Sociedad Mexicana de Bioquímica A.C.

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XXIV Reunión Bioenergética y Biomembranas

October
26 - 30, 2025

Oaxaca, México

Book of Abstracts

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XXIV REUNIÓN DE BIOENERGÉTICA Y BIOMEMBRANAS

ORGANIZING COMMITTEE

CHRISTIAN CORTÉS ROJO
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Bienvenida

Hace 45 años, una docena de investigadores y sus cerca de 30 estudiantes se reunieron dentro de un foro sencillo a discutir cómo los gradientes electroquímicos pueden convertirse en energía química y viceversa. En ese momento nació la Rama de Bioenergética y Biomembranas, la rama más antigua de nuestra Sociedad Mexicana de Bioquímica. Desde su inicio, los investigadores de la Rama siempre colocaron a los estudiantes como los pilares de ésta, convencidos de la estrategia de sembrar para el futuro. Así, cada dos años desde 1979, los estudiantes de esta rama han tenido la oportunidad de exponer sus avances de investigación, de discutir sus resultados con los investigadores, y de escuchar a investigadores reconocidos nacional e internacionalmente, siempre dentro de un ambiente de unión académica, de apoyo y solidaridad científica.

Con el tiempo, nuestra Rama se diversificó en el estudio de diversos fenómenos biológicos relacionados con la obtención de energía celular y su relación con las membranas que la conforman, su interacción con otras vías metabólicas, así como la participación de estos sistemas en procesos fisiológicos y patológicos, dentro de diversos modelos de estudio.

En cada reunión, nuestra Rama se enorgullece en reconocer con La medalla “Dr. José Laguna” a un investigador consolidado y a un investigador joven como reconocimiento a sus contribuciones a esta área del conocimiento, y como forma de motivar a los estudiantes dentro de éste maravilloso mundo de la investigación.

¡Les damos la más cordial bienvenida a Oaxaca, Oax.! Esperamos que todos aportemos lo mejor de nosotros para hacer de esta reunión una experiencia inolvidable.

El Comité Organizador

MEDALLA "JOSÉ LAGUNA GARCÍA"

ESTA MEDALLA SE OTORGA POR LA RAMA DE BIOENERGÉTICA Y BIOMEMBRANAS A DOS MIEMBROS QUE SE HAN DISTINGUIDO POR LA CALIDAD DE SUS TRABAJOS DE INVESTIGACIÓN, SUS CONTRIBUCIONES EN EL ÁREA Y SU PARTICIPACIÓN EN LAS ACTIVIDADES ACADÉMICAS DE LA RAMA.

PREMIADOS EN 2023
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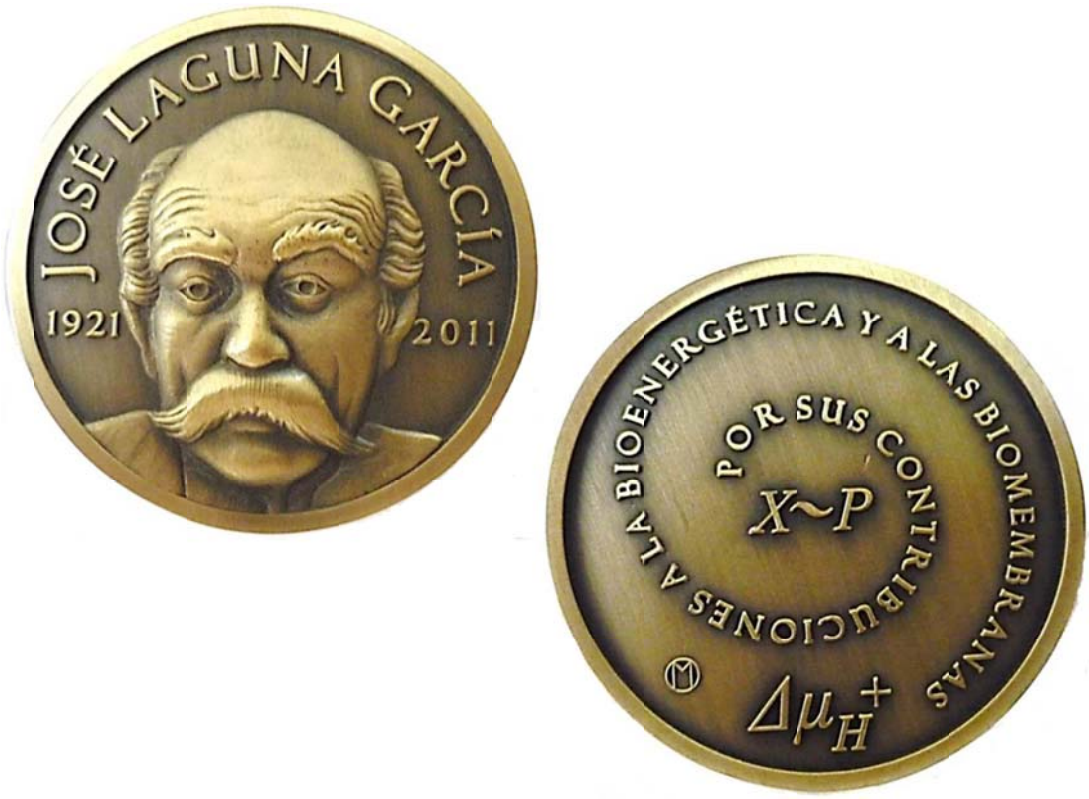

Medalla José Laguna García, acuñada en el 2013

Cuño Anverso: Efigie del Dr. José Laguna, con la leyenda "JOSÉ LAGUNA GARCÍA 1921-2011".

Cuño Reverso: Formando una espiral, la leyenda "POR SUS CONTRIBUCIONES A LA BIOENERGÉTICA Y A LAS BIOMEMBRANAS". En el origen de la espiral, el símbolo *X*~*P*, representando el intermediario fosforilado de alta energía de la hipótesis de E.C. Slater (1953) y en el final de la espiral, el símbolo $\Delta\mu H^+$, representando la fuerza protón-motriz de la hipótesis quimiosmótica de Peter Mitchel (1961).

Metal: Bronce

Peso: 125 g

Diámetro: 6 cm

Diseño: Miguel Gómez Counahan

Troquelado: "ARTE Y ESCULTURA FIDIAS", México

LINEAMIENTOS PARA EL OTORGAMIENTO DE LA "MEDALLA JOSÉ LAGUNA GARCÍA"

La "Medalla José Laguna García" fue acuñada *ex profeso* en el 2013 para honrar la memoria del Dr. José Laguna, quien fuera pionero de la bioquímica moderna en México, uno de los fundadores de la Sociedad Mexicana de Bioquímica (SMB) y mentor de muchos de los miembros que integraron la primera generación de investigadores que cultivaron el estudio de la Bioenergética y de las Biomembranas, generando así la corriente más activa de investigación en esta área en nuestro país.

- **Objeto del reconocimiento**

La "Medalla José Laguna García" fue concebida para preservar y estimular la investigación en el área de la bioenergética y las biomembranas. Este reconocimiento lo otorgará la Rama de Bioenergética y Biomembranas de la SMB (de aquí en adelante denominada como "la Rama") a miembros distinguidos de la comunidad que llevan a cabo investigación en alguna de estas dos áreas del conocimiento. Se otorga en dos modalidades, la primera a la trayectoria de científicos consolidados y la segunda como un estímulo a las contribuciones realizadas por científicos más jóvenes.

