone and FSH at the ages studied. SYCP3 clusters were found with different morphologies in all prepubertal mice, with a peak of appearance at 14 days of age and a higher presence at the zygotene stage. SYCP1, SMC3 and SUMO-2/3/4 showed no differences respect to the adult. Corresponding to the hormones that were quantified, both presented a cyclic periodicity every 4 days, being more evident in testosterone; starting with a higher concentration which decreased after each age and its subsequent increase at the beginning of the following cycle, even though a relationship with SYCP3 cumulus was not found. Also, SYCP3 was demonstrated in round spermatids from 18 days to adult within the chromocenter, demonstrating that SYCP3 could also have an involvement outside the meiotic process.

References

1. A Qiao H, Chen JK, Reynolds A, Höög C, Paddy M, et al. (2012) PLoS Genet 8(6): e1002790.

EFFECTS OF MELATONIN ON HUMAN SPERM IN VITRO CAPACITATION

Gabriela Hernández Silva¹, Jaqueline Ugalde Zavala¹, Yadira Hernández Rueda², Roberto Chavira Ramírez¹, Víctor Torres-Flores², Fernando Larrea¹, Mayel Chirinos¹

¹Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán, Vasco de Quiroga 15, Tlalpan, 14080, CDMX, ²Universidad Nacional Autónoma de México, Av. Universidad 3000, Coyoacán 04510, CDMX.
E-mail: gabriela.hernandezs@incmnsz.mx

Introduction. Capacitation is a series of biochemical and functional changes that occur in the sperm during its passage through the female reproductive tract that enables it to fertilize. This process is characterized by the increase in protein tyrosine phosphorylation and hyperactivated motility, among other changes. Molecules present in the female reproductive tract modulate sperm capacitation. Melatonin (MT) is a hormone contained in the follicular fluid with antioxidant properties that helps to prevent oxidative stress in the sperm. However, it is unknown whether it plays a role during sperm capacitation. **Objective.** To study the effect of MT on *in vitro* capacitation and to identify the presence of MT receptors in the human sperm. **Materials and methods.** Spermatozoa from normozoospermic donors were incubated under capacitating conditions in absence or presence of different concentrations of MT. Changes on motility and protein tyrosine phosphorylation were assessed by manual evaluation under contrast microscopy following WHO manual guidelines (2010), Computer-Aided Sperm Analyzer (CASA), and Western blots with anti-phosphotyrosine antibodies. The effect of luzindole, a MT receptor antagonist, on motility was further studied. The presence and distribution of MT receptors was evaluated by Western blot and immunofluorescence, respectively. **Results.** MT 0.1, 0.5 and 1 mM promoted a decrease in the percentage of non-progressive spermatozoa, accompanied by increase of progressive sperm. Moreover, MT 1 mM decreased hyperactivation and protein tyrosine phosphorylation, and pre-incubation with luzindole inhibited the effect of MT on hyperactivation. MT receptors were detected in human spermatozoa as two proteins of 55 and 70 kDa, and were mostly located in the post-acrosomal region. **Conclusions.** MT modulates sperm function variables that are relevant for sperm capacitation. We postulate that MT could protect sperm from premature capacitation and this effect could be mediated by its specific receptors.

NANOVACCINES DESIGN AGAINST *VIBRIO CHOLERAE*

Lucia Perezgasga¹, Francisca Villanueva-Flores², Alejandro Huerta-Saquero*

¹Centro de Nanociencias y Nanotecnología, Universidad Nacional Autónoma de México. Km. 107 carretera Tijuana-Ensenada. 22860. Ensenada, BC. México. ²Centro de Investigación en Ciencia Aplicada y Tecnología Avanzada, Instituto Politécnico Nacional. Boulevard de la Tecnología, 1036 Z-1, P 2/2, 62790. Atlacholaya, Morelos.

*E-mail: saquero@ens.cnym.unam.mx

Vibrio species are Gram-negative, halophilic, curved-rod, and motile bacteria that naturally inhabit estuarine, marine, and coastal environments. Numerous marine animals can carry *Vibrio cholerae*, and foodborne infections are associated with eating undercooked seafood. *V. cholerae* produces virulence factors that promote the colonization of the small intestine and cause profuse, watery diarrhea. Immunization is an important strategy to control and prevent epidemics or pandemics caused by cholera. *V. cholerae* virulence factors ranging from cholerae toxin (TC), the toxin-coregulated pilus (TCP), lipopolysaccharide (LPS) to outer membrane proteins (OMPs) such as OmpU, OmpW, and other antigenic proteins are ideal candidates for cholera vaccine design. *In silico* prediction is one of the alternatives for developing vaccines, allowing for the preliminary identification of highly immunogenic antigens. This approach reduces the reliance on conventional detection based on animal testing to obtain a potentially suitable antigenic candidate. Antigenic proteins-coupled-Gold nanoparticles (AuNPs) vaccines are an alternative detection method of antigen functionalization for AuNPs to develop highly stable antigens and promote a robust immune response against *V. cholerae*. We used highly immunogenic epitope prediction programs such as Vaxigen, Epitopia, Ellipro and BCpreds to select immunogenic proteins. An additional criterion used was surface proteome analysis, allowing us to select proteins exposed to the extracellular environment to inhibit *V. cholerae*-cell interactions. On the other hand, AuNPs were synthesized to be functionalized with the antigenic proteins. The genes encoding *V. cholerae* antigenic proteins, including *motY*, *ompA*, and *flgH*, were cloned into the pET28 expression vector, overproduced, and affinity purified. This process ensures the production of high-quality proteins for our research. Additionally, outer membrane proteins (OMPs) were obtained and purified, further enhancing the reliability of our results. The purified proteins will be used as free and AuNPs-functionalized proteins for mice immunization. We will evaluate the immune response by producing antibodies against the selected proteins, the blockage of the *V. cholerae*-cell interaction in cell cultures, and antibody functionality assays by opsonophagocytosis. Finally, neutralizing antibodies and infection inhibition assays will be characterized in an *in vivo* model, providing a comprehensive understanding of the vaccine's potential.

References

1. Teschler et al., 2015. <https://doi.org/10.1038/nrmicro3433>
2. Huerta-Saquero et al; 2022. <https://doi.org/10.1128/spectrum.04734-22>
3. Tapia et al., 2021. DOI: 10.1128/mBio.01227-21
4. Sanchez-Villamil et al., 2019. doi: 10.1128/mBio.01869-19.
5. Acknowledgments: AH-S was funded by PAPIIT grant IN221624 from DGAPA-UNAM.

EXPRESSION OF MICAL2 PROTEIN IN ORAL SQUAMOUS CELL CARCINOMA. BIBLIOGRAPHIC REVIEW PILOT STUDY

Huizar Reyes José Manuel, Ronell Eduardo Bologna Molina, Tremillo Maldonado Omar Alejandro, Sanny Mariuxi Alcivar Cedeño, Gamboa Guerrero Leslie Karina

Faculty of Dentistry, Universidad Juárez del Estado de Durango.
Email: jmhr23@outlook.com

Introduction. Oral squamous cell carcinoma is an aggressive malignant tumor with unpredictable biological behavior and poor prognosis. MICAL2, is a protein involved in the control of F-actin cytoskeleton dynamics and therefore also in several processes involved in health and disease. It has been related to different types of cancer, has the function of participating in cell proliferation to accelerate tumor growth, promote proteins that are related to epithelial and mesenchymal transition. In the present study and review we used hematoxylin–eosin staining techniques and immunohistochemical technique, which allows to observe the expression of MICAL2 protein in oral carcinoma. Six cases of oral squamous cell carcinoma were analyzed, of which 2 (33.3%) were well differentiated, 2 (33.3%) were moderately differentiated and 2 (33.4%) were poorly differentiated.

In the results of this study, MICAL2 positivity was observed in all histologic grades of ESCC. In agreement with studies in other types of cancer, positive expression of MICAL2 was associated as a risk factor for overall survival in patients.

General objective. To determine MICAL2 expression in oral squamous cell carcinoma.

Results. Six cases of Oral Squamous Cell Carcinoma were analyzed, of which, those corresponding to well-differentiated OSCC were 2 (33.3%), 2 (33.3%) moderately differentiated and 2 (33.4%) poorly differentiated. Figure 1 shows the percentages of incidence of ESCC.

In all cases, positive (1+) or very positive (2+) expression of MICAL2 protein was observed in the different histological grades.

Discussion and conclusions. In the results of this study, MICAL2 positivity was observed in all histological grades of ESCC. In agreement with studies in other types of cancer, positive expression of MICAL2 was associated as a risk factor for overall survival in patients. In determination to what was observed in the MICAL2 protein, is that it is increased in squamous cell carcinoma, where it promotes growth, migration and invasion. This protein could be very useful for the study of ESCC since it would help us to see how the cancer develops and how it directly affects the patient's life.

Materials and methods. Slides with histological sections of tissues with COCE diagnosis from the Postgraduate and Research Laboratory of the School of Dentistry of the Universidad Juárez del Estado de Durango were analyzed.

References

1. Wang, Y., Min, P., Qi, C., Zhao, S., Yu, M., Zhang, Y., & Du, J. (2021). MICAL2 Facilitates Gastric Cancer Cell Migration via MRTF-A-Mediated CDC42 Activation. *Frontiers in molecular biosciences*, 8, 568868.
2. Ronell Bologna, Yasunori Takeda, Takahisa Kuga Y., & Toshinari Mikami (2015) Expresion del tumor Wilms 1 (WT1) en ameloblastomas.

PARTICIPATION OF ESTROGEN AND PROGESTERONE RECEPTORS, AND PKA SIGNALING IN THE EXPRESSION REGULATION OF UNFOLDED PROTEIN RESPONSE GENES DURING DECIDUALIZATION IN IMMORTALIZED HUMAN ENDOMETRIAL STROMAL CELLS

Jesús Iván Jiménez-Rivera¹, Alejandra Monserrat Retis-Resendiz¹, Miguel Ángel Jiménez-Beltrán¹, Moisés Leon-Juárez² and Edgar Ricardo Vázquez-Martínez^{1*}

¹Unidad de Investigación en Reproducción Humana, Instituto Nacional de Perinatología-Facultad de Química, UNAM.

²Departamento de Inmunobioquímica, Instituto Nacional de Perinatología.

*Corresponding author: vamer@comunidad.unam.mx

Background. The endometrium is one of the most important tissues for reproduction, and its functions are mainly regulated by ovarian steroid hormones¹. Estradiol, progesterone, and an intracellular increase in cAMP induce a series of morphological and biochemical changes in this tissue, termed decidualization. It has been previously reported that the expression of genes involved in the Unfolded Protein Response (UPR) is modulated by decidualization^{2,3}. However, information about the mechanisms involved in this process is unknown. The aim of this study was to determine the role of estrogen receptors (ERs), progesterone receptor (PR) and cAMP-mediated signaling in the expression regulation of UPR genes during decidualization.

Methods. Immortalized Human Endometrial Stromal Cells (t-HESC, ATCC CRL-4003) were treated with a combination of estradiol, medroxyprogesterone acetate (MPA), and cAMP (EMC) for 48 h. In addition, antagonists of ERs (ER α and ER β) and PR, as well as inhibitors of protein kinase A (PKA) and a exchange proteins directly activated by cAMP **2** (EPAC2) were used in combination with EMC. RT-qPCR was performed to quantify the expression levels of decidualization biomarkers and representative UPR genes.

Results. EMC treatment induced the expression of *XBPI* isoforms, *GRP78*, and *PERK*, while decreasing *CHOP* expression. Our findings suggest that *XBPI*s expression is favored over *XBPIu* by MPA via the PR. This nuclear receptor also negatively regulates *GRP78* expression, while PKA signaling is essential for the induction of this gene during early decidualization. Our findings also suggest that PKA signaling participates in the regulation of *XBPI* and *CHOP* expression.

Conclusion. This study provides novel insights into the regulation of UPR-relevant genes during early in vitro decidualization of HESCs, which results from a complex interaction of hormones (E2, MPA) and cAMP signaling.

Acknowledgments: We acknowledge UNAM-PAPIIT (Grant Numbers IA203822 and IA203822).

References

1. Murata H, Tanaka S, Okada H. (2022). *Biomolecules*. 12(9):1275.
2. Anelli T, Dalla Torre M, Borini E, et al. (2021). *Traffic*. 23(1): 4-20
3. Pittari, D., Dalla Torre, M., Borini, E., Hummel, B., Sawarkar, R., Semino, C., van Anken, E., Panina-Bordignon, P., Sitia, R., & Anelli, T (2022), *Frontiers in cell and developmental biology*, 10, 986997.

USO DEL EXAMEN ESCRITO PARA EVALUAR ALGUNAS COMPETENCIAS EN LA ASIGNATURA DE BIOQUÍMICA Y BIOLOGÍA MOLECULAR: UNA NUEVA PERSPECTIVA PARA UN VIEJO INSTRUMENTO

Kevin David Laguna Maldonado¹, Deyamira Matuz Mares¹, Teresa I. Fortoul Van der Goes²,
Juan Pablo Pardo-Vázquez¹

¹Universidad Nacional Autónoma de México, Facultad de Medicina, Departamento de Bioquímica, Av. Universidad 3000 Colonia Universidad Nacional Autónoma de México, Ciudad Universitaria, Alcaldía de Coyoacán, C.P. 04510, Ciudad de México. Teléfono 5556220000. ²Universidad Nacional Autónoma de México, Facultad de Medicina, Departamento de Biología Celular y Tisular, Av. Universidad 3000 Colonia Universidad Nacional Autónoma de México, Ciudad Universitaria, Alcaldía de Coyoacán, C.P. 04510, Ciudad de México. Teléfono 5556220000.
E-mail: deyabq@comunidad.unam.mx

La educación basada en competencias ha emergido como un enfoque predominante en la formación médica contemporánea. En la Facultad de Medicina de la UNAM, esta perspectiva se integró mediante el plan de estudios 2010 de Médico Cirujano. No obstante, los métodos de evaluación institucionales permanecen anclados en prácticas tradicionales, como los exámenes de opción múltiple, lo que pone en entredicho los resultados de la evaluación de las competencias especificadas en el perfil profesional e intermedios de la licenciatura.

Este estudio mixto, de tipo exploratorio, se desarrolló en dos etapas. En la primera fase, se utilizaron grupos focales y entrevistas a profundidad para identificar las actividades profesionales a confiar que los estudiantes de la licenciatura de Médico Cirujano deben dominar al finalizar el curso de Bioquímica y Biología Molecular. La segunda fase, de naturaleza cuantitativa, consistió en la creación de un examen escrito con reactivos de respuesta larga, diseñado específicamente para evaluar las actividades profesionales a confiar (EPA) identificadas previamente.

La fase cualitativa permitió definir tres EPA que permiten traducir el perfil de competencias profesionales del médico en un enfoque medible y evaluable a través de métodos cuantitativos. Con base en estos hallazgos, en la fase cuantitativa se elaboró un examen escrito compuesto por doce reactivos con la intención de buscar un enfoque diferente de los exámenes escritos buscando demostrar que, a diferencia de lo reportado por algunos autores clásicos, el uso de reactivos de respuesta larga permite medir competencias avanzadas y complejas de manera eficaz y válida.

BIOPROSPECTING OF ENDOPHYTIC FUNGI FROM MACROALGAE OF THE MEXICAN CARIBBEAN SEA

Ixchel Adriana Loa Ramírez¹, Patricia Velez Aguilar², Julio Adolfo Acosta³,
José Alberto Rivera Chávez⁴, Judith Sánchez Rodríguez⁵

¹Posgrado en Ciencias del Mar y Limnología, UNAM; Av. Universidad 3000, Ciudad Universitaria Ciudad de México, ²Instituto de Biología, UNAM; Av. Universidad 3000, Ciudad Universitaria Ciudad de México, ³Universidad del Mar, Ciudad Universitaria, Puerto Ángel, San Pedro Pochutla, Oaxaca, ⁴Instituto de Química, UNAM; Av. Universidad 3000, Ciudad Universitaria Ciudad de México, ⁵Instituto de Ciencias del Mar y Limnología, UNAM. Puerto Morelos Quintana Roo.
Ixchel_loa@hotmail.com

The Mexican Caribbean Sea harbors a great diversity of algae; however, to date, there are no reports of their endophytic fungi or bioprospecting studies of their secondary metabolites. In this sense, the main objective of this work is to isolate endophytic fungi from macroalgae of the Caribbean Sea to investigate their metabolic potential and their ability to combat microbial resistance. Algae were collected from three beaches in Quintana Roo: Puerto Morelos, Tulum-Sian Ka'an, and Mahahual. Endophytic fungi were isolated from algae. The fungi were grown under axenic conditions, and organic extracts were prepared. The extracts were evaluated for their ability to inhibit bacterial growth using microdilution methods. Another portion of the extract was analyzed by HPLC-MS/MS. Data processing was performed using the Global Natural Products Social Molecular Networking (GNPS) system to generate a molecular network based on the similarity of MS/MS spectra to facilitate the tentative identification of secondary metabolites present in the extracts. Selected fungi were grown on a medium scale to isolate and identify their specialized metabolites using spectroscopic and spectrometric techniques. 250 fungal microorganisms were isolated from 64 species of algae sampled from three beaches in the state of Quintana Roo. A molecular network was created with 32 extracts that inhibited more than 50% of the bacterial growth of three nosocomial isolates (*Acinetobacter baumannii*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*). Extracts identified to contain mycotoxins were automatically eliminated from the study. These preliminary results highlight the successful isolation of a significant diversity of endophytic fungi from macroalgae of the Mexican Caribbean Sea and the identification of several bioactive compounds.

VIGOR CHARACTERIZATION OF WILD AND CULTIVABLE AMARANTH SEEDS

Alberto A. Lomelí-Bernal¹, Ana Paulina Barba de la Rosa¹ & Alfredo Herrera Estrella²

¹IPICYT, Instituto Potosino de Investigación Científica y Tecnológica, A.C. Camino a la Presa de San José 2055, Lomas 4ta Secc, San Luis Potosí, S.L.P., 78216 México. ²Unidad de Genómica Avanzada-Langebio del Centro de Investigación y de Estudios Avanzados, Libramiento Norte León Km 9.6, Irapuato, Gto., 36821, México.
E-mail: alberto.lomeli@ipicyt.edu.mx; apbarba@ipicyt.edu.mx; alfredo.herrera@cinvestav.mx

Climate change has caused a decrease in world food production. Due to this, different strategies have been proposed to maintain the food supply chain, such as introducing more diverse crops and high-quality seeds measured by their vigor. Seed vigor is a complex agronomical trait; it allows rapid and uniform germination under a wide variety of conditions¹. Amaranth seeds can grow in arid, semi-arid, saline, and low-water soils², but the molecular mechanisms behind amaranth seeds surviving in those conditions are poorly explored. Thus, the objective of this work was to analyze the concentrations of non-structural carbohydrates and raffinose family oligosaccharides (RFOs) accumulation in cultivable (*A. hypochondriacus* nutrisol, *A. hypochondriacus* cristalina and *A. cruentus*) and wild (*A. hybridus*) amaranth species and its relationship with the seed vigor. All species contain similar concentrations of carbohydrates, but *A. hypochondriacus* seeds contain higher glucose concentrations, while *A. cruentus* seeds showed the highest galactinol content, a carbohydrate related to vigor; however, no differences were observed in germination and vigor between those species. In addition, the amaranth seed transcriptome is being analyzed to understand the molecular pathways associated with amaranth seed vigor.

References

1. Reta Alemayehu et al. (2014). J Agron Crop Sci. 201, 321–329. 2. E. Bojórquez-Velázquez, et al. (2019). BMC Plant Biol., 59, 1-17.
2. Work supported by Fronteras de la Ciencia-CONAHCYT, project no. CF-2023-G-67

FLUORACIL'S SENOLYTIC POTENTIAL: TARGETING SELECTIVE CELL DEATH IN SENESCENT CELLS

Samael Olascoaga^{1,2}, Mina Königsberg², Jesús Espinal-Enríquez³, Hugo Tovar³, Félix Matadamas-Martínez⁴,
Jaime Pérez-Villanueva^{5*}, Norma E. López-Díazguerrero^{2*}

¹Posgrado en Biología Experimental, DCBS, UAM-I, Ciudad de México, Mexico. ²Laboratorio de Bioenergética y Envejecimiento Celular, DCBS, UAM-I, Ciudad de México, Mexico. ³Computational Genomics Division. National Institute of Genomic Medicine, Ciudad de México, Mexico. ⁴UIMEIP-UMAE Hospital de Pediatría, CMN S.XXI, IMSS, Ciudad de México, Mexico. ⁵Depto. Sistemas Biológicos, DCBS, UAM-X, Ciudad de México, Mexico.

*Corresponding authors: jpvillanueva@correo.xoc.uam.mx, norm@xanum.uam.mx

Our research delves into the intricate realm of cellular senescence, a phenomenon characterized by irreversible cell cycle arrest in response to diverse stressors capable of cellular damage. Over time, the accumulation of senescent cells within organs and tissues significantly contributes to the development and progression of various age-related chronic degenerative diseases. We explore the mechanisms underpinning the accumulation of these cells, focusing on their evasion of programmed cell death through the activation of survival mechanisms and antiapoptotic resistance (SCAPs). Despite extensive investigation, significant gaps persist regarding these survival mechanisms' precise nature and activation pathways. Employing cutting-edge network-based methodologies, we identified Fluoracil as a promising senolytic candidate. Our study validated its senolytic potential through experimentation across two senescence models: one involving premature senescence induced by hydrogen peroxide stress (SIPS), and the other replicative senescence (RS). We standardized these models using primary cultures of human lung fibroblasts sourced from the CCD8-Lu cell line. To assess Fluoracil's efficacy in inducing selective death of senescent cells, we conducted MTT assays alongside quantification of active caspase-3 protein expression using immunofluorescence techniques. Our findings unequivocally demonstrate Fluoracil's novel senolytic properties, particularly in eliciting selective death of senescent cells within the RS model. This breakthrough underscores Fluoracil's therapeutic potential in addressing diseases stemming from the accumulation of senescent cells, thereby opening promising avenues for exploration in regenerative medicine and anti-aging therapies.

ANTITUMOUR AND ANTIMIGRATORY ACTIVITY OF GLAUCOLID E IN BREAST CANCER CELLS

H. Aramis López-Meléndez¹, Hugo López-Muñoz¹, Ma. Luisa Escobar-Sanchez², Luis Sanchez-Sanchez¹

¹Laboratorio de Biología Molecular del Cáncer, Unidad Multidisciplinaria de Investigación Experimental, FES Zaragoza UNAM. Iztapalapa CD MX 09230, ²Laboratorio de Microscopia Electrónica, Facultad de Ciencias UNAM Coyoacan CD MX 04510. E-mail: hugolm@comunidad.unam.mx, luisss@unam.mx

Introduction. Cancer worldwide has high mortality rates, with breast cancer being one of the highest incidence¹. Current cancer treatments are ineffective in advanced stages of the disease, especially when the cells have metastasized, so it is necessary to find new compounds that focus their effect not only on eliminating cells that have high proliferation rates, rather, they can focus on other characteristics of cancer such as migration and metastasis. In this sense, sesquiterpene lactones, such as glaucolid E, have been shown to have this type of activity in different types of cancer, so in the present study the antitumor and anti-metastatic activity of glaucolid E in breast cancer cells is evaluated².

Method. The antiproliferative activity was evaluated by the crystal violet incorporation technique, the necrotic activity was processed by quantification of the activity of the enzyme lactate dehydrogenase and the apoptotic activity was processed by morphological analysis by phase contrast microscopy and staining with DAPI, as well as the evaluation of active caspase 3 by fluorescence microscopy and flow cytometry, the anti-migratory and anti-metastatic activity were evaluated by wound and transwell techniques, as well as the immunodetection of N cadherin, E cadherin and Vimentin by immunofluorescence and western blot

Results. The results obtained show that glaucolid E affects the proliferation of tumor cells, from breast cancer in a dose-dependent manner. They do not induce death by necrosis; however, they induce tumor cells an apoptotic death, and inhibits the invasiveness and migration of tumor cells.

This work was carried out thanks to the support of the projects PAPIIT IN217424, PAPIIT IN218624 and PAPIIT IN215922.

References

1. Siegel RL, Miller KD, Wagle NS, Jemal A., (2023). CA Cancer J Clin.; 73(1): 17-48.
2. P., P., Dhyani., Priyanka, Sati., Eshita, Sharma., Dharam, Chand, Attri., Amit, Bahukhandi., Bekzat, Tynybekov., Agnieszka, Szopa., Javad, Sharifi-Rad., Daniela, Calina., Hafiz, Ansar, Rasul, Suleria., William, C., Cho. (2022). Cancer Cell International, 22(1)

SUBCELLULAR LOCALIZATION OF PHOSPHOMIMETIC FORMS OF RB

Leonardo Lozoya Lozoya¹, Gabriel Alonso Pérez³, Ana María Peña Balderas³, Karla Grisel Calderón González³,
Vanesa Olivares Illana³ and Jesús Hernández Monge²

¹Laboratorio de Biomarcadores Moleculares, Instituto de Física de la Universidad Autónoma de San Luis Potosí, Av. Chapultepec # 1570, 78295 San Luis Potosí- México, ²Laboratorio de Biomarcadores Moleculares-Investigador por México-Instituto de Física de la Universidad Autónoma de San Luis Potosí, Av. Chapultepec # 1570, 78295 San Luis Potosí- México. ³Laboratorio de Interacciones Biomoleculares y Cáncer, Instituto de Física de la Universidad Autónoma de San Luis Potosí, Av. Chapultepec # 1570 San Luis Potosí- México.
E-mail: jesus.hernandez@conahcyt.mx, phone +52 5514293615

The Rb protein was the first tumor suppressor discovered, it was associated with the development of retinoblastoma tumors by the end of the 80s(1; 2). Since the discovery of this phosphoprotein, two prevalent phospho-states are accepted, the hyper- and hypo phosphorylated forms. Recently, Narasimha et. al. showed an outstanding observation that the Rb hypophosphorylated form is actually an Rb molecule that can only be mono phosphorylated once and only once in any of the 14 phosphorylatable residues at the early G1 cell cycle phase(3).

Few reports have investigated the individual activities of these mono phosphorylated Rb molecules. Our aim in this work is to search the contribution of mono phosphorylation in Rb and its subcellular localization using phosphomimetic mutants. Preliminarily, the results showed that unphosphorylated Rb and the mutant Rb-249D can be localized either in cytoplasm and nucleus in contrast to the strict nuclear localization of Rb wt, suggesting that exist other yet unexplored phosphomimetic sites that may retain Rb in the nucleus.

References

1. Alzahem T, Alsarhani W, Albahlal A, Abu Safieh L, Aldahmash S. 2019. Number of.
2. Martinez-Sanchez M, Hernandez-Monge J, Rangel M, Olivares-Illana V. 2022. FEBS J 289:4371-82
3. Narasimha AM, Kaulich M, Shapiro GS, Choi YJ, Sicinski P, Dowdy SF. 2014. Elife 3

IN SILICO DETERMINATION OF PKA AND LOGD OF NOVEL VOLTAGE-GATED POTASSIUM ION CHANNEL BLOCKERS BASED ON 4-AMINOPYRIDINE

Jair A. Madera-Hurtado², Karen Sofia Hernández-Medina¹, Sofía Rodríguez-Rangel¹, Mario U. Delgado-Jaime², Aikaterini Diamanti² & Jorge E. Sánchez Rodríguez¹

¹Departamento de Física & ²Departamento de Química, CUCEI, Universidad de Guadalajara, Guadalajara, Jalisco 44430, México.
E-mail: jorge.srodriguez@academicos.udg.mx

The reliability of 4-Aminopyridine (4AP) against neurodegenerative diseases such as Alzheimer's and multiple sclerosis has attracted significant attention due to its capability to block voltage-gated potassium (K_v) ion channels present on neurons in the central nervous system. The blocking mechanism of K_v channels by 4AP depends on membrane voltage, basicity, and lipophilicity. Recently, new 4AP derivatives with methyl and fluorinated groups have been studied as potential alternative blockers¹⁻³. Although experimental approaches are the current standard method for obtaining these properties, computational chemistry provides a useful alternative to compute the same properties as a predictive rule. To gain further insights into the basicity and lipophilicity properties of these derivatives, we conducted experimental and computational assays to determine values and at different pHs. Computations were performed using Density Functional Theory implemented in Orca software⁴ to identify levels of theory that could accurately predict the experimental data. Several types of functionals spanning from local and gradient-corrected, hybrid, meta-GGA and hybrid meta-GGA, up to range-separated hybrid, combined with the cc-pVTZ basis set, and the implicit solvation models CPCM and SMD were used. values were obtained based on the computed Gibbs free energies according to the thermochemical cycles for the acid-base titration reactions. The functionals LC-PBE, TPSS0, and BHANDHLYP were the most accurate for predictions, with mean absolute errors (MAE) of 0.71, 0.82, and 0.56, respectively. LC-PBE and TPSS0 performed better for the methylated compound (absolute percentage errors (APE) of 2.03% and 3.36%, respectively), while BHANDHLYP performed better for the fluorinated one (APE of 3.13%). values were calculated based on the solvation energies of the molecules in water/1-octanol at the same levels of theory, which presented larger MAE values, with the best functional being TPSS0 (MAE of 0.56), closely followed by BHANDHLYP and wB97X-D3, both achieving a MAE of 0.57.

References

1. Brugarolas, P., Sánchez-Rodríguez, J. E., Tsai, H. M., Basuli, F., Cheng, S. H., Zhang, X., ... & Popko, B. (2018). Scientific reports, 8(1), 607.
2. Rodríguez-Rangel, S., Bravin, A. D., Ramos-Torres, K. M., Brugarolas, P., & Sánchez-Rodríguez, J. E. (2020). Scientific reports, 10(1), 52.
3. Sun, Y., Rodríguez-Rangel, S., Zhang, L. L., Sánchez-Rodríguez, J. E., & Brugarolas, P. (2024). Scientific Reports, 14(1), 11105.
4. Neese, F., Wennmohs, F., Becker, U., & Riplinger, C. (2020). Journal of Chemical Physics, 152(22).

ANALYSIS OF THE BINDING OF MDM2 PROTEIN WITH RB MRNA

Alondra Mariana Martinez Partida and Vanesa Olivares Illana

Laboratorio de Interacciones Biomoleculares y Cancer. Instituto de Física, Universidad Autónoma de San Luis Potosí, Avenida Parque chapultepec1570, Privadas del Pedregal, 78295, San Luis Potosí, SLP. México.
Tel. 4448262300. Email: vanesa@ifisica.uaslp.mx

This work aims to study the interaction of the protooncprotein MDM2 with the mRNA of the tumoral suppressor Retinoblastoma (Rb). Earlier, a translation promotion of Rb under genotoxic stress conditions and MDM2 presence was detected. In a previous study of our lab, a map of *Rb*'s IRES, locating the bidimensional structures of its mRNA 5' untranslating region (5'-UTR), was proposed. It has been suggested that some of these secondary structures found are fundamental for MDM2 recognition. The general idea is to produce Rb constructions varying the length of its 5' UTR using clonation techniques and expressing them in cells previously knocked out for Rb. With every construction, we compared Rb expression in MDM2 presence or absence by western-blot.

Our results strongly suggest that there is a fundamental structure between nucleotides -210 and -106 that is recognised by MDM2 to regulate Rb expression. On the other hand, previous work noted that genotoxic stress was needed for MDM2 to recognise *Rb* mRNA. Thus, our work implies that there is also a regulatory segment between nucleotides -257 to -210 in *Rb* mRNA

This work demonstrates that MDM2 is an ITAF for the IRES present in the 5' UTR of Rb mRNA.

References

1. Hernandez-Monge, J., Martínez-Sánchez, M., Rousset-Roman, A., Medina-Medina, I., & Olivares-Illana, V. (2021). MDM2 regulates RB levels during genotoxic stress. *EMBO reports*, 22(1), e50615. <https://doi.org/10.15252/embr.202050615>.
2. Komar, A. A., Hatzoglou, M. (2011). Cellular IRES-mediated translation: the war of ITAFs in pathophysiological states. *Cell Cycle (Georgetown, Tex.)* 10(2):229-240. DOI: 10.4161/cc.10.2.14472. PMID: 21220943; PMCID: PMC3048795.
3. Ma, W., Ma, B., Ma, J., & Zhu, R. (2023). RB1 5UTR contains an IRES related to cell cycle control and cancer progression. *Gene*, 887, 147724. <https://doi.org/10.1016/j.gene.2023.147724>
4. Martínez-Sánchez, M., Hernandez-Monge, J., Rangel, M., & Olivares-Illana, V. (2021). Retinoblastoma: from discovery to clinical management. *The FEBS journal*, 10.1111/febs.16035. Advance online publication. <https://doi.org/10.1111/febs.16035>.

SURVEILLANCE OF NATURAL KILLER CELLS IN PATIENTS WITH TICK-BORNE DISEASES

Mariana Ivonne Martínez Rivera¹, Maribel Baquera Arteaga², Mayela Rosario Espinoza Duarte¹,
Gerardo Pável Espino Solís^{1*}

¹Laboratorio de Investigación Traslacional, Facultad de Medicina, Universidad Autónoma de Chihuahua, Circuito 1, Nuevo Campus Universitario, Chihuahua CP 31125, México. ²Hospital General Regional No 1, Unidad Morelos del Instituto Mexicano Del Seguro Social, Calle Ortiz de Campos, Esq. Universidad 500-Sector 5, San Felipe I Etapa, 31203 Chihuahua, Chih.

* Corresponding author: 614 461 8314, gespinos@uach.mx

Tick-borne diseases are infections caused mainly by bacteria of the genus *Rickettsia*. These infections are transmitted through tick bites and present non-specific symptoms that can be confused with other diseases such as the cold. The difficulty in diagnosis based solely on symptoms makes these diseases an emerging public health problem, underscoring the need to better understand their impact on the immune system (1). This study focused on examining alterations in the phenotypic profile of Natural Killer cells in patients with suspected rickettsiosis to improve the understanding of the immune response in these infections. Molecular diagnosis was performed to detect the presence of rickettsial bacteria DNA in patient samples. Subsequently, NK cell immunophenotype analysis was carried out by flow cytometry, using cell surface markers (CD3, CD56, CD16, NKG2A, and NKG2D). The molecular diagnosis confirmed infection by identifying DNA from rickettsial bacteria in the samples. Flow cytometry analysis revealed significant alterations in the phenotypic profile of NK cells, including changes in the expression of their cellular markers, suggesting a perturbation in their population and activity. This study highlights the immunological alterations in patients with *Rickettsia spp*, evidencing changes in the phenotypic profile of NK cells. These findings underscore the importance of accurate molecular diagnosis and detailed analysis of the immune system to improve the understanding and management of these infections.

References

1. Parola, P., & Raoult, D. (2001). Ticks and tickborne bacterial diseases in humans: an emerging infectious threat. *Clinical Infectious Diseases*, 32(6), 897-928.

CHARACTERIZATION OF THE FUNCTIONAL RELATIONSHIP BETWEEN THE RETROGRADE PATHWAY AND CELL LONGEVITY IN *SACCHAROMYCES CEREVISIAE*

Medina Flores, Paola Abril; Ruelas Ramírez, Hilario; Mendoza Martínez, Ariann E.; Funes, Soledad
Instituto de Fisiología Celular UNAM, Circuito Exterior s/n Ciudad Universitaria, Coyoacán, 04510, Ciudad de México.
E-mail: amedina@ifc.unam.mx

Yeast *Saccharomyces cerevisiae* can activate critical signaling pathways to enhance longevity during mitochondrial stress. One such pathway is the retrograde pathway, which typically activates during yeast aging after a finite number of cell divisions.

In retrograde signaling, a pivotal event involves the translocation from the cytoplasm to the nucleus of the Rtg1-Rtg3 heterodimer, a process facilitated by the protein Rtg2. Upon entering the nucleus, this heterodimer triggers the expression of various target genes involved in the retrograde response, with assistance from the transcriptional coactivator complex SLIK. Upstream, TORC1, a complex involved in cell growth regulation, protein synthesis and other cellular functions in response to nutrient availability, serves as a principal inhibitor of the retrograde pathway. Active TORC1 suppresses the activation of the retrograde pathway, particularly under nutrient-rich conditions when the cell is unstressed and does not require activation of rescue pathways like the retrograde pathway. Conversely, TORC1 inactivation induces the retrograde pathway, typically occurring during nutrient starvation when the cell needs to activate this pathway for survival. In addition, the retrograde pathway can be experimentally activated by specific inhibitors of TORC1 such as rapamycin and caffeine.

Previous work in our laboratory has identified the protein Slm35 as a negative regulator of mitophagy which may participate in activating the retrograde response. Deletion of the corresponding gene correlates with a resistance phenotype under oxidative stress induced by hydrogen peroxide (H_2O_2). The presence of reactive oxygen species (ROS) such as H_2O_2 have been previously reported to activate the retrograde response as heightened ROS levels can induce oxidative stress and damage cellular components like proteins, lipids, and DNA, prompting activation of this pathway as an adaptive mechanism.

This study aims to deepen our understanding on the molecular mechanisms underlying how different signals are read in order to activate the retrograde pathway, in particular, how the function of the *SLM35*, *RTG1* and *RTG3* genes is connected during this process.

DINAMIC OF PHOSPHATIDYLSERINE EXPOSURE DURING CAPACITATION IN HUMAN SPERM

Ivonne Medina¹, Claudia Treviño², Takuya Nishigaki³, Iván Oseguera-López⁴, Paulina Torres⁵

¹Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, 62210, México.
E-mail: ivonne.medina@ibt.unam.mx

Phosphatidylserine (PS) is a glycerophospholipid present exclusively in the inner leaflet of the plasma membrane, but it can be translocated by flip-flop scrambling to the outer leaflet. This process is triggered by several stimulus but in general it is related to apoptosis in most cellular types (Copic et al., 2023).

Sperm are one of the most specialized cells in mammals which sole purpose is to fertilize an oocyte. Flip flop of PS has been observed in sperm and some authors proposed that the translocation of PS may be a marker of an important sperm physiological event called capacitation. This process can be defined as a series of biochemical changes that sperm must undergo to acquire the ability to undergo the acrosomal reaction (AR), a requirement for fertilization. During the AR the plasma membrane fuses with the outer acrosomal membrane releasing its content (Zigo et al., 2020). These two processes allow sperm to find and fertilize the egg. In contrast, other authors claimed that the PS translocation to the outer leaflet is a positive marker for sperm death, as reported for other cellular types. The present work is aimed to solve this controversy and eventually, it may help to improve the diagnosis and treatment of male infertility which is a global health problem.

To study the PS dynamic during capacitation we plan to use annexin V coupled to different fluorophores. Annexin V belongs to a family of proteins with high affinity for phospholipids. To be able to combine annexin V with other fluorescent dyes to detect death cells simultaneously with PS translocation, we will produce our own annexin V conjugated initially, with mNeptune 2.5 (Exλ 599–Exλ 643) and mScarlet (Exλ 569–Exλ 594). We will combine this annexin V conjugated proteins with sytox blue (Exλ 405–Exλ 444/480) and sytox green (Exλ 450–Exλ 523) and determine PS translocation and percentage of cell death at different capacitation times using a Accuri BD6 flow cytometer.

We constructed the plasmid pRSET-mScarlet-annexin V from the plasmid pRSET-mNeptuno 2.5-annexin V, which was already constructed in our group, then competent cells DH5α were transformed to amplify both constructions. The proteins were then expressed using BL21(DE3) cells and purified by nickel affinity chromatography. The concentration of the proteins was determined by spectrophotometry. Using this fluorescent tools we will continue our work to attempt to solve the controversy.

References

1. Zigo, M., Maňásková-Postlerová, P., Zuidema, D., Kerns, K., Jonáková, V., Tůmová, L. & Sutovsky, P. (2020). Cell and tissue research, 380, 237-262.
2. Čopič, A., Dieudonné, T., & Lenoir, G. (2023). Current Opinion in Cell Biology, 83, 102-192.

THE E1B-55KDA ONCOPROTEIN BINDS TO THE ADENOVIRAL GENOME AND REGULATES GENE TRANSCRIPTION

Eduardo Mundo, Ramón A. González

Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas. Universidad Autónoma del Estado de Morelos. Cuernavaca, Morelos, México. Av Universidad No. 1001, Chamilpa, 62210 Cuernavaca, Morelos México.

Adenoviruses (AdVs) are infectious agents that cause acute respiratory infections, gastroenteritis and keratoconjunctivitis. These viruses infect numerous animal species, including humans (HAdVs), and are responsible for 5-7% of acute respiratory diseases and about 20% of gastroenteritis in children worldwide, as well as high mortality rates in immunocompromised patients. HAdVs have been used as vectors for gene therapy, vaccines, and anticancer therapies that are based on oncolytic viruses. As one of the main models of tumor virology, studies of the biology of HAdVs have led to key discoveries, including the identification of tumor suppressors, cell cycle regulation, and regulation of gene expression. The HAdV genome encodes oncoproteins that can cooperate to induce cellular transformation and tumor formation interfering with the two main pathways that regulate cellular proliferation and apoptosis, the retinoblastoma (Rb) and p53 pathways. The HAdV E1A gene-products induce entry into the S phase of the cell cycle, activating apoptosis, which is then efficiently inhibited by the E1B-gene products, thus establishing conditions in the infected cell that are conducive to efficient viral replication or to oncogenic transformation. The E1B-55KDa (E1B) oncoprotein is one of the main candidates in the design of oncolytic viruses, as HAdVs recombinants that are null for expression of this protein have shown promising results in combined anticancer therapies. Nevertheless, the molecular activities that make the E1B-null HAdVs work as oncolytic viruses are incompletely understood. The E1B is a multifunctional protein that is necessary for various steps of the adenoviral replication cycle, including viral DNA replication and viral gene expression, but the molecular mechanisms for each of these functions are not yet known. In this work we have demonstrated that the E1B can interact with HAdV promoters and either increase or reduce transcription through each of the HAdV promoters (early, intermediate and late promoters), a feature that has not been reported for any other protein encoded by a DNA virus and was only previously found for the HAdV E1A proteins. Analysis of the E1B interactome suggests that E1B may regulate transcription of the viral promoters through protein-protein interactions with cellular or viral transcription factors that bind directly to the viral DNA, and it should be interesting to determine whether E1B regulates transcription through this mechanism. These novel findings are relevant because they help to better understand the HAdV biology and will contribute to improve the design of AdVs to develop new strategies for anticancer therapies and vaccines.

LIFE AFTER DEATH?

Christopher Navarro Martínez¹ and Gabriel Del Río Guerra²

¹Institute of Cellular Physiology, ad AV. Universidad 3004, Copilco Universidad, Coyoacán, 04510 Ciudad de México, CDMX,

²Institute of Cellular Physiology, AV. Universidad 3004, Copilco Universidad, Coyoacán, 04510 Ciudad de México, CDMX.

E-mail: gdelrio@ifc.unam.mx

Cell death is an essential response for the development, differentiation, homeostasis, and survival of organisms. This process includes distinct biochemical cascades and morphological features(1) differentiation, homeostasis, and survival of organisms. This highly heterogeneous process, which includes apoptotic and autophagy-based cell death, can be activated by distinct biochemical cascades, and can display different morphological features. For this reason, a precise characterization of the numerous cell death modalities described so far in eukaryotes, and of their relationships, constitutes a major challenge for current research. Cell death-associated phenomena occur extensively in the larvae of holometabolous insects (i.e. Lepidoptera. Our understanding of cellular death as an irreversible phenomenon in all living systems has been an irrefutable paradigm for biology until today (2). However, recent research has discovered a recovery phenomenon called “anastasis” that challenges the prevalence of this paradigm. Anastasis is a cellular recovery phenomenon described in mammals and invertebrates whose function is to rescue cells from a state of death (2). A prominent feature of these cells is their ability to survive toxic compounds (STC) that previously killed them (4). This could be due to the damage to the genetic material caused during cellular death, even though there is a proportional correlation between the generation of mutations and the generation of resistance to toxic compounds (5–7)including mitochondrial fragmentation, executioner caspase activation, and DNA damage, it is assumed that cell death inevitably follows. However, this assumption has not been tested directly. Here we report an unexpected reversal of late-stage apoptosis in primary liver and heart cells, macrophages, NIH 3T3 fibroblasts, cervical cancer HeLa cells, and brain cells. After exposure to an inducer of apoptosis, cells exhibited multiple morphological and biochemical hallmarks of late-stage apoptosis, including mitochondrial fragmentation, caspase-3 activation, and DNA damage. Surprisingly, the vast majority of dying cells arrested the apoptotic process and recovered when the inducer was washed away. Of importance, some cells acquired permanent genetic changes and underwent oncogenic transformation at a higher frequency than controls. Global gene expression analysis identified a molecular signature of the reversal process. We propose that reversal of apoptosis is an unanticipated mechanism to rescue cells from crisis and propose to name this mechanism “anastasis” (Greek for “rising to life”). The ability of STC is not exclusive to anastatic cells but has also been widely described in bacteria and fungi (8). We propose that anastasis is a conserved cellular recovery phenomenon in these microorganisms with evolutionary implications contrary to the random nature of mutations, and to validate this, we studied cellular death and recovery in *Saccharomyces cerevisiae* at lethal and sublethal doses of the antifungal peptide Iztli 1 (PI1)(9). The results suggest that *S. cerevisiae* acquires resistance to PI-1 through an adaptive cellular recovery mechanism.

References

1. Franzetti E, Romanelli D, Tettamanti G. (2014). Autophagy: Cancer, Other Pathologies, Inflammation, Immunity, Infection, and Aging Elsevier, 333–49.
2. Ha K-T, (2023). Exp Mol Med, 55(8),1571–2.
3. Tang HM, Tang HL, (2018). R Soc Open Sci, 19;5(9),180442.
4. McDonald PC, Nagel JM, Dedhar S. (2021). Biol Mech Adv Approaches to Overcoming Cancer Drug Resist.251–60.
5. Tang HL, Tang HM, Mak KH, Hu S, Wang SS, Wong KM, et al, (2012). Mol Biol Cell, 23(12), 2240–52.
6. Krašovec R, Belavkin R V, Aston JAD, Channon A, Aston E, Rash BM, et al. (2014). Nat Commun, 29;5(1), 3742.
7. Martinez JL. (2014). Drug Discov Today Technol, 11(1), 33–9.
8. Papaneophytou C, Giannenas I, Dragomir C, (2019). Feed Additives: Aromatic Plants and Herbs in Animal Nutrition and Health. Elsevier Inc, 339–354.
9. Rodríguez-Plaza JG, Villalón Rojas A, Herrera S, Garza-Ramos G, Torres Larios A, Amero C, (2012). PLoS One, 7(7). e40125.

LABELING OF RECOMBINANT LECTIN WITH NANOPARTICLES AND ITS ANALYSIS IN MICROSCOPY

Angélica Núñez¹, Ricardo Cervantes¹, Antonio Topete² and Teresa García-Gasca¹

¹UAQ, FCN, 2CINVESTAV Zacatenco.

E-mail: ricardo.cervantes@uaq.mx

Lectins are glycoproteins that recognize and bind to carbohydrates, including aberrant glycosylation patterns. Visualizing their interactions is crucial, achievable through nanoparticle labeling. Quantum dots (QDs), semiconductor nanocrystals with superior optical properties, offer a promising option. This study aimed to analyze and characterize the labeling of a recombinant lectin (Lr) from Tepary bean (*Phaseolus acutifolius*) with nanoparticles using microfluidic techniques and microscopy. This investigation generated uniform complexes, known as Lr-QD, visualized using multiphoton microscopy and transmission electron microscopy. Conjugation of Lr-QD complexes was performed using microfluidics with two infusion pumps at a flow rate of 1.2 to 2 mL/h. In this process, 10 mg of CdTe-QDs were placed in 1 mL of deionized water (pH 6) with 200 µg of lectin in 10 mL of 0.1 M MES buffer (pH 7). Binding of the complexes was achieved through a chip. The complex exhibited a characteristic spectral emission at 570 nm. The coupling process did not induce significant changes ($p < 0.05$) in the fluorescence emission wavelength, ensuring that the recombinant lectin did not alter the essential optical properties of the QDs. TEM analysis revealed notable morphological differences between the Lr and the Lr-QD complex. When the lectin bound to the QDs, clusters with an icosahedral-like morphology were observed suggesting a specific and ordered organization. A decrease in zeta potential compared to the unlabeled recombinant lectin indicated the presence of carboxyl groups on the QDs, conferring a negative charge and demonstrating the electrostatic interactions and stability of the Lr-QD complex. In conclusion, recombinant lectins were successfully conjugated with quantum dots, creating highly sensitive and specific probes with significant potential for early disease detection.

References

1. Cervantes-Jiménez, R., Sánchez-Segura, L., Estrada-Martínez, L. E... & García-Gasca, T. (2020). Quantum dot labelling of tepary bean (*Phaseolus acutifolius*) lectins by microfluidics. *Molecules*, 25(5), 1041.
2. García-Gasca, T., García-Cruz, M., Hernandez-Rivera, E., López-Matínez, J ...& Blanco-Labra, A. (2012). Effects of Tepary bean (*Phaseolus acutifolius*) protease inhibitor and semipure lectin fractions on cancer cells. *Nutrition and cancer*, 64(8), 1269-1278.

MDM2 CONTROLS THE EXPRESSION LEVELS OF P53 AND RB TUMOUR SUPPRESSORS

Vanesa Olivares Illana

Laboratorio de Interacciones Biomoleculares y Cancer. Instituto de Física, Universidad Autónoma de San Luis Potosí, Avenida Parque chapultepec1570, Privadas del Pedregal, 78295, San Luis Potosí, SLP. México.
Tel. 4448262300. Email: vanesa@ifisica.uaslp.mx

MDM2 is well known as the primary negative regulator of the tumor suppressor p53 due to its E3 ubiquitin ligase activity. Under normal conditions, MDM2 binds to the p53 protein to degrade it; however, under genotoxic stress conditions, MDM2 binds to the *p53* mRNA and increases its translation. Recently, MDM2 has been considered a hub protein because of its capacity to interact with a large number of different proteins and mRNAs. The retinoblastoma protein (RB) is also a target for the E3 ubiquitin ligase activity of MDM2. Moreover, MDM2 can recognize and bind to the *RB* mRNA and induce its translation. The result of this regulation is that MDM2 induces G1 cell cycle arrest by enhancing the translation of the *RB* mRNA under genotoxic stress. These results provide a dual regulatory mechanism by which MDM2 controls cell cycle progression during DNA damage. The mechanisms by which MDM2 regulates the levels of p53 and RB are not the same; however, these two MDM2 activities allow it to control the expression levels of these two crucial tumor suppressor proteins in the cell. We explain the mechanism by which MDM2 controls p53 and RB levels and the differences between them.

IFC-305 TREATMENT INDUCES APOPTOSIS MEDIATED BY MIR92A-3P IN LIVER CANCER CELLS

Bibiana Ortega Domínguez, Enrique Chávez Jiménez, Nora Gabriela Velasco Loyden, Erick Andrés Muciño Olmos, Mariana Domínguez López, Luis Castro Sánchez* and Victoria Chagoya y Hazas

Instituto de Fisiología Celular. Circuito exterior s/n, Coyoacán, 04510, CDMX. (55) 56225614.

*Centro Universitario de Investigaciones Biomédicas, Universidad de Colima.
vchagoya@ifc.unam.mx.

Hepatocellular carcinoma (HCC) is a type of liver cancer, caused by several factors such as cirrhosis, virus hepatitis B, virus hepatitis C, and others. It is necessary to research new treatments because of the lack of effectiveness of current therapies and the development of resistance. Evasion of apoptosis is one of the main resistance mechanisms due to alterations in the signaling pathways that regulate it. Additionally, the overexpression of anti-apoptotic proteins such as Bcl-2 or MCL-1 favors the death resistance, and promoting an imbalance between death and cell proliferation. There are studies about an adenosine derivate (IFC-305) as a hepatoprotective agent. Previous studies have shown that IFC-305 treatment could regulate mitochondrial dynamics and autophagy in experimental models of HCC. Several microRNAs are deregulated in cancer, including the overexpression of miR92a-3p in HCC models *in vitro* and *in vivo*, promoting metastasis, and poor prognosis. However, it is unknown if IFC-305 can regulate the expression of miR92a-3p and its implication in HCC.

For this study, we used HepG2 cells (cells derivated from liver cancer). All assays were performed either without treatment (control) or with 1mM IFC-305. To evaluate the expression of miR92a-3p, we isolated RNA from the cells and obtained cDNA. The expression of miR92a-3p was decreased in response of 1mM of IFC-305 at 48 h. Bioinformatic analysis revealed that Bak is a target of miR-92a-3p. Bak interacts with another pro-apoptotic protein called Bax. This interaction leads to the release of cytochrome C, a key event that triggers the activation of the apoptotic caspase cascade. We evaluated the expression of Bak, which increased after treatment of HepG2 cells with 1 mM of IFC-305. The results suggest that IFC-305 treatment is modulating the expression levels of miR92a-3p and Bak.

We performed a Western blot to analyze the abundance of Bak and other proteins implicated in apoptosis, Bak is increased, while the Bcl-2 protein is decreased with 1mM IFC-305. Other proteins such as Bax and cytochrome C were also evaluated, we found that their abundance protein changed with 1mM of IFC-305. Moreover, we showed that there is a reduction in the formation colony of HepG2 cells in a concentration dependent-manner. Taken together, the results suggest that treatment with IFC-305 could modulate the miR92a-3p levels and promote apoptosis in HepG2 cells. This finding may be related to the previously described effects of IFC-305.

GM1 DIMERIZATION INDUCES AN INCREASE IN THE INTRACELLULAR Ca^{2+} CONCENTRATION AND THE ACROSOME REACTION IN CAPACITATED HUMAN SPERM

Iván Oseguera-López¹, Ignacio López-González^{1*}, Adán Guerrero² and Alberto Darszon^{1*}

¹Departamento de Genética del Desarrollo y Fisiología Molecular, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos 62210, México. ²Laboratorio Nacional de Microscopía Avanzada, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos 62210, México.
E-mail: ignacio.lopez@ibt.unam.mx, darszon@ibt.unam.mx

Ganglioside GM1, is a glycosphingolipid with a ceramide linked to four sugar residues and one molecule of sialic acid, which participates in the acrosomal reaction (AR) in mice by modulating the CaV2.3 channel, thereby increasing intracellular Ca^{2+} .

In this study, we used advanced microscopic techniques, including the ONI nanoimager and inverted confocal microscopy, combined with high-resolution image analysis like Mean-Shift Super Resolution (MSSR)², to evaluate the redistribution of endogenous and exogenous GM1 and their impact in sperm physiology.

We monitored the subcellular distribution of exogenous Bodipy-labeled GM1 (GM1-B) in human non-capacitated (NCap) and capacitated (Cap) sperm. Our preliminary data suggest that GM1-B is incorporated more rapidly into the plasma membrane of NCap sperm compared to Cap sperm. Notably, the kinetics of dimer formation of membrane-incorporated GM1-B (GM1-DII), monitored by a red shift in the Bodipy emission spectrum, were faster in NCap than in Cap sperm. Interestingly, as the presence of GM1-DII increased, we observed a remodeling of the sperm head plasma membrane suggesting the AR induction.

To assess the effect of exogenous GM1 on the intracellular calcium concentrations ($[Ca^{2+}]_{int}$) and the AR, we used Fluo-3 AM and PNA- Alexa 647, respectively. In capacitated cells, we observed that GM1 induces an increase in the ($[Ca^{2+}]_{int}$) and the percentage of AR, in a concentration-dependent manner (>30% AR with 12.5 μ M GM1; 70% of AR with 25 μ M GM1). Nevertheless, GM1 did not trigger an increase in the percentage of AR or the previously observed rise in the ($[Ca^{2+}]_{int}$) in NCap cells, suggesting that GM1 is inducing these processes through a mechanism that is activated during the sperm capacitation.

Regarding the endogenous GM1, we used the beta subunit of cholera toxin (CTB) to monitor its redistribution, and we observed the accumulation of endogenous GM1 in the apical region of the sperm head in the presence of AR inducers such as ionomycin or progesterone, confirming the redistribution of endogenous GM1 during the AR.

References

1. Cohen R, Buttke DE, Asano A, et al. (2014). Dev Cell. 28 310-321.
2. Torres-García E, Pinto-Cámara R, Linares A, et al. (2022). Nat Commun. 13

POST-EMBRYONIC ACTIVATION OF THE PRIMARY ROOT APICAL MERISTEM CELLS IN SOME CACTACEAE SPECIES

Juan Manuel Palacios-Corona^{1,2}, Selene Napsucialy-Mendivil¹, Joseph G. Dubrovsky¹,
Svetlana Shishkova¹, Gustavo Rodríguez-Alonso^{2*}

¹Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México. Cuernavaca, Morelos, México. CP 66210. ²Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México. CP 62210. Telephone: +52 7773 297 020 CP: 62210 * email: gustavo.rodriguezal@uaem.edu.mx

Most Cactaceae species exhibit determinate growth of the primary root, this is, the primary root stops growing shortly after germination as a consequence of root apical meristem (RAM) exhaustion. The RAM, located in the root apex, functions as a reservoir of cells with high proliferation activity, and it provides all cells that participate in longitudinal root growth, which in most species is indeterminate. Previous studies on *Pachycereus pringlei* and *Stenocereus gummosus* have shown mitotic activity in cells of the root apex, which indicates post-embryonic RAM activation and its functional establishment before RAM exhaustion. However, no further studies on post-embryonic RAM activation in other Cactaceae genera have been performed. In this work we studied the primary root growth of several *Mammillaria* species. *Mammillaria* is one of the most widely distributed Cactaceae genus in Mexico, with ca. 250 species. The development of the primary root in these species has not been characterized, although preliminary results showed that it exhibits determinate growth, growing only 1-2 days after germination. In this work we studied the primary root of *Mammillaria* species to assess whether its growth is due to post-embryonic RAM activation, or if it is sustained only by elongation of cells from the radicle. For this, we surface-sterilized seeds and germinated them *in vitro* to monitor root growth. Cell differentiation was evidenced by cleared root preparations analyzed with Nomarski microscopy, which showed cell enlargement, xylem differentiation, and root hair development from all epidermal cells in the apex of the primary root. Moreover, in some *Mammillaria* species, we observed the development of lateral root primordia in the primary root apex. Taken together, these results confirm the determinate root growth in *Mammillaria* species. We also analyzed post-embryonic activation of the RAM of the primary root: we assessed cell cycle progression of the RAM cells using the thymidine analog 5-ethynyl-2-deoxyuridine (EdU), which indicates passage through the S phase of the cell cycle. To discriminate whether the EdU incorporation suggests endoreduplication, cell division cycling, or both, we also evaluated the mitotic index in the RAM post germination using the nucleic acid stain SYTOX Green and confocal microscopy analysis.

We thank DGAPA-PAPIIT-UNAM (grants IN208824 to SS and IN203024 to JGD). We are thankful to Raul Puente and Ulises Rosas for seed donation; and to P.E. Calderón Delgado and S. Esteban Hernández for their collaboration in this project.

CARVACROL IMPROVES THE CONTRACTILE PROPERTIES OF SKELETAL MUSCLE UNDER LOW-INTENSITY ELECTRICAL STIMULATION

Daniel Perea Ruiz¹, Adán Dagnino Acosta¹ & Ricardo A. Navarro Polanco¹

¹Biomedical Research University Center, University of Colima, 25 de Julio Avenue, Villa San Sebastián, Colima, Col., Mexico, 28045. Tel: +52 312 1206419
E-mail: dperea@ucol.mx

Carvacrol, an organic compound found in aromatic plants such as oregano and thyme¹, has shown anti-proliferative properties in cancer cells by modifying calcium homeostasis². It has been proposed that its mechanism of action may involve the inhibition of TRP channels (TRPM7) in HEK cells³, the calcium pump of the sarcoplasmic reticulum (SERCA) in sarcoplasmic reticulum vesicles, as well as the activation of the RyR1, a calcium release channel of skeletal muscle⁴, isolated and incorporated into artificial plasma membrane vesicles. However, its effect on force production and fatigue in intact skeletal muscle or isolated fibers is unknown. In this study, the effects of carvacrol on maximal force generation, fatigue resistance, and force evolution in soleus and EDL muscles were evaluated. Soleus and EDL muscles from C57BL/6J mice (10-12 weeks old) were incubated with 1 mM carvacrol for 20 minutes and placed in organ chambers. Carvacrol did not affect fatigue resistance in EDL muscle, but it did reduce fatigue resistance in soleus muscle starting from the second minute. Changes in relaxation kinetics in presence of carvacrol were observed during fatigue, as well as a leftward shift in the force-frequency curve in both muscles. Taken together, our results suggest a therapeutic potential for improving muscle function during low-intensity physical activity, such as diseases recovery or clinical rehabilitation.

References

1. Suntres, Z., Coccimiglio, J., Misagh A. (2015). *Food Science and Nutrition*, 55, 304-318.
2. Lim, W., Ham, J., Bazer, F., Gwonhwa, S. (2019). *Journal of Cellular Physiology*, 234, 1803-1815.
3. Parnas, M., Peters, M., Dadon, D., Lev, S., Vertkin, I., Slutsky, I. (2009). *Cell Calcium*, 45, 300-309.
4. Sárközi, S., Almássy, J., Luckács, B., Dobrosi, N., Nagy, G., Jóna, I. (2007). *Journal of Muscle Research and Cell Motility*, 25, 167-174.

CHARGE DISTRIBUTION CONTRIBUTES TO IN VITRO PROTEIN DESICCATION PROTECTION OF A GROUP OF HIGHLY CHARGED ANHYDROBIOTIC INTRINSICALLY DISORDERED PROTEINS

Diego Pérez-Villanueva¹, Thomas C. Boothby², César Luis CuevasVelázquez*

¹National Autonomous University of Mexico, Mexico City, Mexico, ²Wyoming University, Laramie, U.S.A.
diegopvillanuev@gmail.com

*Author for correspondence: E-mail: cuevas@quimica.unam.mx

All life forms on the planet need water to survive. There are organisms that can tolerate losing more than 90% of their water content (desiccation), an ability called anhydrobiosis. Examples of anhydrobiotic organisms are some species of tardigrades, arthropods, nematodes, resurrection plants and most plant seeds. To tolerate desiccation, anhydrobiotic organisms share the accumulation of intrinsically disordered proteins (IDPs). *In vitro* experiments have shown that IDPs act by preventing the denaturation/aggregation of other proteins during desiccation. However, little is known regarding the molecular features that drive IDPs protein protection during desiccation. In this study, we found that for a group of five highly charged IDPs from the tardigrade *Hypsibius exemplaris* and the insect *Polypedilum vanderplanki*, the degree of charge distribution correlates with the *in vitro* protein protection efficiency. Using the model enzyme lactate dehydrogenase (LDH) to perform *in vitro* LDH activity protection assays, we found that highly charged IDPs with more segregated oppositely charged residues protect LDH from desiccation more efficiently than the IDPs with well-mix charges. Charge distribution is known to alter the global dimensions of IDPs. *In silico* global dimension analysis revealed that proteins with lower “asphericity” are better LDH protectors. Our results suggest that charge distribution together with asphericity (“spherical” shape) contribute to the protein protection capacity of highly charged IDPs during desiccation *in vitro*. These results point out that sequence-encoded properties such as the global dimensions may conduct the protection capacity of IDPs. This understanding not only broadens our fundamental knowledge of biology under water-limited conditions, but also holds great biotechnological potential. The insights gained into how the dimensions of IDPs direct protein protection could be applied in the creation of stabilizers for biological products, such as proteins, cells, and tissues, and even in the preservation of organisms sensitive to extreme conditions.

Funding: PAPIIT-UNAM IA203422; CONAHCYT CF-2019/252952; PAIP-FQ 5000-9182.

MIR-107 EXPRESSION AS A POTENTIAL PREDICTOR OF CASTRATION RESISTANCE IN PROSTATE CANCER LIQUID BIOPSES

Jonathan Puente Rivera^{1,2}, David Alejandro De la Rosa Pérez¹, José Gadú Campos Saucedo, Omar Hernandez-Leon, María Elizabeth Álvarez-Sánchez¹

¹Posgrado en Ciencias Genómicas, Universidad Autónoma de la Ciudad de México (UACM). ²División de Investigación, Hospital Juárez de México, San Lorenzo 290. Col. Del Valle. Ciudad de México, México CP 03100. Instituto Politécnico Nacional 5160, Col. Magdalena de las Salinas, Mexico City CP 07360, Mexico. Tel. 5537103671.
Correo: jo_puenter@hotmail.com

Prostate cancer (PCa) is the second most common cancer in men worldwide and a leading cause of cancer-related death in Mexican men. Conventional diagnosis relies on the Prostate-Specific Antigen (PSA) blood test, which can lead to overdiagnosis and overtreatment of patients. Androgen deprivation therapy (ADT) is one of the main treatments, but some patients develop castration-resistant prostate cancer (CRPC). Recent discoveries highlight the potential of microRNAs (miRNAs) as diagnostic tools. These small non-coding RNAs play crucial roles in cellular processes like proliferation, differentiation, and apoptosis, processes often disrupted in cancer. Their unique expression profiles in cancer tissues compared to normal tissues make them promising biomarkers. Additionally, their stability in bodily fluids like serum makes them suitable for minimally invasive testing. The discovery of miRNAs unveiled their role in regulating fundamental cellular processes, and dysregulation in their expression has been linked to various human diseases, including cancer. Advances have been made in understanding miRNA stabilization and detection in blood, establishing their presence in various bodily fluids. Notably, circulating miRNAs are protected from degradation by binding to proteins or being encapsulated within vesicles. These features make blood-based miRNAs, including those investigated in this study, attractive candidates for cancer detection, monitoring tumor dynamics, and predicting prognosis. Here, we investigate the potential diagnostic value of circulating microRNA-107 (miR-107) in serum for differentiating CRPC patients from patients with prostate cancer without castration resistance and those who haven't undergone androgen deprivation therapy. A case-control study was designed to assess the potential of circulating miR-107 in serum as a diagnostic tool to distinguish between PCa patients with and without CRPC and androgen deprivation. miR-107 expression was evaluated using qRT-PCR and analyzed statistically. Significantly higher expression ($p < 0.0001$) of miRNA-107 was observed in patients with PCa compared to the control group. However, the expression levels of miRNA-107 among different PCa stages did not show significant statistical differences. Notably, miRNA-107 was overexpressed ($p < 0.05$) in patients with CRPC compared to patients without castration-resistant prostate cancer. These findings suggest that miRNA-107 has potential diagnostic value for identifying castration-resistant prostate cancer patients.

MITIGATION OF EMERGING DISEASES IN WILD ANIMALS THROUGH PHARMACOMICROBIOMICS

Ramírez-Villarreal Jimena¹, Roberto Álvarez-Martínez¹

Universidad Autónoma de Querétaro, Facultad de Ciencias Naturales. Doctorado en Ciencias Biológicas, Laboratorio de Biología Cuantitativa y Sistemas Complejos. Carr. a Chichimequillas S/N, Ejido Bolaños, 76140, Santiago de Querétaro, Qro. Correo: jramirez102@alumnos.uaq.mx, roberto.alvarez@uaq.edu.mx

Bioinformatics tools make it possible to design molecules with antimicrobial potential and even develop vaccines in less time; not only that, but the function of existing molecules can also be improved. In this study, a protein was designed against the pathogenic fungus *Batrachochytrium dendrobatidis* (Bd), which affects amphibians in wildlife. An effective treatment against chytridiomycosis caused by this fungus has yet to be identified, and there are no mitigation strategies related to the design of peptides aimed at eliminating this fungus. In this project, we designed a peptide-peptide to effectively control and combat Bd. In this study, it was found that some short sequences are capable of having functions similar to that of naturally occurring antimicrobial peptides. By joining these sequences, peptides with multiple molecular targets could be generated. This study identified a unique and specialized receptor within the Bd genome as a target for the designed peptide. The result was a small molecule with specific antifungal potential for Bd, with three different binding sites, which was supported by tests with programs that identified AMPs with antifungal/antimicrobial function, quality, and molecular docking tests. We believe that this peptide could be effective against the Bd fungus that causes chytridiomycosis in amphibians, looking for a suitable procedure for its implementation in vitro tests and later in the field, and that, in conjunction with conservation activities such as the improvement of their ecosystems, could have a longer life expectancy for these animals that are in danger of extinction. On the other hand, this method can be helpful in the development of new drugs with antimicrobial potential. Even with the change of some variables such as the net charge, binding amino acids, aliphatic amino acids, or changes in hydrophobicity within the sequence, with the search for less specific and more conserved molecular targets among pathogens, its usefulness could be extended to more disease-causing microorganisms. This project aims to generate comprehensive treatments against infections with minimal impact on the host and its microbiota by including aspects such as microbial ecology, that is, implementing other tools for the study of the microbiota present in each organism so that together with the designed peptide, the structure of these microbial communities is not altered, which in most cases has great benefits for the health of their hosts.

THE COMMON BEAN MUTANT (*PHASEOLUS VULGARIS*) NON-NODULATING MUTANT: *NNOD(2114)* IS AFFECTED IN THE INFECTION THREAD DEVELOPMENT DURING SYMBIOSIS WITH RHIZOBIA

Rocío Reyero-Saavedra*, Gladys E. Jiménez-Nopala*, Karina San Vicente Trujillo, Mario Ramírez Yáñez, Sara I. Fuentes Membreño, Alfonso Leija Salas, Litzy Ayra Pardo, Pablo Peláez Hernández, Lourdes Girard Cuesy, José Erick Cruz Valderrama, Rosana Sánchez López, Georgina Hernández Delgado°

Centro de Ciencias Genómicas, UNAM. Avenida Universidad s/n, Col. Chamilpa 62210. Cuernavaca, Morelos.
°gina@ccg.unam.mx

Legumes are capable of establishing symbiosis with nitrogen-fixing bacteria called rhizobia, through the symbiotic N₂ fixation (SNF). Proceeding the initial step of the symbiotic interaction, rhizobia infect the legume root penetrating into the root hairs through an infection thread (IT)-an invasive invagination of the plant cell- the cortex and triggering nodule formation. Advances in legume genetics and genomics, especially during the last 20 years, has enhanced our understanding of legume genes relevant for SNF. For this, the characterization of symbiotic mutants, mainly from the model legumes *Medicago* and *Lotus*, has been instrumental.

Despite the agricultural importance of common bean, to date there is only one symbiotic (hypernodulating) mutant that has been molecularly characterized (Ferguson *et al*, 2014). Our group has screened for common bean symbiotic non-nodulating mutants, from an EMS mutant population from the BAT93 (WT) genotype. From this analysis, the non-nodulating *nnod(2114)* mutant was selected for characterization. This mutant is able to initiate rhizobia infection and to form ITs; however, the development of IT is blocked and these do not reach the root cortex cells. Occasionally, very few, small, non-infected pseudo-nodules were formed in inoculated *nnod(2114)* roots (Reyero-Saavedra *et al.*, 2013).

We performed a genome sequence analysis between WT and *nnod(2114)* to identify the responsible gene of the non-nodulating phenotype. The *LIN/CERBERUS* gene showed a “high impact variant”: a premature stop codon that, hypothetically, will produce a truncate protein. The *LIN/CERBERUS* gene, encodes a Ubiquitin E3 ligase, that is part of an exocytosis complex in the tip of the IT, important for guiding the development of the IT. Transgenic hairy roots from *nnod(2114)* expressing *LjCERBERUS* were able to nodulate, thus showing the genetic complementation of the mutant. BAT93 plants silenced for *PvLIN*, through RNAi, were also complemented for effective nodulation with *LjCERBERUS*. Complementation with the *PvLIN* gene is in progress. Current research is aimed to confirm the *PvLIN* affection in *nnod(2114)*.

Reference

1. Ferguson, et al. (2014). Nature. 417: 962-966.
2. Reyero-Saavedra, et al. (2023). Plants, 12, 1310
3. Murray, et al., (2011). Plant J. 65: 244–252.
4. Liu, et al. (2019). Nat Commun 10: 2848.

UNVEILING THE PROTEOMIC IMPACT OF CALCIUM CARBIDE RIPENING ON MARADOL PAPAYA SEEDS

Francisco Antonio Reyes-Soria¹, José Elizalde-Contreras¹, Esaú Bojórquez-Velázquez¹ and Eliel Ruiz-May^{*1}

¹Red de Estudios Moleculares Avanzados, Instituto de Ecología A.C. (INECOL),
Carretera Antigua a Coatepec No. 351, Xalapa, Veracruz, México.

*E-mail: eliel.ruiz@inecol.mx

Papaya (*Carica papaya* L.), a prominent tropical fruit, boasts a rich composition of health-promoting compounds. Mexico, a key player in the global papaya market, cultivates the prized Maradol variety known for its exceptional qualities¹. Understanding papaya ripening is crucial for optimizing postharvest shelf life (PSL) and consumer preferences².

Ethylene orchestrates papaya ripening, influencing fruit quality³. Artificial ripening agents like calcium carbide (CaC₂) are employed to control marketability⁴. While cost-effective⁵, CaC₂ raises safety concerns⁶. CaC₂ releases acetylene, mimicking ethylene's effects. Acetylene expedites ripening but may compromise flavor, aroma, and nutrient content.

Our study investigates the impact of CaC₂-induced ripening on the Maradol papaya seed proteome. We employ a multifaceted approach to understand the dynamics of seed proteins during green maturation to commercial maturity under CaC₂ exposure. This is the first report on CaC₂-induced changes in papaya seed proteome.

Preliminary results revealed an increasing trend of 391 proteins, that enrich primary metabolism, particularly those involved in pyruvate metabolism and glycolysis. This suggests CaC₂-induced ripening may accelerate energy production in papaya seeds. We are currently investigating the functional roles of these increased proteins to understand their impact on seed viability and germination under CaC₂ exposure. By elucidating the interplay between physiological processes and biochemical pathways, this research offers valuable insights into the dynamics of CaC₂-induced papaya ripening.

References

1. Alcántara J, J. Á., Castro H, E., Ayvar S, S., Nava, A. D., & Guadarrama B, T. (2010). Revista Venezolana de Ciencia y Tecnología de Alimentos, 1(1), 035–046.
2. Chen, T., Qin, G., & Tian, S. (2020). New Phytologist, 228(4), 1219–1226.
3. Gray, W. M. (2004). PLOS Biology, 2(9), e311.
4. Cissé, M., Silue, Y., Cissé, M., Kouadio, A. D. S., & Nindjin, C. (2020). Current Journal of Applied Science and Technology, 24–30.
5. Adeyemi, M. M., Bawa, M. H., & Muktar, B. (2018) J of Chemical Society of Nigeria, 43(2), Article 2.
6. Chandel, R., Sharma, P. C., & Gupta, A. (2018). J of Food Processing and Preservation, 42(2), e13420.

COMPARATIVE INSIGHTS INTO GLUTATHIONE TRANSFERASES IN *TAENIA SOLIUM*: TS24GST, TS25GST AND TS26GST

Cinthyia Alejandra Rocha Sánchez¹, Abraham Landa², Enrique Rudiño Piñera¹ & Ricardo Miranda Blancas¹

¹Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología, UNAM, Cuernavaca, Mexico,

²Departamento de Microbiología y Parasitología, Facultad de Medicina, UNAM, México.

E-mail: ale.rocha42@ciencias.unam.mx

The study of glutathione transferases (GSTs EC 2.5.1.18) is important due to their role in detoxifying xenobiotics. Cytosolic GSTs the most abundant, can be classified into different classes¹. Cytosolic GSTs have been identified in *Taenia solium*, a medically significant parasitic worm that has developed resistance to anthelmintics¹. In this work, three GSTs from *T. solium* were purified: Ts24GST, Ts25GST, and Ts26GST using affinity chromatography (AC), ion exchange chromatography (IC), and size exclusion chromatography (SEC). The activity of the three purified enzymes was measured using kinetic assays with reduced glutathione (GSH) and 1,2-dichloro-4-benzene (CDNB) as substrates². Their kinetic parameters (V_{max} , K_M , K_{cat} , and K_{cat}/K_M) were determined for each GST, revealing variations in enzyme activity. Ts26GST exhibited the highest activity towards GSH and CDNB substrates, while Ts24GST and Ts25GST showed lower activity, suggesting that detoxification might not be their primary function. These variations could be related to their different classes: Ts24GST (κ class), Ts25GST (μ class), and Ts26GST (α - μ class)³. A structural analysis, including secondary structures and tertiary structure prediction using AlphaFold2, revealed structural differences among the three enzymes. These differences suggest that variations in enzymatic activity could be partly explained by structural differences. This initial analysis aids in understanding how the three different *T. solium* GSTs interact with various molecules, providing useful information for the development of potential specific inhibitors and drug design.

References

1. Vibanco-Pérez, N., Jiménez, L., Merchant, M. T., & Landa, A. (1999). *The Journal of Parasitology*, 85(3), 448–453.
2. Torres-Rivera, A., & Landa, A. (2008). *Archives of Biochemistry and Biophysics*, 477(2), 372–378.
3. Roldan, A., Torres-Rivera, A., & Landa, A. (2013). *Parasitology Research*, 112(11), 3865–3872.

MEASURING MACROMOLECULAR CROWDING IN ARABIDOPSIS ROOT CELLS FROM DIFFERENT DEVELOPMENTAL ZONES

Delia L. Rodríguez-Bustos¹, Sterling Field², Seung Y. Rhee², César L. Cuevas-Velázquez¹,
Javier Plasencia de la Parra^{1*}

¹Depto. de Bioquímica, Facultad de Química, UNAM, Cd Mx 04510, México delialuisa2@gmail.com

² Plant Resilience Institute, Michigan State University, East Lansing, MI, USA.