- **Académicos que pueden hacerse acreedores a este reconocimiento**

En su primera edición la medalla se otorgó a investigadores que cultivaron la Bioenergética en sus inicios y que colaboraron con la creación y desarrollo de la Rama. Bianualmente se entregan dos reconocimientos "Medalla José Laguna García", una de ellas a un miembro de la Rama que cuente con una trayectoria fructífera de investigación, que haya contribuido de manera notable a profundizar el conocimiento en el campo y que haya mantenido una actividad académica sostenida en la Rama. La segunda medalla se le otorga a un miembro de la Rama menor de 50 años que haya participado con constancia en las actividades académicas de ésta y que haya realizado contribuciones relevantes en este campo.

- **Procedimiento para la concesión del reconocimiento**

- No se emitirá convocatoria alguna para concursar por el reconocimiento "Medalla José Laguna García", tampoco se recibirán solicitudes institucionales o personales apoyando a un determinado candidato.
- Los merecedores de este reconocimiento honorario serán seleccionados directamente por el jurado, en función de los estudios, investigaciones y/o entrevistas que el propio jurado realice sobre la calidad de las contribuciones

científicas que han llevado a cabo los miembros de la Rama, tomando en cuenta asimismo su probidad académica y moral.

- La entrega del reconocimiento "Medalla José Laguna García" se hará exclusivamente en la Reunión o Congreso bianual de la Rama. Quedará a juicio del Comité Organizador de la Reunión de la Rama escoger, dentro del programa académico, el momento apropiado en que se efectuará la ceremonia de entrega de las medallas. Será prerrogativa del Comité Organizador invitar o no, a alguno de los premiados a impartir una conferencia dentro del programa científico de la reunión.
- El jurado que otorgará la medalla en su siguiente edición será elegido durante la Reunión previa de la Rama, de la misma forma en que se elige al Comité Organizador para la siguiente Reunión.
- El jurado quedará conformado por tres personas, todos ellos integrantes reconocidos y asiduos a las reuniones de la Rama. Al menos uno de ellos deberá haber participado en un Comité Organizador de alguna Reunión. Los miembros del Comité Organizador de la Reunión en turno no podrán ser a la vez miembros del jurado durante ese mismo periodo.
- Los miembros del jurado desarrollarán sus actividades de forma honoraria, sin percibir retribución económica alguna. Además, organizarán sus sesiones y deliberaciones de la manera que mejor convenga a sus intereses. En caso de que algún miembro del jurado esté incapacitado para realizar sus funciones, el Comité Organizador de la Reunión en funciones será el encargado de seleccionar e invitar a algún otro integrante de la Rama para que ocupe el puesto vacante.
- Uno de los miembros del jurado deberá permanecer como integrante en el siguiente jurado, con el fin de preservar los antecedentes de los procedimientos y criterios de selección. El integrante que permanecerá en funciones en una segunda ocasión será seleccionado por mutuo acuerdo del jurado anterior. En la reunión subsecuente de la Rama se elegirán a dos nuevos miembros para integrar el siguiente jurado y así sucesivamente.
- El jurado dispone de más de un año y medio para llevar a cabo sus deliberaciones y deberá emitir su fallo al menos tres meses antes de la fecha en que se lleve a cabo la Reunión de la Rama, haciéndolo del conocimiento únicamente del Comité Organizador. Así mismo, será el propio Comité Organizador el encargado de informar a los colegas seleccionados que se hicieron acreedores al reconocimiento y convocarlos a la ceremonia de entrega.
- El fallo del jurado será inapelable.
- Ninguno de los dos firmantes de este documento puede hacerse acreedor al reconocimiento.
- No podrá otorgarse la medalla en forma póstuma.

- No podrá otorgarse este reconocimiento a científicos externos a la Rama que no hayan tenido una participación relevante y sostenida en la vida académica de la propia Rama.
- No se le podrá otorgar la "Medalla José Laguna García" a una persona que ya fue distinguida con este reconocimiento.
- En las memorias de la Reunión de la Rama ulteriores se incluirán en orden alfabético, por una parte, los nombres de los científicos distinguidos con la medalla, tanto de la Reunión correspondiente como de las anteriores y por otra, los nombres de los miembros del jurado del año correspondiente.
- El financiamiento, la responsabilidad de la manufactura y la disponibilidad de las medallas para cada ceremonia de entrega, estarán a cargo de los abajo firmantes. Por lo tanto, la responsabilidad de elaborar las medallas no recaerá sobre los miembros del Comité Organizador de la Reunión de la Rama.
- El reconocimiento "Medalla José Laguna García" no podrá otorgarse en ninguna otra instancia académica o social fuera del ámbito de la Rama.
- Estos lineamientos se publicarán, en lo sucesivo, en las memorias de cada Reunión de la Rama. Cualquier situación no contemplada en éstos será resuelta por el pleno del jurado, consultando, si así lo consideran necesario, con los miembros del Comité Organizador de la Reunión de la Rama o con integrantes de jurados anteriores.

México D.F., Febrero de 2015

Georges Dreyfus
Diego González Halphen

XXIV Reunión Bioenergética y Biomembranas

October 26 - 30, 2025

SMB

TIME	SUNDAY October 26th		MONDAY October 27th	TUESDAY October 28th	WEDNESDAY October 29th	THURSDAY October 30th
7:30 – 9:00	Arrival		Breakfast	Breakfast	Breakfast	Breakfast
9:00 – 10:00			Plenary Lecture Dra. Alicia Kowaltowski	Plenary Lecture Dr. Pierre Cardol	Plenary Lecture Dr. Héctor Valdivia	
10:00 – 11:30			Oral Session I Electron Transport Chains	Young Career Researcher´s Symposium	Oral Session VI Regulation of Energy Metabolism	Check out
11:30 – 12:00			Coffee Break	Coffee Break	Coffee Break	
12:00 – 13:00			Oral Session II ATP synthases, ATPases and pyrophosphatases	Oral Session IV Photosynthesis	Simposium ALACF	Departure
13:00 – 14:00	Registration	Check in		Oral Session V Lipids and membranes		
14:00 – 16:00		Lunch	Lunch	Lunch	Lunch	
16:00 – 16:30	Welcome ceremony		Plenary Lecture Dr. Alfredo Cabrera Orefice	Plenary Lecture Dr. José Manuel Pérez Aguilar	Oral Session VII Mitochondrial pathologies	
16:30 – 17:00	Cultural Conference Dr. Abraham Jahir Ortiz Nahon					
17:00 – 17:30			Oral Session III Structure and function of membrane proteins	Poster Session	Plenary Lecture Dra. Valentina Parra Ortiz	
17:30 – 18:00	Coffee Break					
18:00 – 19:00	Plenary Lecture Dr. John Lemasters		Free Time		Closing Ceremony	
19:00 – 19:30	José Laguna´s Medal Award			Business Session	Free Time	
19:30 – 20:00						
20:00 – 22:00	Dinner		Dinner	Dinner	Dinner and Farewall Party (20:30 – 1:30)	

Scientific Program

Códice Tonindeye. Cultura mixteca

Sunday, October 26th

16:00 – 16.30 Welcome Ceremony

16:30 – 17:30 **Cultural Conference**

Fotografía en comunidades afromexicanas: documentación sociohistórica, diversidad pluricultural y memoria visual

Abraham Jahir Nahón
Universidad Autónoma Benito Juárez de Oaxaca

Chair: Luis Alberto Luévano
Tecnológico de Monterrey

17:30 – 18:00 Coffee break

18:00 – 19:00 **Plenary Lecture**

Mitochondria in Pathobiology

John Lemasters
Medical University of South Carolina, USA

Chair: Christian Cortés Rojo
Universidad Michoacana S.N.H.