*Authors for correspondence: cuevas@quimica.unam.mx, javierp@unam.mx

The cell cytoplasm is densely packed with macromolecules such as proteins, nucleic acids, and polysaccharides. The resulting macromolecular crowding (MC) is crucial for efficient cellular functions, influencing molecular transport and the kinetics of biochemical reactions. Changes in MC occur in response to environmental perturbations like hyperosmotic stress. Monitoring MC levels and dynamics in living cells is essential to understanding its impact on cell functions. One method to estimate MC *in vivo* is passive microrheology, which tracks tracer particle movement within the cell. The genetically encoded multimeric (GEM) nanoparticles are homomultimeric scaffolds fused to a fluorescent protein that self-assemble in the cell into bright particles of defined shape and size used as tracer particles. Using GEMs, we estimated MC through the diffusion coefficient of these nanoparticles in different developmental zones of 5-day post-germination *Arabidopsis thaliana* primary roots under standard and hyperosmotic stress conditions. Here, we show that GEMs diffusion coefficients varied between 0.01 to 0.1 $\mu\text{m}^2 \text{s}^{-1}$ in root cells under standard conditions, but significant differences in this parameter were observed among the meristematic, elongation, and differentiation zones. We found that cells in the meristematic zone have lower diffusion coefficients (higher MC) than those in elongation and differentiation zones (lower MC). Our results show that the microrheological properties of the cytoplasm are different across cells from different developmental zones of the *Arabidopsis* primary root, suggesting that MC levels are tuned to carry out specialized biochemical processes in each developmental zone. Our next step is studying how MC of the root cells' cytoplasm is affected by hyperosmotic stress and during acclimation. Quantifying MC changes in plant cells during stress will enhance understanding of how plant cells sense, respond to, and acclimate to environmental stress conditions.

Funding: CONAHCYT CF-2019/252952; (PAPIIT-UNAM) IA203422; (PAIP-FQ) 5000-9182 and 5000-9121.

COMPARATIVE ANALYSIS OF THE SEPTIN 2 INTERACTOME IN MOSQUITO CELLS (AAG2) INFECTED AND NOT INFECTED WITH DENGUE VIRUS

¹Jose Ángel Rubio Miranda, ²Antonio Celestino Montes, ¹Valeria Calderón Frontana, ¹Carlos Vázquez Calzada, ¹Leticia Cortez Martninez, ¹Febe Elena Cazares Raga, ¹Fidel De la Cruz Hernández

¹Departamento de Infectómica y Patogénesis Molecular. ²Centro de Investigación Especializada en Microbiología-Universidad Autónoma de Guerrero. Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Av. Instituto Politécnico Nacional No. 2508. Col. San Pedro Zacatenco. C.P. 07360 México, Ciudad de México. Tel: (55) 57473800 ext.5646
jose.rubio@cinvestav.mx cruzcruz@cinvestav.mx

Septins are a family of GTP-binding proteins, which participate in different cellular processes such as: vesicular trafficking, cytokinesis, chromosome segregation, autophagy, as well as in innate immunity and in recent years, it has been shown that some septins such as septin 6, participate in the replication of the Hepatitis C Virus (HCV) in mammalian cell lines (Huh7), while mosquito septin 2 interacts with viral proteins of the dengue virus (DENV) replicative complex and possibly also participate in virus replication. Considering that Aedes mosquitoes are the main vectors of the dengue virus and that septin-type proteins could help its replication, it is important to know that other molecules can interact with septin 2 during infection with DENV4. The main objective of the work was to analyze the septin 2 interactome in both mosquito cells infected and uninfected with DENV4. Aag2 cells cultured in 6-well plates in Schneider medium and incubated at 28°C until a confluency of 80%, were infected with DENV4 at a multiplicity of infection of 3 (MOI:3) for 48 hours, subsequently, both uninfected cells as infected, they were subjected to protein extraction and used for immunoprecipitation assays using the Pierce co-immunoprecipitation kit and the mosquito anti-septin 2 antibody. Immunoprecipitation products were analyzed by Western blotting and mass spectrometry. We observed that, under basal conditions, septin 2 interacted with proteins related to vesicular trafficking, cytoskeleton, RNA processing and proteasome and in cells infected with DENV4, septin 2 interacted with proteins related to translation, oxidative stress, cytoskeleton, metabolisms, and proteasome. Within all the data obtained, we particularly observed that during infection with DENV, septin 2 interacts with the protein Glyceraldehyde 3-phosphate dehydrogenase and RPL7, molecules that have been shown to be essential to increase the metabolism of the cell during virus replication. and to translate cellular proteins that are used by the virus, while under basal conditions the aforementioned interactions were not identified. These data suggest that septin 2 is a protein that is most likely related to cellular mechanisms that help DENV viral proliferation.

PROTEIN CONFORMATIONAL DYNAMICS AND THERMAL ADAPTATION

Jorge Emiliano Salinas López, Alejandro Sosa Peinado

Laboratorio de Físicoquímica e Ingeniería de Proteínas, Departamento de Bioquímica, Facultad de Medicina, Universidad Nacional Autónoma de México, Ciudad de México, Mexico. Edificio de Investigación de la Facultad de Medicina, 2º piso. Circuito Escolar S/N, Ciudad Universitaria, CDMX 04510. Tel. 55 5623 2275.
asosa@bq.unam.mx

Proteins perform a central role in almost every biological process by controlling a wide array of functions. For this, proteins fold in three-dimensional structures determined by its amino acid sequence. Knowing the protein 3D structure provides information about the structural elements needed for it to function. Nonetheless, as proteins are marginally stable, they are not rigid, and energetically compensate for their interactions with other regions thanks to their flexibility. Thus, proteins are highly dynamic entities, which means that their static conformations are not enough to carry their functions, and their dynamic properties are directly related to the biological activity¹. In the case of enzymes, its adaptability, evolvability and capacity to acquire novel functions arises from the enzyme ability of sampling different thermally accessible conformations^{2,3}, *i. e.* the enzyme conformational dynamics, which plays a key role in the promiscuity, regulation, inhibition, and essential steps during the enzyme catalytic cycle. In order to understand the functional aspects of the enzyme dynamics, the comparative study of homologous with different thermal stabilities (psychrophilic, mesophilic, and thermophilic) have proven useful⁴. Enzyme flexibility/rigidity has an important role, since the thermophilic ones tend to be more rigid and less active than its mesophilic counterpart at ambient temperature, and following this trend, the psychrophilic homologs display increased flexibility and a narrower thermal stability window. In spite of this, the relationship between enzyme conformational processes and enzymatic activity and thermodynamic stability has proven difficult to characterize. As mentioned earlier, a decrease in flexibility tends to come with an increment in thermal stability, which leads to the hypothesis that the relevant motions for the enzyme activity can be identified by comparing the conformational dynamics of homologs adapted to different thermal environments. To explore this topic, we performed molecular dynamics (MD) simulations with a psychrophilic, mesophilic and thermophilic variant of glucosidases from the glycoside hydrolase family 5 (GH5). This enzymes' catalytic domain adopts a $(\beta/\alpha)_8$ barrel fold, with two conserved Glu residues at the end of the β -strands four and seven. Despite the highly conserved topology of these enzymes, the sequence identity between its members is rather low, with the main differences being in the loops connecting the α -helix and β -strands secondary structures. In addition, the elements that contributes to their activity and stability at different temperatures are not so clear, given that the relative flexibility of each homolog is similar between them, as well as the propensity to form different types of non-covalent interactions that tend to modify the thermal adaptation of enzymes, such as salt bridges, hydrogen bonds, hydrophobic interactions and so on. The MD simulations analysis provides a structural (RMSD, RMSF, secondary structure analysis, etc.) and dynamical (free energy landscapes, non-covalent interaction networks, PSN, ENM, etc.) depiction of the dynamical processes inside the GH5 glucosidases, and by the comparison between homologs, a new insight into the role of this elements in its thermal adaptation.

References

1. T. Saleh, C. G. Kalodimos, *Science*. 355, 247–248 (2017).
2. N. Tokuriki, D. S. Tawfik, *Science*. 324, 203–207 (2009).
3. G. G. Hammes, S. J. Benkovic, S. Hammes-Schiffer, *Biochemistry*. 50, 10422–10430 (2011).
4. K. A. Stafford, P. Robustelli, A. G. Palmer, *PLoS Comput. Biol.* 9, e1003218 (2013).

THE COMMON BEAN (*PHASEOLUS VULGARIS*) NON-NODULATING MUTANT *NNOD(1895)* IS AFFECTED IN THE INITIAL STEP OF RHIZOBIAL INFECTION DURING THE N-FIXING SYMBIOSIS

Karina San Vicente Trujillo*, Rocío Reyero-Saavedra*, Gladys E. Jiménez-Nopala, Mario Ramírez Yáñez, Sara I. Fuentes Membreño, Alfonso Leija Salas, Mishael Sánchez-Pérez, Lourdes Girard Cuesy and Georgina Hernández Delgado^o

Centro de Ciencias Genómicas, UNAM. Avenida Universidad s/n, col. Chamilpa 62210. Cuernavaca, Morelos.
^ogina@ccg.unam.mx

Legumes are capable of establishing symbiosis with nitrogen-fixing bacteria generally called rhizobia. The first step in this interaction is the communication between the two symbionts through molecular signaling, namely flavonoids/isoflavonoids secreted by the plant root to the rhizosphere that trigger rhizobia biosynthesis of lipochitooligosaccharides, known as nodulation factors (NF). The symbiotic N-fixation (SNF) symbiosis has been studied since a long time ago. Advances in legume genetics and genomics, especially during the last 20 years, has enhanced our understanding of legume genes relevant for SNF. For this, the characterization of symbiotic mutants, mainly from the model legumes *Medicago* and *Lotus*, has been instrumental.

Despite the agricultural importance of common bean, to date there is only one symbiotic (hypernodulating) mutant that has been molecularly characterized (Ferguson et al, 2014). Our group has screened for common bean symbiotic mutants, from an EMS mutant population from the BAT93 (WT) genotype. From this analysis, the non-nodulating *nnod(1895)* mutant was selected for characterization. *nnod(1895)* plants showed a defect in root hair deformation, required to trap the bacteria and trigger rhizobial root infection. As compared to WT, *nnod(1895)* plants present increased non-effective root hair deformation. The latter suggests that *nnod(1895)* is affected in the first steps of the SNF (Reyero-Saavedra, et al., 2023).

We followed the comparative -WT vs. *nnod(1895)*- whole genome sequence analysis to identify candidate genes that could be responsible for the mutated phenotype. From our analysis, the *ISOFLAVONE SYNTHASE (ISF)* gene of *nnod(1895)* presented a “high-impact variant”, lost of start codon. Other legumes mutated in *ISF* are incapable to nodulate because the presence of isoflavones is necessary to initiate the symbiosis (Subramanian et al. 2006). We observed that root exudates from *nnod(1895)* plants did not induce rhizobial nod genes. In addition, we achieved to revert the incapacity of *nnod(1895)* plants to nodulate by inoculating these with a rhizobial strain that constitutively secretes NF -without isoflavonoids presence-. These results suggest that *ISF* could be the responsible gene of the *nnod(1895)* non-nodulating phenotype. Current research of the characterization of this mutant is in progress

Reference

1. Ferguson, et al. (2014). Nature. 417: 962-966.
2. Reyero-Saavedra, et al. (2023). Plants, 12, 1310
3. Subramanian, et al. (2006). Plant J. 48: 261-273.

DEVELOPMENT OF A SINGLE-CYCLE NON-CLASSICAL HASTV REPLICON SYSTEM FOR STUDYING VIRAL REPLICATION AND INHIBITORS

Guadalupe Sanchez Flores¹, Carlos F. Arias², Susana López², Carlos Sandoval Jaime²

¹Centro de Investigación en Dinámica Celular, UAEM, Cuernavaca, Morelos. ²Instituto de Biotecnología, UNAM, Cuernavaca, Morelos.

guadalupe.sanchezflo@uaem.edu.mx / carlos.sandoval@ibt.unam.mx

Astroviruses are etiological agents of gastrointestinal infections worldwide, affecting children, the elderly, and immunocompromised patients. Among astroviruses, the non-classical human astrovirus (HAstV) group VA1 has been associated with central nervous system infections in humans and other mammals.

Replicon systems have been utilized for high-throughput screening of viral replication inhibitors and understanding the mechanisms of viral replication. Here, we report the development of a single-cycle infectious non-classical HAstV replicon system with a green fluorescent protein (GFP) reporter. This system allows for the development of both RNA-based and DNA-based replicons with initiation translation controlled in an internal ribosome entry site (IRES)-dependent manner.

To study the replication biology of the VA1 strain, we designed a replicon based on a full-genome clone of the VA1 strain in the Pt7CFEIRES vector. This vector contains all the viral machinery necessary for replication, ORF1 between the positions 1 – 2699, ORF1b between 2657 – 4222, and ORF2 between the positions 4211 – 6488, flanked by 5' and 3' untranslated regions (UTRs) between the positions 1 – 38 and 6489 – 6586, respectively. The GFP gene was cloned, replacing 849 bp of the ORF2 corresponding to the viral capsid between positions 4823 and 5672. The quantification of the GFP protein was performed using the CYTATION 1 automated fluorescence quantification system.

Our replicon system may facilitate studies on viral replication and the discovery of antiviral compounds.

EFFECT OF ZINC OXIDE NANOPARTICLES AGAINST ROTAVIRUS INFECTIVITY

Juan Diego Siary Leyva, Guadalupe González Ochoa, Jaime Manuel Ibarra Hurtado, Mario Eduardo Almada Ortiz, José Guadalupe Soñanez Organís, Luis Alberto Zamora Álvarez

Universidad de Sonora-Campus Navojoa, Blvd. Lázaro Cárdenas No. 100 Col. Francisco Villa, Navojoa, Sonora, CP 85880.
Cell phone: +1 52 6424259950. E-mail: guadalue.gonzalezchoa@unison.mx

Introduction: Viral gastroenteritis is a worldwide problem and one of the main causes of death in children under 5 years of age. The main etiological agent of this disease is Rotavirus. Nanotechnology is a relatively new science that offers us an alternative to find a treatment against this pathology. Previous studies have shown that nanoparticles have antiviral properties, which is of great interest because these properties can be used and applied in the medical area against this pathology. **Objective:** this work aimed to evaluate, quantify and determine the antiviral effect of ZnO nanoparticles (NP's-ZnO) against Rotavirus infectivity. **Methodology:** The synthesis and characterization of ZnO nanoparticles (NP's-ZnO) was done using a biogenic method. Subsequently, Rotavirus (MOI 0.1) was combined NP's-ZnO in several concentrations (100, 75, 50 and 25ug/mL) and incubated at 37 °C 1 h. The rotavirus infectivity was determined by immunoperoxidase assay. **Results:** In assays with rotavirus plus NP's-ZnO at 100, 75, and 50 the infectivity dropped to 73, 72 and 71% respectively; but a significative reduction of infectivity was observed in assays with rotavirus with NP's-ZnO at 25ug/mL where the rotavirus infectivity dropped to 58%. Our results indicate that NP's-ZnO might block rotavirus infectivity.

References

1. Bellido, J., Varela, M., Rius i Gibert, C., Pañella, H., Sanz, J., Sánchez, A., Hernandez, G., Alemany, L., Moraga, F., García, S., Puig, J., & Godoy, P. (2007). Epidemiología de las gastroenteritis agudas víricas aspectos actuales. EMISA.
2. Bishop, R. (2009). Discovery of rotavirus: Implications for child health. In *Journal of gastroenterology and hepatology*: Vol. 24 Suppl 3 (pp. S81–S85). <https://doi.org/10.1111/j.1440-1746.2009.06076.x>
3. Estes, M., & Greenberg, H. (2013). *Fields Virology: Rotaviruses* (D. Knipe & Howley P, Eds.; Rotaviruses).
4. Mirzaei, H., & Darroudi, M. (2017). Zinc oxide nanoparticles: Biological synthesis and biomedical applications. In *Ceramics International* (Vol. 43, Issue 1, pp. 907–914). Elsevier Ltd. <https://doi.org/10.1016/j.ceramint.2016.10.051>
5. Ghaffari, H., Tavakoli, A., Moradi, A., Tabarraei, A., Bokharaei-Salim, F., Zahmatkeshan, M., Farahmand, M., Javanmard, D., Kiani, S. J., Esghaei, M., Pirhajati-Mahabadi, V., Ataei-Pirkooh, A., & Monavari, S. H. (2019). Inhibition of H1N1 influenza virus infection by zinc oxide nanoparticles: Another emerging application of nanomedicine. *Journal of Biomedical Science*, 26(1). <https://doi.org/10.1186/s12929-019-0563-4>

THE FOLLICLE STIMULATING HORMONE MODULATES PROTEIN TYROSINE PHOSPHORYLATION, HYPERACTIVATION AND ACROSOME REACTION DURING HUMAN SPERM CAPACITATION

Viridiana Estefany Solis Ayala¹, Gabriela Hernández Silva¹, Jaqueline Ugalde Zavala¹,
Yadira Hernández Rueda², Víctor Torres-Flores², Mayel Chirinos¹

¹Departamento de Biología de la Reproducción Dr. Carlos Gual Castro, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán, Ciudad de México. ²Laboratorio de Biomembranas, Facultad de Medicina, Universidad Nacional Autónoma de México, Ciudad de México.
E-mail: fanysa.96@hotmail.com

Capacitation is the process by which mammalian spermatozoa acquire the ability to fertilize the oocyte. During this process, spermatozoa experiment a series of biochemical and physiology changes that initiate a signal transduction cascade that ultimately promotes protein tyrosine phosphorylation, changes in the motility pattern (hyperactivation), and prepare cells to undergo the acrosome reaction. These changes are modulated by molecules present in the female genital tract. One of these molecules is the follicle-stimulating hormone (FSH), which reaches its maximum concentrations in serum during the woman's fertile window and is also present in the follicular fluid. Since sperm preparing to fertilize can be exposed to FSH, our aim was to evaluate its participation in sperm capacitation.

Semen samples were obtained from normozoospermic volunteer donors and spermatozoa were separated from seminal fluid by discontinuous density gradients. We evaluated FSH receptor (FSHr) expression and the effect of FSH in protein tyrosine phosphorylation (pY), motility and acrosome reaction (AR) of spermatozoa incubated under capacitating conditions for one or four hours.

We found that FSHr is expressed in the post-acrosomal segment, midpiece and tail of human spermatozoa. Moreover, incubation with FSH at a concentration of 50 ng/mL increased pY, hyperactivation and AR, while a concentration of 200 ng/mL only increased pY and AR. We conclude that FSH may play a role in sperm capacitation, and therefore must be further investigated to understand the mechanisms that regulate human fertilization.

MOLECULAR MODELING OF THE EFFECT OF NON-SYNONYMOUS ABCC4 MUTATIONS OVER MRP4 CONFORMATION AND SUBSTRATE BINDING

Valeria Jacqueline Soto Ontiveros^{1,2,4}, Giovanny Aguilera Durán³, Guadalupe García-Alcocer^{1,2}, Edgardo Becerra Becerra¹, Alicia Irasema Mendieta Trejo^{1,2}, Antonio Romo Mancillas^{1,4*} and Laura Cristina Berumen Segura^{1,2*}

¹Posgrado en Ciencias Químico Biológicas, Facultad de Química, Universidad Autónoma de Querétaro, Cerro de las Campanas S/N, Querétaro 76010, México; ²Unidad de Investigación Genética, Facultad de Química, Universidad Autónoma de Querétaro, Centro Universitario, Querétaro 76010, Mexico; ³Laboratorio de Química Cuántica y Modelado Molecular, Unidad Académica de Ciencias Químicas, Universidad Autónoma de Zacatecas, Zacatecas 98160, México; ⁴Grupo de Diseño Asistido por Computadora y Síntesis de Fármacos, Facultad de Química, Universidad Autónoma de Querétaro, Centro Universitario, Querétaro 76010, México.

*Correspondence: ruben.romo@uaq.mx (A.R.M.); lcbsq@yahoo.com (L.C.B.S.)
Tel.: +52-442-192-1200-75032 (A.R.M.)

Multidrug resistance protein-4 (MRP4) is ubiquitously expressed in most human tissues and has a broad substrate specificity, including many chemotherapeutic agents. Dysregulation of MRP4 has been reported in several pathological conditions, particularly cancer, its overexpression is associated with poor prognosis. The non-synonymous MRP4 mutations Phe368Trp (F368W) and Arg375Ser (R375S) variants produce altered transport pattern, with a greater effect on the transport of the substrate drug methotrexate and to a lesser extent on the transport of endogenous substrates; however, a detailed understanding of the structural and mechanistic basis of their substrate recognition and the effect on the transport of other drugs and endogenous substrates is still not fully understood. Thus, the present work aimed to compare the effect of two MRP4 variants (F368W and R375S) on the structure and ligand affinity between endogenous and drug substrates compared to the wild-type (WT) protein, using computational chemistry tools such as threading modeling, molecular docking and molecular dynamics simulations. The results showed significantly different conformations between MRP4 variants and WT-MRP4. The structure of the F368W model is more similar to that of the WT, but with reduced movement of transmembrane passes 6 and 12, which are responsible for ligand recognition and transport. On the other hand, the structure of the R375S model adopted a significantly more compressed/closed conformation compared to the WT and a null movement of transmembrane passes 6 and 12. Despite the conformational changes in the structure of the F368W and R375S mutants, docking scores (DS) values for endogenous ligands remain steady compared to WT model. However, most of the drug substrates significantly decreased their DS in the F368W model compared to the WT model; for the R375S variant, only leucovorin followed this behavior, hydrochlorothiazide and furosemide obtained a significantly more positive DS. In conclusion, the F368W mutant impairs the transport of drug substrates. This demonstrates the mechanistic relevance of this amino acid in differential transport and highlights its importance in the development of specific MRP4 inhibitors with therapeutic potential.

IN SILICO ANALYSIS OF STRUCTURAL MOTIFS IN ANTIBODIES WITH BIOTHERAPEUTIC POTENTIAL AGAINST SARS-COV-2

Wanda Yael Tlalmis Guzmán¹, Jesús Agustín Badillo Corona², Martha Pedraza-Escalona^{*3}

¹Unidad de Desarrollo e Investigación en Bioterapéuticos, Escuela Nacional de Ciencias Biológicas IPN, Ciudad de México, Mexico; ²Unidad Profesional Interdisciplinaria de Biotecnología, IPN, Ciudad de México, México; ³CONAHCyT- Unidad de Desarrollo e Investigación en Bioterapéuticos, Escuela Nacional de Ciencias Biológicas, IPN, Ciudad de México, Mexico.

*E-mail: martha.pedraza@udibi.com.mx

In this study, we analyzed the molecular characteristics of the human monoclonal antibodies (mAbs) that bind to the Receptor Binding Domain (RBD) of SARS-CoV-2, a crucial element for interaction with the ACE2 receptor in human host cells. Our goal was to identify structural motifs associated with the biotherapeutic activity of mAbs and public clonotypes, which are sets of common antibody sequences in different individuals. This information help us to understand how the somatic hypermutation process in B cells generates broad neutralizing antibodies against the immune evasion strategies of the SARS-CoV-2 variants. We worked with the open database CoV-AbDab¹, which includes 12,916 records of neutralizing antibodies against betacoronaviruses like SARS-CoV-2, updated as of February 8, 2024. We selected 356 mAbs capable of neutralizing more than seven variant of concern from SARS-CoV-2, according to CDC classification. The data from these antibodies were curated and validated through literature review. The sequences were analyzed for V(D)J gene use, the pairwise association, the CDR3 length, and amino acid frequency using R, IMGT server and Clustal Omega. We identified the presence of sequence motifs using Python and MEME. Furthermore, we determined the structural role of these motifs in the interaction with the RBD. From these analyses, the most common IGHV genes are 1-69, 3-9, and 3-53; whereas the IGHJ3, IGHJ4 and IGHJ6 genes were most frequent, where IGHJ4 showed the highest incidence; for the D genes, the most common are the IGHD6-13.01 and IGHD6-25.01. Some of these results were identified previously^{2,3}. Interestingly, we found at least eight sequence motifs conformed by four residues and three motifs of five residues in the CDR-H3. Whereas we identified seven sequences motifs of four residues and three of five residues in the CDR-L3, which are present in more than 5% of the analyzed sequences coming from different patients around the world. The identification of these structural motifs in antibodies that retain efficacy against multiple variants can guide the development of more robust and universal treatments against SARS-CoV-2.

References

1. Matthew I J Raybould, Aleksandr Kovaltsuk, Claire Marks, Charlotte M Deane. (2020). CoV-AbDab: the Coronavirus Antibody Database, *Bioinformatics*, 37, 734–735.
2. Jiang, J., Boughter, C. T., Ahmad, J., Natarajan, K., Boyd, L. F., Meier-Schellersheim, M., & Margulies, D. H. (2023). SARS-CoV-2 antibodies recognize 23 distinct epitopic sites on the receptor binding domain. *Communications Biology*, 6,
3. Zhang, Q., Ju, B., Ge, J., Chan, J. F., Cheng, L., Wang, R., Huang, W., Fang, M., Chen, P., Zhou, B., Song, S., Shan, S., Yan, B., Zhang, S., Ge, X., Yu, J., Zhao, J., Wang, H., Liu, L., Lv, Q., ... Zhang, Z. (2021). Potent and protective IGHV3-53/3-66 public antibodies and their shared escape mutant on the spike of SARS-CoV-2. *Nature communications*, 12(1), 4210.

EXPRESSION OF THE CCD4-1, CCD4-2, CCD4-3, CCD4-4 GENES IN DIFFERENT TISSUES OF THE N4 MORPHOTYPE OF *BIXA ORELLANA* L. BY PCR

Diego Torres-Pech¹, Margarita Aguilar-Espinosa¹, Renata Rivera-Madrid¹, Sara Nahuat-Dzib²

¹Unidad de Biología Integrativa. Centro de Investigación Científica de Yucatán, A.C. Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo, Mérida, Yucatán, México. CP 97205. ² Instituto Tecnológico de Mérida, Km 5 Carretera Mérida Progreso A.P 911 C.P. 97118. Mérida Yucatán México.
Contact renata@cicy.mx

Bixa orellana L. is a shrubby plant native to the American tropics belonging to the bixaceae genus. One of the characteristics that distinguish this plant is that it contains a red-orange pigment of an apocarotenoid nature called bixin, which accumulates in the aril of the seeds.¹ In the analysis of the transcriptome of *Bixa orellana* L., three key enzymes, a carotenoid cleavage dioxygenase (CCD), an aldehyde deoxygenase (ALDH) and a methyl transferase (MET)² were proposed, which could potentially be involved in the biosynthetic pathway of this pigment. The present work focuses on the first of these enzymes (CCDs) whose biological and chemical functions have been confirmed by *in vitro* assays in *E. coli* using achiote leaf and immature/ripe seed tissues as a study model.^{1,2,3} With the aim of knowing the expression of the CCD4-1, CCD4-2, CCD4-3, CCD4-4 subfamily in different tissues of *B. orellana*. In this research, a comparative analysis of gene of the CCDs transcript was carried out in 6 tissues, root, stem, leaf, flower and fruits of achiote in morphotype N4. The analysis was performed with polymerase chain reaction (PCR) technique, taking as reference the expression of the 18S ribosomal gene. The data obtained show a differential expression of the CCDs gene in all tissues analyzed in this study, indicating that CCD4-1 it tends to be expressed preferentially in photosynthetic tissues: leaf and stem. CCD4-2 it tends to be expressed in root, stem, leaf, flower. CCD4-3 and CCD4-4 were expressed in root, stem, leaf and fruit. These results confirm that CCD4-1, CCD4-2, CCD4-3, CCD4-4 have different substrate specificity as well as and activity specific to tissue type and developmental stage.

Reference

1. Carballo-Uicab, V. M., Cárdenas-Conejo, Y., Vallejo-Cardona, A. A., Aguilar-Espinosa, M., Rodríguez-Campos, J., Serrano-Posada, H., Rivera-Madrid, R. (2019). doi:<https://doi.org/10.7717/peerj.7064>, <https://doi.org/10.7717/peerj.7064>
2. Cárdenas-Conejo, Y., Carballo-Uicab, V., Lieberman, M., Espinosa, M. A., Coma, L., & Madrid, R. R. (2015) doi:10.1186/s12864-015-2065-4
3. Us-Camas, R., Aguilar-Espinosa, M., Rodríguez-Campos, J., Vallejo-Cardona, A. A., Manuel, C.-U. V., Serrano-Posada, H., & Rivera-Madrid, R. (11 de February de 2022). . Plant Sci. doi:<https://doi.org/10.3389/fpls2022.82908>

BENEFICIAL ELEMENT OR NOT? EXPLORING THE EFFECTS OF TiO₂ NANOPARTICLES ON COMMON BEAN (*PHASEOLUS VULGARIS* L.)

Jessica Denisse Valle-García¹, Edgar Vázquez-Núñez², Fabián Fernández-Luqueño³

¹Transdisciplinary Doctoral Program in Scientific and Technological Development for the Society, Cinvestav-Zacatenco, Mexico City, ²Division of Sciences and Engineering, University of Guanajuato, Leon, Guanajuato, ³Sustainability of Natural Resources and Energy Programs, Cinvestav-Salttillo, Ramos Arizpe, Coahuila de Zaragoza, 07360, Mexico City.
E-mail: jessicad.valleg@cinvestav.mx

Plants need essential elements for their growth and development, according to Arnon and Stout¹ these elements satisfy the following conditions: the plant cannot complete its life cycle without the element, the function of each element cannot be performed by another, and it must be involved in the metabolism. There are other elements that, although not considered essential, present a positive response in certain crops or under specific conditions, one of them is Ti. Lyu *et al.*² mention that Ti can participate in the biological fixation of nitrogen (BNF) in legumes and can increase the absorption of other nutritive elements such as Fe. For legumes, the process of BNF is essential. The common bean is a legume that is distributed worldwide; for Mexico it represents 36% of the daily protein intake and is ecologically important due to its ability to associate with bacteria (Lara-Flores³; Nosengo⁴). In this research, the effect of TiO₂ nanoparticles (NPs) on common bean variety pinto Saltillo in the presence of *Rhizobium phaseoli* was analyzed. A completely randomized experiment was carried out using the following treatments: T1= soil + plant, T2=soil + plant + 150 mg NPs TiO₂ kg⁻¹ dry soil, T3= soil + plant + 300 mg NPs TiO₂ kg⁻¹ dry soil, T4= soil + plant + *Rhizobium*, T5= soil + plant + *Rhizobium* + 150 mg NPs TiO₂ kg⁻¹ dry soil, T6= soil + plant + *Rhizobium* + 300 mg NPs TiO₂ kg⁻¹ dry soil. Two destructive samplings were performed at 45 and 120 days after emergence (DAE) to determine plant length, dry biomass, number of leaves, number of nodules, number of pods and seeds; chlorophyll content in leaves was also determined. The results were analyzed using an ANOVA ($p < 0.05$). The highest number of nodules was observed found in treatments T1 and T4, i.e., without the presence of NPs. The presence of NPs and *Rhizobium* to increase some variable at 120 DAE, for instance, T6 presented the largest plant length (68.25±12.31cm); a similar trend was seen in amount of leaves (16.50±2.88), pods (8.75±4.03) and seeds (19.00±6.58), compared to the control treatment (T1). At 45 DAE chlorophyll A was always higher with respect to chlorophyll B and the A/B ratio was *ca.* 2. These results allow to open a panorama toward the study of the effect of NPs on crops that could benefit the agricultural sector or to propose methodologies that broad further investigation of the advantages and drawbacks of nanotechnology in edible or industrial crops.

References

1. Arnon DI, Stout PR (1939). *Plant Physiol.* 14, 371–375.
2. Lyu S, Wei X, Chen J, Wang C, Wang X, Pan D (2017). *Front Plant Sci*, 8, 597
3. Lara-Flores M (2015). *Revista Digital Universitaria*, 16(2). Recuperado de <http://www.revista.unam.mx/vol.16/num2/art09/#>
4. Nosengo N (2003). *Nature*, 425, 894–895.

TOWARDS THE ALLOTOPIC EXPRESSION OF A CHIMERIC COX3 GENE IN THE YEAST *SACCHAROMYCES CEREVISIAE*

Miriam Vázquez-Acevedo¹, Patrice P. Hamel² and Diego González-Halphen¹

¹Departamento de Genética Molecular, Instituto de Fisiología Celular, UNAM. Apartado Postal 70-242 CP 04510.
Tel. (5255)-5622-5620. E-mail: mvazquez@ifc.unam.mx

²Department of Molecular Genetics and Department of Biological Chemistry and Pharmacology,
Ohio State University, Columbus, Ohio, USA

According to the endosymbiotic theory, most of the genes originally present in the genome of the alpha-proteobacterium ancestor that gave rise to mitochondria, were lost, or transferred to the nucleus. During evolution, mitochondrial genomes reduced their size and nowadays only carry a subset of genes that encode OXPHOS proteins, ribosomal RNAs and tRNAs. Cytochrome c oxidase (CcO) is the terminal complex of the mitochondrial respiratory chain. The three genes encoding the core subunits of this complex, Cox1, Cox2, and Cox3 are usually located in the mitochondrial genome. However, in some chlorophycean algae like *Polytomella parva*, the *COX3* gene is encoded in the nucleus. This gene naturally migrated to the nucleus, acquired polyadenylation signals, a nucleotide sequence encoding a mitochondrial targeting sequence (MTS), and suffered changes in its genetic code and in its codon usage. Furthermore, its cognate protein product exhibits a diminished hydrophobicity and is readily internalized into mitochondria and assembled into CcO (1). The artificial relocation of a mitochondrial gene to the nucleus is called allotopic expression, a technique with potential applications for the development of gene therapies for human mitochondrial diseases. In this study, we explored conditions to carry out the allotopic expression of the *COX3* gene in *Saccharomyces cerevisiae*. We used a yeast null strain lacking the mitochondrial *cox3* gene, that has also lost its respiratory capacity. We then introduced a cDNA construct encoding the Cox3 protein fused to the mitochondrial targeting sequence of the Oxal1 insertase. Additionally, we made modifications in the amino acid sequence of three transmembrane segments (TMS) of the yeast Cox3 subunit, replacing them with the corresponding TMS of the *P. parva* subunit (Cox3q). We also built an additional cDNA construct (based on Cox3q) to which we added a sequence encoding the mitochondrial targeting sequence of the hydroxy-benzoate polyprenyl-transferase of maize (ZMCox3q), which is known to facilitate the internalization of several cytosol-synthesized proteins into mitochondria (2). Finally, we evaluated the respiratory growth of our yeast transformants in non-fermentative carbon sources (growth on ethanol, glycerol and lactate).

Reference

1. The Journal of Biological Chemistry (2000) Vol. 275, No. 39 pp 30144-30152
2. Cell Reports (2018) Vol. 22, pp 2818-2826

TARGETING CONSERVED EPITOPES OF INFLUENZA VIRUS PROTEINS TO DENDRITIC CELLS FOR GENERATING A HETEROTYPIC IMMUNE RESPONSE

B. M. Felipe de Jesús Vázquez Alba, M. in C. Herbey Oswaldo Padilla Quirarte,
Dra. María de Lourdes Gutiérrez Xicotencatl, Dra. Delia Vanessa López Guererro,
Dr. Fernando Roger Esquivel Guadarrama

Faculty of Medicine, Autonomous University of the State of Morelos, Leñeros S/N Los Volcanes, 62350, Cuernavaca, Morelos.
Contact: 7343460454, fernando.esquivel@uaem.mx

The Influenza A virus (IAV) continues to pose a global health threat. However, annual vaccines offer limited homotypic protection, and there is always the risk of new strains emerging each year, causing pandemics. Thus, it is necessary to explore new vaccination strategies. Targeting antigens to dendritic cells (DCs) using monoclonal antibodies (mAbs) has proven to be an interesting strategy to enhance T and B cell responses. In this study, we analyzed whether synthetic peptides representing highly conserved B and T cell epitopes from different IAV proteins, chemically conjugated to a mAb specific for the DEC-205 molecule present in DCs, induce a superior protective immune response against infection compared to free peptides. Groups of BALB/c mice were immunized three times subcutaneously (s.c) with hemagglutinin peptides 322-356 (B cell epitope) and 96-104 (Th cell epitope), nucleoprotein 182-205 (Th cell epitope), or the matrix 2 ectodomain (M2e) 1-24 (Th and B cell epitope) in the presence of poly I:C as an adjuvant. Alternatively, another group of mice were immunized with each of the peptides conjugated to an anti-DEC-205 mAb. Immunized mice were challenged intranasally (i.n) with 30LD50 of IAV H1N1/New Caledonia/20/99 or H3N2/NT/60/68, and weight loss and survival were monitored for 10 days. It was found that only the M2e1-24 peptide conjugated to the anti-DEC-205 mAb induced a higher level of protection against IAV infection compared to the free peptide. This protection was heterotypic and antibody-dependent. On the other hand, unexpectedly, it was found that HA and NP peptides conjugated to the mAb, which are Th cell epitopes, induced higher mortality than the free peptides. These results indicate that targeting epitopes derived from influenza virus proteins to DCs can enhance their immunogenicity, but this will depend on the characteristics of the epitope used.

FUNCTIONAL ROLE ASSESSMENT OF DELTASATELLITES IN THE ACCUMULATION DYNAMICS OF BEGOMOVIRAL TITLES IN HOST PLANTS

José Manuel Vázquez Rodríguez¹ and Yair Cárdenas-Conejo²

^{1,2}Laboratorio de Biología Sintética Estructural y Molecular de la Universidad de Colima.
Carretera los limones-Loma de Juárez CP. 28629, Colima, México.
E-mail: ycardenasco@conahcyt.mx

Begomoviruses (Geminiviridae family) are a diverse group of single-stranded DNA (ssDNA) plant viruses posing a significant threat to global crop production. Their compact genomes encode all essential functions for completing their viral replication cycle. However, the complexity of begomoviral infections often increases with the presence of satellite molecules. These small, circular ssDNA elements can modulate viral pathogenicity and are classified into four main types: alpha, beta, delta satellites, and SEGS-21 (sequence-enhancing geminivirus symptoms). Alpha, beta, and SEGS-2 satellites encode proteins with well-established functions. In contrast, the functions of delta satellites remain unclear, despite being unique non-coding entities devoid of protein-coding genes and their demonstrated influence on begomovirus titer accumulation in the model plant *Nicotiana benthamiana*². This study delves into the molecular mechanisms underlying this intriguing phenomenon. Through a comprehensive molecular characterization of delta satellites, we aim to identify genetic regions harboring non-coding regulatory RNAs, such as microRNAs (miRNAs), long non-coding RNAs, or siRNAs. We will also focus on small open reading frames (<100 codons), which are important sources of functional peptides in begomoviruses³. Analysis of satellite sequences revealed a subset encoding small proteins subsequently grouped into four distinct clusters. Additionally, a subset of satellites was identified as potentially harboring hypothetical miRNAs. Notably, one such miRNA candidate exhibited sequence complementarity to the RDR2 gene, encoding a key component of the RNA-directed silencing pathway implicated in plant defense against viral infections. To elucidate the functional roles of these newly identified small proteins and the RDR2-targeting miRNA, we will employ heterologous expression systems and gain-of-function assays in planta. These experiments will provide valuable insights into the potential mechanisms by which delta satellites modulate begomovirus infection. Understanding these regulatory pathways could pave the way for the development of novel strategies to counteract begomoviral diseases and protect crop production.

References

1. J Virol. 2021 Oct 13;95(21):e0043221. doi: 10.1128/JVI.00432-21.
2. New Phytol. 2016 Nov;212(3):680-692. doi: 10.1111/nph.14071
3. Nat Commun. 2021 Jul 13;12(1):4278. doi: 10.1038/s41467-021-24617-4.

DETERMINATION OF THE MOLECULAR MECHANISM OF INTERACTION BETWEEN HGOS2 PROTEIN AND THE ANTIAPOPTOTIC PROTEIN BCL2

Ligia Vega-Becerra¹ and Samuel Lara-González¹

¹Instituto Potosino de Investigación Científica y Tecnológica A.C, División de Biología Molecular, San Luis Potosí 78216.
E-mail: samuel.lara@ipicyt.edu.mx

GOS2 is a 103 amino acid protein first described in mononuclear cells where it was differentially expressed during the drug-induced cell cycle transition from G0 to G1 phase.¹ This protein has only been identified in vertebrates and is highly conserved between mouse and human species with 78% global identity.^{2,3} GOS2 is a protein involved in multiple cellular processes in vertebrates. Its best known function is the inhibition of the enzyme triglyceride lipase of adipose tissue, resulting in the negative regulation of lipolysis.⁴

In this work we focus on the participation of GOS2 in apoptosis, where upon interaction with Bcl-2 it releases the Bax protein disrupting the formation of the Bcl-2/Bax heterodimer. The release of Bax initiates the apoptosis process inhibiting cell proliferation. The aim of this work is to describe the molecular mechanism by which GOS2 interacts with Bcl-2 and dissociates the Bcl-2/Bax complex by means of biochemical and biophysical techniques. For this purpose, SUMO-hGOS2, hBcl-2, mBcl-2 and hBcl-2 XL constructs were cloned and expressed in the pET28pps vector with the restriction enzymes NdeI and HindIII. Subsequently, each protein was purified separately by nickel affinity chromatography (Ni-NTA) and a second purification by molecular exclusion chromatography (SEC). hGOS2 protein yielded a total production of 1.64 mg/l, for hBcl-2 a total yield of 0.56 mg/l was obtained, for mBcl-2 a total yield of 7.3 mg/l was obtained and finally for hBcl-2 XI a total yield of 9.954 mg/l was obtained. The identity of each protein was confirmed by western blot. The evaluation of the interaction of the hGOS2/hBcl-2 complex by molecular exclusion chromatography was successful obtaining a complex with a weight of 48.9 kDa. With this we conclude that there is a strong and stable interaction between hGOS2 and hBcl-2 proteins since the complex can be observed from co-purification with Bcl-2 by Ni-NTA, as well as with SEC where the complex elutes in a single peak. With the previously purified samples, crystallization trials of the complex were carried out. However, the hBcl-2 protein shows self-degradation in a matter of days, so it is still necessary to test the complex formation with the mBcl-2 and hBcl-2 XL proteins, since the total production of these two proteins is much higher and they are easier to work with compared to hBcl-2.

References

1. Russell, L., & Forsdyke, D. R. (1991). DNA and cell biology, 10(8), 581-591.
2. Cerk, I. K., Wechselberger, L., & Oberer, M. (2018). Current protein & peptide science, 19(2), 221-233.
3. Páez-Pérez, E. D., Llamas-García, M. L., Benítez-Cardoza, C. G., Montero-Morán, G. M., & Lara-González, S. (2020). ACS omega, 5(40), 25841-25847.
4. Yang, X., Lu, X., Lombès, M., Rha, G. B., Chi, Y. I., Guerin, T. M., Smart, E. J., & Liu, J. (2010). Cell metabolism, 11(3), 194-205.

INVOLVEMENT OF HUMAN PAPILLOMAVIRUS 16 E5 PROTEIN IN THE ACTIVATION OF THE WNT/ β -CATENIN PATHWAY

Velásquez-Hernández Jimena Yadira^{1,2}, Muñoz-Bello J. Omar¹, Contreras-Paredes Adriana¹ y Lizano Marcela^{1,3}

¹Instituto Nacional de Cancerología CP 14080, ²Programa de Maestría y Doctorado en Ciencias Biológicas, Universidad Nacional Autónoma de México. ³Instituto de Investigaciones Biomédicas, UNAM, C.P. 04510 CdMx
*Cel: 5516570573. E-mail: jimnavh15@ciencias.unam.mx

High-risk human papillomavirus infection, especially with HPV16, is a determining factor in the development of cervical cancer. Although the role of HPV E6 and E7 oncoproteins in cell transformation has been widely recognized, the importance of E5 protein as an oncogenic factor has been highlighted in recent years. It has been shown that HPV16 E5 activates the PI3K/Akt signaling pathway, thereby stimulating cell proliferation. Additionally, the active form of Akt has been found to inactivate GSK3 β kinase, which could potentially have a positive impact on β -catenin function. The Wnt/ β -catenin signaling pathway, crucial in the regulation of cell proliferation and differentiation, is altered in HPV-related cancers, where E6 and E7 play a decisive role. However, the effect of E5 protein on this pathway has not been explored until now. Therefore, the aim of this study is to investigate how HPV16 E5 affects the activation of the Wnt/ β -catenin pathway and its impact on processes related to cervical cancer. Our results indicate that in HaCaT cells stably transfected with HPV16 E5, stimulation with epidermal growth factor (EGF) and EGFR recycling by E5 protein lead to an increase in β -catenin protein and mRNA levels. Additionally, a notable decrease in the expression of negative regulators of the pathway, such as Axin 2, is observed. In parallel, an increase in the expression of Wnt/ β -catenin pathway target genes, such as Cyclin D1, cMyc, and cJun, is evident.

We thank CONACYT for the financial support provided during the realization of this project with grant (CVU) 1191125 and the support of PRONAI-7-Virus and Cancer 303044.

IN SILICO STUDIES OF THE SUBSTRATE SCOPE OF GLYCEROL DEHYDROGENASE FROM *E. COLI*

Julian L. Wissner¹, Diego A. Del Castillo Copland¹, Wendy Escobedo-Hinojosa^{1*}

¹Laboratorio de Productos Naturales Marinos. Unidad de Química en Sisal, Yucatán. Facultad de Química de la Universidad Nacional Autónoma de México. *Corresponding author: Wendy Escobedo-Hinojosa. Unidad de Química en Sisal, Yucatán. Puerto de Abrigo S/N, UNAM, 97355 Sisal, Yucatán. Tel. +527771594447. E-mail: wendyitzeleh@quimica.unam.mx

Recently, we reported that glycerol dehydrogenase (GldA) from *Escherichia coli* strain BW25113 is capable to rearomatize *cis*-dihydrocatechol derivatives.^[1,2] Encouraged by this results, our group performed *in silico* docking studies of the different, commercially not available enantiomers of C₆-C₁₀ *cis*- and *trans*-dihydrocatechols. The results of the present study support the previously reported findings, that *cis*-dihydrocatechol, *cis*-(1*S*,2*R*)-3-methylcyclohexa-3,5-diene-1,2-diol and *cis*-(1*S*,2*R*)-3-ethylcyclohexa-3,5-diene-1,2-diol are forming reactive conformations with GldA and are thus converted.

While *cis*-(1*S*,2*S*)-1,2-dihydroxy-3-bromocyclohexa-3,5-diene and *cis*-(1*R*,2*S*)-1,2-dihydronaphthalene-1,2-diol were not forming reactive conformation with GldA and could not be converted *in vitro*^[2], their other enantiomers *cis*-(1*R*,2*R*)-1,2-dihydroxy-3-bromocyclohexa-3,5-diene and *cis*-(1*R*,2*S*)-1,2-dihydronaphthalene-1,2-diol, respectively, exhibited *in silico* reactive conformations. Interestingly, none of the docked *trans*-dihydrocatechol substrates are able to form a reactive conformation in GldA's active site, due to the disrupting proximity of one of the substrates hydroxyl groups and the co-substrate NAD⁺. In conclusion, we predict that GldA should be capable to transform halogenated C₆ *cis*-dihydrocatechols as well as larger C₁₀ *cis*-dihydrocatechols, both reactions that have not been previously reported.^[2]

Reference

1. J. L. Wissner, J. Ludwig, W. Escobedo-Hinojosa, B. Hauer, J Biotechnol 2021, 325, 380–388.
2. W. Escobedo-Hinojosa, M. A. Vila, J. L. Wissner, N. Härterich, P. Horz, C. Iglesias, B. Hauer, J Biotechnol 2023, 366, 19–24.

EFFECT EVALUATION OF CURCUMIN AND RESVERATROL COMPOUNDS ON CANCER STEM CELL-ENRICHED CULTURES DERIVED FROM CANCER CELL LINES

Jorge Aguilar Meza^{1,2}, Heriberto A. Valencia González², Marcela Lizano Soberón³, Alejandro García Carranca^{2,4}

¹Ing. en Biotecnología, Universidad Abierta y a Distancia de México, ²Laboratorio "Virus y Cáncer", Instituto Nacional de Cancerología (INCan), ³Laboratorio "Virus Oncogénicos", INCan, ⁴Laboratorio "Virus y Cáncer", INCan – Instituto de Investigaciones Biomédicas – UNAM, San Fernando 22, Sec. XVI, Tlalpan, CDMX, México. C.P. 14080.
E-mail: carranca@biomedicas.unam.mx Tel.: 5536935200 X 226

Cervical cancer is a disease that affects women worldwide. Conventional treatments involve chemotherapy and/or radiotherapy. However, these treatments may have a low success rate in treating the disease. Recently, special attention has been paid to developing adjuvant treatments using natural compounds such as curcumin and resveratrol. The experiment was conducted in three-dimensional (3D) cultures with non-adherent conditions, where a higher proportion of cancer stem cells has been found than monolayer cultures. Using Western Blot, the presence of transcriptional factors Nanog and Sox-2, which are markers of stemness, was higher expressed in three-dimensional cultures. Pre-treatment with curcumin or resveratrol polyphenols in cervical cancer cell lines such as HeLa and SiHa did not significantly increase sensitivity of cancer cells to chemotherapy. However, it was possible to identify that polyphenols, at very low doses as Inhibitory Concentration 25 (IC25), calculated from monolayer cultures, could increase sensitivity treatment of cancer stem cells. Data were analyzed using Test-T, where significance variation was found between treated with polyphenols and untreated 3D cultures data. On the other hand, no significant difference was obtained between treatments. Data suggest that adjuvant treatments using curcumin and/or resveratrol could play a crucial role in the cervical cancer therapy due their ability to increase the sensitivity of cancer cells, even stem cells, to chemotherapy, as well as their selective cytotoxic effect, makes them valuable allies in oncological care.

Acknowledgment. CONAHCYT PRONAI 303044 & Instituto Nacional de Cancerología.

References

1. Santos-López, G., *et al.* (2015). *Rev Med Inst Mex Seguro Soc*, 53 Suppl 2, S166–S171.
2. Sun, X., *et al.* (2022). *Int J Mol Med*, 47, 335–345.
3. Teymouri, M., *et al.* (2017). *BioFactors*, 43: 331–346.

PHYTOCHEMICAL PROFILE OF MEXICAN MYRTLE (*PSIDIUM SARTORIANUM*) LEAVES EXTRACTS AND INFUSION BY TRADITIONAL METHOD

José Guadalupe Torres Salazar¹, Juan Paulino Segoviano León¹
and Xiomara Patricia Perea Domínguez^{1*}

¹Departamento de Salud-Licenciatura en Ciencias Biomédicas, Universidad Autónoma de Occidente, Unidad Regional Guasave, Av. Universidad S/N, Fraccionamiento Villa Universidad, C.P. 81048, Guasave, Sinaloa, México.
E-mail: xiomara.perea@uadeo.mx

Myrtle (*Psidium sartorianum* (O. Berg) Nied Myrtaceae) is a multi-purpose plant native to Mexico and tropical America. Its fruit is often used in traditional medicine in different regions to treat problems such as diarrhea, dysentery, scabies, intestinal parasites and as a healing agent ¹. Belongs to the Plantae kingdom, Magnoliophyta division, Magnoliopsida class, Myrtales order, Myrtaceae family, *Psidium* genus and *P. sartorianum* species.

In Mexico, myrtle plant is distributed in several states, predominantly in those with a tropical climate, mainly in the wild ². The uses it has in Mexico range from firewood, the construction of houses, the production of sweets, flavored waters, syrups, ice cream, tamales and popsicles, the leaves are also used for their tonic and astringent properties, it is used to treat diarrhea, cough, vomiting and stomach pains, the macerated fruits and leaves are used topically on ulcers, wounds and infected parts of the skin, in Sinaloa the stem is used in the treatment of dental diseases^{2,3}, It is worth mentioning that also in Sinaloa in some rural areas of the northwest of the state they use an infusion of myrtle leaves to control sugar levels in diabetic people who usually do not have easy access to modern medicine. Pío-León et al., (2013) carried out a study on the nutraceutical and bactericidal potential of some endemic fruits such as nanchi (*Byrsonima crassifolia*), myrtle (*Psidium sartorianum*) and ayale (*Crescentia alata Kunth*) finding that myrtle fruit has bactericidal potential and a high nutraceutical value. On the other hand, Fuentes et al., (2016) carried out studies on another variety also known as myrtle native to South America (*Luma apiculata*), it is worth mentioning that this has several similarities with the myrtle distributed in Mexico such as the shape of the tree and the fruit. , with the difference that in South America it has a reddish color ⁴. In this study they found an important antioxidant capacity and, above all, a high capacity of myrtle extract as a protector of vascular damage induced by high glucose. This last finding is of great relevance to us, due to the previously mentioned use that is given to it. to the plant in rural communities of Sinaloa. Despite the multiple studies carried out in Mexico on medicinal plants, the profile of metabolites responsible for many of the properties attributed to them is still unknown, the myrtle is one of these cases.

Some studies have focused on the fruit, however, the phytochemical profile of the leaves that are used for therapeutic purposes is still unknown. In the present research, a phytochemical screening (phenolic compounds, flavonoids, tannins, saponins, triterpenes and alkaloids) was carried out on three myrtle leaf extracts in which solvents with different chemical

affinities were used (methanol, hexane) are presented. and dichloromethane) assisted with sonication⁵, additionally, an infusion was made following the instructions of inhabitants of the region about the preparation used for therapeutic purposes. Phytochemical screening gives us a first look at the possible actors involved in the bioactive potential of the plant. It is a test that, although it provides qualitative results, these can be extremely informative and fundamental in decision making to define which groups of metabolites focus the search and identification.

References

1. Pío-León, J. F.; Díaz-Camacho, S. P.; López-López, M. A.; Uribe-Beltrán, M. de J.; Willms, K.; López-Angulo, G.; Montes-Avila, J.; Delgado-Vargas, F. Actividad antibacteriana de extractos de frutos de nanchi (*byrsonima crassifolia* (L.) Kunth), arrayán (*Psidium sartorianum* (O. Berg) Nied.) y ayale (*Crescentia alata* Kunth). *Bol. Latinoam. y del Caribe Plantas Med. y Aromat.* 2013, 12 (4), 356–364.
2. Delgado-Vargas, F.; Diaz-Camacho, S. P.; Salazar-Zamora, G.; Uribe-Beltran, M. J.; Vega-Avina, R. *Psidium sartorianum* (O. Berg) Nied., an indigenous plant to Mexico, from biology to biological activity. *Sect. Title Pharm. Anal.* 2006, 13, 81–114.
3. Valera Montero, L. L.; Enríquez Nava, S.; Silos Espino, H.; Padilla Ramírez, J. S.; Perales Segovia, C.; Flores Benítez, S. Propiedades fisicoquímicas de guayabilla ("*Psidium guineense*"), arrayán ("*Psidium sartorianum*") y guayaba ("*Psidium guajava*"). *Rev. Mex. ciencias agrícolas* 2018, 9 (6), 1099–1108.
4. Fuentes, L.; Valdenegro, M.; Gómez, M.-G.; Ayala-Raso, A.; Quiroga, E.; Martínez, J.-P.; Vinet, R.; Caballero, E.; Figueroa, C. R. Characterization of fruit development and potential health benefits of arrayan (*Luma apiculata*), a native berry of South America. *Food Chem.* 2016, 196, 1239–1247. <https://doi.org/https://doi.org/10.1016/j.foodchem.2015.10.003>.
5. Schinor, E. C.; Salvador, M. J.; Turatti, I. C. C.; Zucchi, O. L. A. D.; Dias, D. A. Comparison of classical and ultrasound-assisted extractions of steroids and triterpenoids from three *Chresta* spp. *Ultrason. Sonochem.* 2004, 11 (6), 415–421. <https://doi.org/10.1016/j.ultsonch.2003.09.005>.

A KALANCHOE FLAMMEA EXTRACT IN COMBINATION WITH DRUGS CAN HELP REDUCE THE TUMOR GROWTH OF CANCER CELL LINES IN AN *IN VIVO* MODEL

Heriberto A. Valencia González¹, Ana K. Pablo-Gómez², Edgar A. Celis-Hernández², Cynthia Ordaz-Pichardo^{2†}, Jorge Cornejo Garrido², Alejandro García Carranca³

¹Laboratorio "Virus y Cáncer", Instituto de Investigaciones Biomédicas UNAM – Instituto Nacional de Cancerología. Av. San Fernando 22, Tlalpan. CDMX, 14080, México. ²Laboratorio de Biología Celular y Productos Naturales, Escuela Nacional de Medicina y Homeopatía del IPN. Guillermo Massieu Helguera 239, Gustavo A. Madero, 07320 CDMX, Mexico. E-mail: acarranca@iibiomedicas.unam.mx & jcornejog@upn.mx Tel. 55 3693 5200 Ext 227

WHO estimates project that by 2030 the incidence of cancer in general could increase by 50% with approximately 2.4 million new cases, due to exposure to risk factors for developing it. In Mexico, cervical cancer (CaCu) occupies 2nd place in deaths from neoplasia in women in 2022. While prostate cancer (CaPr) is the most common malignant tumor in men over 50 years of age, the 2nd cause of death in Hispanic men with an estimate in 2020 of around 1.4 million new cases and 375,000 deaths in the world. Despite advances in chemotherapy, surgery, radiotherapy, targeted therapy, hormonal therapy, and their combinations, the disease has not been overcome. Therefore, they look for alternatives in natural sources that can provide a greater response to the treatment¹. Mexico has a great biological diversity of plants used in traditional medicine, it is possible to obtain compounds or extracts with antitumor activity². This is the case of plants of the Kalanchoe genus endemic to the state of Tabasco in Mexico such as Kalanchoe flammula (Kf) (Belladonna) used as an anti-inflammatory, antiseptic, antitussive and in some burns³. The evaluation in vivo of antitumor activity was performed on xenografts of SiHa and PC3 cells in NOD-SCID mice. Doses of a Kf extract were administered orally, alone or in combination with drugs, and the follow-up for 14 days showed that the combination of the extract can help reduce tumoral growth, however, low doses of the Kf extract also influence the proliferation of tumor cells. The treatments with the best outcomes were 0.9 mg/kg of Kf with 1 mg/kg of Docetaxel for the PC3 cell tumor and 1 mg/kg of Kf with 1 mg/kg of Paclitaxel for the SiHa cell tumor. Agreement to CONAHCYT Pronaii-7-Virus y Cáncer 303044.

References

1. Gordon M. Cragg, John M. Pezzuto (2016). Med Princ Pract, 25(Suppl 2): 41-59
2. Casimiro Cárdenas García (2009). Encuentros en la Biología, 2: 1134-8496
3. Iván Arias-González et al. (2018). J Ethnopharmacol 10:222:133-147

MOLECULAR CHARACTERIZATION OF BINDING SITE OF β -CCB IN OLIGODENDROGLIAL GABAA RECEPTOR $\alpha 3\beta 2\gamma 1$

María Berenice Varela Correa[®], Teresa Edith Garay Rojas & Rogelio O. Arellano¹

Laboratorio de Neurofisiología Celular, Departamento de Neurobiología Celular y Molecular, Instituto de Neurobiología, UNAM, Campus Juriquilla, Querétaro, México.

[®]bvarela@comunidad.unam.mx

Calle Las Ranas #118, Santa Rosa Jaureguí, Querétaro, Querétaro, C.P. 76220
di.bere95@gmail.com Tel: 4731211913

The GABA_A receptor (GABA_AR) is the ionotropic receptor activated by γ -aminobutyric acid (GABA). GABAergic signaling modulates the myelination process through GABA_AR activation expressed in oligodendrocytes (OLs). Our data show that the GABA_AR in OLs is mainly composed by $\alpha 3\beta 2\gamma 1$ subunits with a specific pharmacological signature; in particular for a group of molecules named β -carbolines. Originally described as inverse agonists acting on the neuronal GABA_AR through the benzodiazepine (BZD) binding site, some β -carbolines such as β -CCB is instead a potent potentiator of the GABA_AR in OLs. This differential β -CCB effect in OLs versus neurons would be explained by a different binding site in each conformation.

Previously, we have shown that a mutation that eliminates the effect of BZD maintains intact the modulatory effect of β -CCB, this suggested that its positive action in OLs depends on a different binding site. In this study is explored the participation of either the loreclezole or the neurosteroids binding site in the effect of β -CCB on the GABA response in OLs.

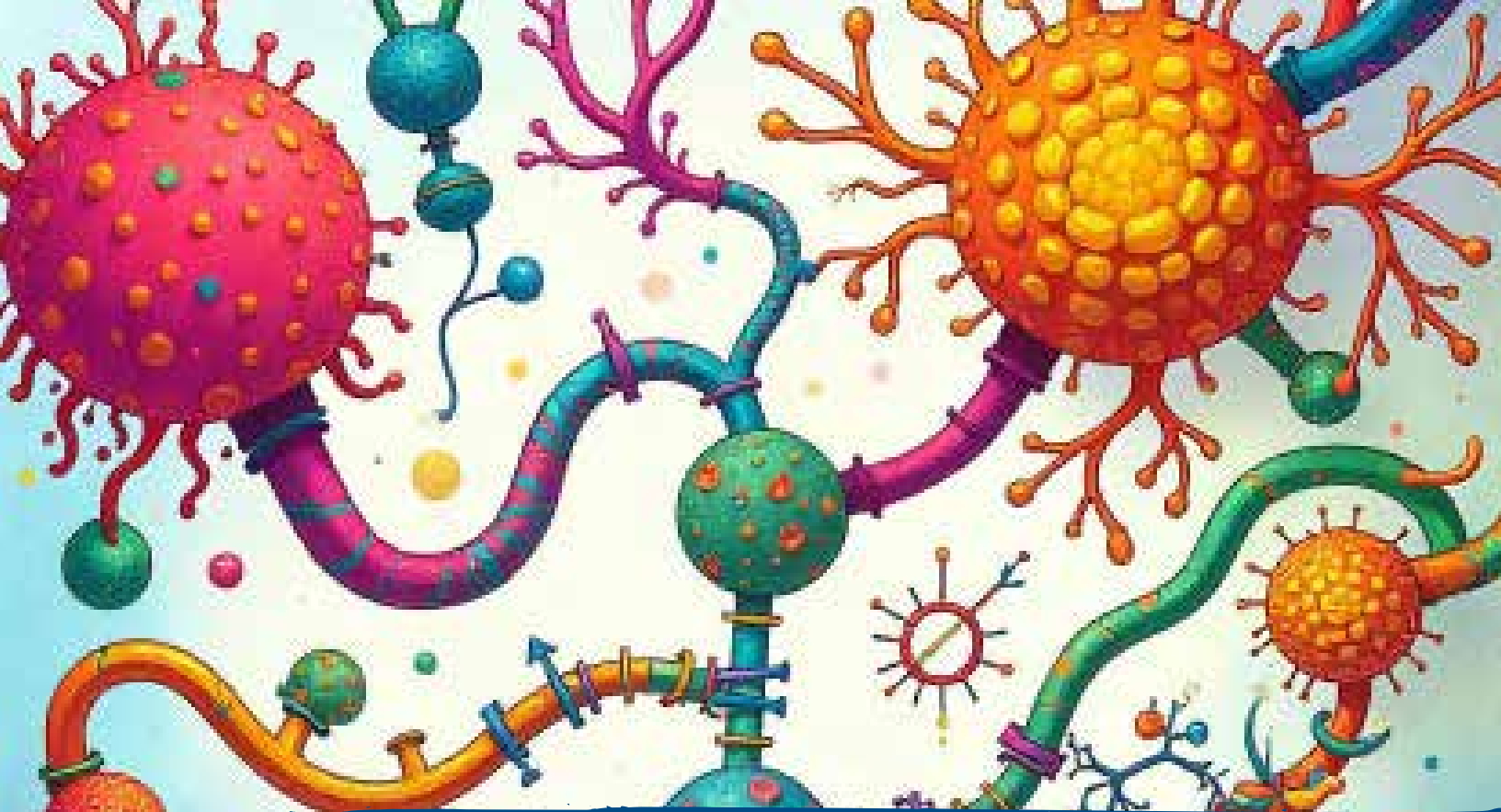
In here, using heterologous expression in *Xenopus laevis* oocytes, the mRNA of rat $\alpha 3\beta 2\gamma 1$ GABA_AR subunits were studied electrophysiologically and its characteristics were compared with those receptors in which specific mutations eliminated the effect of loreclezole ($\alpha 3\beta 2N265S\gamma 1$) or neurosteroids ($\alpha 3Q266L\beta 2\gamma 1$).

Results showed that in $\alpha 3\beta 2N265S\gamma 1$ GABA_AR was completely eliminated the sensitivity to loreclezole. However, the mutation decreased only partially the positive effect of β -CCB. This suggested that loreclezole binding site participated for that of β -CCB although the structural determinants for this site are not identical.

Neuropharmacology

Keywords. GABA_A receptor, Oligodendrocytes, β -ccb, *Xenopus* oocyte.

Acknowledgement. This work was supported by PAPIIT UNAM-México No. IN205822 and CONAHCYT CF-319740 to R.O.A. We Thank Martín García S., Alejandra Castilla L., María A. Carbajo M., Leopoldo González S., Ramón Martínez O. and Felipe Ortiz Cornejo for their technical assistance.



AUTHOR INDEX

XXXIV NATIONAL MEETING OF BIOCHEMISTRY



A

- Aarón Domínguez López** 554-563
Aarón Iván Vélez Ramírez 670
Aarón Mena-Rodríguez 250
Aarón Saúl López Carrasco 1106
A. Arturo Guevara-García 265
A. Báez-Rogelio 910
Abdú Orduña Díaz 411
Abelardo Camacho Luis 704-832
A. B. Fariás 1081
Abigail Betanzos Fernández 225-677
Abigail González Valdez 177-428-883
Abigail Moreno-Pedraza 249
Abigail Roldan-Salgado 838
Abraham Heriberto García-Campoy 751
Abraham Landa 221-1242
Abraham Madariaga Mazón 1094
Abraham Méndez-Fernández 220
Abraham Said Arellano Buendía 732-784
Abril Bernardette Martínez Rizo 848
Abril Carbajal-García 1022-1058
Abril Gijbers 376
A.C. Cepeda 319
Acela Cuamatzi de Gante 411
Acevedo Whitehouse Karina Alethya 628
A. Cortes-Morales 694
Ada Agustina Carrillo 766
Adamary M. Hernandez P. 337
Adán Dagnino Acosta 1194-1211-1236
Adán Guerrero 174-1234
Ada Sandoval-Carrillo 623
Adda Agustina Sandoval Carrillo 292-721
Addí Rhode Navarro Cruz 953-1000
Adela Guadalupe Rosas Rosas 355
Adelfo Escalante Lozada 428
Adelma Escobar Ramírez 285-294-986
Adilene Gonzalez Silva 849-913
Adolfo Gracia 378
Adolfo I. Ruiz-Ballesteros 561-570
Adolfo Padilla Mendiola 479-548
Adolfo Sierra-Santoyo 757
Adriana Alarcón Aguilar 182-553-643-961
Adriana Castillo Villanueva 339-731-793
Adriana Contreras Paredes 749-779
Adriana del Carmen Aguilar Lemarroy 232-545-590
Adriana Gallardo-González 882
Adriana Garay-Arroyo 173
Adriana González Gallardo 574-588
Adriana Guadalupe Orozco García 610
Adriana Gutiérrez-Hoya 1030-1031-1062
Adriana Inclán Popoca 627
Adriana Jazmín Legorreta-Castañeda 450-839
Adriana López-Barradas 713-728
Adriana Moreno-Rodríguez 338
Adriana M. Salazar-Montes 778
Adriana M. Salazar Puente 299
Adriana Pacheco 477
Adrián Ávalos-Rangel 387-556
Adrián Camacho-Ortiz 842-908
Adrián Díaz Pacheco 449-462
Adrián Gómez-Baltazar 154
Adrián González 847
Adrián Islas Ortíz 248
Adrián J. Rivera 1167
Adrián Ochoa-Leyva 406-475
Adrian Rivas Muñoz 1166
A. Fabiola Camacho-Gamiño 879-912
Aguayo Morales Hilda 400-957
Aguilar Melo C. M. 874-880
Aguilera-Méndez A. 397
Agustina Cano Martínez 745
Agustín Guerrero-Hernández 276-293-318-330-346
Agustín López-Munguía 475-509
Agutín Guerrero 359
Ahiezer Rodríguez-Tobón 1025
Aida Gabriela Mora Acebedo 885
Aída Guadalupe Díaz Martell 715
Aida Hamdan Partida 821
Aidán Alejandro Ortega García 969-971
Aideé Saray López Torres 1060

- Aikaterini Diamanti** 304-1224
Aketzalli Martinez Cruz 1052
Alain Raimundo Rodríguez Orozco 790
Alain R. Rodríguez Orozco 756
Alan A. Pedraza-Ramírez 1021
Alan Carrasco Carballo 918
Alan Hernández-Melgar 316-713-1150
Alan Jesé Rodríguez Almaraz 225
Alan Meneses-Carbajal 820
Alba Adriana Vallejo Cardona 533
Alba Adriana Vallejo-Cardona 386-764
Albany Reséndiz Mora 527
Albert Guskov 223
Alberto Alatorre 939
Alberto A. Lomelí-Bernal 1220
Alberto Camacho-Morales 931
Alberto Darszon 138-1234
Alberto Guevara Flores 235
Alberto Hernández-Eligio 566
Alberto Kousuke De la Herrán Arita 1082
Alberto Ramírez 814
Alberto Ramírez-Mata 555-825-826-893-898-900-920-963-1112
Alberto Tenorio-Torres 634
Alberto Zamora-Bello 243
Albros H. Poot 1091-1101
Alday Montañez F.D. 744
Aldo Agustín Pérez Fonseca 921
Aldo E. García Guerrero 171-295
Aldo Moreno-Ulloa 713-1150
Aldo O. González-Cruz 213-1173
Aldo R. Camacho-Zarco 201
Aldo R. Hernández Zamarripa 585
Aldo Yael Miranda Muñoz 672
Alejandra Cedillo Baños 663
Alejandra Contreras Valladares 739
Alejandra Figueroa Ríos 695
Alejandra Gabriela Valdez Lara 147
Alejandra García-Núñez 504
Alejandra. G. Valdéz-Lara 1140
Alejandra Hérnandez-García 482
Alejandra Mandujano Chávez 601
Alejandra Monserrat Retis-Resendiz 236-591-1217
Alejandra Ochoa-Zarzosa 433-482-500-658-1069
Alejandra Paola García Hernández 192
Alejandra Rangel-Mendoza 1008
Alejandra Romero-Utrilla 1082
Alejandra Vargas-Caraveo 683
Alejandra Vázquez Raygoza 704
Alejandro Aguayo-Reyes 153
Alejandro Aguilar-Vera 1182
Alejandro Alagón 278-514
Alejandro Alvarado Gutiérrez 520
Alejandro Angel-Cuapio 535
Alejandro Avilés-Salas 779
Alejandro Barrios Nolasco 554
Alejandro Camacho-Zamora 1082
Alejandro Canizales-Ontiveros 982
Alejandro De Las Peñas 181-567-571-580-583-596-599-649-811-812
Alejandro Delgado Rey 1195
Alejandro Elliot Flores-Oliva 941
Alejandro García Carrancá 645-1260-1263
Alejandro Garcíarrubio 480
Alejandro Hernández-Morales 386-836
Alejandro Huerta-Saquero 1215
Alejandro Israel Castillo Montoya 1179
Alejandro Juárez-Reyes 571-596
Alejandro López Hernández 1042
Alejandro López-Saavedra 609
Alejandro Olvera-Rodríguez 514
Alejandro Ordaz Ramos 1052
Alejandro Padilla-Trejo 1039
Alejandro Pereira-Santana 399-1111-1124
Alejandro Rubén Martínez Espinoza 520
Alejandro Salazar-Salgado 964
Alejandro Sánchez-Flores 378-961
Alejandro Sánchez Gonzalez 513
Alejandro Sosa Peinado 374-1245
Alejandro Vázquez-Reyes 809

- Alejandro Zentella Dehesa 320-761
Alejandro Zugasti Cruz 476
Aleph A. Corona Morales 966
Alessandra Cataldo 774
Alethia Muñiz-Ramirez 751
Alexander De Luna 163-603-615-896
Alexander E. Braun 242
Alexey Llopiz 308-314
Alexia Ramírez Ávila 342
Aleyda G. Zamudio-Martínez 232-545
Alfonso Daniel Díaz Fonseca 953-955-959
Alfonso Díaz 958-1000
Alfonso Jiménez Adón 424-448
Alfonso Leija 228
Alfonso Leija Salas 137-1186-1240-1246
Alfonso Méndez Tenorio 1117
Alfonso Miranda Molina 509
Alfonso Ragnar Torres Jiménez 587
Alfonso Salgado Aguayo 700
Alfredo Ayala-Ham 403-407
Alfredo Cabrera-Orefice 220-231
Alfredo Hernández 1182
Alfredo Herrera Estrella 426-459-484-610-828-1220
Alfredo Hidalgo-Miranda 632-634-774
Alfredo Ibarra-Sánchez 1033-1039-1050
Alfredo Martínez Jiménez 164-534
Alfredo Miranda-Martínez 1133
Alfredo Saavedra Molina 753-756-791-992-1001
Alfredo Téllez Valencia 285-294-714-731-781-793
Alicia Beltrán-Soto 1019
Alicia Gonzalez-Manjarrez 269-270-334-850-1115
Alicia Irasema Mendieta Trejo 1250
Alicia Kowaltowski 306
Alicia María Reveles-Espinoza 764
Alicia N. Aguilar Fuentes 804
Alicia Ortega-Aguilar 487-930
Alicia Reveles Espinoza 533
Alicia Vega-Segura 302-371-376
Alí Fernando Ruiz-Higareda 842-908
Alina Barquet-Nieto 693
Alina Elizabeth Torres Aguirre 526
Allison B. Herman 152
Alma Beatriz Peregrino Uriarte 200-622
Alma Cristina Salas-Leal 623
Alma Delia Campos Parra 716-723
Alma Dolores Pérez Santiago 282
Alma Karina Tamez Castellón 429
Alma Laura Díaz Pérez 307
Alma Laura Rodríguez-Piña 593
Alma Lilia Hernández-Olvera 215-617
Alma Luz Carmona 396
Alma Marlene Guadrón-Llanos 1082
Almanelly Agabo-Martínez 1129
Alma Ortiz-Plata 938
Alma Rosa Agapito-Ocampo 379
Alma Rosa Corrales Escobosa 448
Alma Rosa González Pérez 868-1144
Alma Ruth Reyes González 902
Alondra Mariana Martinez Partida 1225
Alonso Lopez-Zavala 406-475
Álvaro Marín Hernández 734-801-1108
Alvin Noé Suarez Castillo 886
Amador Campos-Valdez 393-399
Amanda Gálvez-Mariscal 726
Amanda Gayosso-Vázquez 487
Amanda Mixon-Blackwell 171-295
Amanda Renté 346
Ambar López Macay 564-595
Amelia López-Albores 315
Amelia López-Herrera 272
Amelie V. August 170
Amellalli Bazan-Cordoba 634
América Padilla Viveros 203-366-532-1193
America Selene Gaona Mendoza 210
América Vanoye-Carlo 1191
Amira Klip 143
Ana Alicia Sánchez Tusie 715-1123
Ana Belén Vargas Antillón 181
Anabel Félix Arredondo 286
Anabella Handal Silva 1165