19:00 – 19:30 **José Laguna's Medal Award**

Chair: Manuel Gutiérrez Aguilar
Facultad de Química, UNAM

19:30 – 21:30 Welcome cocktail

Monday, October 27th

9:00 – 10:00

Plenary Lecture

Diets, mitochondria and calcium transport

Alicia Kowaltowski
Institute of Chemistry, University of São Paulo

Chair: Luis Alberto Luévano Martínez
Tecnológico de Monterrey

10:00 11:30 *Oral Session I*

Electron Transport Chains

Chair: Ariann Elizabeth Mendoza
Instituto de Fisiología Celular, UNAM

The NDi1 starts the electron flux in the Saccharomyces cerevisiae respirasome
Oscar Flores Herrera. Facultad de Medicina, UNAM

Identification of oxidative phosphorylation complexes of Paramecium multimicronucleatum
María Guadalupe Quintanar Solis. Instituto de Investigaciones Biomédicas, UNAM

Kinetic characterization of Bos taurus respirasome
Carolina Guerrero Teudosio. Facultad de Medicina, UNAM

Ancient respirasome: Kinetics and polypeptide composition
Mercedes Esparza Perusquía Facultad de Medicina, UNAM

Characterization of Mitochondrial Oxidative Phosphorylation Complexes from Auxenochlorella protothecoides
Tóshiko Takahashi Íñiguez. Instituto de Investigaciones Biomédicas, UNAM

Kinetic characterization of respirasomes and free complex I from Yarrowia lipolytica
Giovanni García-Cruz García Cruz. Facultad de Medicina, UNAM

11:30 – 12:00 Coffee break

12:00 – 14:00 *Oral Session II*

ATP synthases, ATPases and pyrophosphatases

Chair: Mercedes Esparza Perusquía.
Facultad de Medicina, UNAM

Kinetic characterization and polypeptide composition of the dimer and monomer of the FoF1-ATP synthase from Yarrowia lipolytica
Alejandro Cruz-Cárdenas. Facultad de Medicina, UNAM

Does the δ-subunit of F_1-ATPase from Polytomella parva has a role in regulating the hydrolytic activity of this enzyme?
Marcos Ostolga Chavarría. Instituto de Fisiología Celular, UNAM)

Kinetic characterization of dimeric F_1F_O-ATP synthase
Anaiza Rico Luna. Instituto de Investigaciones Biomédicas, UNAM

Distribution of the F_1F_O-ATP synthase regulatory ζ subunit in alphaproteobacterial
Fidel Serrano López. Facultad de Ciencias, UABC

Cysteine Oxidation as a Regulator of the A Subunit of the Vacuolar ATPase in Saccharomyces cerevisiae
Mariana Michell Garcia Reyes. Instituto de Fisiología Celular, UNAM

Exploring the druggability of the binding site of exogenous allosteric inhibitors of F_1F_O -ATP synthase
Enrique García Hernández. Instituto de Química, UNAM

Gene expression modulation of SERCA3 in MCF-7 cells treated with the extract of Capsicum annuum L. var. Fascinato
Roberto Jorge García Mendoza. Facultad de Ciencias Naturales, UAQ

Epigenetic regulation of ATP2A2 and ATP2A3 genes and their potential role in hepatocellular carcinoma progression
Guadalupe Hernández Martínez. Universidad Veracruzana

14:00 – 16:00 Lunch

16:00 – 17:00 **Plenary Lecture**

Exploring the mitochondrial complexome: from protein complexes to nucleoprotein assemblies

Alfredo Cabrera Orefice
Biochemical Institute. Justus Liebig Universität Giessen

Chair: Salvador Uribe Carvajal
Instituto de Fisiología Celular, UNAM

17:00 – 18:00 *Oral Session III*

Structure and function of membrane proteins

Chair: Luis González de la Vara
Cinvestav Irapuato

The role of the ubiquitn Ligase Gzl in apical-basal transytosis during the development of Drosophila
Víctor Ángel Urbieta Ortiz. Instituto de Investigaciones Biomédicas, UNAM

Study of the function of Pet494 in Cox3 biogenesis in mitochondria of Saccharomyces cerevisiae
Juan Pablo Berry Leon. Instituto de Fisiología Celular, UNAM

Estrogen Deficiency Aggravates Mitochondrial and Cardiac Dysfunction in a Female Mouse Model of Cardiometabolic Injury Through Impaired Phospholamban Signaling
Silvia Araceli López Morán. Escuela de Medicina y Ciencias de la Salud, ITESM

Bioenergetics in aging yeast: the interplay between sirtuin2 and caloric restriction
Carolina Ricardez García. Instituto de Fisiología Celular, UNAM

18:00 Free evening

Tuesday, October 28th

9:00 – 10:00 **Plenary Lecture**

Evolutionary Remodeling of Respiration and Photosynthesis in Euglena gracilis

Pierre Cardol
Université de Liège, Belgium

Chair: Héctor Miranda Astudillo
Instituto de Investigaciones Biomédicas, UNAM

10:00 – 11:30 **Young Career Researcher's Symposium**

Chair: Alfredo Cabrera Orefice
Justus Liebig Universität Giessen

Light harvesting regulation and photodamage interplay in Chlamydomonas reinhardtii
Wojciech J Nawrocki
Institut de Biologie Physico-Chimique, CNRS, France

The physiology of methane-producing archaea under stress
Michel Geovanni Santiago Martínez
University of Connecticut

11:30 – 12:00 Coffee break

12:00 – 13:00 *Oral Session IV*

Photosynthesis

Chair: Tóshiko Takahashi Íñiguez.
Instituto de Investigaciones Biomédicas, UNAM

Far-Red Component Enhances Paramylon Production in Photoautotrophic Euglena gracilis
Zhaida Itzel Aguilar González. Instituto de Investigaciones Biomédicas, UNAM

Quantifying the photoprotective effect of qE NPQ in Chlamydomonas
Felix Vega de Luna. Institut de Biologie Physico-Chimique, Sorbonne Université

Ammonia detoxication via photosynthetic reactions: a tale about a natural solar powered ammonia scrubbing system in salamander eggs
Alonso Zavafer. Brock University

The protein import machinery in the colorless plastids of the chlorophycean alga Polytomella parva
Sergio Fuentes Hernández. Instituto de Fisiología Celular, UNAM

13:00 – 14:00 *Oral Session V*

Lipids and Membranes

Chair: Miriam Vázquez Acevedo.
Instituto de Fisiología Celular, UNAM

Contribution of sphingolipids to the regulation of plasma membrane H+-ATPase activity in Arabidopsis
Laura Carmona Salazar. Facultad de Química, UNAM

Design and movement: Sphingolipid influence on membrane fluidity
Marina Gavilanes Ruíz. Facultad de Química, UNAM