- Anabelle Manzo-Sandoval 870
 Anabel Ortiz Caltempa 379
 Ana Bonavides-Aguilar 1096
 Ana Carolina González Trillo 1099
 Ana Carolina Tirado-Garibay 1069
 Ana Cecilia Sánchez Trujillo 633-651-768-1064
 Ana C. Murrieta 1136-1159
 Ana Cunningham 152
 Ana Gabriela Cordova Mejía 825
 Ana Gabriela Hernández Puga 1123
 Ana Gabriela Leija Montoya 289-752
 Anahí Chavarría-Krauser 182-949
 Ana Hilda Figueroa Gutiérrez 798
 Ana I. Chávez-Martínez 174-565-592-600-1114
 Anaiza Rico Luna 341-348
 Ana Karen Álvarez Contreras 810-854-862-881-914-923-927
 Ana Karen Avendaño Gutiérrez 835
 Ana Karen Díaz-Martínez 1092-1100
 Ana Karen Gómez-García 445
 Ana Karen Ponce Quiroz 895
 Ana Karen Villa Merlán 918
 Ana Karina Segovia Parra 781
 Ana K. Pablo-Gómez 1263
 Ana Laura Alonso-Nieves 474
 Ana Laura Guzmán Ortiz 226-684
 Ana Laura Valdez Arellanes 488
 Ana L. Encinas-Montes 284
 Ana Leticia Arriaga-Guerrero 930
 Ana Lidia Arellano-Ortiz 683
 Ana Lilia Hernández Alba 732-784
 Ana Lilia Martínez-Rocha 218-434-504-853
 Ana Lilia Roa Espitia 1056-1060-1063
 Ana Lilia Torres-Machorro 598-631-641
 Ana Lizbeth López-Marmolejo 596
 Ana L. Quintero-Ramírez 457
 Ana L. Roa-Espitia 1040
 Ana Lucía López-Gurgua 1107
 Ana Marcela González Bedoy 1211
 Ana Margarita Balderas Peña 555
 Ana María Castillo González 355
 Ana María García Montelongo 436-1206
 Ana María López Colomé 1013-1045-1070
 Ana María Peña Balderas 1223
 Ana María Rincón-Murillo 491
 Ana Mayra Reséndiz Corona 763
 Ana M. Rivas-Estilla 842-908
 Ana O. Quintana-Escobar 1125
 Ana Paola Balderrama Carmona 985
 Ana Paola Rivera Torres 1135-1177
 Ana Paulina Barba de la Rosa 543-1220
 Anareli Mota Morales 750
 Ana Romarowski 138
 Ana R. Rocha-Vallejo 498
 Ana Silvia González-Méndez 870
 Ana Sofía García-García 936
 Ana Sofía Ramírez-Pelayo 234-538
 Ana Sofía Salmerón Oms 625
 Ana Sonia Silva-Ramírez 936
 Ana S. Sandoval-Rodríguez 729
 Ana Valeria Martínez-Silva 976
 Ana V. Coria-Téllez 1192
 Ana Victoria Zermeño-Granados 870
 Anayansi Molina-Hernández 942
 Ana Yasmín Martínez-Ocampo 592
 Andrade Valdez Paola Nataly 549
 Andrea Barajas Aguilar 742
 Andrea Bayona-Hernández 593
 Andrea Castillo-Campos 935
 Andrea Duarte Elías 417-423
 Andrea Gala Varona-Velázquez 769
 Andrea Margarita Rivas Castillo 448
 Andrea Martinez Marroquin 578-581
 Andrea Molina-Pineda 232
 Andrea Montserrat Sanchez Barajas 775
 Andrea Moyao 605
 Andrea Muñoz Ayala 752
 Andrea Navarro-Zazueta 232
 Andrea Ramón-García 489
 Andrea Regina de Souza Baptista 844
 Andrea Román Delgado 610
 Andrea Romero-Reyes 284

- Andrea Ruiz-Betancourt** 505
Andrea Sánchez-Ríos 1122
Andrea Terán Ramos 1068
Andrea Yamile Rivas Escobedo 766
Andrés Burgos-Palacios 296-310
Andrés Cota-Huizar 406-475
Andrés Eliu Castell Rodríguez 325
Andrés Hernández-Oliveras 584
Andrés Méndez-Zamora 399-1111
Andrés Moya 878
Andrés Opazo-Capurro 153
Andrés Paredes-Rocha 970
Andrés Reyes Chaparro 279-305
Andrés Rivera Chávez 533-764
Andrés Rodríguez Rodríguez 733
Andrés Salas Casas 798
Andrés Silva 967
Andrés Usiel Rodríguez Rodríguez 240-817
Andrés Zárate-Romero 250-517
Andrey Salazar-Mendoza 511-540
Andrómeda Liñán Rico 909-981
Anette Ramírez-Carrera 1204
Angela Daniela Saldaña Alvarado 773-1147
Ángel Alfonso Zaráin Herzberg 1070
Angela María Chapa Oliver 455
Angela María Jaramillo-Granada 147
Angel Andrade 205-926
Angela Patricia Moreno Londoño 244
Angela Raquel García Martínez 1205
Angela Regina Castillo Jimenez 271
Ángela Rodríguez Padrón 716-723
Angel Díaz Duarte 1178
Angeles C. Tecalco-Cruz 613
Ángeles Fortis-Barrera 1004
Ángeles García-Vicente 786-1032
Ángeles Rangel-Serrano 655
Angeles Rivera Román 495
Ángeles Tecalco Cruz 1049-1067
Ángel Fernández-Mora 220
Ángel Flores-Alcántar 676
Ángel Gabriel Alpuche Solís 1089
Angel Giovanni Arietta García 1131
Angel Gustavo Romo Mireles 770
Angélica Coyoy-Salgado 937-940-950-951
Angélica Eulalia Pérez Cid 1164
Angélica Feregrino-Pérez 574-588
Angélica Godínez-Oviedo 154
Angélica González Hernández 899
Angélica López-Izquierdo 511-540
Angélica López-Rodríguez 434-504
Angélica Nallehy Rodríguez-Ocampo 386-836-848
Angélica Núñez 1231
Angélica Pérez Juárez 1143
Angelica Piedra-Ramírez 941
Angélica Rueda 185-318-350
Angélica Samantha Santos Reyes 1015-1071
Angelica Santana Calderón 680
Angélica Saraí Jiménez Osorio 718-735
Angélica Silva-Olivares 677
Angelica S. Martinez-Ramirez 1047
Angélica Soberano Nieto 638
Angelica Sofía González Garibay 332
Angélica Vallejo Giraldo 534
Angel Medina Reyes 877
Ángel Miliar García 554-563
Ángel Pulido Capiz 289
Ángel Radamés Rábago-Monzón 1082
Angel R. Díaz Duarte 414-420
Ángel Salmerón Hernández 633-651-768-1064
Ángel T. Tello-López 654
Ángel Zaráin Herzberg 584-651-768-1064
Anibal H. de la Vega Pérez 222
Anna Bella Handal Silva 953
Annete Itzel Apodaca-Medina 550
Annie Pardo 261-703-705-765-775-1032
Antonia Encina 153
Antonino Báez Rogelio 416-422-471
Antonio Andrade-Meza 656
Antonio Ávila Guerrero 554-563
Antonio Basurto-Muñoz 1187
Antonio Celestino Montes 1244
Antonio De León-Rodríguez 543

- Antonio García Esteva 275
 Antonio García-Esteva 272-326
 Antonio Laville-Conde 969
 Antonio Peña 229-263-358
 Antonio Pérez Carillo 900
 Antonio Ramses Vargas Lara 868
 Antonio Romo Mancillas 1250
 Antonio Ruperto Jiménez Aparicio 469
 Antonio Topete 1231
 Antonio Zorzano 186
 Anuar Salazar Gómez 1171
 Anya A Valdez-Hernandez 788
 Araceli Martínez-Larios 873-894
 Araceli Montoya Estrada 579
 Araceli Posadas Mondragón 720-1143
 Araceli Rojas-Bernabé 1191
 Araceli Rosales-Cruz 820
 Araceli Ruiz Silvestre 594
 Arantxa Melissa Aguilar López 261
 Aranza C. Martínez-Zacarias 1074
 Aranzazú López-Jaime 367-995
 Areli Duron Castellanos 899
 Arely Morales-Vaca 673-690
 Ariadna Zelanda Salazar 994
 Arijita Ghosh 139
 Arisbeth G. Almeida-Juárez 191-365
 Arlene Calderón 473
 Arline Fernández-Silva 314
 Armando A. Ambrosio-Huerta 1076
 Armando Cruz-Rangel 204
 Armando García-Velasco 1031
 Armando Hernández-García 446-758
 Armando Hernández-Mendoza 378
 Armando Luna-López 182-553-961-962-1004
 Armando R. Tovar 713
 Armando Valenzuela Peraza 932-1134
 Armando Vilchis-Ordoñez 771
 Armendáriz-Toledano, F. 997-1007
 Arnaud Ronceret 621
 Arnulfo Albores Medina 757-759
 Arroyo-Sánchez Beatriz Irene 194
 Arturo Aguilar-Rojas 572-767-1181
 Arturo Cadena-Ramirez 391
 Arturo Edgar Zenteno Galindo 727-783
 Arturo Flores-Pliego 579-1191
 Arturo Galván Hernández 682
 Arturo Hernández-Cruz 964
 Arturo Jiménez Sánchez 280
 Arturo Orozco-Barocio 706
 Arturo Rojo-Domínguez 217
 Arturo Tlelo-Reyes 1185
 Arturo Valle-Mendiola 1030-1062
 A. Said Arellano-Buendía 797
 Asdrúbal Aguilera Méndez 753-818
 Astrid Karyme Polanco López 767
 Astrid N. Espino-Vázquez 863
 Atahualpa Guzmán de Casa 375
 Atl Emiliano Tarbuck Valle 364
 Audrey Bravo Prado 759
 Augusto Franco 1088
 Augusto José González Canto 673-690
 Aurelio Álvarez Vargas 921
 Aurora Espejel-Nuñez 1191
 Aurora Lara-Núñez 311
 Aurora Montes 967
 Aurora Osorio 178
 Axel Fernando Ramírez Rubio 1142
 Axel Reyes Coatl 901
 Ayari Fuentes 195
 Ayixon Sánchez Reyes 517-610
 Aylin del Moral Morales 245-582-625-626-637-1090
 Azhiel Contreras-Chávez 714
 Azucena Herrera-González 719
 Azul Castrillo Sanchun 821
 Azul Vázquez López de la Fuente 647
- B**
- Babatunde O. Adetuyi 227
 Barbara B.Moguel 1088
 Bárbara Cúmez Caté 180
 B. Areli Carranza Correa 280
 B. Cisneros 694

- B. Domínguez-Ortega** 1067
Beatriz Aguirre-Lopez 1055
Beatriz Alejandra Ávila-Castañeda 1017-1043
Beatriz E. Baca 555-814-819-900-920
Beatriz E. González-Contreras 608
Beatriz Eugenia Baca 825-826-893-898-918-1112
Beatriz Gómez-González 182-949-980
Beatriz Hernández-Estrada 1102
Beatriz King-Díaz 310
Beatriz Laug 1201
Beatriz Rufina González-Pérez 984
Beatriz Sosa-Pineda 1047
Beatriz Velazquez-Cruz 373-491-1093
Beatriz Xóchitl Ávila Curiel 727-783
Beatriz Xoconostle Cázares 203-366-453-532-1193
Beltrán-Peña E. 397
Benito Flores-Chávez 441-535
Benito Minjarez 757
Benjamín Bustamante Gómez 594
Benjamin Cartes Saavedra 139
Benny Weiss-Steider 1062
Berence Calderón Pérez 203-532
Berence Fernández-Rojas 338
Berence Franco, Susana Castro 1188
Berence García Ponce de León 265
Berence Mondragón Zúñiga 739
Berence Otero Díaz 1029
Berence Palacios González 728-754
Bernabe-Allende Alejandra 816
Bernardo Franco Bárcenas 655-1083
Bernhard Drotleff 163
Bernhard Hauer 1198
Bertha Campos-López 561-570-747
Bertha Espinoza Gutiérrez 682
Bertha Fenton-Navarro 960
Bertha González Pedrajo 577
Bertha Isabel Carvajal Gámez 628
Bertha León 993
Bertha Rueda Zarazúa 1119
Betsabe Guadalupe Najera Ruiz 674
Bexidui Castillo-Escoto 1191
Bianca Nieblas-León 220
Bianca Vianey Rivera-Serrano 1082
Bibiana Ortega Domínguez 1233
Birte Höcker 208-242
Blanca A. Delgado-Coello 320-541
Brandon Araujo-Mendoza 831
Brandon Jorge Romero Rodríguez 795
Brandon Ortiz Pérez 963
Braulio Trujillo-Santoyo 457
Bravo Madrigal Jorge 867
Brayan Alexis Jacinto-Arellano 612
Brenda Avelina López Muñiz 751
Brenda Chimal-Vega 289-752
Brenda Hernández 1036
Brenda Hernández Juárez 953
Brenda Hernández Pérez 955
Brenda Paola Gutiérrez Soto 731
Brenda Sofía Loaeza Cruz 862
Brenda Tellez de la Garza 915
Brenda Yarely Fernández Madrid 429
Brenda Yazmín Vargas Hernández 453
Brian I. Morales-López 945
Brigitte V. Vargas-Morales 1125
Brito-Elías Luis 630
Bruno Guillermo Hidalgo-Hernández 218
Bryan Adrián Priego Parra 723
Bryan Alexis Rivera-Suárez 322
Bryan Hernández 477
Bryan Rivera 1059
Bulmaro Cisneros 638-643
Burgos Dueñas, O. 997-1007
Burgos-Palacios Andrés 198
Burgos Solorio, A. 997-1007
Bustillos-Cristales, M.R. 910
- C**
- Camila Barrio Santoyo** 447
Camilla Koolmeister 170
Camille Caldeira 306
Camilo Pérez-Salazar 224-484-828
Campos Ornelas José Miguel 1211

- Candelario Vázquez Cruz** 233-614-627-804-872-876-905-906-1095
- Carina Becerril Berrocal** 598-631-700-742
- Carina López Leyva** 739
- Carla Angulo-Rojo** 1082
- Carla Isabela Cuevas Bañuelos** 279
- Carla Isabel Tena Fuentes** 857
- Carla O. Contreras Ochoa** 1156
- Carlos Aguilar Salinas** 241
- Carlos Alberto Brizuela Rodríguez** 458
- Carlos Alberto Matias-Cervantes** 370
- Carlos Alberto Molina-Vera** 882
- Carlos Alejandro Rocha Martinez** 1003
- Carlos Alfonso Sánchez-López** 1120
- Carlos Alfredo Barrón-Gallardo** 590
- Carlos Alvarez** 205
- Carlos A. Matias Cervantes** 794-1118
- Carlos Anerillas** 152
- Carlos Ángel Guzmán** 939
- Carlos Antonio Alba-Fierro** 401-432
- Carlos Arámburo de la Hoz** 954
- Carlos A. Rocha Martínez** 361
- Carlos Bravo** 252
- Carlos Calderón-Vázquez** 506
- Carlos Campero-Basaldúa** 269-850-1115
- Carlos Chávez Castañeda** 253
- Carlos Cortés** 173
- Carlos C. Patiño-Morales** 572-618
- Carlos Cruz-Cárdenas** 1187
- Carlos Daniel Bautista Olivier** 197
- Carlos Daniel Cordero-Rivera** 920-1082
- Carlos De Jesus-Quiroz** 1026
- Carlos de la Rosa Ureña** 1186
- Carlos Emilio Galindo Corona** 722
- Carlos Enrique Orozco Barrios** 1160
- Carlos F. Arias** 1210-1247
- Carlos F. Méndez-Cruz** 1096
- Carlos Getzael Vazquez-Osorno** 441-535
- Carlos G. Pérez Plascencia** 288
- Carlos Guadalupe Pérez Plascencia** 1163
- Carlos Javier López-Victorio** 986
- Carlos Jesús Ventura-Martínez** 685
- Carlos Jhovani Pérez-Amado** 634
- Carlos J. López Victorio** 285-294
- Carlos Josué Solórzano Mata** 294-727-783
- Carlos Lazcano-Arce** 1187
- Carlos Manuel Contreras Romero** 573
- Carlos Mújica-Jiménez** 314-372
- Carlos Noe Farfan-Morales** 1082
- Carlos Orozco-Barrios** 937-940-950-951
- Carlos Palma Flores** 230
- Carlos Peña** 396
- Carlos Peña Malacara** 907
- Carlos Pérez-Plascencia** 206-246-454-573-609-1061
- Carlos Ramón Vázquez Quiñones** 862-881-914-923-927
- Carlos Ricardo González Ruiz** 181-580
- Carlos Romero-Díaz** 1118
- Carlos Ruiz-Suárez** 1140
- Carlos Saldaña** 345-574-586-588-647-831-882-915
- Carlos Sandoval Jaime** 1247
- Carlos Vázquez Calzada** 660-1244
- Carlos Vázquez Salinas** 810-854-862-881-914-923-927
- Carlos Wong Baeza** 527-659-666-678-679
- Carmen Aitana López Zequeira** 1017-1043
- Carmen Alejandra Luque-Camacho** 456
- Carmen Cano Canchola** 921
- Carmen Carolina Alvarado González** 1153-1166
- Carmen Clapp** 1128-1145
- Carmen I Rodríguez A.** 1168
- Carmen Julia Figueredo-Urbina** 858
- Carmen M. Gurrola Díaz** 778
- Carmen Quinto** 228
- Carmen Vargas Lagunas** 902
- Carolina Alvarado-González** 382
- Carolina Chetirquen-Silva** 712
- Carolina del Carmen Murúa López** 537
- Carolina González Torres** 578-581
- Carolina Morales-Ochoa** 748
- Carolina Ortiz Murillo** 890
- Carolina Ramírez-López** 465
- Carolina Ricardez-García** 260-324-347-352
- Carol Siseth Martínez Caballero** 364-1094

- Carol S. Martínez-Caballero** 1076
Cassandra A. Alvarado-Verdín 809
Catalina De la Rosa-Juárez 392-394
Catalina Flores-Maldonado 1021
Cecilia Beatriz Peña Valdivia 272-275
Cecilia Gonzales Calixto 674
Cecilia Guerrero-Siancas 719
Cecilia Mata-Cruz 1175
Celia Robles Murguía 466
César Abdiel Cárdenas Villanueva 395
Cesar A. Esquivel Chirino 698
César A. Godoy 512
César Alberto González Guzmán 845
César A. Mendoza Calles 966
Cesar Arturo Nava-Valdivia 877
César Bonavides-Martínez C. 1096
Cesar Cardona-Felix 1093
César Díaz Pérez 307
Cesaré Ovando-Vázquez 249-787-1113-1121
César Femat-Castañeda 719
César Francisco Pacheco-Tena 711
César Hernández-Rodríguez 805-822-849-873-894-913-914
Cesar Lam Chung 241
César L. Cuevas-Velázquez 1243
César López-Camarillo 192-609-632
César Luis CuevasVelázquez 1237
César Muñoz Bacasehua 286
César Pacheco-Tena 212-760
Cesar Sanchez 221
Cesar V. F. Batista 892
Cesia Daniela Pérez Aguilar 615
Chang-Yi Cui 152
Charbel Giuseppe Solano García 739
Charlotte Olivares Yañez 746-754
Charmina Aguirre Alvarado 262-587
Christian Cortés-Rojo 189-756-785-790-791
Christian Daniela Ortiz-Robles 656
Christian Daniel Meza-Juárez 1050
Christian D. Ortiz-Robles 1162
Christian González-Reyes 386-836-848
Christian Guerra-Araiza 940-950-951
Christian Silva-Platas 741-748-772
Christopher Navarro Martínez 1230
Christopher P. Baines 169
Christopher V. Wright 1047
Christyna Rosales-González 1140
Cinthia Berenice Romano Martín 636
Cinthia E. Núñez 815
Cinthia Itzel Landa Moreno 753-992-1001
Cinthia V. Soberanes Gutiérrez 519
Cinthy Alejandra Rocha Sánchez 1242
Cinthy Paola Rangel-Chávez 417-423-1100
Cintya Atonal Águila 627
Citalli Gamboa Esteves 963
Citlali A. Suárez-Rangel 972
Citlali Figueroa Guzmán 271
Citlally Condado-Huerta 713
Claire Vielle 534
Clara Espitia 1076
Clara Garcia-Chavez 646
Clara Luz Sampieri Ramírez 716
Claudia Albany Reséndiz Mora 659-666-678-679-687
Claudia Avitia Domínguez 714-781
Claudia Castillo 936
Claudia Cecilia Ramírez Ramírez 916
Claudia Cecilia Vega-García 234-538
Claudia Desíree Norzagaray-Valenzuela 1082
Claudia Espinosa-García 980
Claudia G. Benítez-Cardoza 313
Claudia González-Espinosa 1033-1039-1050
Claudia Guadalupe Benítez Cardoza 795
Claudia I. Avitia Domínguez 285-294
Claudia Isabel García Berumen 753
Claudia Isela Avitia Domínguez 731-793
Claudia Ivonne Flores Pucheta 264-381-472-478-507
Claudia J. Bautista 787-1113
Claudia Lydia Treviño Santa Cruz 1205-1207
Claudia Mancilla Simbro 825-898-963
Claudia Mondragón-Albarrán 820
Claudia Patricia Hernández Mosqueira 1023
Claudia Sánchez-Cárdenas 138

- Claudia Segal-Kischinevzky** 243-536-889-919
Claudia Treviño 1228
Claudio Adrián Ramos Cortés 977
Claus Krogh Madsen 461
Clemente Mosso González 668
Concepción Gutiérrez Ruiz 246
Connie Gallegos-Rivera 434
Consuelo Lobato-Calleros 451
Contreras-Paredes Adriana 1258
Coral Y. Jorge-Cruz 1040
Corina-Diana Ceapă 823
Crisalejandra Rivera-Pérez 494
Cristian Eduardo Vázquez-Anguiano 658
Cristian J. Arriero-Carrillo 185-318
Cristian Salcedo-Cuaran 512
Cristian Torres León 388-516-722
Cristina Castañeda Patlán 1068-1069
Cristina García-Viguera 950
Cristina Garcidueñas Piña 437-468
Cristina Mencias-Castillejo 951
C. Rosales-González 681
Cruz Eugenia Martínez Palacios 868
Cuauhtémoc Alvarado Sánchez 285
Cuauhtémoc Villarreal Treviño 668-1110
Cynthia Margarita Concepción Acosta 824
Cynthia Ordaz-Pichardo 1263
Cynthia Paola Rangel-Chávez 409-474-503-1092
Cynthia Sámano Salazar 626-637-1090
- D**
- Dagoberto Tapia-Ramírez** 969
Daisuke Kihara 144
D. Alejandro Fernández Velasco 354-374
Dalia Flores-Molina 683
Dalia Isabel Torres Aguirre 365
Dalia Molina Romero 922
Damián Ramírez-Robles 223
Damien Formay 808
Damien Maurin 201
Danaí Montalván-Sorrosa 541
Dana Montserrat Galarza Ávila 164
Dana Vázquez-de los Reyes 536-889
Danelia Ramírez Ramírez 1056-1063
Daniela Alejandra Alvarado Fernández 1013
Daniela Camacho-Manriquez 328-639
Daniela Delneri 603
Daniela Del Toro 248
Daniela Elysa Solorio-Escamilla 913
Daniel A. Estrada Bárcenas 660
Daniela Fernanda Sánchez-Sánchez 631
Daniela Granados Carrasco 1128-1145
Daniel Agustin Bravata Alcantara 684
Daniel A. Mendoza-Magaña 464
Daniela Pérez-López 227
Daniela Rodríguez-Benito 350
Daniela Rodríguez-Cruz 767-1181
Daniel A. Rodríguez-García 464
Daniel Cantú-Ruiz 707-709
Daniel Cortés-Avalos 838
Daniel Corzo-Toledo 938
Daniel Cuevas Ramos 241
Daniel David Pacheco Rodríguez 331
Daniel Eduardo Sandoval-Colin 712
Daniel Ernesto Noyola Cherpitel 1089
Daniel Favato 255
Daniel Francisco Ramos-Rosales 552-569-648
Daniel Guerrero-Morán 1033
Daniel Hurtado-González 350
Daniel Isaac Alcántara Jara 1130
Daniel Juárez Serrano 955-958-959-1000
Daniel López-Cifuentes 729
Daniel López Pastrana 866
Daniel Martínez Anaya 1109
Daniel Ortega Zambrano 147
Daniel Padilla-Chacón 272-326-330
Daniel Perea Ruiz 1194-1236
Daniel Pérez-Calixto 786
Daniel Pérez-Vega 485
Daniel R. Browne 601
Daniel Reyes-Haro 238

- Daniel Ríos Barrera 1029-1046-1066
 Daniel Segura González 179-196-396-605-907
 Daniel Talamás Lara 660-677
 Daniel Uribe-Ramírez 368
 Daniel Uriostegui Escoto 1025
 Danna Cruz Rico 410
 Darién Huerta-González 1148
 Darío Rafael Olicón-Hernández 450-834-839-855
 David A. Camarena-Pozos 392-405-498
 David Acosta Muro 259
 David Alejandro De la Rosa Pérez 1238
 David A. Lizcano 705-765
 David Andrés Benavides Fernández 699
 David Barrera 736-738-799
 David Calderón Guzmán 1134
 David Cortés-Sotres 826
 David Eduardo Pérez Calderas 1037
 David F. Aguilar-Ávila 929-945
 David F. Barrera-Gómez 265
 David Guillermo Pérez Ishiwara 604-749
 David Hernández 305
 David Mauricio Cañedo-Figueroa 1082
 David Olvera 330
 David Reyes Corona 1160
 David Romero Camarena 157
 David Sebastián Hernández Toledano 256
 David Tirado Torres 916-917
 David Toledo-Padilla 1050
 David Valle-García 646
 David Weaver 139
 Dayren Díaz Gómez 396
 del Arrenal Mena, I.P. 990-991-996-997-1002-1007
 Delia L. Rodríguez-Bustos 1243
 Delia Vanessa López Guererro 1255
 Delina G. Montes Sanchez 698
 Del Prado-Audelo, M.L. 1002
 Denis Castro-Bustos 425
 Denisse Clavijo Cornejo 595
 Denisse García Sánchez 989
 Denis Yarazetl Romero Maldonado 768
 Dennis Adrián Infante Rodríguez 1164
 Dennis Cabrera Munguía 400
 Deyamira Matuz Mares 267-1218
 Deyanira Judith De Anda Alejandrez 1089
 D.G. Ríos-Lopez 1067
 Diana A. Millán-Aldaco 964
 Diana Baeza 813
 Diana Barrera Oviedo 695
 Diana Emilia Martínez-Fernández 877
 Diana Escalante-Alcalde 976-978
 Diana Flores Percino 835
 Diana Gabriela Domínguez Lazcano 716-723
 Diana Guerra Ramírez 355
 Diana Itzel López-Monterrubio 451
 Diana Laura Cortés Figueroa 909
 Diana Laura Flores García 1200
 Diana Laura Sierra Ulín 636
 Diana L. Esparza-Reyes 1039
 Diana Moroni González 934
 Diana M. Torres Cifuentes 958
 Diana Pérez-Barrios 683
 Diana Reyes Martínez 739
 Diana Rubalcava-Gracia 170
 Diana Sarahi De la Merced-García 942
 Diana Solano-Arguello 639
 Diana Soriano Becerril 579
 Diana Villarreal-Huerta 243-919
 Diana X. Robledo-Cadena 1108
 Dianey Celeste Cruz Muñoz 408
 Diaz-Ramos Leticia Itzel 1196
 Dickens Terrazas D. 744
 Diego A. Del Castillo Copland 1259
 Diego A. Moreno 950
 Diego Armando Martínez Cruz 530
 Diego de Jesús Pantoja Gutiérrez 297-333
 Diego Delfín López 759
 Diego Eduardo Sánchez Jasso 629
 Diego Estrada Luna 718
 Diego Estrada-Luna 735
 Diego Flores-Bautista 174
 Diego García González 922
 Diego González-Halphen 142-290-1254

- Diego Jeovani Huerta-Flores 888
 Diego Ornelas 173
 Diego Pérez-Villanueva 1237
 Diego Reyna Colín 618
 Diego Ruiz Ruiz 241
 Diego Torres-Pech 1252
 Diego Valdez-Tapia 311
 Diego Velázquez Trejo 248
 Dimitrios Tsitsipatis 152
 Dinorah Nashely Martínez-Carrillo 657
 Divanery Rodríguez-Gómez 389-503-1120
 D. Martínez-Carrera 774
 D. Martínez-Pastor 1067
 Dolores Gutiérrez-Alanis 387
 Domancar Orona-Tamayo 394-1085
 Donají A. García-Ortiz 296
 Dony Karent Mayor-Villeda 769
 Dora María Velázquez Hernández 236
 Dulce Andrade- Pavón 840
 Dulce Andrea Montes Perez 1112
 Dulce Desireé Izar Marín 669
 Dulce Estefanía Nicolás Álvarez 279
 Dulce Hernández Sacramento 958
 Dulce Jazmín Alvarez Olvera 692
 Dulce Malvaez-Luis 671
 Dulce Palmerín-Carreño 831
 Dulce Yamileth Medina Valenzuela 877
 Durairaj Ragu Varman 238
- E**
- Ebandro Uscanga-Mortera 272-275
 E. Carrillo-Flores 397
 Echeverría-Martínez Olga M. 611
 Edaena Benitez-Rangel 372
 Edda Klipp 163
 Eddy Sánchez-León 384
 Edén Noé Cervantes Leal 865
 Eder Arredondo-Espinoza 213-1173
 Eder U. Arredondo Espinoza 488
 Edgar A. Celis-Hernández 1263
 Edgar Adrian Ramirez-Gonzalez 334
 Edgar Alejandro Turrubiartes Martínez 642
 Edgar A. Ramírez González 1115
 Edgar Balcázar López 426-459-610
 Edgar Bonales-Alatorre 1206
 Edgar Daniel Barreras Duran 697
 Edgardo Becerra Becerra 763-1250
 Edgardo Galán Vásquez 209-519
 Edgardo Madrid-Paulino 1104
 Edgar D. Páez-Pérez 313-608
 Edgar Iván Galarce Sosa 666-678-679-687
 Edgar José Rundquist Sánchez 527-666-678-679-687
 Edgar López-Pérez 1074
 Edgar Martínez-Fernández 379-425
 Edgar Morales Ríos 327-524-699
 Edgar Neri 278
 Edgar Octavio Bonales-Alatorre 981
 Edgar Quezada-Ruiz 654-677
 Edgar Ramírez 269-850
 Edgar Ricardo Vázquez-Martínez 236-591-1217
 Edgar Robles-Rodríguez 349
 Edgar Vázquez-Núñez 1253
 Edgar Yebrán Villegas Vázquez 762-777-1106-1142-1169-1176-1179
 Edgar Zenteno Galindo 285-294-338
 Edith Arenas-Ríos 1025
 Edith Catalina López Hernández 1013-1045
 Edith C. Rivera 1167
 Edith Sánchez-Jaramillo 979
 Edmar de Jesus Díaz-García 282
 Edmundo Lozoya Gloria 601
 Edoardo Yañez 173
 Edson I. Rubio-Hernández 936
 Edson Mosqueda-Martínez 324
 Eduardo Alexis Valencia Dorantes 519
 Eduardo Alvarado Ortiz 244
 Eduardo A. Robleto 998
 Eduardo Cardona-Guerrero 270
 Eduardo Carrillo Tapia 692-1077
 Eduardo Castañeda-Saucedo 1149

- Eduardo Chaparro-Barrera 711
 Eduardo García Huerta 689
 Eduardo Hernández Vázquez 364
 Eduardo Jahir Gutiérrez Alcántara 916
 Eduardo López-Urrutia 454
 Eduardo Lorenzo Pérez-Campos 370
 Eduardo L. Pérez-Campos 1118
 Eduardo Mandujano García 911
 Eduardo Minto-González 820
 Eduardo Monjaraz Guzmán 184-1014-1016-1037-1048-1051
 Eduardo Mundo 1229
 Eduardo Padilla Camberos 332-533
 Eduardo Palomino Reyna 890
 Eduardo Perez Arteaga 1163
 Eduardo Pérez Campos 794
 Eduardo Pérez Salazar 720
 Eduardo Sánchez-Cisneros 1066
 Eduardo Trillo-Hernández 412-418
 Eduardo Vázquez-Garza 748-1044
 E. Estrada-Muñiz 681
 Ehekatzin García-Valdés 710-725-1141-1154
 Eiby Grisell Benitez Ibarra 701
 Eiko Osawa-Martínez 757
 Ekta Pathak 951
 Elahe Mirzaeichesmeh 617
 Eleazar Aguirre-Mandujano 451
 Eleazar Martínez-Barajas 176-326
 Eleazar Ramírez-Hernández 338
 Elena Álvarez-Buylla 173
 Elena Aréchaga Ocampo 572-618-767-1181
 Elena Aréchaga-Ocampo 797
 Elena Berezhnaya 139
 Elena Lizbeth Garcia-Villegas 298
 Eliakym Arámbula-Meraz 452
 Eliana Valencia-Lozano 1088
 Elianeth Amaro 383
 Elías Isidro Magallán 659
 Elías Nieto Zaragoza 223-268-988
 Elias Uriel Avila-García 1018
 Elidé Avilés Berzunza 402
 Eliel Ruiz-May 266-323-771-1241
 Elihú Bautista 1131
 Elisa Analí Camacho-Ureta 550
 Elisa I. Azuara Liceaga 597
 Elisa Irene Azuara Liceaga 225
 Elisa M. Valenzuela Soto 286
 Eliseo Cristiani-Urbina 368
 Elizabeth Estrada-Muñiz 227-1140-1146
 Elizabeth Calderón-Cortés 785-790
 Elizabeth Carreón Torres 718-745
 Elizabeth Cisneros-Lozano 404
 Elizabeth Ernestina Godoy-Lozano 378
 Elizabeth Estevez 939
 Elizabeth García Gómez 236
 Elizabeth Irasema Antuna-Salcido 559
 Elizabeth J. Castañeda-Ortiz 225
 Elizabeth Ochoa-Muñoz 213-1173
 Elizabeth Quintana-Rodríguez 394-498
 Elizabeth Sánchez Duarte 756-792
 Elizabeth Selene Gómez Acata 222-1097
 Elizarrarás-Rodríguez 485
 Elliot Flores-Oliva 976
 Elodia Nataly Díaz-De la Cruz 987
 Eloy-Andrés Pérez Yépez 573-609-1061
 Eloy Rodríguez-de-Leon 1005
 Elsa Irma Quiñones Ramírez 810-854-862-881-914-923-927
 Elsa Maribel Aguilar-Medina 407-1110
 E. Luis 1148
 Elva Adán-Castro 1128-1145
 Elva Jiménez Hernandez 578
 Elvira Galarraga-Palacio 969
 Emanuel Bojórquez-Quintal 1187-1192
 Emanuel Hernández Núñez 488
 Emanuel Osmar Flores Camargo 287
 Emiliano Aparicio Trejo 700
 Emiliano Bueno González 445
 Emiliano Osorio-Osorio 441
 Emilio E. Muñiz Reynoso 954
 Emilio Espinoza 736-738-799
 Emilio Espinoza-Simon 223

- Emilio García-Torres Caffarel 568
 Emilio Romero Rangel 717
 Emily Jocelyn Gómez Flores 579
 Emily L. Castillo-García 950
 Emma Arévalo-Salina 308
 Emma Berta Gutiérrez Cirlos Madrid 342
 Emmanuel Severino Quintana 716-723
 Emma Saavedra 1108
 Emma S. Calderón-Aranda 656-1162
 Enah Montserrat Fonseca Ibarra 135
 Enikar Patlán 1158
 Enoch Luis 1137-1158
 Enoc Mariano Cortés Malagón 674
 Enoc M. Cortés-Malagón 1021
 Enrico Alejandro Ruiz Castillo 985
 Enrique Castaño 593
 Enrique Chávez Jiménez 742-1233
 Enrique García Hernández 204-211
 Enrique González Pérez 455
 Enrique Ibarra-Laclette 864-1086-1087
 Enrique I. Oliver 138
 Enrique Maldonado Cervantes 851
 Enrique Mendez-Bolaina 230
 Enrique Merino-Pérez 596
 Enrique O. Hernández-González 1040
 Enrique Othón Hernández González 1056-1060-1063
 Enrique Querejeta 939
 Enrique Ramírez-Chávez 249
 Enrique Reyes Muñoz 579
 Enrique Rudiño Piñera 191-216-221-298-365-370-845-1190-1242
 Enríquez Medrano Francisco Javier 499
 E. Perez-Rueda 1081
 E. Ramos 345
 Erasmo Negrete Abascal 233-614-804-872-876-884-895-905-906
 Erendira Perez-Hernandez 320
 Eria Alaide Rebollar Caudillo 824
 Erica Jasmín Sánchez Hernández 226
 Erica Karime Ventura García 375
 Erica K. Ventura García 361
 Eric Alejandro García López 761
 Eric Alejandro Sánchez Rosas 515
 Erica L. Hagman Aguilar 519
 Ericel Hernández García 837
 Ericka Moreno-Méndez 603
 Erick Andrés Muciño Olmos 1233
 Erick Antonio Rojas-Pacheco 941
 Erick de la Cruz Hernández 779
 Erick Hernández-Hipólito 303
 Erick Israel Pérez-García 676
 Erick López 736
 Erick López-Cruz 738-799
 Erick Nereu Chávez Fonseca 848
 Erick R. Corzo-Toledo 938
 Erick Rojas-Pacheco 976
 Erick Sierra Campos 309-312-361-375-1003
 Eric Nahum Jiménez-Vázquez 979
 Eric Suriel Cruz Ruiz 1138
 Erika Bueno González Bosc 1160
 Erika Castaño-Moreno 707-709
 Erika Chavira-Suárez 215-712
 Erika Cruz-Bonilla 1088
 Erika Flores Loyola 388
 Erika Gomez-Chang 303-726-1129
 Erika Martínez-López 706-778
 Erika Melchy-Pérez 676
 Erik Andrade-Jorge 693
 Erika Rodríguez-Martínez 1133
 Erika Rubí Luis García 631-700-742
 Erik R Farquhar 376
 Erin L. Seifert 139
 Ernestina Hernández García 1134
 Ernesto Enríquez-Enríquez 487
 Ernesto Oyervides Muñoz 476
 Ernesto Prado Montes de Oca 533
 Ernesto Soto-Reyes 245-553-582-625-626-637-1090
 Ernesto Tapia-Campos 813
 Esaú Bojórquez-Velázquez 266-771-1241
 Escalante-Alcalde, Diana 947
 Esdras Enoc Pacindo Cabrales 478
 Esly Alejandra Euceda-Padilla 264

- Esmeralda Mónica Peña González 821
 Espinoza-Simón Emilio 611
 Esteban Acosta-Ramos 1011
 Estefania Odemaris Juárez Hernández 424
 Estefanía Ramírez Meraz 1008
 Estefania Valle Velasquez 732-784
 Estefani Gaytan-Nuñez 1208
 Estefany Damaris Guerrero Molina 299-301
 Estela Ruiz-Baca 218-414-420-434-504-853
 Estrada Georgina 427-443
 Estrada Gutiérrez G. 644
 Estrada-Rodríguez C.A. 833
 Etzel Garrido-Espinoza 831
 Eucario León Rodríguez 761
 Eugenio Contreras-Castillo 667
 Eugenio Leyva-Figueroa 1150
 Eugenio Mancera 896-1187
 Eulogio Orozco 539
 Eunice Dalet Farfan 939
 Eunice López-Ahumada 670
 Eunice López-Muñoz 767-1181
 Eva Cristina Sosa-Sillas 521
 Eva Guadalupe Palacios-Serrato 613
 Eva Mariela Lunar-Mata 455
 Evangelina Beltrán-López 398-445-452-456-521
 Evarista Arellano García 513
 Eva Ruvalcaba-Limón 634
 Eva Sosa-Sillas 398
 Evelin Aylin Valenzuela-Villalobos 531
 Evelyn de Dios Vásquez 285
 Everardo Hernández-Plata 631
 Everardo López-Romero 207-602
 Exsal Albores-Mendez 892
 Eyra Judith Hernández-Hernández 202
- F**
- Fabián Fernández-Luqueño 1253
 Fabián Galindo Ramírez 1048
 Fabiola Claudio Piedras 640
 Fabiola Cortés-Mendoza 247
 Fabiola del Razo Olvera 241
 Fabiola Hernández Vázquez 971
 Fabiola Mallon Mercado 1154
 Fabiola Salgado 153
 Fabiola V. Borbolla-Jiménez 496
 Facundo Barraque 1053
 Fanny Flores-Gallegos 1096
 Farida Korobova 1047
 Fátima Berenice Ramírez-Montiel 655
 Fátima Clarita Cota Ruiz 1193
 Fátima Sánchez-Ramírez 514
 Fátima Tornero-Gutiérrez 655
 Fatima Yexalen Bermudez Urquiza 579
 Fausto Arellano Carvajal 628
 Fausto Roberto Méndez de la Cruz 1025
 Faviola Tavares-Carreón 205-926
 F. Castellanos López 1007
 Febe Elena Cazares Raga 660-938-1244
 Federico Ávila 551-1036
 Federico Ávila Moreno 1119
 Federico Castro-Muñozledo 350
 Felicitas Calderón Vega 916
 Felipe Alonso Barredo-Pool 363
 Felipe Cruz-García 568
 Felipe de Jesús Salazar-Ramírez 772
 Felipe de Jesús Vázquez Alba 1255
 Felipe Leon-Garcilazo 529
 Felipe Morales-León 153
 Felipe Salazar-Ramírez 220
 Felipe Vadillo-Ortega 215-617-712-746
 Felipe Villegas-Carlos 634
 Félix Matadamas-Martínez 1221
 Fernanda Cornejo-Granados 406-475
 Fernanda Lizeth Gutiérrez-Palomeque 1092-1100
 Fernanda Marlen Enríquez Sánchez 1139
 Fernanda Medina-Flores 949
 Fernanda Padrón-Rodríguez 202
 Fernanda Toscano-Marquez 705-765-786
 Fernando Anaya-Velázquez 655
 Fernando Becerra-Velez 933
 Fernando Calzada Bermejo 1131
 Fernando Cortes Camacho 702-732-797