The plasma membrane of beetroots submitted to waterlogging
Luis E. González de la Vara. Cinvestav Irapuato

Effect of killer conjugated Ag nanoparticles over biological systems
Carlos Alberto Molina Vera. Facultad de Ciencias Naturales, UAQ

14:00 – 16:00 Lunch

16:00 – 17:00 **Plenary Lecture**

Application of computational methods to understand the function of membrane proteins

José Manuel Pérez Aguilar
Benemérita Universidad de Puebla

Chair: Christian Cortés Rojo. IIQB - UMSNH

17:00 – 19:00 Posters Session

19:00 – 20:00 Bussines session SMB

Wednesday, October 29th

9:00 – 10:00 **Plenary Lecture**

Cardiac Ryanodine Receptor Channelopathies: From Membrane Bioenergetics to Clinical Phenotypes

Héctor Valdivia
University of Wisconsin Medical School

Chair: Gerardo García Rivas
Tecnológico de Monterrey

10:00 – 11:30 *Oral Session VI*

Regulation of Energy Metabolism

Chair: Francisco Guillermo Mendoza Hoffmann
Universidad Autónoma de Baja California

Mitochondrial ROS induces lysosomal dysfunction impairing autophagic flux in human cells carrying the ApoE4 allele
Sandra Aurora Esquivel Niño. Cinvestav Zacatenco

Effect of cancer cells-derived conditioned medium on cardiomyocyte energy metabolism
Fernando Emiliano Jiménez Mondragón. Instituto Nacional de Cardiología "Ignacio Chávez"

Exercise combined with metformin and tert-butyl hydroquinone improves hepatic mitochondrial bioenergetics and redox status in middle-aged obese female Wistar rats
Mina Konigsberg Fainstein. Universidad Autónoma Metropolitana – Iztapalapa

Upregulation of oxidative metabolism through HIF-1a is related to the high cytokine and chemokine levels in peripheral blood mononuclear cells from patients with pulmonary arterial hypertension
Rodrigo de Jesús López Velázquez. Escuela de Medicina y Ciencias de la Salud, ITESM

Cannabidiol modulates via PPARγ endocrine activity of white-like adipocytes
Omar Lozano García. Escuela de Medicina y Ciencias de la Salud, ITESM

Muscle-type–specific mitochondrial stress responses reveal dissociation between function and structure in a murine cardiometabolic HFpEF model
Bianca Nieblas León. Escuela de Medicina y Ciencias de la Salud, ITESM

11:30 12:00 Coffee break

12:00 – 14:00 **Symposium ALACF**

Biophysical and bioenergetic alterations in skeletal and cardiac muscle in different pathological situations

Chair: Carmen Valdivia
University of Wisconsin

Mitochondrial adaptation mechanisms as a possible target of non-pharmacological interventions in the treatment of sarcopenic obesity
Andrea Del Campo
Facultad de Química y de Farmacia. Pontificia Universidad Católica de Chile

Postnatal Environment and Cardiovascular Adjustments in Hypertension
Luciana Venturini Rossoni
Institute of Biomedical Sciences, University of São Paulo

Alterations in cardiomyocyte ionic currents in maladaptive hypertrophy. Possible therapeutic strategies
Alejandro Aiello
Facultad de Ciencias Médicas, UNLP-CONICET

K2p Channels in cellular biophysics
Carlos Saldaña
Facultad de Ciencias Naturales, UAQ

14:00 – 16:00 Lunch

16:00 – 17:00 *Oral Session VII*

Mitochondrial Pathologies

Chair: Manuel Alejandro Vargas Vargas
Universidad Michoacana de San Nicolás de Hidalgo

Lysine hyperacetylation impairs the mitochondrial ATP synthase complex in the cardiometabolic HFpEF heart
Abraham Méndez Fernández. Biomedicina cardiovascular, ITESM

Cannabidiol prevents pathological cardiac hypertrophy via activation of PPARs and preservation of mitochondrial function
Carolina Alejandra Morales Ochoa. Biomedicina cardiovascular, ITESM

MCU inhibition via AAV9 transfection confers cardiac protection by maintaining mitochondrial health
Felipe de Jesús Salazar Ramírez. Escuela de Medicina y Ciencias de la Salud, ITESM

4H-benzo[d][1,3]oxazines inhibits Proliferation, Migration, and Invasion Cervical Cell Lines
Jesús Adrián López. Universidad Autónoma de Zacatecas

17:00 – 18:00 **Plenary Lecture**

LEAP-2 and cardiac dysfunction in MASLD: exploring the mitochondria–lipid droplet axis in metabolic steatotic liver disease

Valentina Parra
Universidad de Chile

Chair: Luis Alberto Luévano
Tecnológico de Monterrey

18:00 – 19:00 Closing Ceremony

20:30 – 1:30 Dinner and Farewell Party

POSTER SESSION

Tuesday October 28, 2025
17:00 – 19:00

ATP synthases, ATPases, Phosphatases y pyrophosphatases

1. *Gene context and location of the ζ subunit from the F_1F_O-ATP synthase of the different alphaproteobacterial*
   Francisco Guillermo Mendoza Hoffmann. Universidad Autónoma de Baja California
2. *Identification of F_1F_O ATP synthase from Phaeodactylum tricornutum*
   Rafael Suárez Torres. Instituto de Investigaciones Biomédicas. UNAM

Biophysics of channels and transporters

3. *Electrochemical Gradient and K^+ Transporters Shape Killer Sensitivity in Saccharomyces cerevisiae*
   Brenda Téllez de la Garza. Universidad Autónoma de Querétaro

Electron Transport systems

4. *STAT5 regulates mitochondrial energy metabolism genes in cervical cancer cells stimulated with Interleukin 2*
   Rubén Alejandro Fuentes Pascacio. FES - Zaragoza, UNAM
5. *Role of STAT3 on the regulation of mitochondrial activity in cervical cancer cells after IL-2 treatment*
   Rodrigo Rojas-Mercado. FES - Zaragoza, UNAM
6. *Insights in activation of COX1 mRNA translation in yeast mitochondria*
   Yolanda Margarita Camacho Villasana. Instituto de Fisiología Celular, UNAM
7. *Distinct roles of NAC and RAC complexes in mitochondrial protein import and cytosolic proteostasis*
   Ariann E. Mendoza Martínez. Instituto de Fisiología Celular, UNAM
8. *Allotopic expression of the COX2 gene lacking the sequence encoding the leader peptide*
   Miriam Vázquez Acevedo. Instituto de Fisiología Celular, UNAM
9. *Impact of respirasome organization on species-specific inhibition of complex III in human parasites*
   Anaiza Rico-Luna. Instituto de Investigaciones Biomédicas. UNAM

Lipids and membranes

10. *Interaction of Amphotericin B with Lipid Bilayers via Molecular Dynamics*
Miriam Merari García Ronces. Centro de Investigación en Ciencias. UAEM

11. *Lipid Antigen-Driven Activation of T γδ Cells via NPA-Containing Liposomes in a Murine Model of Lupus*
Edgar Iván Galarce Sosa. Escuela Nacional de Ciencias Biológicas, IPN

12. *Mitochondrial dynamics and mtROS regulate B cell differentiation in response to non-bilayer phospholipid arrangements*
Giovanna Berenice Barrera Aveleida. Escuela Nacional de Ciencias Biológicas, IPN