- Fernando Daniel García López** 1179
Fernando Daniel Urbieto Villalobos 543
Fernando Díaz-Otero 870
Fernando Díaz-Partida 415-421
Fernando Emiliano Jiménez Mondragón 734
Fernando González-Zozaya 1187
Fernando Guzmán-Chávez 328-357
Fernando I. Ramírez Luna 963
Fernando Lara-Rojas 592
Fernando Larrea 1214
Fernando Montelongo-Martínez 177
Fernando Montes García 804
Fernando Ochoa-Cortes 981
Fernando Roger Esquivel Guadarrama 1255
Fernando Uriel Rojas Rojas 904-1171
Fernando Verduzco-Valdez 511-540
Fernando Zamudio 1157-1168
Fernando Zamudio Zuñiga 1161
Fidel De la Cruz Hernández 1244
Fidel de la Cruz Hernández-Hernández 660-938
Fidelia Romero 935
Fidel O. Ramírez-Amador 308
Fidel Ramírez Amador 509
Figueroa Gutierrez Ana Hilda 974
Filiberto Gutiérrez Gutiérrez 773-1147
Filiberto Sánchez 475
Filippo Bedani 254
Flavio F. Contreras-Torres 772-1136-1159
Flor de Fátima Rosas Cárdenas 430
Flores Rosetti 671
Flores Benavides-Flor 430
Flores-Oliva, Alejandro Elliot 947
Flores Pliego A. 644
Flores Tizapa Hazel Rayel 739
Flores Valdez Mario Alberto 867
Floria Lizárraga Sánchez 594
Flor Yohana Flores 533
Frances H. Arnold 141
Francisca Trujillo Jimenez 1134
Francisca Villanueva-Flores 148-1202-1215
Francisco A. Aguilar-Alonso 782
Francisco Antonio Reyes-Soria 1241
Francisco A. Reyes-Soria 266
Francisco Castañeda-Gómez 662
Francisco Correa 801
Francisco Cruz Sosa 505-522
Francisco Flores-Murrieta 713
Francisco Héctor Chamorro Ramírez 821
Francisco Javier Alarcón Aguilar 1131
Francisco Javier Avelar-González 501
Francisco Javier Enríquez Medrano 476
Francisco Javier Gaytán Cervantes 578-581
Francisco Javier Juárez Castañeda 309
Francisco Javier Lievanos-Ruiz 960
Francisco Javier Luján-Méndez 1204
Francisco Javier Ríos-Fránquez 401-432-1135-1177-1178
Francisco Javier Torres-López 925
Francisco Josué Carrillo-Ballesteros 877
Francisco J. Plou-Gasca 719
Francisco Limón Salvador 1209
Francisco Padilla-Garfias 229-263-358-850-889
Francisco Torres-Quiroz 223-268-988
Francisco Villarreal 230
Francisco Xavier Castellanos-Juárez 557-559-623-1184
Francisco Xavier Guerra Castillo 587
F. Recillas-Targa 1067
Fred Luque-Ortega 452
Fredrik Levander 170
Frida Azucena Valentín García 725-1141
Frida I Barcenas-Robles 788
Frida Paulina Muñiz-Ruvalcaba 968
Frida Rodríguez-Izquierdo 1061
Frida Santillán Valenzuela 668
Frida Sofía Doroteo Platas 416-422
Fritz D. Herrera Salcedo 963
Fuentes-Ramírez, L.E. 910
Fulgencio Alatorre-Cobos 547-1078-1087
Funes, Soledad 1227

G

- Gabriela Alemán-Escondrillas** 712
Gabriela Carmona Castro 442

- Gabriela Carolina Morales Sandoval 749-779
 Gabriela Contreras González 1025
 Gabriela Delgado 618
 Gabriela Figueroa González 762-777-1106-1142-1169-1176-1179
 Gabriela Galan-Ramírez 241
 Gabriela Guerrero-Palomo 1146
 Gabriela Hernández Silva 1214-1249
 Gabriel Alarcón-Carranza 1096
 Gabriel Alonso Pérez 1183-1223
 Gabriela López Herrera 692
 Gabriela Mellado Sánchez 699
 Gabriela Meneses Ruiz 527
 Gabriela Montero-Morán 313
 Gabriela Moralí 980
 Gabriela Olmedo-Álvarez 386
 Gabriela Pérez-Flores 1132-1152
 Gabriela R. Luna-Palencia 283
 Gabriela Rodríguez Rodríguez 633-651-768-1064-1070
 Gabriel Betanzos Cabrera 718
 Gabriel Corkidi 174
 Gabriel Del Río Guerra 312-458-1091-1101-1230
 Gabriel Emilio Herrera-Oropeza 151
 Gabriel Guillermo Luna Arvizu 580
 Gabriel Isaías Torres Vernet 300-339
 Gabriel Marcelino Pérez 458
 Gabriel Muciño 703
 Gabriel N. Aviña Martínez 436
 Gabriel Sánchez Cruz 837
 Gabriel Trejo Sánchez 527-659-678-679-687
 Gabriel Vazquez Arredondo 792
 G. Aleph Prieto 972
 Galindo-Albarrán, A. 150
 Gamaliel Ortiz Sarabia 292-721
 Gamboa Guerrero Leslie Karina 1216
 García Alcludia Vanessa 294
 García-Bautista Gabriel Alejandro 1196
 García Castillo Verónica 619-630
 García-de los Santos, A. 910
 García-Merino, Gabriela 1012
 García-Ramírez B 508
 García Rivas G. de J. 319-356-361
 García-Sáinz, J. Adolfo 1012
 Gareth Omar Rostro Alonso 1179
 Garza-Velasco Raúl 675-688-1084-1196
 G. Carrión Hernández 708
 Genaro J. Rosales-Muñoz 1203
 Genaro Patiño-López 977
 Genaro Vázquez-Victorio 786-1067
 Georgina Gurrola-Briones 1157-1168
 Georgina Hernández 195-847
 Georgina Hernández Chavez 534
 Georgina Hernández Delgado 137-1186-1240-1246
 Georgina Quintana-González 322
 Georgina Victoria Acosta 650
 Geovanni Romero Quintana 403-407
 Gerardo A. Anguiano Vega 414-420
 Gerardo Alfonso Anguiano-Vega 401-432-1135-1177-1178
 Gerardo Antonio Ramírez Paz y Puente 233
 Gerardo Barragán Mejía 1134
 Gerardo Contreras-Ruiz 277
 Gerardo Corzo 382-427
 Gerardo Corzo Burguete 1153
 Gerardo de Jesús García Rivas 962
 Gerardo Díaz-Godínez 425
 Gerardo García Rivas 741
 Gerardo García-Rivas 188-220-748-772-1044-1159
 Gerardo Gonzalez-Rocha 153
 Gerardo Javier Chaidez-Gurrola 401
 Gerardo Mejia 806-878
 Gerardo Orta 138
 Gerardo Pável Espino Solís 382-1153-1166-1226
 Gerardo Ramirez-Mejia 245
 Gihovani Ademir Samano Barbosa 1025
 Gilberto Juárez Munguía 897
 Gilberto Velázquez 539
 Gilberto Velázquez Juárez 435
 Giovanna Barrera Aveleida 527-659
 Giovanna Berenice Barrera Aveleida 666-678-679-687
 Giovanny Aguilera Durán 1102-1250
 Girian Jaguer-García 369
 Gisela Altés 152

- Gisela Ceballos Cancino** 594
Giulia Cosentino 774
Giuliana Zanella Braña 875
Gladis Anelsi Fernández Lozano 1178
Gladys E. Jiménez-Nopala 137-1186-1240-1246
Gladys Romero-Corona 500-658
Gloria Altamirano-Cruz 808
Gloria Angélica González Hernández 390-841-885
Gloria Beatriz Molina Valdés 881
Gloria Fernández-Tilapa 657
Gloria Hernández-Alcántara 302-343-371
Gloria Martina Yepiz Plascencia 622
Gloria Saab-Rincón 308-431-480-509-1190
Gloria Soberón-Chávez 177-428-883
Gloria Soldevila 1050
Gloria Stephanie Villa-Jaimes 541
Gloria Vargas-Romero 369
Gloria Yepiz-Plascencia 200
G. Martinez 1081
Gochar-Mora A. 644
Gokabeth Zarco Hernández 921
Goldie Oza 764
Gómez-Velasco H 508
Gonzaga-Sánchez José G. 702-797
González Ortiz Rebeca 1098
González Ramírez D. F. 874-880
González Torres Luis Fernando 739
González-Zacarías Jacqueline Carolina 675
Gonzalo Martínez de la Escalera 1128-1145
Grace X. Terrazas Quiñones 361
Graciela Castro-Escarpulli 822-873-894
Graciela Jesed Reyes-Trujillo 1116-1122
Graciela Villeda Gabriel 884
Graciela Zambrano Galván 704
Grecia Díaz 654-677
Grecia Elena Hurtado-Nuñez 987
Grecia Hernández-Hernández 181-583-649
Gregorio Godoy Hernández 402
Gretel Mendoza Almanza 594
Gricelda Sanchez Sanchez 632
Griselda Rodríguez-Martínez 769
G. Rosas 433
G. Tapia-Urzúa 1067
Guadalupe Abigail Flores Pérez 430
Guadalupe A. Chávez-Melendrez 273
Guadalupe Ayala Rodríguez 1080
Guadalupe Domínguez-Gómez 1197
Guadalupe Elizabeth Jiménez-Gutiérrez 262
Guadalupe Espín 196-396-605-829-907
Guadalupe García-Alcocer 763-1005-1204-1250
Guadalupe García Arenas 375
Guadalupe González Ochoa 686-701-737-1248
Guadalupe Guerra Sánchez 353-834-855
Guadalupe Gutiérrez-Escobedo 567-580-583-596-812
Guadalupe Hernández-Martínez 584
Guadalupe Montserrat Leyva Olvera 739
Guadalupe Morquecho Ramos 604
Guadalupe Reyes Cruz 1072
Guadalupe Sanchez Flores 1247
Guadalupe Vázquez Rodríguez 917
Guerra-Rosas M.I. 833
Guevara-Flores, A. 990-991-996-997-1002-1007
Guevara Salazar Juan Alberto 1138
Guillaume Bouvignies 201
Guillén-Chablé Francisco A. 427
Guillermo Ceballos 230-788
Guillermo Elizondo 197-560-1020
Guillermo Gosset Lagarda 534
Guillermo Moreno-Gándara 504
Guillermo M. Zúñiga-González 706-778
Guillermo Salcedo-Barrientos 371
Guillermo Torre-Amione 220-748-772
Gustavo Acosta Altamirano 558-589-720
Gustavo Alexis Niño Vega 1083
Gustavo A. Valero-Pibara 314
Gustavo G. Medina-Mendoza 283
Gustavo Guevara-Balcazar 788
Gustavo J. Fabian- Rodríguez 929-945
Gustavo Pedraza-Alva 360
Gustavo Rodríguez-Alonso 1235
Gustavo Sánchez-Chávez 796
Gutierrez Hoya A. 1035

- Gutiérrez-Reyna, D.Y. 150
 György Csordas 139
 György Hajnóczky 139
- H**
- H. Angélica Martínez Becerril 964
 H. Aramis López-Meléndez 1222
 Harumi Shimada-Beltrán 904
 Haydeé Rosas Vargas 578-925
 Héctor Aaron Vázquez-Vázquez 969
 Héctor Abdel Vázquez Lechuga 793
 Héctor Daniel Hermenegildo Rosas 442
 Héctor E. López-Valdés 929-945-946-954
 Héctor Frayde-Gómez 289-322-755
 Héctor Gerardo Frías Ureña 466
 Héctor Guillén-Alonso 224-249
 Héctor Hernández Escoto 390
 Héctor Iván Saldívar Cerón 977-1015-1041-1065-1071
 Héctor Manuel Acuña Ramírez 865
 Héctor Manuel Mora Montes 344-844-1083
 Héctor Quezada Pablo 226-684
 Héctor Ruiz-Ortega 506
 Héctor Samuel López-Moreno 398-445-452-456-521-537
 Héctor Serrano 1197
 Héctor Vázquez-Meza 368
 Héctor V. Miranda-Astudillo 172-341-348-369-374-380
 Heidi Paola Galván Antonio 837
 Heidy Martínez Pacheco 966
 Heladia Salgado 1096
 Helena Cobos Justo 627
 Helen Yarimet Lorenzo-Anota 1044
 Helia Bello-Toledo 153
 Herbey Oswaldo Padilla Quirarte 1255
 Heriberto A. Valencia González 1260-1263
 Herlinda Clement 382
 Hermelinda Salgado Ceballos 937-940
 Hernández-Acevedo, G.N. 150
 Hernández-Alba Ana L. 702-797
 Hernández-Díaz Raquel Fabiola 443
 Hernández-Espinosa, David A. 1012
 Hernandez Poblano Joan Aldae 957
 Hernández Vidales Karen 802
 Herrera-Garduño, C.P. 996
 Hervé Degand 369
 Hilda C. Leyva-Sánchez 1104
 Hilda Mabel Sosa Esquivel 520
 Hilda Martina Gracia Valenzuela 865
 Hilda Martínez-Coria 929-945-946-954
 Hilda Mercado Uribe 147
 Hilda Montero L. de Guevara 716
 Hilda Ramos-Aboites 1187
 Hipólito Castillo Ureta 1110
 Hiram Hernández López 299
 Hiram Lozano-Ruiz 318
 Honza Ameli Cazarin-Mendoza 270
 Horacio Cano-Camacho 207-602
 Horacio Osorio Alonso 732-784
 Hueman Jaimes Díaz 1117
 Huerta-Venegas Pedro Iván 1038
 Hugo Alejandro Bojórquez Armenta 770
 Hugo Alejandro Méndez Hernández 1078-1125
 Hugo Antonio Hernández Pérez 856
 Hugo Castelán Sánchez 440
 Hugo G. Castelán Sánchez 1080-1095
 Hugo Hernández Fragoso 952
 Hugo López-Muñoz 1222
 Hugo Nájera 217-352
 Hugo Saavedra-Bonilla 238
 Hugo Sánchez-Hernández 938
 Hugo Serrano-Posada 373-491-1093
 Hugo Tovar 1221
 Hugo Villamil-Ramírez 329
 Hui-Ying Tsai 1053
 Huizar L. Díaz-Ceniceros 349
 Huizar Reyes José Manuel 1216
 Humberto Flores-Soto 431
 Humberto García Ortiz 1119
 Humberto González Márquez 1063
 Humberto Gutiérrez 215-248-635-1103-1119
 Humberto Herrera-Ubaldo 417-423-474-568-1092-1100
 Humberto Lanz-Mendoza 654-677
 Humberto L. Mendoza Figueroa 849

- Humberto Peralta 902
Humberto R. Medina 333
- I**
- Ian García Aguirre 643
Ian Ilizaliturri Flores 617
Iasha Jocelyn Ferreyra-Luna 925
Ibáñez-Hernández Miguel Ángel Antonio 1084
Ibarra-Estrada, I.E. 991
Ibrahim Ramirez-Soto 930
Ignacio Camacho-Arroyo 236-973
Ignacio López-González 1234
Ignacio Martínez Martínez 682
Ignacio Pacheco-Bernal 782
Ignacio Patricio Cáceres Salazar 466
Ignacio Rosas-Román 249
Igor Garcia-Atutxa 1202
Ilane Hernández-Morales 670
Ildelfonso Roberto de la Peña López 761
Ileana Castillo Tobias 400
Iliana Barrera-Martínez 393-399
Iliana Geraldine Barreto-Flores 389
Iliana Herrera 631-703
Ilse Araceli Careaga Rojas 447
Ilse Berenice Espinoza Hernández 830
Imelda Juárez-Avelar 656
Imelda Martínez-Ramírez 1019
Imperio Sánchez-Pérez 241
Ingrid Cuevas-Cárdenas 569-648
Ingrid Melissa Gómez-Vázquez 904
Ingrid Simón Lara 716-723
Ingrid Yaritzit Carreon 661
Irais Poblete Naredo 757-759
Irán Alía Tejacal 379
Irasema Mendieta 763-1005
Irene Castaño-Navarro 156-181-567-571-580-583-596-649-811-852
Irene de la Concepción Perea Arango 469
Irene Lee Rivera 1013-1045-1070
Irene Perea Arango 442
Irina del Rosario López Gámez 759
- Iris Grisel Galván-Escobedo 326
Iris K. Madera-Salcedo 671-1039
Iris-Rocío Tirado-Lucas 239
Irlanda Camacho 383-473
Irma A. Jiménez-Ramírez 593
Irma Romero 303-726-1129
Irving Israel Vega-Juárez 981
Isaac Chairez 533-764
Isaac Virgilio Martínez Cárdenas 597
Isabel Arrieta-Cruz 929-940-945-946-954
Isabel Baeza Ramírez 527-659-666-678-679
Isabel M. López-Lara 178
Isabel Sinaí Meza Abarca 789
Isabel Soto-Cruz 1030-1062
Isabel Velázquez-López 374
Isaías Balderas-Rentería 213-1173
Isela Esther Juárez Rojop 325
Isela Hernández-Plata 956
Isela Montufar Robles 674
Isela Parra-Rojas 561-570-747
Ishar Arad Retana-Arellano 617
Isidro X. Perez-Añorve 572-618
Isis Del Mazo-Monsalvo 1108
Islas Sánchez Selene Rubí 802
Ismael Antonio García 794
Ismael Bustos Jaimes 281-1212
Ismael Gonzales Anaya 750
Ismael Jimenez 473
Ismael Luna-Reyes 320-541
Ismael Mancilla Herrera 579
Ismael Secundino Velazquez 868-911
Israel Cañas-Villamar 580
Israel Enrique Padilla Guerrero 390-841-885-899
Israel Muñoz 736
Israel Parra Ortega 771
Israel Ramirez-Sanchez 230-788
Itzel Abigail Loredo-Alonso 936
Itzel Adriana Cabildo Delgado 560
Itzel Astrid Aviña-Ávalos 1185
Itzell A. Gallardo-Ortiz 693
Itzel Lievano 293

- Itzel Páramo-Pérez** 655
Itzel Patricia Vásquez Martínez 794
Itzel Peralta Salguero 578-581
Itzel Plascencia-Ortiz 873-894
Itzel Rodríguez Guerra 924
Itzhel García-Torres 302
Iván Alejandro Banda-Flores 1132
Iván Antonio García Montalvo 370
Iván Arenas Sosa 1153-1166
Ivan Cesar Arteaga 953-958-959-1000
Ivan Estrada-Mota 546
Iván Galarce Sosa 659
Iván Ignacio-Mejía 943
Iván Martínez Duncker 844
Iván Meneses-Morales 434-504-531-562
Iván Natividad-Bonifacio 854-881-914-923-927
Iván Ortega Blake 682
Iván Oseguera-López 1228-1234
Iván Rodríguez-Cruz 176
Ivan Rubio-Gayosso 788
Iván Salgado 533
Iván Salgado Ramos 764
Ivan Salido-Guadarrama 774
Ivan Sánchez Hernández 332
Iván Uriel Valladares-Hernández 979
Iván Velasco-Velázquez 933-970-975
Ivette Andrea Martínez-Santiago 673-690
Ivette Mendoza-Salazar 485
Ivonne María Olivares-Corichi 986
Ivonne Medina 1228
Ivón Salazar-Martínez 603
Ixchel Adriana Loa Ramírez 1219
Ixsoyen F. Vázquez Sandoval 280
Izmary Delgado-Waldo 206
- J**
- Jackeline Lizzeta Arvizu-Gómez** 386-836-848
Jacqueline Ayza Ríos Garces 723
J. Adolfo García Sáinz 1018
Jaime Adrian Hurtado Solorzano 859
Jaime Bustos Martínez 866
Jaime Juan Huerta Delgado 917
Jaime Manuel Ibarra Hurtado 1248
Jaime Mas-Oliva 199-320-541
Jaime Mora 902
Jaime Morales Romero 723
Jaime Ortega López 264-381-478-479-507-508
Jaime Pérez-Villanueva 1221
Jaime Rosas Díaz 502
Jaime Sánchez-Meza 706-778
Jaime Santoyo-Salazar 227
Jaime Zamora Chimal 682
Jair A. Madera-Hurtado 1224
Jair Santiago Salazar 956
J. Alfredo Hernández García 849
J. A. Martínez Yerena 460
James González 243-367-536-889-919-995
Jan Baumbach 1090
Janet Flores Lujano 578
Janeth Lucero Hernández Gaytán 520
Janet Ledesma-García 764
Janet Palacios-Martínez 174-565-592-600-621-1114
Janett Ríos-González 607
J. Ángel Rubio-Miranda 938
Janintzitic López-Niño 969
Janis Aislinn García Flores 573
Janisse Michelle Ramírez García 1015-1071
Jannú R. Casanova Moreno 764
J. Antonio Ibarra 838
Jaqueline Gómez 1210
Jaqueline Ugalde Zavala 1214-1249
Jason Enrique Vazquez-Navarro 569-648
Jatziry Daniela Ocampo Ulloa 225
Javier A. Arcibar-Orozco 392
Javier Acuapan Henández 1077
Javier de Jesús Vasconcelos-Ulloa 322
Javier De Las Rivas 151
Javier Fernández Torres 564-595
Javier López Miranda 492
Javier Luis Solís Martínez 766
Javier Montalvo-Arredondo 228
Javier Placido Arrizon-Gaviño 719

- Javier Plasencia de la Parra 879-912-1243
 Javier Raya-González 387-1054-1057
 Javier Solís Martínez 750-770
 Jazmín García Machorro 1041
 J. Campos Guillen 586
 J. Carlos Fernández 589
 JC Rodríguez-Sierra 457
 J. David Villeda-González 796
 Jean Louis Charli 935
 Jeannette Barba León 867
 J. Eleazar Martínez-Barajas 277
 Jenaro Lemus De La Cruz 753-992-1001
 Jen-Hao Yang 152
 Jeniffer Espinoza-Hernández 1199
 Jennifer L. Martindale 152
 Jennifer Vianey Fernández Dodero 1095
 Jens Riedel 249
 Jeshua Johayra Garcia-Soberanes 724
 Jessamyn Reyna Crespo Sandoval 1143
 Jessica Aviléz-Aviléz 949
 Jessica Cholula-Calixto 566
 Jessica Denisse Valle-García 1253
 Jessica Díaz Salazar 371-376
 Jessica E. Rodríguez-Rodríguez 693
 Jessica F. Contreras-Mojica 405
 Jessica Hernández Pineda 579
 Jessica Jazmin Saucedo-Rivas 634
 Jessica K. Suastes-Rivas 889
 Jessica Ortega Pérez 756
 Jessica P. Campos-Blázquez 1021
 Jessica Pita Aquino 171
 Jessica Romero Reyes 236
 Jessica Rubí Morán Díaz 1138
 Jessica Ruiz Escobedo 907
 Jessica Ruvalcaba Galindo 909
 Jesus Adelmo Rosas Anaya 622
 Jesús Adolfo García Sainz 1072
 Jesús Agustín Badillo Corona 287-1251
 Jesús Agustín Loyola Alonso 868
 Jesús Alberto Olivares-Reyes 183-214-1011-1026-1034
 Jesús Alberto Segovia-Cruz 517
 Jesús Alberto Urbar-Ulloa 399
 Jesús Alejandro Claudio Rizo 400
 Jesús Alejandro Gabutti Thomas 761
 Jesús Alfredo Rosas Rodríguez 286-697-701-737-985
 Jesús Antonio Arroyo García 812
 Jesús Antonio Oria Hernández 300-339-731
 Jesús Antonio Pescador Álvarez 770
 Jesús Antonio Rauda-Ceja 211
 Jesús Azahel Piña López 1165
 Jesús Bernardino Velázquez-Fernandez 533
 Jesús Bernardo Páez Lerma 492-548
 Jesús Campos García 271-307-818-857-859-861-869
 Jesús Carlos Ruiz Suárez 147
 Jesús Daniel López Loera 865
 Jesús-Daniel Zambrano-Romero 667
 Jesús Eduardo Castañeda-Betancourt 562
 Jesús E. Gutiérrez-Hernandez 671
 Jesús Elías Isidro Magallán 666-678-679-687
 Jesús Emiliano Cantador-Campos 394
 Jesús Emiliano Covarrubias-Lobatón 1137
 Jesús Ernesto Ramírez Nieto 899
 Jesús Espinal-Enríquez 1221
 Jesús Fabián Cervantes Meneses 564
 Jesús García-Gómez 545
 Jesús García Pereyra 436
 Jesús Garza 1168
 Jesús Hernández Monge 733-1183-1223
 Jesús Iván Jiménez-Rivera 591-1217
 Jesús López 1201
 Jesús Medina 1059
 Jesús Montiel González 1186
 Jesús Morales-Jiménez 822
 Jesús Omar García-Jiménez 550
 Jesús Omar Medina Cruz 322
 Jesús Oria Hernández 793
 Jesús Ortiz-Saucedo 446
 Jesús Páez Lerma 429
 Jesús Pérez-Hernández 690
 Jesús Ramírez-Mercado 683
 Jesús Ramon Castillo Hernández 851
 Jesús Ricardo Pérez Velázquez 339

- Jesús Rojas 352
 Jesús Rubén Rodríguez Núñez 455-916
 Jesús Salvador López Bucio 175
 Jesús Salvador López-Bucio 864-1042
 Jesús Sánchez Sánchez 779
 Jesús Urbar Ulloa 470-1124
 Jesús Valdés Flores 225
 Jesús Zepeda-Cervantes 613
 J. Félix Gutiérrez-Corona 218-853
 J. Francisco Zambrano-Zaragoza 809
 J. Gustavo Vázquez 1059
 Jhonny Aguilar-Grajales 540
 Jimena Alejandra Méndez Castro 745
 Jimena Carolina Enríquez Fonseca 424
 Jimena Cid-Uribe 1157
 Jimena Cris Campos-Arce 1020
 Jimena Isaias Cid Uribe 1161
 Jimena Meneses-Plascencia 603
 Jimena Ortiz Laris 474
 Jimena Reyes-Nicolau 820
 Jiménez-Arteaga Vanessa 808
 Jinshui Fan 152
 J.L. Morán-López 1121
 J. Mareš 460
 J. Montesinos 694
 Joana Sánchez Medina 777
 Joan Sebastian Salas-Leiva 212
 Joaquín Cordero Martínez 262-1056-1060
 Joaquín López-Carrillo 316
 Joaquín Manzo-Merino 1019
 Joaquín Omar Chávez Santiago 1083
 Joaquín Zúñiga 1119
 Jocelyn C. Pérez-Lara 283
 Jocelyne del Rosario Calvo-Ramírez 783
 Jocelyne Martínez Martínez 300-339
 Joel Edmundo López-Meza 433-482-500-658-696-1069
 Joel Hurtado Patiño 574-588
 Joel Ireta 167
 Joel Ríos Alvarado 492
 Johan Olalde-Hernández 328
 John Carlos Botia Becerra 390
 Jonathan Alejandro Zúñiga-Domínguez 376
 Jonathan García Hernández 840
 Jonathan García-Román 716-1209
 Jonathan Gerardo Sánchez Peralta 1156
 Jonathan J. Magaña 496
 Jonathan Padilla-Gómez 178
 Jonathan Puente Rivera 1238
 Jonathan Ricardo Robles Reyes 680
 Jonathan Sánchez-Yepéz 956
 Jonathon E. Mohl 212
 Jorge Adrián Islas 1103
 Jorge Aguilar Meza 1260
 Jorge Aguirre Joya 516
 Jorge Alfonso Martín Trejo 578
 Jorge Alfredo Campoy-Ramírez 888
 Jorge Antonio García-Álvarez 961
 Jorge Armando López Gutiérrez 407-1110
 Jorge Arturo Cisneros Martínez 793
 Jorge Arturo Torres Juárez 1123
 Jorge Benavides 1044
 Jorge Brito Sánchez 776
 Jorge Chávez-Servín 882
 Jorge Cime-Castillo 654-677
 Jorge Cisneros Martínez 714-781
 Jorge Cornejo Garrido 554-1263
 Jorge Díaz Blancas 1052
 Jorge Eduardo Hernández-Espinosa 218-853
 Jorge Emiliano Salinas López 1245
 Jorge Enrique Palomino Carrillo 739
 Jorge E. Sánchez Rodríguez 304-1170-1224
 Jorge Gutiérrez-Cuevas 729
 Jorge Gutiérrez-Franco 809
 Jorge Humberto Limón Pacheco 197-560-1020
 Jorge Humberto Mundo Ariza 469
 Jorge Humberto Ramírez Prado 1083-1107
 Jorge I. Castañeda-Sánchez 797
 Jorge Iván Castillo-Arellano 946
 Jorge López-Gutiérrez 403
 Jorge Luis Álvarez Zapata 921
 Jorge Luis Araujo Sánchez 1078
 Jorge Luis Blé Castillo 325

- Jorge Luis Copado-Romero 782
 Jorge Luis Medrano-Cerano 211
 Jorge Manuel Almanza Torres 1176
 Jorge Manuel Vázquez Ramos 301
 Jorge Marcos Víquez 410
 Jorge Meléndez Zajgla 594-1052-1119
 Jorge Molina-Torres 249
 Jorge Noé García-Chávez 474-1113
 Jorge Pacheco Rosado 932
 Jorge Reyes-García 1022-1058
 Jorge Rodríguez 234
 Jorge Rojas-Vargas 378
 Jorge Romo-Tena 1027
 Jorge Verdín 234-384-470-538-813
 Josahandy Jiménez-Peralta 308-431
 Joscelyn Macías Ríos 794
 José A. Domínguez-Rosales 706-778
 José A. González Garrido 285-294
 José A. Hernández 1167
 José Alberto Narváez-Zapata 363
 José Alberto Parra Espinoza 480
 José Alberto Patiño Medina 789
 José Alberto Rivera Chávez 1219
 José Alberto Verduzco Palafox 686
 José A. Lorenzo Alcudia 285
 José A. Narváez-Zapata 493
 José Ángel Cueva Villanueva 837
 Jose Angel Regla-Nava 877
 Jose Ángel Rubio Miranda 660-1244
 José Angel Santiago Cruz 1143
 José Antonio Arias-Montaño 140-944
 José Antonio Guerrero Analco 323
 José Antonio Magaña Lizárraga 865
 José Antonio Martínez Villalba 875
 José Antonio Munive Hernández 897
 José Antonio Ocampo Cervantes 626-637-1090
 José Antonio Pedroza-García 328-357-639
 José Antonio Ramírez-Rafael 151
 José Antonio Velázquez Domínguez 795
 José Arcadio Farías-Rico 208
 José Armando Hernández Soto 743
 José Arnold González-Garrido 986
 José Ascención Martínez Álvarez 344-844
 José Augusto Ramírez Trujillo 442
 José Bargas Díaz 969-971-973-978-980
 José Bonilla Delgado 674-1021
 José Bruno Apodaca Fuentes 985
 Jose Carlos Campero-Basaldúa 270-334
 José Carlos González Arce 1143
 José Carlos Parada-Fabián 881-914-923-927
 José C. Crispín 671
 José Cisneros 261-631-786
 José Cisneros-Lira 742
 José Cristian Pérez Gómez 893
 José Cruz Jiménez Galindo 275
 José David Torres-Tirado 1132-1152
 José de Jesús Arellano García 442-469
 José de Jesús Esparza-Claudio 394
 José de Jesús García-Trejo 302
 José de Jesús Martínez González 235
 José de Jesús Olivares-Trejo 226-888
 José de Jesús Peralta Romero 739
 José Díaz-Chávez 1197
 José Dolores Solano-Becerra 782
 José Edwin Dolores-García 1197
 José Eleazar Barboza Corona 439-463
 José Elizalde-Contreras 1241
 José Erick Cruz Valderrama 1240
 José Esparza-López 1027
 José Everardo Avelino Cruz 1051
 Josefa Espitia-López 441-535
 Jose Fernando García-Rodrigo 1128-1145
 Josefina Atrisco-Morales 657-1149
 Josefina Dalia Ramírez-Ramírez 545
 Josefina Guzmán Aparicio 196-396-605-815-907
 Josefina Rodríguez González 388
 José Francisco Morales Domínguez 437-468
 José Francisco Muñoz Valle 686
 José Gadú Campos Saucedo 1238
 José G. Trujillo-Ferrara 693
 José Guadalupe de la Cruz Hernández 285
 José Guadalupe Rendón Maldonado 865

- José Guadalupe Soñanez Organis** 686-985-1248
José Guadalupe Torres Salazar 1261
José Guadalupe Trujillo Ferrara 1138
José Guillermo Gonzaga Sánchez 732-784
José Hugo Castorena-García 465
José Israel Torres Avendaño 1110
Jose J Olivares-Trejo 496
José Juan Pérez-Trujillo 213
Jose Juan Torres Martinez 1121
José Leopoldo Aguilar Faisal 1143
Joseline Suhail Alejo Guerra 175-807
José López-Bucio 175-387-556-807-860-864-1042-1054-1057
José López Hernández 175-864
José Luis de la Vega-Beltrán 138
José Luis González-Alfonso 719
José Luis Hernández-Flores 386
José Luis Herrera Celis 764
José Luis Morales Velázquez 716-723
José Luis Morán Perales 955-1000-1165
José Luis Ramos-Balderas 667
José Luis Rodríguez Mejía 378-909
José Luis Rodríguez Olivares 761
José Luis Velasco-Bolom 1136-1159
José Luis Ventura Gallegos 761
José Luis Vique-Sanchez 289
José Manuel Hurtado Capetillo 716-723
José Manuel Reyes-Ruiz 1082
José Manuel Salas Pacheco 292-552-557-569-575-623-648-721-766
José Manuel Sánchez López 594
José Manuel Vázquez Rodríguez 1256
José Martínez-Álvarez 655
Jose M. Cervantes 980
José M. Elizalde-Contreras 266-771
José Miguel Cuaxospa 979
José O. Saucedo-Lucero 392
José Pablo Romero-López 711
José Pedraza Chaverri 700
Joseph G. Dubrovsky 607-1235
José Rafael Irigoyen Campuzano 530
José Ramón Barraza Amaya 388
José Raúl Junco Carmona 449
Jose Raymundo Cruz-Perez 677
José Refugio Torres Nava 578
José Romo Yáñez 579
José R. Palacios-Valladares 1162
José Rundquist Sánchez 659
José Salas Pacheco 770
José Salud Rodríguez-Zavala 801
José Vázquez Prado 1072
José Velasco-Bolom 772
José Villalobos-Escobedo 506
Joshua Fernández Jasso 834
Joshua Imanol Cervantes Andraca 710-725-1154
Josephlyn Hernández Alcántara 626
Jossie Fernanda Espinoza Ávila 426-459
Jossimar Coronel Hernández 246-288
Jossue Ortiz-Álvarez 822
Josué Camberos-Barraza 1082
Josué Raymundo Solís Pacheco 610
Joyce Trujillo 787-1113
J. Sergio Casas-Flores 331
J. Trujillo-Barrientos 345
Juana Elizabeth Reyes Martínez 868-1144
Juan Alberto Ascacio Valdés 773
Juan Alfredo Hernández-García 913
Juan Antonio Laville Conde 971
Juan-Antonio Rodríguez de la Cruz 1117
Juan Antonio Rojas Contreras 395-429-492-548
Juan Antonio Sanpedro Luna 627
Juan A. Osuna-Castro 373
Juan Armendariz-Borunda 729
Juan Campos-Guillén 386-836-882
Juan Carlos Almagro 485-1175
Juan Carlos Baltazar Vera 917
Juan Carlos Cabrera Angeles 732-784
Juan Carlos De Lima Mar 827
Juan Carlos Estrada Mora 366-660
Juan Carlos Fernández Hernández 720
Juan Carlos Fragoso Jiménez 534
Juan Carlos Gallardo-Pérez 800
Juan Carlos García Acosta 1142