Mitochondria and diseases

13. *The unsaponifiable fraction of avocado oil improves non-alcoholic fatty liver disease, insulin resistance, and mitochondrial dysfunction in rats fed a high-fat, high-fructose diet*
Marcela González Montoya. Universidad Michoacana de San Nicolás de Hidalgo

14. *Effect of the unsaponifiable fraction of avocado oil on electron transport chain function in liver and kidney mitochondria of rats fed a high-fat and fructose diet*
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15. *Mitochondrial Dysfunction in Menopause: A Journey Through the Liver, Kidney, Muscle, Heart, and Brain*
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16. *ZmVDAC and ZmHXK4 are involved in the modulation of cell death during drought stress*
Sobeida Sánchez Nieto. Facultad de Química, UNAM

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María Concepción José Núñez. Instituto de Fisiología Celular, UNAM

20. *Neonatal neglect increases the risk of the cardiometabolic disorder in adulthood: a pre-clinical model*
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Structure and function of membrane proteins

21. *Predicting the entry routes of nuclear-encoded mitochondrial proteins into yeast mitochondria*
Diego González Halphen. Instituto de Fisiología Celular, UNAM

22. *Methanolic Extracts of Capsicum annuum var. Fascinatum (MECaF) Disrupt Calcium Homeostasis in MCF-7 Breast Cancer Cells*
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23. *Role of sodium-hydrogen exchanger 1 in cardiometabolic injury vs. pressure overload pure model*
Julieta Palomeque. Institute for Obesity Research. Tecnológico de Monterrey

24. *Evaluation of cellular changes and cell membrane-associated proteins (metalloproteinases and ABCB1) in etoposide and paclitaxel resistant cells*
Jesús Adrián López. Universidad Autónoma de Zacatecas

25. *New real-time method (Nar JJ) to measure the activity of membrane nitrate reductase (Nar) of denitrifying bacteria*
José J. García-Trejo. Facultad de Química. Universidad Nacional Autónoma de México

Physiochemistry and transport phenomena through membranes

26. *The use of thioflavin T to estimate the plasma membrane potential (PMP) in different yeast strains and the effect of pH*
Norma Silvia Sánchez S. Instituto de Fisiología Celular, UNAM

Free radicals and antioxidants.

27. *Administration of the aqueous extract of the aerial part of Eryngium carlinae and its combination with silver nanoparticles improves mitochondrial function in a model of type 2 diabetes*
Jenaro Lemus de la Cruz. Instituto de Investigaciones Químico Biológicas. UMSNH

28. *MERCs and regulation of sulforaphane-modulated ER stress in cardiomyocytes subjected to chemical hypoxia*
Gabriela Navarrete Anastasio. Instituto Nacional de Cardiología "Ignacio Chávez"

29. *Evaluation of Phaseolus vulgaris L. extracts in breast cancer cells*
Angelica Judith Granados López. Universidad Autónoma de Zacatecas

Regulation of energy metabolism.

30. *Evaluation of L-glutaminase activity in the extremophile yeast Rhodotorula mucilaginosa*
Paola I. Acosta-Valdelamar. Instituto de Fisiología Celular, UNAM

31. *Adenosine derivative compound IFC-305 reverses epithelial-mesenchymal transition induced by palmitic acid and TGF-β1 in HepG2 cells*
Irina Cardoso-Lezama. Instituto de Fisiología Celular, UNAM

32. *Bioenergetic disruptions in neutrophil-like differentiated HL-60 cells caused by high glucose culture*
Jorge Andrés Cazares-Preciado. Tecnológico de Monterrey

33. *Cannabidiol as an anti-inflammatory and mitochondrial function-protective therapy in cardiorenal syndrome*
Héctor Chapoy Villanueva. Tecnológico de Monterrey

34. *Wolbachia metabolism after incubation in an axenic medium*
Natalia Chiquete Félix. Instituto de Fisiología Celular, UNAM

35. *Effects of IFC-305 on mitochondrial function in the B-cell precursor acute lymphoblastic leukemia (pre-B ALL) cell line NALM-6*
Ana María Hernández Jiménez. Instituto de Fisiología Celular, UNAM

36. *An unexpected alliance: riboflavin production by Wolbachia in Saccharomyces cerevisiae*
Ofelia Alejandra Méndez Romero. Instituto de Fisiología Celular, UNAM

37. *Disrupted mitochondrial calcium handling impairs pancreatic β-cell function under palmitate-induced lipotoxic stress*
Nora Greys Zamora Benavides. Tecnológico de Monterrey

38. *The methylome transcriptional regulatory network activated by copper: impact on basal metabolism and energy production genes*
Mauricio Latorre. Universidad de O'Higgins, Chile

39. *Bioenergetic Architecture of a Decoupled Aquaponic System: Metagenomic Characterization of Microbial Flux and Nutrient Cycles*
Saulo E. Andrade-Rincón. Universidad Autónoma de Baja California

40. *Intermittent fasting improves cardiorenal function in a murine model of heart failure.*
Emanuel Adrián Guajardo-Correa. Tecnológico de Monterrey

41. *FKBP51 disrupts the insulin signaling pathway and impairs mitochondrial bioenergetics in HepG2 cells*
Rodrigo Troncoso. INTA. Universidad de Chile

Plenary Abstracts

Ciudad de Tilantongo

Fotografía en comunidades afromexicanas: documentación sociohistórica, diversidad pluricultural y memoria visual

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Universidad Autónoma "Benito Juárez" de Oaxaca (Oaxaca, México)
https://AbrahamNahon

La intención es dar a conocer la relevancia que tiene la fotografía como fuente documental e histórica para conocer diversas culturas en nuestro país, a partir de una investigación detallada que rastree imágenes del pasado así como significativas narraciones visuales de nuestro presente. En este caso, me concentraré en la investigación que he realizado sobre fotografías en comunidades afromexicanas, incluyendo especialmente comunidades de Oaxaca. Mostraré algunos proyectos visuales que escapan a la espectacularidad de los medios modernos, confrontando una narrativa identitaria totalizadora y homogénea —anclada a los estereotipos promovidos por esta sociedad del espectáculo, el exotismo y la superficialidad—, abriendo el campo visual a otras comunidades, rostros, posturas, elementos materiales, culturas populares, formas de vida y de persistencia. Se activan distintas perspectivas para construir la historia y la memoria en estos pueblos negros, reafirmando la importancia política de su representación visual en el reconocimiento de su identidad y en la lucha contra la discriminación.

Mitochondria in Pathobiology

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Mitochondria frequently contribute to disease. Mitochondrial calcium loading, iron uptake, and oxidative stress induce the mitochondrial permeability transition (MPT) and consequent mitochondrial swelling. Mitophagy removes such damaged mitochondria, but with greater stresses (ischemia/reperfusion, oxidative stress, acetaminophen hepatotoxicity), MPT onset induces both necrotic cell death due to ATP depletion and apoptosis activated by cytochrome *c* release after outer membrane rupture. Global suppression of mitochondrial metabolism can occur in various pathological settings. Closure of voltage-dependent anion channels (VDAC) in the outer membrane can account for mitochondrial metabolic suppression consistent with a role for VDAC as a dynamic regulator, or governator, of mitochondrial function. In alcohol-associated and metabolic dysfunction-associated steatohepatitis (ASH and MASH [formerly called nonalcoholic steatohepatitis or NASH]), aldehyde generation promotes VDAC closure. In addition, reversible mitochondrial depolarization (mtDepo) occurs, together with VDAC closure causes selective and more rapid detoxifying mitochondrial aldehyde oxidation. Chronically, however, mtDepo induces mitophagy with release of mitochondrial damage-associated molecular patterns (mtDAMPs) that produce inflammation and fibrosis. In proliferating cancer cells, a switch from electrogenic to non-electrogenic mitochondrial ATP/ADP exchange leads to lower cytosolic ATP/ADP ratios and stimulation of glycolysis as the molecular basis for the Warburg phenomenon. Overall, mitochondrial alterations promote pathogenesis by a variety of different mechanisms.

Calorie restriction and aging: the mitochondrial connection

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In humans, obesity is associated with increased incidence of a variety of age-related diseases. Similarly, laboratory rodent lifespans are limited by obesity, including overeating promoted by *ad libitum* access to standard chow diets. Indeed, a daily limitation of caloric intake (calorie restriction) has been widely shown to enhance lifespans and prevent age-related diseases in rodents. We will discuss the metabolic effects of caloric restriction, and show that mitochondrial bioenergetics, redox state, and calcium homeostasis are regulated by caloric restriction, with a impact on tissue stress responses.

Exploring the mitochondrial complexome: from protein complexes to nucleoprotein assemblies.

Alfredo Cabrera-Orefice[1]

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Mitochondria host a highly organized network of multiprotein complexes essential for energy production, metabolic regulation and gene expression. Understanding the composition, assembly and interactions of these complexes, i.e., the mitochondrial complexome, is fundamental to deciphering mitochondrial function in health and disease.

Over the past decade, complexome profiling (CP) has emerged as a powerful strategy for mapping the landscape of mitochondrial protein complexes [1]. By integrating native separation techniques, quantitative mass spectrometry and computational clustering, CP enables systematic exploration of protein assemblies in diverse biological systems. Our research combines CP with complementary methods (e.g., cross-linking mass spectrometry and SILAC) as a comprehensive strategy to map mitochondrial protein assemblies, track their dynamics and identify new players in their biogenesis. These integrative approaches have broadened our understanding of mitochondrial proteostasis and the molecular basis of mitochondrial disorders.

Focusing on mitochondrial complex I (CI), the largest and most intricate enzyme of the respiratory chain, we have investigated its modular assembly, the role of accessory subunits, and the involvement of species-specific assembly factors. Using models such as *Yarrowia lipolytica*, mammalian tissues, cultured cells, and patient-derived fibroblasts, we uncovered novel assembly factors, characterized key intermediates and shed light on maintenance mechanisms such as subunit turnover and repair of the holoenzyme.

We have recently expanded CP to study mitochondrial nucleoprotein assemblies by optimizing workflows that preserve DNA- and RNA-associated interactions [2]. These adaptations enable comprehensive profiling of both protein and nucleic acid-bound complexes within a single experiment, offering a broader view of mitochondrial organization.

CP has matured into a versatile tool, advancing our understanding of mitochondrial biology and providing unique opportunities to unravel the dynamic architecture of protein complexes and their regulatory networks. As the complexome field evolves, CP will remain at the forefront of dissecting cellular complexity and elucidating molecular mechanisms.

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[2] Potter A, Cabrera-Orefice A, Spelbrink JN. Let's make it clear: systematic exploration of mitochondrial DNA- and RNA-protein complexes by complexome profiling. *Nucleic Acids Res*. 2023 Oct 27;51(19):10619-10641. doi: 10.1093/nar/gkad697.

Evolutionary Remodeling of Respiration and Photosynthesis in *Euglena gracilis*

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Euglena gracilis is a photosynthetic unicellular eukaryote from the Euglenozoa clade, related to trypanosomes but distinguished by its secondary green plastid acquired through endosymbiosis. This complex evolutionary history has driven distinctive remodeling of both respiratory and photosynthetic systems, resulting in unique bioenergetic strategies.

Mitochondrial oxidative phosphorylation in *E. gracilis* is mediated by canonical complexes I–IV and ATP synthase (complex V), but with extensive divergence. Proteomic and structural analyses have identified over 40 Euglenozoa-specific subunits in addition to ~50 conserved eukaryotic components. While maintaining overall architectural similarities to classical oxidases, cryo-EM revealed lineage-specific features, including an extended peripheral arm in complex I, a helmet-like domain in complex IV, and a modified membrane domain and peripheral stalk in complex V.

Photosynthesis in *E. gracilis* is equally divergent. Both PSI and PSII lack several canonical core subunits and incorporate a unique LHCE antenna family that binds red-shifted chlorophyll a. Cryo-EM analysis showed a PSI–LHC supercomplex with a reduced core surrounded by 14 LHCE and 2 LHCBM proteins. Notably, the PSI subunit PSAD was acquired via lateral gene transfer from cyanobacteria and is essential for antenna integration. Functional knockouts of genes such as *CAO* (chlorophyll b biosynthesis) and *CP29* (antenna stabilization) confirmed their role in light harvesting and photoprotection under high light.

Respiration and photosynthesis in *E. gracilis* are tightly coupled, with trophic conditions modulating their interaction. In mixotrophy, this coupling is regulated by chloroplast redox state, while in photoautotrophy it is influenced by CO_2 limitation and photorespiration. Mitochondria–chloroplast contact sites suggest strong metabolic integration. Under dark anoxia, *E. gracilis* maintains photosynthetic activity through PSI cyclic electron flow, likely supported by fermentation pathways such as wax ester fermentation, despite the absence of hydrogenase activity.

Application of Computational Methods to Understand the Function of Membrane Proteins

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Computational methods applied to investigate the structure and function of proteins have shown significant progress in recent years driven by the development of scientific software and computational infrastructure. Such development has allowed to study more complex biomolecular systems providing information at the molecular level that complements information from different experimental techniques. Here we discuss how computational methods could be applied to investigate not only the function of membrane proteins but also how molecular entities, including peptides, lipids, ions, and ligands modulate such function. In particular, two cases of membrane proteins will be discussed in detailed.

In the first case, the investigation of the ADAM17 and iRhom protein complex is going to be discussed with emphasis in the use of artificial intelligence methods to potentiate the protein characterization. In particular, the development of AlphaFold to facilitate the structure-function investigation of proteins will be underlined. ADAM17 is a well-known enzyme that controls the release of soluble pro-inflammatory cytokines including the Tumor Necrosis Factor alpha (TNF alpha).

In the second case, the investigation of the interacting mode of the PI(4,5)P2 lipid at the TMEM16A channel will be presented. TMEM16A is a calcium-activated chloride channel involved in important physiological processes including neuronal excitation. Molecular details of the chloride permeation along the pore channel will be also discussed in the context of experimentally known information.

Cardiac Ryanodine Receptor Channelopathies: From Membrane Bioenergetics to Clinical Phenotypes

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The cardiac ryanodine receptor (RyR2) is a massive calcium release channel of the sarcoplasmic reticulum, central to the bioenergetics of excitation-contraction coupling. By gating the flux of Ca^{2+} across an intracellular membrane system, RyR2 links electrical activity to the energetic demands of contraction. Precise control of this channel is therefore critical not only for rhythm and contractility, but also for preserving mitochondrial function and overall cellular homeostasis.