- Juan Carlos Gómora Martínez 193
 Juan Carlos Juárez-Cruz 657-1149
 Juan Carlos Martínez Morales 244
 Juan Carlos Muñoz Herrera 669
 Juan Carlos Núñez Enríquez 578
 Juan Carlos Torres Guzmán 390-841-885-899
 Juan C. Cabrera-Ángeles 702-797
 Juan Cristóbal García García 447
 Juan Cuauhtémoc Díaz Zagoya 325
 Juan de Dios 473
 Juan de Dios Ortega Alonso 519
 Juan Diego Siary Leyva 1248
 Juan E. Olivares 174
 Juan Esteban Bello Lara 917
 Juan Fernando Montes García 872-884
 Juan Fidel Osuna-Ramos 1082
 Juan Francisco Sánchez López 515-921
 Juan Gabriel Juárez Rojas 732-784
 Juan Guillermo Cruz Castillo 355
 Juan Jair Santillán-Cigales 975
 Juan Ludert 654-677
 Juan Luis Cota Quintero 407
 Juan Luis Monribot Villanueva 323
 Juan Luis Rendón 235
 Juan M. Agraz-Cibrian 809
 Juan Manuel Arias 359-979
 Juan Manuel Hernández-Martínez 979
 Juan Manuel Mejía Arangure 578
 Juan Manuel Méndez-Contreras 542-652
 Juan Manuel Palacios-Corona 1235
 Juan Manuel Ruíz-Ruíz 925
 Juan Manuel Sánchez Contreras 776
 Juan Manuel Sandoval Valverde 780
 Juan Manuel Serna Velázquez 527
 Juan Martín Hernández Castillo 1101
 Juan M. Gallardo 1021
 Juan M. Hernández Castillo 1091
 Juan Pablo Ochoa-Romo 475
 Juan Pablo Pardo-Vázquez 243-367-368-372-373-995-1218
 Juan Pablo Pintor-Ríos 487
 Juan Pablo Reyes Grajeda 199-204
 Juan Pablo Robles 1128-1145
 Juan Paulino Segoviano León 1261
 Juan P. Cabral-Miramontes 504
 Juan Pedro Luna Arias 733
 Juan Ramiro Pacheco-Aguilar 836
 Juan Santiago-García 584
 Juan Santiago Salas Benito 1079
 Juan S. Pardo-Tamayo 512
 Juan Unzueta Mendoza 643
 Juan Vázquez-Martínez 389-409-1120
 Juárez-Gonzalez Víctor Rivelino 816
 Juárez Rojas Juan G. 702
 Judith Bernal-Ramírez 188-748-772-962
 Judith Hernández-Aranda 1011-1026
 Judith Sánchez Rodríguez 1219
 Judith Santos-Ceseña 1005
 Judith Ulloa-Calzonzin 603
 Juliana Marisol Godínez-Rubí 706-778
 Juliana Rojo 829
 Julian L. Wissner 1198-1259
 Julián Ramírez-Bello 634-674
 Julie Martin 494
 Julieta Griselda Mendoza-Torreblanca 943
 Julieta López-Cuevas 1016
 Julieta López Vazquez 723
 Julieta Palomeque 741-1053
 Julieta Rosas-Vallejo 503
 Julio Adulfo Acosta 1219
 Julio Altamirano Barrera 741
 Julio Armando Massange Sánchez 210-408-461
 Julio C. Cordero Olivares 963
 Julio César Chávez Zamora 1205
 Julio César García Rodríguez 297
 Julio César Garza-Cabello 842-908
 Julio César Ontiveros-Rodríguez 791
 Julio Cesar Ríos Guzman 391
 Julio Cesar Sánchez-Lemus 487
 Julio César Villagómez-Castro 483
 Julio Collado-Vides 1096
 Julio C. Vega-Arreguín 904
 Julio García Cordero 283-699

Julio Guerrero Castro 909
 Julio Guerrero-Castro 981
 Julio Morán 940
 Juventino García-Vilchis 658

K

Kame Alberto Galán-Huerta 842-908
 Karem Sánchez Martínez 357-362
 Karen Abundiz-Yáñez 984-1010
 Karen Bahena-De Dios 247
 Karen Carreto Meneses 934
 Karen E. Álvarez Castillo 1151
 Karen Elizabeth Franco Reyes 721
 Karen Estefes-Carranza 505
 Karen Julissa Loaeza-Reyes 338
 Karen Larrauri 993
 Karen M. Dahlman Cabrera 280
 Karen Olivia Rojas 533
 Karen Onofre Rentería 476
 Karen Pelcastre Guzmán 1015-1065
 Karen Pesqueda-Cendejas 561-570-747
 Karen Sofia Hernández-Medina 304-1224
 Karen Stephania González Ponce 439
 Karen Virginia Pineda-Hidalgo 452
 Karina Anastacia Solís Labastida 578
 Karina García-Gutiérrez 450-839-855
 Karina López-Olvera 1199
 Karina Manzo Valencia 528
 Karina Martínez Flores 564-595
 Karina Robledo-Márquez 269
 Karina San Vicente Trujillo 1240-1246
 Karina Torres-Ortiz 241
 Karla Berenice Sánchez Huerta 932
 Karla Carvajal 748
 Karla Concepción López Pacheco 317
 Karla Daniela Hernández González 214
 Karla de Anda-Mora 926
 Karla Estephania Ávila-Galicia 553-961
 Karla Fernandez-González 586
 Karla Grisel Calderón González 733-817-1223
 Karla G. Serrano-Dávila 311
 Karla Liseth Macías-Sánchez 394
 Karla Lizbeth Macías Sánchez 424-448
 Karla María Morales-Zamora 673-690
 Karla Mayolo-Deloisa 1044
 Karla Miranda López-DeLucio 641
 Karla Morelia Díaz-Tapia 207-602
 Karla P. Guzmán-Barrenechea 671
 Karla Pineda-Medina 247
 Karla Ramírez Estrada 488-1173
 Karla Susana Vera Delgado 792
 Karla Vazquez Santillan 1052
 Kassandra-Selene Reyna-Gaeta 239
 Kate Ariadna Rossano Gutiérrez 906
 Katia Aviña-Padilla 151
 Katia Daniela Hernández Morfin 353
 Katya Carrasco-Urritia 683
 Katya Manzanita Quintero 1045
 Katya Rodríguez-Vázquez 209
 Katy Juárez 566
 Katzumy Lizbeth Blancas-Olvera 559
 Kayley Aileen Hernández Ramírez 444
 Keila M. Gómez-Castellanos 1175
 Kempis-Calanis, L.A. 150
 Kenia Franquez Gómez 836
 Kenya Lizbeth Mena-Garcés 598
 Keren Tovar-Presas 683
 Kevin Daniel Llanos-Vargas 386
 Kevin David Laguna Maldonado 1218
 Kevin E. Lugo García 311
 Kevin E. Villa-Vuelvas 1194
 Kevin Ledesma Muñiz 861
 Keyla Gómez-Castellano 485
 K. Helivier Solís-González 1018
 Kimberly Arisbeth Casal-Rodríguez 398-456-521
 Kimberly Flores-Rivera 431
 Kimberly Quezada Cardenas 704
 Kirán Rubí Jiménez-Vázquez 175-807-860-864
 Koré Montserrat Moreno Calderón 756
 Kotb Abdelmohsen 152
 Kristina Bubb 170
 Kristy Elizabeth Jiménez Meráz 909

Krystyna Mazan-Mamczarz 152

Kumari Rashmi 616

L

Labastida-Conde, R.G. 150

Laila Espinosa Rentería 664

Laila Pamela Partida-Martínez 224-249

La-Llave León Osmel 559

Lanz-Mendoza, H. 640

Lara-Ochoa, C. 910

Laura Álvarez 1105

Laura-Angélica Vera-Salazar 649

Laura A. Palomares 1202

Laura A. Velazquez-Villegas 713

Laura A. Vera-Salazar 583

Laura Barragán Ledezma 704

Laura Camacho-Jiménez 200

Laura Cano 1188

Laura Carmona-Salazar 265

Laura C. Berumen 763-1005

Laura C. Bonifaz Alfonzo 650

Laura C. Flores García 288

Laura C. Fuentes-Vázquez 291

Laura Cristina Berumen Segura 1204-1250

Laura Cristina García Carnero 344

Laura Denise López Barrera 1151

Laura E. García-Cortés 838

Laura Elizabeth Merino Pasaye 578

Laura Espinosa-Barrera 373-491-1093

Laura E. Zavala García 528

Laura Gabriela Sánchez Lozada 732-784

Laura G. Hernández Aragón 963

Laura Gómez-Romero 634

Laura González-González 184

Laura Guadalupe Stopani Quiñones 623

Laura Hernández-Andrade 870

Laura Isabel Vázquez-Carrillo 226-684

Laura Itzel Quintas Granados 692-762-777-1106-1142-1169-1176

Laura Jaramillo-Meza 870

Laura López-Bañuelos 1146

Laura Mejía Teniente 455

Laura N. Balarezo-Cisneros 603

Laura Palomares 383-473

Laura Pérez-Campos Mayoral 370-794

Laura Q. López-Hernández 671

Laura Rodas-Zuluaga 477

Laura Silvia González Valdez 832

Laura S. Rangel-Cova 771

Laura V. Sánchez Orozco 706-778

Laura Yamamoto-Flores 501

Laurence A. Marchat 609

Lauren L. Zhang 1170

Leandro Alberto Núñez Muñoz 203-453-532-1193

Leena Paul 1047

Lefan Yu 201

Leisy M. Santiago-Hernández 963

Leobardo Ibarra-Sánchez 504

Leobardo Reyes Velasco 837

Leonardo del Valle Mondragón 745

Leonardo Gallegos 293-346

Leonardo Josué Castro-Muñoz 1019

Leonardo Lozoya Lozoya 733-1223

Leonardo Munguía Aguirre 606

Leonardo Naredo-Rojas 886

Leonardo Peraza Reyes 846-1055

Leonel Armas 551

Leonel Valencia-Estudillo 1096

León Francisco Ruiz-Herrera 556-1057

Leonor Galera 230

Leonor Pérez Martínez 360-1104

Lérida Liss Flores-Villavicencio 483

Leslie Gamboa Guerrero 292-721

Leslie Karina Gamboa Guerrero 575

Leslie Mariana Apodaca Ayala 1184

Leslie Sánchez Pérez 777

Lesly Yohana Parrado Quintana 971

Leticia Bucio-Ortiz 1203

Leticia Casas-Godoy 399-412-415-418-421-510-511-1111-1124

Leticia Cedillo Barrón 283-699

Leticia Cortés Martínez 660-938

Leticia Cortez Martninez 1244

- Leticia León-Luna** 216
Leticia Olvera 480
Leticia Olvera-Rodríguez 308-431-509
Leticia Ramírez-Silva 302-343-371
Leticia Vega-Alvarado 1114
Leyva-Gómez, G. 1002
Libertad Adaya García 196-378
Libia Vega 227-1140-1146
Lidia F.E. Huerta-Nuñez 771
Lidia Huerta-Nuñez 892
Lidia Itzel Castro Rodríguez 1023
Ligia Vega-Becerra 1257
Lihui Song 169
Lilia Angélica Bernal-Gracida 277
Lilia González Cerón 668
Lilia Leyva-Carrillo 200
Lilia Lopez-Canovas 597
Lilia Martina Velez-Velez 623
Liliana Alejandra Hernández Espinosa 854
Liliana Argueta-Figueroa 338
Liliana Elizabeth García-Valencia 1185
Liliana Hortencia Méndez Barredo 323
Liliana López 1201
Liliana López Pliego 820-897-922-924
Liliana Lugo-Zárate 735
Liliana Mercado Díaz de León 468
Liliana Montes Aguilar 742
Liliana Pardo-López 191-298-378
Liliana Quintanar 166
Liliana Valdez Jiménez 1139
Lilian Gabriela Valencia-Turcotte 351-529
Lilian González-Segura 302-315
Lilian Karem Flores Mendoza 686
Lilia Noriega-López 774
Lina R. Riego-Ruiz 269-608
Linda Aimara Kempis-Calanis 685
Linley Pammely Prado Celis 730-780
Lino Javier Martínez-Soto 1116-1122
Lino Sánchez Segura 528-601
Lira Reyes A. I. 874-880
Lisethe Palomo Ligas 773-1147
Lisset Esther Martínez Magaña 998-1006
Lisset Torres Martinez 682
L. Itzel. Quintas Granados 1077
Litzzy Ayra Pardo 137-1186-1240
Lizano Marcela 1258
Lizano Soberón M 1019
Lizbeth Guadalupe Chávez-Ramos 403
Lizeth Abad Alvarez 533
Lizeth Anahí López Castro 701-737
Lizeth De Anda 205
Lizeth Enid López-Rodríguez 1172
Lizeth Guadalupe Campos Múzquiz 1147
Lizeth Sandoval 293-346-359
Lizue B. Mancilla-Infante 796
López-Bucio José 1038
López-Franco O. 708
López-Ramírez, V. 481
López-Urrutia Eduardo 619-630
López-Vancell Rosario 673
Loranda Calderon Zamora 1082
Lorena Amaya-Delgado 234-719
Lorena De la Cruz-Mendoza 989
Lorena Martínez Alcantar 271-818-859-861-869
Lorena Orozco 1119
Lorena Paredes 162
Lorena Rodríguez Páez 262
Lorenzo Rodriguez Luis Manuel 867
Lorenzo Segovia 336-1208
Lourdes Ballinas-Casarrubias 382
Lourdes Girard Cuesy 137-195-847-902-1240-1246
Lourdes Gutierrez Xicotencatl 892
Lourdes Martínez-Aguilar 178
Lourdes Millán Pérez Peña 1000
Lourdes Pérez Peña 993
Lourdes Rojas 804
Lourdes Siqueiros-Marquez 1128-1145
Lourdes Vadillo-Pérez 617
Lourdes Villa-Tanaca 805-822-840-849-873-894-913-914
Lourival Domingos Possani Postay 1157-1161-1168
Lozano García O. 356
Luc Dendooven 222-1097

- Lucero Romero-Aguilar** 243-367-372-919-995
Lucero Valladares Cisneros 1168-1174
Lucía Mendoza Viveros 161
Lucía Perezgasga 1215
Lucia Ramirez 230
Lucía Soto-Urzúa 886-1116-1122
Lucila Méndez Morán 466
Lucio Antonio Ramos Chávez 1165
Luévano Martínez, L.A. 319-356
Luigui Gallardo-Becerra 406-475
Luis Acosta 473
Luis Adolfo Cenicerros-Morales 1135-1177-1178
Luis Adrián De Jesús-González 1082
Luisa Durán-Pasten 1137
Luisa Escobar 736-738-799
Luis Alberto Ayala-Ruiz 362-696
Luis Alberto García Casillas 435
Luis Alberto Ibarra-Sánchez 831
Luis Alberto Sánchez Briones 792
Luis Alberto Zamora Álvarez 1248
Luisa López Ochoa 636
Luis A. Luévano-Martínez 220
Luis Amsteins-Romero 153
Luis Ángel León-Cholula 940
Luis Antonio Buendía Sánchez 1188
Luis Antonio Ortiz López 575
Luis Antonio Pérez-García 1132-1152
Luis A. Rivera Rivas 585
Luis Arturo Baiza-Gutman 340
Luis Arturo Torres Castañón 530
Luis A. Valencia-Flores 1199
Luis Briebe 328
Luis Brito-Elías 454
Luis Cárdenas 174-565-592-600-1114
Luis Caspeta Guadarrama 164-502
Luis Castro Sánchez 1233
Luis D. Alcaraz 806-878
Luis Delgado-Olivares 735
Luis E Arias-Romero 977
Luis E. Florencio Martínez 576-585-606-620-672-689
Luis E. Gómez-Quiroz 1203
Luis Enrique Aguirre-Gil 565
Luis Enrique Ambrosio-Castillo 783
Luis Enrique Arias Romero 1015-1041-1071-1072
Luis Enrique Cobos Puc 400-476-499-957
Luis Enrique Florencio Martínez 664-665
Luis Enrique Jiménez Vázquez 817
Luis E. Pérez-Córdova 308
Luis Ernesto Fuentes Ramírez 897
Luis Felipe Jave Suárez 590
Luis Felipe Jave-Suárez 232-545
Luis Felipe Padilla-Vaca 655
Luis Fernando Cofas-Vargas 211
Luis Fernando García Ortega 202
Luis Fernando Hernández Zimbrón 1171
Luis Fernando Jacinto-Alemán 613
Luis Fernando Montelongo-Martínez 428-883
Luis Figueroa-Yáñez 533-764
Luis Fuentes 1201
Luis Gabriel Briebe-de-Castro 882
Luis Gerardo Arriaga 764
Luis Gerardo Ortega-Pérez 696
Luis Gerardo Zepeda Vallejo 849
Luis Herrera-Estrella 387
Luis Javier Martínez-Morales 886-1116-1122
Luis Javier Solís Martínez 721-750
Luis Jesús Martínez Tozcano 219
Luis Luevano 1044
Luis Luévano-Martínez 772
Luis Marcelino Ramírez Maldonado 909
Luis M. Montañó 1022-1058
Luis Pablo Guzmán Vargas 1213
Luis Rojas-Abarca 1187-1192
Luis Sanchez-Sanchez 1222
Luz Adriana Pichardo-Macías 943
Luz Areli Gorostieta-Nava 440
Luz del Carmen Valerio Jácome 645
Luz Edith Casados Vázquez 210-439-463
Luz Hernández-Esquivel 801-1108
Luz Idalia Valenzuela-García 404-413-419-467-486-530-531
Luz María Fuenzalida 153

- Luz María Martínez 534
 Luz María Rico 1168
 Luz Victoria Flores Villegas 578
 L. Vega 681
 L. Zdvoráková 460
- M**
- Ma. Adriana Martínez Prado 492
 Ma. Azul del Rocío Martínez Vázquez 885
 Mabel Rodríguez 383-473
 Macías-Silva Marina 1049
 Ma. Cristina Amada Vélez del Valle 1023
 Ma. de la Paz Salas 195-847
 Ma. Del Carmen Gutiérrez Villafuerte 1168-1174
 Ma. de Lourdes Acosta Urdapilleta 379-425
 Madhurya Lutikurti 231
 Ma. Elena Cobos Justo 876
 Ma. Fernanda Meneses-Hernández 879-912
 Magali Espinosa Castilla 594
 Magdalena Bezanilla 174
 Magdalena Zamora 1128-1145
 Magdiel Salas-Montenegro 510
 Magnolia del Carmen Ramírez Hernández 650
 Magnolia Estrada-Rivera 202
 Ma. Guadalupe Gutiérrez Escobedo 181-599
 Maidoly Batista Fernández 699
 Maikel González Matos 1034
 Ma. Irene Morelos-Martínez 207-602
 M.A. Juárez-Verdayes 228
 Ma. Luisa Durán-Pastén 541
 Ma. Luisa Escobar-Sanchez 1222
 Manasés Gonzáles Cortazar 469
 Manlio Alejandro González-Rivera 529
 Manuel A. Castro-García 706-778
 Manuela Gómez Gaviria 344-844
 Manuel Alejandro Herrera 306
 Manuel Alejandro Vargas-Vargas 785-790-791
 Manuel Arias 293-330-346
 Manuel de Jesús Castillejos-López 612
 Manuel de Jesús Ochoa-Valdez 608
 Manuel Gutiérrez Aguilar 169-280-830
- Manuel Martínez-Hernández 1152
 Manuel S. Rodríguez 308
 Manuel Vargas Vargas 189
 Ma. Patricia Sanchez Alonso 895
 Marcela Ayala Aceves 165-526
 Marcela Cilia García 275
 Marcela González-Montoya 189-785-790-791
 Marcela Lizano Soberón 779-1024-1028-1033-1260
 Marcela Robles-Machuca 239-399-412-415-418-421-510-511-1111
 Marcel Arturo Gómez García 1209
 Marcela Sosa-Garrocho 613-1050-1067
 Marcelo Barraza-Salas 404-548-552-557-562-569-648
 Marcelo Gomez Palacio Gastélum 292
 Marcelo Gómez Palacio Gastelum 766
 Marcelo Victorio-De Los Santos 809
 Marco A. Loza-Mejía 1065
 Marco Antonio Espina Ordóñez 261
 Marco Antonio Juárez Oropeza 717
 Marco Antonio Loza Mejía 1015-1071
 Marco Antonio Piñón-Chávez 758
 Marco Antonio Sánchez Medina 282
 Marco Antonio Sánchez Villa 739
 Marco Antonio Valdez-Flores 1082
 Marco Antonio Vargas-Hernández 943
 Marco A. Obregón-Mendoza 1129
 Marco A. Perez C. 337
 Marco A. Ramírez-Martínez 335
 Marco A. Ramos-Ibarra 273-289-321
 Marco Espina-Ordoñez 786
 Marco González 330
 Marco Ibrim Rodríguez Sánchez 381-507
 Marco I. Valencia-Sánchez 369
 Marco Iván Valle Maldonado 789
 Marco J. Hernández Chávez 811
 Marco Josué Hernández Chávez 852
 Marco Ramsés Cota-Pineda 1085
 Marco Rito-Palomares 1044
 Marcos D. Couoh-Cauch 1086-1087
 Marcos Francisco Hernández-Robles 881-914-923-927
 Marcos Morales Reyna 684

- Marco Velasco-Velázquez 247-1175
 Margareht Reyes Aldana 975
 Margarita Aguilar-Espinosa 493-546-1252
 Margarita Bahena Román 1156
 Margarita de Lourdes Aguilar-Espinosa 363
 Margarita Isabel Palacios-Arreola 612
 Margarita Jacaranda Rosendo Pineda 193
 Margarita L. Alvarado-Noguez 227
 Margarita Montoya-Buelna 747
 Margarito Martínez Cruz 370
 María Adriana Martínez Prado 395
 María Angélica Santana-Calderón 657-661-662-663-685
 María Berenice Varela Correa 1264
 María C. Ávila Arcos 136
 María Cristina Cardona Echavarría 221-1190
 María Cristina Castañeda Patlán 244-727
 María Cristina Franco-Arellanes 783
 María Cristina Ortiz León 716
 María Cristina Sánchez Aguirre 516
 María Cristina Villa-Medina 456
 María de Jesús Ibarra-Sánchez 1027
 María de la Luz Miranda Beltrán 1139
 María de la Paz Sánchez 173
 María del Carmen González-López 202
 María del Carmen Gutiérrez Galván 730
 María del Carmen Gutiérrez Villafuerte 469
 María del Carmen Oliver Salvador 287
 María del Consuelo Gómez García 604-749
 María de los Ángeles Rangel Rosales 753-992-1001
 María De Lourdes Gutiérrez River 578
 María de Lourdes Gutiérrez Xicotencatl 1255
 María del Rocío Bustillos Cristales 897
 María del Rocío Reyero-Saavedra 195
 María del Rosario Abraham Juárez 916
 María del Socorro Pina Canseco 282
 María Denisse Tejeda Ramírez 914
 María Elena Báez Flores 865
 María Elena Cobos Justo 614-895
 María Elena Ibarra-Rubio 782
 María Elizabeth Álvarez-Sánchez 684-888-1238
 María Elvira Galarraga 971
 María Encarnacion-García 713
 María E. Santana-Román 360
 María Esther Olvera Cortés 1139
 María Esther Rubio Ruiz 745
 María Eugenia Gonsebatt Bonaparte 1165
 María Eugenia Hidalgo Lara 453
 María Eugenia Jaramillo Flores 563
 María Eugenia Jaramillo Torres 554
 María Eugenia Meneses 774
 María Eugenia Núñez-Valdez 440
 María F. Casas-Hernández 1058
 María Fernanda Alvarado-Jáquez 212
 María Fernanda Ballesteros-Barrera 556
 María Fernanda Chávez Jacobo 922
 María Fernanda Frutis Osorio 1029
 María Fernanda Mendoza-Acosta 463
 María Fernanda Pérez Hernández 739
 María Fernanda Shehin 635
 María Guadalupe Cuiniche-Mendez 791
 María Guadalupe Figueroa Torres 866
 María Guadalupe Guerra Sánchez 450-839
 María Guadalupe López Rodríguez 442
 María Guadalupe Martel Gallegos 851
 María Guadalupe Miranda-Novales 925
 María Guadalupe Nieto-Pescador 401-432-1135-1177-1178
 María Guadalupe Pérez-Esquivel 482
 María Guadalupe Quintanar Solís 341
 María Guadalupe Zavala-Páramo 207-602
 María Imelda López Villaseñor 317
 María Isabel Baeza Ramírez 666-687
 María Isabel Pérez Vega 1139
 María Isabel Soto-Cruz 1031
 María Islas-Osuna 506
 María Jatziry Camacho Campos 268
 María José Reyes Chávez 479
 María Jose Toledo Ramírez 998-1006
 María Julieta López Cuevas 1014-1037-1048-1051
 María Karina Manzo Valencia 297-871
 María Luisa Durán-Pastén 1158

- María Luisa Pérez Saldivar** 578
María Luisa Xiqui Vázquez 555-814-825-826-893-898-900-920-1112
María Marcela Robles Machuca 393-1124
María Martha Pedraza Escalona 669
María M. Banda 195-847
Mariam Gasga Tehuintle 1207
María Montserrat Rosendo-Vargas 616
Mariana Aline González Manríquez 846
Mariana Avendaño-Félix 403-407
Mariana Domínguez López 1233
Mariana Flores-Torres 215
Mariana García 551
Mariana Ivonne Martínez Rivera 1226
Mariana J. Kaplan 1027
Mariana Maldonado 263
Mariana Michell García-Reyes 988
Mariana Montserrat Morales Avila 599
Mariana Ochoa López 887
Mariana Río de la Loza 705-765
Mariana Romo-Castillo 769
Mariana Sánchez Ramos 505-522
Mariana Saucedo-García 265
Mariana Segovia-Mendoza 1011
Mariana Suárez 858
Mariandrea Aranda-Barba 384
Mariani Guadalupe Rodríguez López 620
Mariano Buffone 138
Mariano E. Cebrian 1162
Marian Valladares Coyotecatl 1109
María Olívia Medel Flores 604-749
María Patricia Sánchez Alonso 614-627-876
María Raquel Miranda Madrazo 578
María Rosete Enríquez 901
María Sinisterra-Sierra 393-399
María Soledad Mendoza-Trejo 956
María Teresa Alarcón-Herrera 413-419-467-530
María Teresa Álvarez Bañuelos 716
María Teresa Castromonte 139
María Teresa Espinosa García 717
María Teresa Hernández Huerta 370-794
María Teresa Lara Ortiz 458
María Teresa Orta Ledesma 889
María Teresa Percino 330
María Teresa Viana 475
María T. Hernández-Huerta 1118
María Zambrano-Astorga 1148
Maribel Aguilar-Medina 403
Maribel Baquera Arteaga 1226
Maribel Cervantes-Flores 404-479-557
Maribel Hernández-Rosales 151-1088
Maribel Ibarra-Sarlat 925
Maribel Lucila Herrera Ruiz 469
Maribel Ortiz Herrera 1134
Maricarmen Meneses-Juárez 465
Maricarmen Saavedra-López 465
Maricela Aguilar-Duran 434
Maricela Cabrera-Castro 933
Maricela Esteban Méndez 704-832
Maricela Luna Muñoz 954
Maricela Olvera 383-473
Maricruz García-Barrera 1203
Maricruz Gómez Zavala 776
Mariela Judith Domínguez Domínguez 716-723
Mariel Alexis Quiven Feria 999
Mariel Maldonado 261-631-742
Marie S. O'Neill 712
Marilena Iorio 774
Marilen Federico 1053
Marina Campos-Valdez 706-778
Marina Franco-Herrera 384
Marina Gavilanes-Ruiz 265
Marina Macías-Silva 613-1050-1067
Marina Murillo Villicaña 753-992-1001
Marina Ramírez-García 769
Mario A. Bolaños Carrillo 698
Mario Alfredo Urbina Mata 837
Mario Eduardo Almada Ortiz 1248
Mario García Solís 604
Mario Gil Moreno Pérez 1160
Mario I. Gutiérrez Velasco 759
Mario Israel Oregel-Cortez 755

- Mario J. Aguilar-Méndez** 1164
Mario Néquiz-Avedaño 673-690
Mario Pedraza-Reyes 486-530-984-998-1006-1008-1010
Mario Quezada-Aguiluz 153
Mario Rafael Reyes-Torres 925
Mario Ramírez Yáñez 137-1186-1240-1246
Mario Rodríguez Monroy 442
Mario U. Delgado-Jaime 304-1224
Marisa Rodríguez 195-847
Marisela González-Ávila 719
Marisol Báez-Magaña 482-500-658
Marisol Castillo-Castrejón 712
Marisol De la Vega-Tinajar 949
Marisol Mir García 1156
Marisol Olivares-Arévalo 329
Marisol Orozco Ibarra 700
Marisol Ubaldo-Aguilar 367-995
Mark R. Chance 376
Marlen Delgado 228
Marlene Areli Gaytán Gómez 977
Marlene Estefania Campos Morales 818
Marlen Valdez Fuentes 989-1133
Marlon De Ita Ley 638
Martha Calahorra 229-263-358
Martha Dolores Bibbins Martínez 219
Martha Elena Espin-Reza 425
Martha Estrella García-Pérez 987
Martha Imelda Maldonado Cervantes 851
Martha Isela Ramírez Díaz 789
Martha Itsel Cayetano-Marcial 272
Martha Itzel Ramírez Alvarado 739
Martha Lizeth Pérez-Méndez 483
Martha Monzerrath Orozco Sifuentes 1147
Martha Paloma Domínguez Mora 236
Martha Patricia Sierra-Vargas 612
Martha Pedraza-Escalona 485-1199-1251
Martha Robles Flores 244-1068-1069
Martha Rosales-Castro 504
Martha Salcedo Álvarez 804
Martina Rossi 152
Martin Blackledge 201
Martínez-Anaya Claudia 808
Martínez-González, J. J. 990-991-996-997-1002-1007
Martínez Martínez A. 744
Martínez-Pacheco H. 965
Martínez-Rojas R. 874
Martínez-Silva, Ana Valeria 947
Martín González Andrade 342-346-1105
Martin Gustavo Pedraza-Alva 1104
Martin Hernández Lucas 954
Martín Mora-García 608
Martín Pallares-Perez 316
Martin Salamini-Montemurri 152
Martín Salas Paniagua 698
Martin Tiznado-Hernández 506
Marvín Antonio Soriano 939
Ma. Soledad Moreno 396-815
Mate Katona 139
Ma. Teresa Lara Ortiz 312
Ma. Teresa Romero-Ávila 1018
Ma. Teresa Silva-Guzmán 1173
Matías Ramirez-Noguez 1055
Matthew Schipma 1047
Maura Téllez-Téllez 425
Mauricio A. Trujillo-Roldán 438
Mauricio Domínguez Castro 579
Mauricio Flores-Fortis 572-618-767-1181
Maurício S. Baptista 306
Mauro Manuel Martínez-Pacheco 785
Max Alejandro Maximino Rojas 1037-1048
Maximiliano Díaz-Inzunza 511-540
Maximiliano Loredo-Rosales 710-725-1141-1154
Maximiliano Ramírez Vela 1197
Máximo B. Martínez Benítez 226-597
Max Paulfranz 242
Maya Topf 1090
Maydelín Espadín 980
Mayela Rosario Espinoza Duarte 1226
Mayel Chirinos 1214-1249
Mayra Martínez Alfonso 872
Mayra Mejía Sánchez 1110
Mayra Murillo Villicaña 753

- Mayra Patricia Gaspariano Cholula 843
 Mayra Pérez-Tapia 247-1175
 Mayra Quiñonez 473
 M. Carmen Castillo Hernández 702
 Medina-Abreu Karen H. 1049
 Medina Flores, Paola Abril 1227
 Medina-Mendoza Gustavo Gerardo 194
 Medina-Sánchez A. 708
 Medrano-Roldan, H. 490
 Mejia-Carmona G. E. 744
 Melani León Martínez 994
 Melissa Nuñez-Valdés 471
 Melissa Rivera-Escoto 561-570
 Mellado-Rojas M.E. 397
 Méndez-Hernández Edna-Madaí 559
 Méndez-Romero Ofelia 274
 Mendoza-Calles C.A. 965
 Mendoza Martínez, Ariann E. 1227
 Meneses Sánchez A. A. 874-880
 Mercedes Bermúdez-Cortés 711
 Mercedes Edna García Cruz 943
 Mercedes G. López 601
 Mercedes Olvera-Sanchez 632
 Mercedes Olvera-Valencia 609-1061
 M. G. Martinez-Hernández 345
 M. Guadalupe Frías Muñoz 299
 Michelle Chávez-Hernández 805-822-873-894-913
 Michelle Maxson 145
 Michel López-Teros 182-961
 MichelLópez-Teros 553
 Miguel A. Ávalos Rangel 920
 Miguel A. Avila-Rodriguez 320
 Miguel A. Becerril 205
 Miguel Aguilera Ortiz 1003
 Miguel Angel Ávalos Rangel 895
 Miguel Ángel Castillo Martínez 203
 Miguel Angel De La Torre-Arellano 413-419
 Miguel Ángel Hernández-Oñate 1085
 Miguel Ángel Jiménez-Beltrán 591-1217
 Miguel Ángel Juárez Mancera 594
 Miguel Ángel Leyva-López 557-562
 Miguel Ángel López Valle 767-1181
 Miguel Ángel López Vázquez 1139
 Miguel Ángel Mendoza-Catalán 657-1149
 Miguel Angel Orozco-González 536-889
 Miguel Ángel Palomo-Colli 771
 Miguel Ángel Reyes-López 1099
 Miguel Ángel Rodríguez Fernández 768
 Miguel Ángel Rosas-Paz 919-995
 Miguel Angel Sarabia Sánchez 244
 Miguel Ángel Valle Yañez 532
 Miguel Antonio Franco-Vázquez 432
 Miguel A. Rosas-Paz 367
 Miguel Castañeda Lucio 820-827-835-887-897-910-922-924-1201
 Miguel Castañeda-Vaquera 827
 Miguel Cocotl Yañez 177-428-883
 Miguel De Jesús Piña-Mena 486
 Miguel Emiliano Hernández-Fernández 855
 Miguel Froylan Gómez Laguna 1051
 Miguel Gaxiola 775-1032
 Miguel Hernández-Oñate 506
 Miguel Ramírez Aragón 193
 Miguel Rosas-Paz 243-536-889
 Miguel Serrano-Reyes 969
 Mildred Jiménez Báez 307
 Mildred Y. Manríquez-Zamora 457
 Mina Königsberg 182-553-643-961-962-1004-1221
 Minerva Araiza-Villanueva 229-263-358
 Minerva Frutis-Murillo 433
 Minerva García Rangel 851
 Minerva Mata Rocha 578
 M. Inés Hernández-Castellanos 586
 Miranda Desireé López Olivera 650
 Miranda M. Mojica-Prisciliano 311
 Mireya Cisneros Villanueva 192-632
 Miriam F. Ayón-Pérez 809
 Miriam Guadalupe Mateo-Cruz 264
 Miriam Joceline Cruz-Ramos 409
 Miriam L. Llamas-García 313
 Miriam R. Bueno-Topete 706
 Miriam Rodríguez-Sosa 656

- Miriam Vázquez-Acevedo** 290-1254
Miroslava A. Silva-Goujon 392
Misael Josafat Fabian del Olmo 177-428
Misael Ríos-Ortiz 831
Mishael Sánchez-Pérez 137-195-847-926-1121-1246
Mitchel Daniela Berman-Flores 343
Mitzuko-Dautt Castro 202
M. Leticia Santos Martínez 1195
M. Lizbeth Saucedo Bello 978
Mohammed El-Hafidi 919
Moisés Leon-Juárez 236-591-1217
Moisés Selman 261-703-705-765-775-1032
Molina-Jiménez T. 708
Mónica Andrea Valdez Solana 309-375-1003
Mónica A. Ramírez-Cabrera 213
Mónica A. Valdez Solana 361
Mónica Azucena Ramírez Cabrera 488
Mónica Flores Muñoz 966
Mónica García Montelongo 436
Mónica Griselda Arellano Mendoza 549-1138
Mónica Jasso-Romo 1132-1152
Monica Lizbeth Salgado Lucio 1056-1060-1063-1064
Mónica Michele Hernández-Cruz 710-725-1141-1154
Monica Peña-Luna 774
Mónica Ramírez 1149
Mónica R. Calera 608
Mónica Rodríguez-Bolaños 369
Mónica Rosalía Jaime Fonseca 725-1141
Mónica Sánchez-Tapia 713
Mónica Sarzo Gómez 317
Mónica Sierra Martínez 558-589-720
Mónica Torres-Ramos 929-945
Mónica Vianney Ayala Martínez 695
Montserrat Llaguno-Munive 941
Montserrat Mijares Rodríguez 754
Montserrat Quiroga 153
Montserrat Trejo Valencia 723
Montserrat Vázquez-Sánchez 326
Montserrat Guadalupe Vera Delgado 792
Montserrat Hernández-Iturriaga 154
Montserrat Mijares Rodríguez 746
Montserrat Padilla Orozco 971
Montserrat Solé 180
Montserrat Torres-Vanda 951
Montserrat Padilla-Orozco 969
Morales-Castro, J. 833
Morales-Ríos Edgar 194
Morales-Tlalpan V. 624
Moreno-Eutimio Mario Adán 675-688-1084-1196
Mou Pal 886
M. Schwark 460
Munive-Hernández, J.A. 910
Muñoz-Agüero Jessica-Daniela 559
Muñoz-Bello J. Omar 1258
Muñoz Castillo Juan Alejandro 802
Muñoz-Flores M. 708
Muñoz-Herrera JC 508
Muñoz-Pérez Gilberto Andrés 427
Myriam Gorospe 152
Myriam Michell García Luján 875

N

- Nadia Caram-Salas** 316-1150
Nadia F. Esteban Román 954
Nadia Jacobo Herrera 245-246
Nadia Judith Jacobo Herrera 1163
Nadia Luisa Islas Madrid 180
Nadia Mariela Varela Pérez 534
Nadia Naffakh 201
Nadia Tzayaka Castillo Mendieta 937
Nagamani Balagurusamy 375
Nahid Rafiei 621
Nahuel Matias Ko 1198
Nailea Zambrano Pérez 911
Nain Gabriela Pedroza Viveros 614
Nancy Edith Lozoya- Pérez 394
Nancy Fabiola Pérez Herrera 267
Nancy Judit Quintana Rodríguez 896
Nancy Martínez Montiel 843-906
Nancy Paola-Puga 786
Nancy Patricia Rodríguez-Flores 1197
Nancy Sarai Ortiz Ventura 1161