Mutations in *RYR2* destabilize this delicate balance, producing a heterogeneous group of disorders now recognized as *cardiac ryanodinopathies*. The best-characterized of these is catecholaminergic polymorphic ventricular tachycardia (CPVT), where gain-of-function variants create diastolic Ca^{2+} leak, delayed afterdepolarizations, and triggered arrhythmias in structurally normal hearts. More recently, loss-of-function mutations have been identified, giving rise to calcium release deficiency syndrome (CRDS), a condition that reveals the dual requirement for both sufficient and restrained Ca^{2+} flux across the sarcoplasmic reticulum membrane.

Beyond CPVT and CRDS, *RYR2* mutations have been implicated in arrhythmogenic right ventricular cardiomyopathy (ARVC) and long QT syndrome (LQTS), expanding the phenotype beyond pure arrhythmia to include membrane remodeling and metabolic stress. In these contexts, chronic Ca^{2+} dysregulation appears to drive structural changes, activate cell death pathways, and perturb cellular energy balance.

A major challenge in the field remains the unpredictable expressivity of RYR2 variants: the same mutation may produce divergent outcomes depending on genetic background, environment, or gene dosage. This variability underscores the need for integrative approaches that combine structural, biophysical, and bioenergetic perspectives to inform diagnosis, risk stratification, and therapeutic strategies. By examining RyR2 through the lens of bioenergetics and biomembrane physiology, we gain a deeper understanding of how disruption at the molecular level can resonate outward to shape the clinical spectrum of human disease.

LEAP-2 and Cardiac Dysfunction in MASLD: Exploring the Mitochondria–Lipid Droplet Axis in Metabolic Steatotic Liver Disease.

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Cardiovascular diseases (CVDs) remain the leading cause of death worldwide, with mitochondrial dysfunction recognized as a central mechanism driving cardiomyocyte hypertrophy, impaired energetics, and heart failure. Our group has shown how mitochondrial dynamics and function determine the balance between physiological and pathological remodeling, highlighting mitochondria as nodes not only for ATP production but also for signaling and organelle crosstalk shaping cardiomyocyte fate. In hepatocytes, fatty acids remodel the mitochondria–lipid droplet (LD) axis, influencing whether mitochondria channel energy toward ATP synthesis or LD expansion. This crosstalk has emerged as a determinant of lipid handling, oxidative stress, and organelle fitness, suggesting that its dysregulation contributes to systemic disease. Metabolic dysfunction-associated steatotic liver disease (MASLD), the most common chronic liver disorder, provides a context where these mechanisms are particularly relevant. Beyond lipid accumulation, MASLD alters hepatokine secretion that systemically influences cardiac metabolism and function. Among them, liver-expressed antimicrobial peptide 2 (LEAP-2), an endogenous antagonist of ghrelin, is markedly elevated in MASLD and may act as a mediator linking liver metabolism to cardiovascular dysfunction. Although evidence on its cardiac role is limited, our recent findings suggest that LEAP-2 modulates lipid metabolism and mitochondrial dynamics in cardiomyocytes, promoting mitochondrial biogenesis but reducing membrane potential and counteracting ghrelin's protective effects against lipid overload. Altogether, disruption of the mitochondria–LD axis in MASLD, combined with elevated hepatokines such as LEAP-2, may help explain the increased cardiovascular risk in metabolic liver disease and highlight this axis as a promising therapeutic target.
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Symposia Abstracts

Escritura zapoteca

Young Career Researcher's

Light harvesting regulation and photodamage interplay in Chlamydomonas reinhardtii

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Photosynthesis is a critical process to all life on Earth, as it allows conversion of light energy and CO_2 into biologically-available compounds. Its integration in the metabolism of the cell and the variable nature of sunlight requires photosynthesis to be tighly regulated on various timescales. The faliure to do so will result in transient or prolonged efficiency losses.

In our group, we investigate the regulatory processes in photosynthesis, in particular concerning excitation energy transfer and electron flow. One of such mechanisms, of a particularly large extent in the green microalga Chlamydomonas reinhardtii, is state transitions. This process of antenna exchange between Photosystem I and II (PSII), upon various metabolic and light stimuli allows to balance light harvesting in the cells. I will present a combination of functional and ultrastructural data from a collaborative effort aiming at a description of the memebrane-scale changes upon state transitions.

Under extensive high light conditions this and other regulatory processes fail, leading to photodamage. Particularily concerning PSII, photoinhibition is a conserved process that is exacerbated by environmental stresses and limits photosynthesis; however, its molecular origins are still poorly understood. I will present methodologies developped in our group which aim at quantifying photoinhibition across photosynthetic phyla.

The physiology of methane-producing archaea under stress

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Methane-producing microorganisms from the Domain Archaea (methanogenic archaea) are strict anaerobes with highly specialized metabolisms and produce methane as an end-product of their oxygen-independent respiration. All methanogens produce methane through methanogenesis, their primary energy conservation pathway, making them significant contributors to greenhouse gas emissions during anaerobic digestion of nutrients and, in turn, influencing Earth's climate and biogeochemical processes. Methanogenic archaea are free-living organisms that typically live in sediments in oxygen-depleted environments; however, few species have recently been found in host-associated microbiomes, such as the gut, skin, mouth, and respiratory tract of humans and other animals. But what is the role of methanogenic archaea in the health of humans, animals, and ecosystems (One Health)? How do methanogenic archaea survive the stressful conditions present in their habitats? These are the main questions that have motivated us to investigate the physiology of methanogenic archaea in our Microbial Ecophysiology Laboratory at UConn. We use our expertise in microbial physiology, genetics, and biochemistry to study the biology of methanogenic archaea, a group of specialized microorganisms underrepresented in experimental research, to study the mechanisms that regulate cellular processes in methanogenic archaea and their interactions with ecosystems, microbiomes, and hosts. During my talk, I will present results that address our main research questions and contribute to our understanding of how methanogenic archaea conserve energy under optimal and stressful conditions, as well as their relevance to One-Health.

ALACF

Biophysical and bioenergetic alterations in skeletal and cardiac muscle in different pathological situations

Alterations in cardiomyocyte ionic currents in maladaptive hypertrophy. Possible therapeutic strategies.

Alejandro Aiello

Centro de Investigaciones Cardiovasculares “Dr. Horacio Cingolani”, Facultad de Ciencias Médicas, UNLP-CONICET, La Plata, Argentina.