- Nancy Vanessa Iglesias Vázquez 589
Nanziuan Napaketzalli Tiznado-Parra 445
Naomi Vargas Martínez 1070
Napoleón Navarro Tito 657-1149
Natalia Chiquete-Félix 274-324-347
Natalia Copitin-Niconova 274
Natalia Herrera Castillo 1048
Natalia Martínez González 718
Natalia Mata de los Rios 1153
Natalia Nuño-Lámbarri 1203
Natali Domínguez-Velázquez 283
Nataly Morales Galeana 317
Nathan Basisty 152
Nathiely Ramírez Guzmán 388-516-722
Nava Balderas, G. 990-991-997-1002
Nayeli Shantal Castrejón-Jiménez 735
Nayeli Torres-Ramírez 242-243-736-738-799-919-1213
Nazarett Tabita Mendoza-Centeno 710-725-1141-1154
N. Caram-Salas 1148
Neftalí Esteban Guzmán-Mendoza 441
Neimi Estrella-Casanova 546
Nelly Aburto 776
Nelly del Socorro Cruz-Casino 735
Nelly Molina Frechero 575
Nely Gisela López Desiderio 1041
Néstor Fabián Díaz-Martínez 942
Néstor Herrera-Chávez 153
Nicolás Addiel Serafín Higuera 724
Nicolás Gómez-Hernández 583
Nicolás Serafín Higuera 752
Nicolas Villegas 572
Nicole Justine Moreno Licona 327
Nidia Samara Rodríguez Rivera 695
Nils-Göran Larsson 170
Nimbe Torres 713
Nina Montoya Ciriaco 222-1097
Nina Pastor 335-337
Noé Arroyo Mozo 902
Noé de Jesús Miranda-Breceda 467
Noelia de Yaira López Maya 723
Noelia Ferruz 242
Noemí Cárdenas-Rodríguez 943
Noemí García 220
Noemí Marchena Echeverría 426-459
Noemí Meraz Cruz 746-754
Noé Valentín Duran Figueroa 287
Nohely Aimeé Pizano Sosa 922
Nohemí Salinas-Jazmín 247
Nohé Vargas Chávez 750
Nora Barrera 814
Nora Camila Franco Guerra 792
Nora Gabriela Velasco Loyden 742-1233
Noreide Nava 228
Norma Alejandra Vallejo-Cantú 542-652
Norma Angélica Oviedo de Anda 629
Norma Aurora Stephens Camacho 286-697-701-737
Norma A. Valdez-Cruz 438
Norma C. Calderón-Rosas 392
Norma Cecilia Morales-Eliás 326
Norma E. Herrera Gonzalez 1077
Norma Elena Rojas Ruiz 1095
Norma E. López-Díazguerrero 182-1004-1221
Norma Espinosa 577
Norma Estela Herrera González 589-692-1143
Norma Gabriela Barraza Mendez 557
Norma Gabriela Rojas Avelizapa 448
Norma Heredia 890
Norma Judith Cruz Ortega 955
Norma L. Cruz-Ortiz 392
Norma Lilia Salaiza-Suazo 673-690
Norma Osnaya Brizuela 932-1134
Norma Paola Meléndez Rentería 773
Norma Patricia Adán Bante 697
Norma Ramírez 998
Norma Ramírez-Ramírez 984
Norma Silvia Sánchez 229-263-358-850-919
Norma Urtiz Estrada 413-429-492-493-548
Nubia Francélica Hernández-Sánchez 445

Nubia Leticia López García 716-723

Nuria Sanchez-Puig 223-376-603



Obdulia Vera López 953

Octavio Daniel Reyes Hernández 762-777-1106-1142-1169-1176-1179

Octavio Galindo 1059

Octavio Galindo-Hernández 289-322-724-752-755

Octavio López-Méndez 1021

Octavio Montes-Flores 264-381-472-478-507

Octavio Zambada-Moreno 151

Ofelia Méndez-Romero 243-260-324-347

Olga A. Callejas-Negrete 863

Olga Echeverría 736-738-799

Olga Livier Jasso Durón 447

Olga Margarita Echeverría Martínez 1213

Olga Villamar Cruz 977-1015-1041-1065-1071-1072

Olin-Sandoval Viridiana 194

Olin Torres-Isidro 189-785-790-791

Oliva Briz Tena 267

Oliver Millan Catalan 246-1061

Oliver Rafid Magaña-Rodríguez 743

Olufemi Bolaji 163

Omar Alejandro Aguilar-Vera 153

Omar Alejandro Sepúlveda Robles 578-925

Omar Alejandro Tremillo Maldonado 766-770

Omar B. Rivera-Maya 1162

Omar Elind Arroyo Helguera 723

Omar Granados Portillo 713-728

Omar Hernandez-Leon 1238

Omar Lozano García 188-748-962-1044

Omar Muñoz Bello 1024-1028-1033

Omar Páez 891

Omar Rojas-Gutiérrez 1199

Omar Tremillo Maldonado 292-575-721-750-1184

Oriana Landa-Cansigno 392

Orlando Ángeles Martinez 1077

Orlando Vizuet Uscanga 856

Oropeza-Martinez Enrique 1049

Ortega-Gómez K. 644

Ortiz-Castro Randy 1038

Ortiz Garrido I. 1035

Ortiz-Hernández Rosario 611

Osbaldo López Charcas 193

Oscar Abraham Flores-Amaro 501

Oscar Almeda Ojeda 292-721-766-770

Oscar Arce-Cervantes 385-495-535

Oscar Daniel Aranda Moreno 811

Oscar Eduardo Almeida-Ojeda 559

Oscar Gerardo Arrieta Rodríguez 581

Óscar J. Parada-Parra 964

Oscar-Kevin Reyes 539

Oscar Linares-Vergara 1093

Oscar Lithgow-Serrano 1096

Oscar Martín García Rosales 575

Óscar Pérez Méndez 718

Oscar René Zambrano Vásquez 732-784

Osiris Evelyn Aparicio Carrillo 1014

Osmel La Llave León 623-1184

Osney Leiva- Peláez 583

Osorio-Alonso Horacio 702-797

Oswaldo Bivian Paredes 798

Oswaldo I. Hernández Abreu 285-294

Otoniel Rodríguez Jorge 661-662-663-680-685

Otto Geiger 178

Oyervides Muñoz Ernesto 400-499



Pablo A. Madero-Ayala 273-321

Pablo Cruz-Morales 828

Pablo Edson Bustamante Nieves 932

Pablo E. Visconti 138

Pablo Gómez González 633

Pablo Peláez Hernández 137-1240

Pablo Sinuhé Gómez González 651-768-1064

Pablo Valentín Navarro-Enguilo 470-813

Pacheco-Guillen, S. 640

Pacheco-Gutierrez Sebastian 611

Palacios-Serrato Eva 1049

Palma-Flores Carlos 788

Paloma C. Gil-Rodríguez 216

- Paloma Lara 1096
Pamela Barragán-Reséndiz 1133
Pamela Flores-Torres 1004
Pamela Gutiérrez 703
Pamela Jiménez-Chávez 592
Pamela Romo-Rodríguez 501
Paola Acosta-Valdelamar 274
Paola Bocanegra-Ibarias 842-908
Paola Briseño Díaz 1068
Paola del Valle 736-738-799
Paola Eugenia Agustin Vélez 381-507
Paola Gisell Guillén Herrera 932
Paola Isabel Angulo Bejarano 417-423
Paola Itzel Acosta-Valdelamar 260
Paola Maycotte 993
Paola Mendoza-Montiel 1039
Paola Trinidad Villalobos Gutiérrez 1139
Paola V. León-Mimila 329
Pastelin-Palacios Rodolfo 675-688-1084-1196
Patrice P. Hamel 1254
Patricia Areli Cuellar Silva 225
Patricia Coello 176
Patricia Coello-Coutiño 1017-1043
Patricia Curiel-Muñiz 782
Patricia del Arenal Mena 235
Patricia García-López 1204
Patricia Grether-González 617
Patricia Joseph-Bravo 935
Patricia L.A. Muñoz-Muñoz 273-321
Patricia Lozano Zarain 835
Patricia Pérez Vera 1109
Patricia Ponce-Noyola 483
Patricia Ponce-Peña 414-420-467-562-1135-1177-1178
Patricia Ramírez Noguera 1151
Patricia Ríos-Chávez 362-696-743
Patricia Rueda 621
Patricia Sánchez Alonso 1095
Patricia Velez Aguilar 1219
Patricia Vergara-Aragón 710-725-1141-1154
Patricia Victoria Torres Durán 717
Patricio Gariglio Vidal 645
Patricio Suazo 153
Patrick Chinnery 170
Paula González-Hernández 582
Paula J. Klutho 169
Paula Licon-Limón 667
Paul Gaytan 838
Paulina Castañeda-Tamez 260-274
Paulina E. Mora-García 561-570
Paulina Esmeralda Mora-García 747
Paulina Flores Galicia 946
Paulina González-Muñoz 153
Paulina Torres 1228
Paulina Torres Rodríguez 1207
Paul Jacobs 242
Paul Misael Garza-López 441-535
Paul Montaña-Silva 384
Paul Pfluger 951
Paul Sigala 171-295
P. Cambranis-Boldo 1081
Pedraza-Escalona MM 508
Pedro Alberto Marmolejo Cruz 1046
Pedro A. Lopez-García 863
Pedro Brugarolas 1170
Pedro Javier Escobar-Turriza 1124
Pedro Ronel Vásquez-Díaz 370
Pelcastre-Gómez AY 508
Peña-Chora Guadalupe 379
Pérez-Barrón, B.I. 481
Pérez-Cabeza de Vaca, R. 319
Pérez-Carreón J. I. 644
Pérez-Hernández Jesús 673
Pérez Huerta Lexie 802
Pérez-Plasencia Carlos 619-630
Pérez-Tapia SM 508
Perla A. Bejar 671
Perla Azucena Ramos-Parra 1185
Perla del Carmen Niño Moreno 642
Perla Guadalupe Vázquez Ortega 395-492
Perla Jaqueline Vargas Garcia 824
Peter Rehling 1189
Petra Laura Calzada Contreras 832

P. Hrouzek 460
 Pierre Cardol 369
 Pierre Morsomme 369
 Ponciano García Gutierrez 221
 Porfirio Juárez-López 379
 Porfirio Nava 713
 Praxedis Félix-Alcalá 550
 Priscila Pineda 551
 Prisciluis Caheri Salas Navarrete 164
 P. Sarahy Soto-Juárez 311

Q

Quintero-Gallegos, Juan Carlos 973
 Quintero-Hernández Veronica 816-910

R

Rachel Munk 152
 Rafael Arturo Zubillaga 221
 Rafael Bello Bedoy 513
 Rafael Montiel 1187
 Rafael Salgado-Garciglia 482-753-992-1001
 Rafael Torres-Rosas 338
 Rafael Vázquez-Duhalt 149-250
 Rafael Villalobos-Molina 693
 Raghavendra Singh 139
 Ramirez Cipriano Luis Angel 974
 Ramírez-Villarreal Jimena 1239
 Ramiro Perusquia-García 340
 Ramón A. González 1200-1229
 Ramona Icedo García 701-737
 Ramón Castillo Correa 563
 Ramón Cervantes-Rivera 482
 Ramón Pelagio-Flores 1054
 Ramón Rodríguez-Villanueva 1136-1159
 Ramón Suarez Rodríguez 442
 Rangel-Castañeda Itzia Azucena 675-1196
 Rangel-Chávez, C.P. 481
 Raúl Alcalde-Vázquez 224-249
 Raúl De la Torre-Baez 713
 Raúl Enríquez Habib 717

Raúl Galindo Hernández 209
 Raúl García-Morales 250
 Raúl G. Enríquez 303-1129
 Raúl Iván Pérez Bermúdez 336
 Raúl Librado-Osorio 553-945-961-962-1004
 Raúl Osvaldo Pedraza 825
 Raúl Rodríguez Herrera 773-1147
 Raúl Vladimir Ramírez Olmos 1000
 Raya-González Javier 1038
 R. Carreño-López 910
 Rebeca Chávez 939
 Rebeca D. Martínez Contreras 843-906
 Rebeca García Román 716-723
 Rebeca Manning Cella 576-585-606-620
 Rebeca Santes-Palacios 1172
 Rebecca Marvin 171-295
 Recio-Tótoro, B. 640
 Reema Banarjee 152
 Regina Reyes Montelongo 875
 Regina Ruiz-Barrios 586
 Remedios Ramírez 261-705-765
 Renan Vergara-Gutiérrez 374
 Renata Jordá Chávez 954
 Renata Rivera-Madrid 363-493-546-636-1107-1252
 Renata Salcedo-Sánchez 568
 Rendón, J.L. 990-991-997
 Rene Homero Lara-Castro 414-420-1135-1177-1178
 René Torres Ricario 1099
 Renteria-Ortiz, K. S. 490
 Retana-Arellano I. A. 644
 Reyes-García, L. 1002
 Reyes-Jaquez, D. 490
 Ricardo A. Escalona-Villalpando 764
 Ricardo A. Navarro Polanco 730-780-1236
 Ricardo Carreño 1201
 Ricardo Cervantes 1231
 Ricardo Cocoltzi-Gutierrez 505
 Ricardo Cuatlayotl Olarte 898
 Ricardo Félix 184
 Ricardo Gaspar Lino 577
 Ricardo González-Ramírez 1158

- Ricardo Jasso-Chávez 1108
 Ricardo Mejía-Zepeda 340
 Ricardo Miranda Blancas 221-1190-1242
 Ricardo Reyes-Chilpa 946
 Rigel Quintero Luis 897
 Risk Díaz Castillejos 727-783
 Rivera Madrid Renata 443
 Rivera Soto Maria Ana 496
 Roberta Salinas-Marin 338
 Roberto Álvarez-Martínez 831-1075-1239
 Roberto Arreguín-Espinosa 316-919
 Roberto Briones Gallardo 414-420
 Roberto Chavira Ramírez 1214
 Roberto Cruz-Balam 1087
 Roberto-De la Rosa-Mora 412-418
 Roberto Diaz Torres 1151
 Roberto E. Mercadillo 162
 Roberto Esquivel-García 831
 Roberto Ferriz-Martínez 1204
 Roberto Fidencio González Amaro 642
 Roberto Flores López 221
 Roberto Galindo Ramírez 312
 Roberto I. Cuevas-Hernandez 693
 Roberto Jhonatan Olea-Ozuna 178
 Roberto Joaquín Robles-Ramírez 925
 Roberto Jorge Garcia-Mendoza 574-588
 Roberto Labastida-Cuevas 311
 Roberto Martínez 1076
 Roberto Richeimer-Wohlmuth 617
 Roberto Ruiz Medrano 203-366-453-532-1193
 Roberto Sánchez-Olea 608
 Roberto Santín-Márquez 949-1004
 Roberto Sealtiel Farías Gaytán 1094
 Roberto Velasco-García 372
 Robert Winkler 224-249
 Robles Escajeda E. 744
 Rob W. H. Ruigrok 201
 Rocío Alcántara Hernández 1012
 Rocío Alejandra Silva Contreras 921
 Rocío Bustillos 1201
 Rocío del Carmen Barajas-Ornelas 1008
 Rocío del Carmen Montoya-Pérez 790
 Rocío D. Feregrino-Mondragón 1108
 Rocío Díaz de la Garza 477-707-709
 Rocío Guizar Heredia 728
 Rocío I Díaz de la Garzal 299
 Rocío Jiménez Jiménez 1143
 Rocío Juárez Velázquez 1109
 Rocío Montoya Pérez 756
 Rocío Reyero-Saavedra 137-1240-1246
 Rocío Salceda-Sacanelles 796-968-982
 Rocío Tamayo-García 363
 Rocío Valle-Bautista 942
 Rodolfo Benjamín Ocadiz-Delgado 604
 Rodolfo Bernal Reynaga 865
 Rodolfo Gamaliel Avila Bonilla 1079
 Rodolfo García Contreras 339-926
 Rodolfo Lizárraga 1105
 Rodolfo Miranda-Espino 1157
 Rodrigo Aguayo-Ortiz 169-1076
 Rodrigo Contreras-Gaytán 276
 Rodrigo González-Barríos 1090
 Rodrigo López Valor 739
 Rodrigo Martínez-Acosta 367-995
 Rodrigo Miranda Zamora 325
 Rodrigo Monteagudo Ramos 659
 Rodrigo Prieto-Carrasco 760
 Rodrigo Ramos Monteagudo 527-666-678-679-687
 Rodrigo Reyes-Lamothe 489
 Rodrigo Rojas-Mercado 1062
 Rodrigo Romero Nava 549-739
 Rodrigo Troncoso 187
 Rodrigo Vera 153
 Rodrigo Zamora Cárdenas 730-780
 Rodríguez-Aboy Lil 619
 Rodríguez Amaya Tania Milena 499
 Rodríguez-Flores SN 508
 Rodríguez Jorge, O. 150
 Rodríguez López Yanet Eunice 1169
 Rodríguez-Romero A 508
 Rodríguez Salazar María del Carmen 400-957
 Rodríguez Torres Angelina 628

- Rogelio A. Alonso-Morales** 487
Rogelio de J. Treviño 205
Rogelio D. Gaytan-Castro 438
Rogelio González González 721-1184
Rogelio Hernández Pando 1104
Rogelio Morales-Sotelo 600
Rogelio O. Arellano 1264
Rogelio O. Corona Núñez 759
Rogelio Rodríguez-Sotres 351-374-529
Rogelio Rosas 354
Roger Alexis Maldonado Ruiz 962
Roger A. Maldonado-Ruiz 188
Roger Emmanuel Sales-Pérez 542-652
Rogério Sotelo-Mundo 406-475
Rohit Jain 376
Rojas-Contreras J.A. 833
Rojas-Contreras, J.A. 490
Romero Hernández C.B 1035
Romero-Ibarra Josué 496
Rommel A. Carballo-Castañeda 713-1150
Rommel Sánchez-Hernández 350
Ronal Pacheco 228
Ronell Eduardo Bologna Molina 1216
Rosa Bertha Rubio Graciano 436
Rosa Elda Barbosa Cobos 674
Rosa E. Mares-Alejandre 273-321
Rosa Irma Obando Gonzalez 1193
Rosalba Desiré Romero-Palma 710-725-1141-1154
Rosa Lilian Díaz-Chávez 571
Rosalío Ramos-Payán 403-407-609-1110
Rosa López-Castillero 736-738-799
Rosa María Bermúdez Cruz 629-999
Rosa María Camacho Ruiz 384
Rosa María Chávez-Santos 1076
Rosa María del Ángel 1082
Rosa María Galaz-Avalos 291-1086-1125
Rosa María Ordoñez Razo 1131
Rosa María Uribe 935
Rosa M. Bocanegra-García 299
Rosa M. García-Hernández 1162
Rosana Pelayo-Camacho 545-771
Rosana Sánchez López 1240
Rosa Pamela Flores-Torres 182-949
Rosario A. Muñoz-Clares 168-314
Rosario Castro Oropeza 1052
Rosario López-Vancell 690
Rosario Morales Espinosa 618
Rosario Ortiz 736-738-799-1213
Rosario Peralta-Pérez 382
Rosa R. Mouriño-Pérez 863
Rosas-Flores, W. 833
Rosas-Ramírez ED 508
Rosina Sánchez-Thomas 1108
Rosina Torres-Ortega 249
Rossana Arroyo 264-381-472-473
Rossana C. Zepeda 966
Rosy Yunuen Velázquez Jiménez 564
Roxana Olguín Alor 1153
Roxana Silerio Espinoza 297
Rubén-Alejandro Fuentes-Pascacio 1030
Rubén Antonio Romo-Mancillas 1102
Rubén Ávalos López 1014-1015-1016
Rubén G. Contreras 1021
Ruben Piña-Cruz 232
Rueda Munguía M. 356
Ruelas Ramírez, Hilario 1227
Ruth Elena Soria Guerra 1121
Ruth Gutiérrez-Aguilar 951
Ruth Reyes Cortes 868-1144
Ruth Rincón-Heredia 1137-1158
Ruth Trejo Martínez 642
- S**
- Saacnicteh Toledo-Patiño** 208
Sabrina Wischt 242
Sac Nicté Gómez-Barrera 1212
Sadoc Gamaliel Pérez Medina 437
Saé Muñiz 533
Saé Muñiz Hernández 581
Saira Karina Ramírez Thomé 727-783
Sairy Yarely Andrade-Guillen 655
Salas Casas Andres 974

- Salas-Limón, M.J. 910
 Salas-Pacheco-José Manuel 559
 Salas-Pacheco Sergio-Manuel 559
 Saldaña C. 624
 Salgado Ramos I. de J. 880
 Salinas-Celestino J. 624
 Salomón J. Alas-Guardado 1074
 Salvador Hernández-Martínez 677
 Salvador Manzo Avalos 992
 Salvador Mena-Munguía 757
 Salvador Uribe-Carvajal 260-274-324-347
 Salvador Velarde Félix 537
 Samael Olascoaga 1221
 Samantha Andrea Porras Vázquez 1176
 Samantha Barahona 205
 Samantha E. Ryken 174
 Sammy Gutiérrez-Terrazas 891
 Samuel Alejandro Chan Poot 402
 Samuel Arroyo Carranza 552-648
 Samuel Canizales-Quinteros 329
 Samuel Celaya- Herrera 457
 Samuel Enoch Estrada Soto 442
 Samuel García Arellano 686
 Samuel G. Meléndez-López 273-321
 Samuel Lara-González 313-1195-1257
 Samuel Madera-González 461
 Samuel Treviño Mora 934-952-1130
 Samuel Trujano Camacho 246-609
 Sánchez Bocanegra César Augusto 628
 Sánchez-Cach Lucila A. 427-443
 Sánchez Flores Fidel Alejandro 1098
 Sánchez-Lozada Laura Gabriela 702
 Sánchez-Lozada L. Gabriela 797
 Sánchez-Nieto Sobeida 198
 Sánchez-Pérez Julio Alejandro 630
 Sánchez-Villanueva, J.A. 150
 Sandoval-Carrillo Ada 559
 Sandra Cabrera 703-775-1032
 Sandra Calixto-Tlacomulco 199-541
 Sandra Fabiola Velasco Ramírez 435
 Sandra Guadalupe Sánchez-Ceja 987
 Sandra Inés Garzón-Miguel 783
 Sandra J. Velázquez-Huizar 504
 Sandra L. Guerrero-Rodríguez 247
 Sandra Lizeth Alvarado Ortigoza 192
 Sandra Nicole Sánchez Mejía 1213
 Sandra Parra-Hernández 1191
 Sandra Reyes Carmona 555-814-819-825-826-893-898-900-920-963-1112
 Sandra Ríos-Carrasco 568
 Sandra Romero-Córdoba 241-774
 Sandra Romero-Hidalgo 406
 Sandy Lucero Cabanillas-Salcido 1082
 Saniger Blesa José Manuel 802
 Sanny Mariuxi Alcivar Cedeño 1216
 Santana Calderón, M.A. 150
 Santiago Calvillo Martínez 664
 Santiago Castillo-Ramírez 153-155-1182
 Santiago Martínez Calvillo 576-585-606-620-665-672-689
 Santos García 890
 Santos López 217
 Santos Ramírez Carreto 892
 Sara Centeno-Leija 373-491-1093
 Sarai C. Rodríguez-Reyes 706-778-1132-1152
 Saraí Esparza-Reynoso 828
 Sara I. Fuentes Membreño 137-1186-1240-1246
 Sara Kathrin Goetz 208
 Sara K. Schroder 846
 Sara Landeras-Bueno 877
 Sara Luz Morales-Lázaro 1137-1158
 Sara Margarita Garza Aguilar 299
 Sara M. Garza-Aguilar 1185
 Sara Nahuat-Dzib 1252
 Saúl A. Ríos Cano 963
 Saúl Fraire Velázquez 520
 Saúl Jijón Moreno 202-331-335
 Saúl Villalobos Pérez 917
 Sayuri Salgado-López 925
 Scarlett Gallardo-Arriaga 184
 Sebastián Cervera-Pereyra 986
 Sebastián E. González Villarreal 436
 Sebastián Pacheco Gutiérrez 1213

- Sebastián Palacios-Martínez 1055
Sebastian Poggio 178
Selene Napsucialy-Mendivil 607-1235
Selene Yunuén Mena 1103
Selin Dedeoglu 201
Selva Rivas Arancibia 989-1133
Sendar Daniel Nery Flores 773-1147
Sergio Aarón Tinajero-Vargas 524
Sergio Abraham Gutiérrez Payan 865
Sergio Alberto Matías Vera 876
Sergio Alland Colorado-Cortés 1084
Sergio Alonso Durán-Pérez 550
Sergio Andrés Martínez Hidalgo 911
Sergio A. Román-González 1148
Sergio Casas-Flores 202-616
Sergio Cerpa-Cruz 561-570-747
Sergio De La Fuente 1053
Sergio Enríquez-Flores 302
Sergio Fuentes-Hernández 290
Sergio Galindo Najera 292
Sergio García De la Cruz 576
Sergio G. Hernandez-Leon 349-354
Sergio Grinstein 145
Sergio Hernández-Herrera 1136
Sergio Hernández-León 495
Sergio Juárez-Méndez 764-943-1163
Sergio Manuel Salas Pacheco 292-575-721-766-1184
Sergio Márquez Gamiño 792
Sergio Romero-Romero 242-374-489
Sergio R. Zamudio 943
Sergio Salas Pacheco 770
Seung Y. Rhee 1243
S. Hernandez 694
Sheyla Cruz-Cruz 941-947-976-978
Shey-Shing Sheu 1053
Shirish Chodankar 191
Shirley Mora-Godínez 477
Silva Belmares Sonia Yesenia 400-499-957
Silverio Soto-Alvarez 771
Silvia Araceli López Morán 741
Silvia Armenta 385-410-489-490-495-496-858
Silvia Barrios Aguilar 698
Silvia C. Pacheco-Velázquez 1108
Silvia Díaz-Sandoval 515
Silvia Edith Valdés Rodríguez 333-871
Silvia Gómez Jiménez 200-622
Silvia J. Becerra-Anaya 1076
Silvia Jiménez Morales 578-634
Silvia Juárez 736-799
Silvia López 1044
Silvia Luna Suárez 411
Silvia Valdés Rodríguez 297-528
Simona Mosco Aquino 162
Sitali Olguín-Reyes 1146
Sobeida Sánchez Nieto 259-296-310
Socorro Gama-Castro 1096
Sofía Alanís-Muñoz 654
Sofía Bernal Silva 642
Sofía Bernal-Vega 931
Sofía Martínez-Absalón 195
Sofía Plata-Burgos 245
Sofía Rodríguez-Rangel 304-1170-1224
Sofía Tamara Gutiérrez Trejo 385
Soledad Funes 603
Soledad Moreno 196-605-815-829-907
Soley Berenice Nava Galicia 219
Solís, K. Helivier 1012
Sonia Dávila Ramos 1080
Sonia Luquin 747
Sonia Mayra Pérez-Tapia 485-669-1199
Sonia Nava-Salazar 1191
Sonia Yesenia Silva Belmares 476
Sooruban Shanmugaratnam 208-242
Sosa-Garrocho Marcela 1049
Soto Cruz M.A. 1035
Spencer Freeman 145
Stefan de Folter 568
Stefany Rodríguez-Luna 485
Stefhany Segoviano Correa 879-912
Stephania Galaviz- Hernandez 725
Stephanie Hereira Pacheco 222
Stephanie Iraiz Núñez Olvera 192

- Stephanie M Sánchez-Torres 937-940
 Stephany Abigail Tadeo Cuenca 522
 Stephen Burr 170
 Stephen Cusack 201
 Sterling Field 1243
 Supriyo De 152
 Suresh K. Joseph 139
 Suria Gisela Vásquez Morales 868-1144-1164
 Susana Aideé González-Chávez 212-711-760
 Susana Bernardo Hernández 558-589
 Susana Castro-Obregon 160-312
 Susana López 1210-1247
 Susana Valencia Díaz 442
 Suzette Juárez Contreras 821
 Sven Dennerlein 1189
 Svetlana Dokudovskaya 206
 Svetlana Shishkova 607-1235
- T**
- Takuya Nishigaki 1228
 Tamayo Ordoñez María Concepción 957
 Tania Angeles Floriano 771
 Tania Corkidi 278
 Tania Diaz-Vidal 1111
 Tania Ferrer Villada 730-780
 Tania Marieel Ovalle-Ruiz 501
 Tania Reyes Miguel 1060
 Tania Valdivia-Herrera 705-765
 Tania Zenteno-Savín 180
 Tanya Paola Hidalgo Romero 587
 Tapia-Rodríguez, A. 640
 Tatiana Romero-García 755
 Tatiana Torres Loera 366
 Taurino Amilcar Sosa-Velasco 783
 T. Bañuelos-Olivares 708
 Téllez-Téllez Maura 379
 Telma O. Pariente-Pérez 782
 Teresa Edith Garay Rojas 1264
 Teresa García-Gasca 831-1204-1231
 Teresa Hernández Sotomayor 158
 Teresa I. Fortoul Van der Goes 1218
- Teresita Paz Maldonado 851
 Thalía Barrientos Millán 196
 Thalia E. Sánchez-Correa 245
 Theodor Alexandrov 163
 Thibaut Crépin 201
 T. H. J. Niedermeyer 460
 Thomas C. Boothby 1237
 Thomas L. Borders 1047
 Tiany Miranda 473
 Tim Krischuns 201
 Timo Kossendey 242
 Timothy G. Porch 137
 Timothy P. Devarenne 601
 Tomás López 1210
 Tomás Nepomuceno Mejía 576-585-606-620-664-665-672-689
 Tomas Villamar Duque 233-872-905
 Tomás Villaseñor-Toledo 1104
 Tonantzin Guadalupe Anguheven Ledezma 193
 Tonatiuh Ramírez 383-473
 Torres-Ramírez Nayeli 611
 Tremillo Maldonado Omar Alejandro 1216
 Trieu Le 145
 Tzindilú Molina Muñoz 1050
 Tzvetanka Dimitrova Dinkova 287
- U**
- Ubaldo García 979
 Ulises De la Cruz-Mosso 561-570-747
 Ulises Edgardo de León-Covarrubias 213
 Ulises González González 727-783
 Ulises Juárez Figueroa 1109
 Ulises Vega-Castillo 398-445-452-456-521-537
 Ulisses Moreno-Celis 831
 Ulrich Brandt 231
 Ulrik Pedroza-Dávila 231-1189
 Uriel Camacho-Silverio 705-765
 Uriel Cardona-Baltazar 819
 Uriel Colín Camacho 823
 Uriel Noé Solano-Candia 362
 Uriel Ramírez Sotelo 344
 Ursula Torres 579

V

- Vadillo Ortega F. 644
 Vadillo-Pérez L. 644
 Valeria Alexandra López Toledo 453
 Valeria Calderón Frontana 660-1244
 Valeria Fernanda Avila-Mendoza 675
 Valeria Jacqueline Soto Ontiveros 1250
 Valeria Madrigal Pineda 869
 Valeria Martínez-Silva 941
 Valeria Mateo-Estrada 1182
 Valeria Piazza 967
 Valeria Punzo Mora 761
 Valery Domínguez Villegas 522
 Valle-Velázquez Estefanía 702-797
 Vanegas-Flores Claudia Leticia 675-1196
 Vanesa Olivares Illana 240-733-1183-1223-1225-1232
 Vanessa Dehennaut 285-294
 Vanessa Iglesias 558
 Vanessa Labrada-Martagón 180
 Vanessa N. Alcántara-Garduño 805
 Vanessa Olivares Illana 817
 Vanessa Wendi Suarez Barrios 732-784
 Vannia Daniela Merced-Canales 710-725-1141-1154
 Varinia López-Ramírez 389-409-503-1120
 Vásquez-Lima, C. 990-1007
 Vázquez-Leyva KS 508
 Vázquez-Martínez, J. 481
 V.E. Bolado-García 965
 Vega-Marín, C. 874
 Velasco-Loyden Gabriela 1049
 Velásquez-Hernández Jimena Yadira 1258
 Velazquez-Sámamo Guillermo 675-688-1196
 Ventura Gallegos José Luis 802
 Venus Melissa Gómez Lagunas 851
 Verónica Aranda-Chan 472
 Verónica Bautista-Piña 634
 Verónica Dayali Gutiérrez Martínez 479-557
 Verónica Díaz Hernandez 1022
 Veronica Diaz-Villegas 943
 Verónica Edith Gallegos-Hernández 710-1141-1154
 Verónica García-Betancourt 936
 Verónica García Castillo 288-454
 Verónica Garcia Radilla 1104
 Veronica Jiménez-Jacinto 961
 Verónica Judith Picos-Cárdenas 1082
 Verónica Morales-Tlalpan 574-586-588-647-882-915
 Verónica M. Rodríguez Córdova 956
 Veronica Najera 230
 Verónica Rocío Vásquez Garzón 723
 Verónica Rosales Islas 905
 Verónica Salas-Venegas 182-949-1004
 Verónica-Yutsil García-Rasilla 667
 Vianey Flores González 262
 Vianey Marín 1201
 Vianey Ramírez-Andoney 487
 Vianey Ramírez Lechuga 890
 Vianney Ramírez-Adoney 930
 Vicente Castrejón Téllez 745-994
 Vicente Madrid Marina 1156
 Vicente Sandoval Herrera 1160
 Víctor D. Gonzalez-Fernandez 487
 Víctor Enrique Sarmiento-Ortega 1130
 Víctor Eric López y López 449-462
 Víctor García González 289-292-752
 Víctor Gómez Calvario 958
 Víctor Guadalupe Garcia González 724
 Víctor Henández 967
 Víctor H. Robledo-Zacarias 405
 Víctor Hugo Avilés Rosas 971
 Víctor Hugo Rosales García 1068
 Víctor Hugo Sánchez-Vázquez 139
 Víctor Humberto Bustamante Santillán 824-866-903
 Victoria Chagoya y Hazas 742-1233
 Victoria E. Bolado García 966
 Victoria Palafox Sánchez 188-962
 Victoria Pando-Robles 892
 Victoria Ramírez 787-1113
 Víctor Lázaro-Vidal 1075
 Víctor Manuel Ayala-García 395-404-413-419-467-486-530-531-548

Victor Manuel Carballo-Uicab 546
Víctor Manuel García Vera 841
Victor Manuel Hodgkin-Bastard 441
Víctor Manuel Luna Pineda 204-769
Víctor M. Ayala-García 984-1010
Victor Meza Carmen 789
Víctor M. Loyola-Vargas 291-1086-1125
Víctor Olalde Portugal 297-528-871-1088
Víctor Rodríguez-Escobar 1118
Víctor Torres-Flores 1214-1249
Víctor Treviño 220-707-709
Victor V. Barrios 815
Vijna Illesca 153
Vilma Araceli Maldonado Lagunas 716
Vilma Carolina Bekker Méndez 587
Vilma Maldonado Lagunas 594-1052
Vincent Cerbantez-Bueno 568
Violeta Guadalupe Silva Díaz 637
Virginia Mandujano González 505-522
Viridiana Alejandre Castañeda 789
Viridiana Estefany Solis Ayala 1249
Viridiana Navarrete-Yañez 230
Viridiana Olin Sandoval 327-331
Viviana Escobar-Sánchez 243-536-889-919
Viviana Estefanía García Vázquez 745
V.J. Carrasco Urrutia 744
Vladimir Flores Benavides 430
Vladimir Juarez Arellano 312
Vladimir Morales Erasto 876
V. M. Arenas-Luna 694
V. Morales-Tlalpan 345

W

Walid Kuri Harcuch 1023
Walter Ángel Trujillo-Rangel 987
Wanda Yael Tlalmis Guzmán 1251
Wendy Argelia García Suastegui 1165
Wendy Escobedo-Hinojosa 1198-1259
Wendy Xolalpa-Villanueva 308-431-509
Wendy Zárata-Hernández 542-652
Wilhelm Hansberg 891

William Meza-Morales 303-1129
Willy Angel Delgado Tapia 281
Wilma A. González Kantun 593

X

Xareni Zinereth Herrera Valero 773-1147
Ximena Carranza-Trinidad 351
Ximena Escalera-Fanjul 163
Ximena Hernandez Ramos 857
Ximena Paola Becerril Vigueras 1176
Ximena Valdés Sánchez 868
Ximena Valeria Sánchez Nava 1015
Ximena V. Sánchez-Nava 1065
Xinkun Wang 1047
Xiomara Patricia Perea Domínguez 1261
Xóchitl Flores-Ponce 970
Xochitl Lucia Ramirez Hernandez 977
Xochitl Morales de la Cruz 601
Xochitl Pérez-Martínez 231-1189

Y

Yadira Hernández Rueda 1214-1249
Yadira Ramírez-Rodríguez 1113
Yael Efrén Díaz-López 951
Yael González Tinoco 269-850
Yagul Pedraza Pérez 897-1201
Yair Álvarez-Ricardo 303-1129
Yair Cárdenas-Conejo 443-464-493-909-1206-1256
Yair Romero 705-765
Yamile Paredes-Chiquini 270-334
Yaneli Martínez Sotelo 226
Yang Sun 1170
Yanin Chávarri Guerra 761
Y. Aranda-López 1067
Yaraymi Ortiz Reyes 890
Yareli Guadalupe Ramos Herrera 766
Yarely Mabell Beltrán Navarro 1072
Yarely Ramos Herrera 770
Yarely Rocio Herrera Sánchez 1037
Yazmín Contreras-Bravo 567

Yazmin Melendez-Xicohtencatl 462
Yazmín Mercedes Amador Segovia 548
Yazmín Ramiro-Cortés 358
Yazmith Briseño 754
Yendi Ebenezer Navarro Noya 1097
Yendi E. Navarro Noya 222
Yesenia Cid Cruz 236-1024
Yeshua-Eliezer González-Jiménez 969
Yessica Eduvigis Zamudio Cuevas 564-595
Yobana Pérez-Cervera 338
Yohana Parrado Quintana 969-978-980
Yolanda Camacho-Villasana 231-1189
Yolanda Herrera Arrieta 1099
Yolanda Patricia Alemán-García 769
Yolanda Reboloso-Gómez 608
Yosahandy Palacios Castañon 911
Yoselin-Karely Rodríguez-Reyes 239
Yoselin Xiomara Rodríguez Muñoz 903
Yovani Aguilar-Carrillo 1116
Yrving Daniel Díaz-De Lucio 944
Yuxing Yuan 1053
Yulan Piao 152
Yuma Ávila-Lopez 510
Yuma-Citlalli Ávila-López 415-421
Yumiko De la Cruz Rodríguez 520

Yunuen Moreno-López 972
Yunuen Velázquez Jiménez 595
Yuridia Cruz-Zamora 568
Yuriria Santoyo 1201
Yury Rodríguez-Yañez 757
Yussel F. Pérez Navarro 597
Yvonne Rosenstein 676

Z

Zambrano-Vásquez Oscar R. 702-797
Zazueta-Alvarez, D. E. 490
Zentella Dehesa Alejandro 802
Zepeda R.C. 708-965
Z. Fernandina Nieves-López 224
Zhaida Aguilar-Gonzalez 369
Zhaida I. Aguilar Gonzalez 380
Ziomara Ramos-Jiménez 378
Z. Kameník 460
Zugasti Cruz Alejandro 499
Zuleima Natali Domínguez Velazquez 327
Zumaya-Pérez Luis Carlos 688
Zuribet Baxin-Toto 343
Zurisadai Morales Herrera 1051
Zyanya Patricia Espinosa Riquer 1028



Cinvestav



Coordinación de la
Investigación Científica, UNAM



CCG

Centro de Ciencias Genómicas



CONAHCYT

CONSEJO NACIONAL DE HUMANIDADES
CIENCIAS Y TECNOLOGÍAS



Facultad de Química



Instituto de Biotecnología

UNIVERSIDAD NACIONAL AUTÓNOMA DE MÉXICO



Instituto de
Fisiología
Celular



SECRETARÍA DE
DESARROLLO
INSTITUCIONAL



CTR[®]
SCIENTIFIC

FALCÓN
División Analítica



Instrumentación y Servicios
Analíticos, S.A. de C.V.



RAVER

GRUPO RAVER APLICACIONES SA DE CV

uniparts

Opciones Integrales para los Ciencias de la Vida



**QUIMICA
VALANER**

CIENTÍFICA
SENNA

UNA DIVISIÓN DE EL CRISOL