The development of several cardiac pathologies is closely associated with maladaptive cardiac hypertrophy (MCH), particularly those resulting from persistent hemodynamic overload. The most common conditions include hypertension, ischemic cardiomyopathy, and valvular diseases. If untreated, MCH leads to thickening of the left ventricular wall, resulting in increased cardiac mass and/or ventricular dilation. MCH is characterized by fibrosis, cell death, dysregulation of calcium handling, metabolic reprogramming toward glucose utilization, re-expression of fetal genes (such as ANP and BNP), and alterations in sarcolemma structure, among other changes. Collectively, these processes favor the progression to heart failure and malignant arrhythmias, ultimately predisposing to sudden death. From an electrophysiological perspective, MCH is associated with altered activity of ion channels, leading to prolongation of the cardiac action potential (AP). Among the channels affected are the L-type calcium current (I_{CaL}) and various potassium currents. Consistently, we observed AP prolongation together with a reduction in I_{CaL} and in the inward rectifier potassium current (I_{K1}) in 12-week-old male C57 mice subjected to transverse aortic constriction (TAC) with a titanium clip to induce hemodynamic overload. The G protein-coupled estrogen receptor (GPER) has emerged as a potential therapeutic target for the prevention of MCH. Selective activation of GPER by its synthetic agonist G1 has been shown to prevent and even regress MCH in cardiac tissue. However, the underlying mechanisms of these cardioprotective effects remain incompletely understood. Therefore, the aim of our study was to investigate the role of GPER in cardiac electrical activity. We found that selective activation of GPER by G1 decreased action potential duration (APD) and increased resting membrane potential (RMP) in both Sham and TAC groups. Moreover, in hypertrophic myocytes, G1 reduced I_{CaL} while enhancing I_{K1} and the transient outward potassium current (I_{to}), partially explaining the cardioprotective changes in AP. In conclusion, our findings suggest that selective activation of GPER exerts non-genomic effects in hypertrophic hearts, mediated in part by decreased I_{CaL} and increased potassium currents. These electrophysiological changes may contribute to the cardioprotective role of GPER activation.

Mitochondrial adaptation mechanisms as a possible target of non-pharmacological interventions in the treatment of sarcopenic obesity.

Briones-Manríquez, Fernanda; Ibarra-Barahona, Iván; Almarza, Gonzalo; Araya, María Jesus; Luz-Crawford, Patricia; Pérez-Leighton, Claudio; **del Campo, Andrea.**

Sarcopenic obesity describes the confluence of low muscle mass and/or strength with obesity in older adults. Despite the remarkable similarity between the onset of sarcopenia and obesity, little is known about the underlying mechanistic drivers of this accelerated aging of skeletal muscle. Mitochondrial network plays an essential role in skeletal muscle and may be regulated by adaptive mechanism such as mitochondrial unfolded protein response and mitophagy. C57Bl6 mice were submitted to high fat diet for 12 weeks. Exercise or time-restricted feeding were the two non-pharmacological interventions done, each on its own. Measurements of physical and physiological parameters were done together with mitochondrial protein level detection by western blot, histology and macrophage detection by flow cytometry. Our results show mitochondrial modifications before the appearance of clinical symptoms in sarcopenic obesity, which can be reversed by exercise upon increase of UPRmt, whereby time restricted feeding can also revert muscle function decline probably through mitophagy regulation.

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K2p Channels in Cellular Biophysics

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Two-pore domain potassium (K2P) channels constitute a diverse family of background K^+ channels that regulate membrane potential and cellular excitability across a wide range of tissues in both vertebrates and invertebrates. These channels are characterized by their structural motif of four transmembrane domains and two pore-forming regions per subunit, functioning typically as dimers. K2P channels are involved in processes such as neuroprotection, cardiac rhythm control, and anesthesia response, and are regulated by factors including mechanical stretch, temperature, pH, and lipid signaling molecules. Among the yeast K2P-like channels, the TOK1 (tandem outward rectifying K^+ channel) represents a unique eukaryotic K^+ channel composed of eight transmembrane segments and two pore regions, structurally homologous to mammalian K2P channels. TOK1 functions as an outward rectifier, allowing K^+ efflux and contributing to the regulation of membrane potential in *Saccharomyces cerevisiae*. A particularly interesting biological interaction involves TOK1 and the *K*1 *killer* toxin, a protein secreted by certain strains of *S. cerevisiae* that confers competitive advantage by targeting sensitive yeast cells. The *K*1 toxin binds to the β-glycan receptor on the cell wall and subsequently interacts with membrane components, including the TOK1 channel. This interaction facilitates uncontrolled K^+ efflux from the target cell, leading to membrane depolarization, loss of cytoplasmic content, and ultimately cell death. Importantly, cells that produce the *K*1 toxin express an immunity protein that prevents the toxin's action by inhibiting the TOK1 channel from the cytoplasmic side, thus protecting the producer cell from self-destruction. This interaction highlights the dual role of TOK1 as both a physiological potassium channel and a molecular target in toxin-mediated microbial competition, underscoring its importance in yeast ecology and as a model for studying ion channel–toxin interactions.

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Postnatal Environment and Cardiovascular Adjustments in Hypertension.

Luciana Venturini Rossoni.

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Hypertension is a major global public health challenge, affecting more than one billion people worldwide. Often referred to as the silent killer, it typically presents with no symptoms, but significantly increases the risk of heart disease, stroke, and kidney failure. Hypertension is a multifactorial disease in which both prenatal and postnatal periods play a critical role in the development of high blood pressure levels. Adverse conditions during fetal life, such as poor maternal nutrition, stress, or placental insufficiency, can lead to permanent changes in the structure and function of the cardiovascular system, a process known as fetal programming. In addition, after birth, environmental factors like early-life nutrition, exposure to stress, and physical inactivity can also further influence cardiovascular regulation and exacerbate the risk of developing hypertension later in life. Some researchers have demonstrated that adult spontaneously hypertensive rats (SHR), a classical model of primary hypertension, breastfed by normotensive mothers, exhibit a 26 mmHg reduction in blood pressure compared to naturally reared SHR and have related the association between the genetic background and maternal postnatal environment to the development of hypertension. At this moment, the blood pressure adjustment induced by cross-fostering was associated with the delivery of milk and sodium balance. However, the mechanism by which postnatal cross-fostering reduces blood pressure and impacts the cardiovascular system in SHR remains unclear. In recent years, our laboratory has focused on the role of the maternal environment and the influence of cross-fostering on the cardiovascular system in SHR. In this context, the talk will explore the impact of normotensive and hypertensive postnatal environments on the structure, composition, and functional capacity of the intestinal microbiota, as well as on the cardiovascular system's structure and function.

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Oral Abstracts

Códice Vindobonensis. Cultura mixteca

Far-Red Component Enhances Paramylon Production in Photoautotrophic *Euglena gracilis*

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Recently, microalgae have gained significant biotechnological importance as sustainable source of various metabolites. Among this group of organisms, *Euglena gracilis*, a secondary green flagellate, stands out by its ability to grow photoautotrophically, heterotrophically and mixotrophically. Its reserve polysaccharide, paramylon, becomes important by its diverse applications in biomedicine and pharmaceuticals. Paramylon production in this species can represent up to 80% of its dry weight under heterotrophic conditions. Previously, it has been shown that mixed red and blue light ratios affects biomass and paramylon production, during photoautotrophic growth [1].

To further investigate the effects of varying illumination, we designed and built a modular photobioreactor that allowed us to evaluate simultaneously the photoautotrophic growth of *E. gracilis* under twelve different light conditions: seven single spectrum lights (Ultraviolet, Royal Blue, Blue, Green, Red, Far-Red, and Infrared) and five composite spectra lights (3,000K, 10,000K, 30,000K white lights, Amber, and "Full-spectrum"). 24-day growing kinetics were recorded, and growth parameters calculated for each light regime. Additionally, pigment composition, photosystem II oxygen evolution, and paramylon production were determined under each light condition. The designed *Ankaa* photobioreactor enabled us to follow the adaptation of *E. gracilis* to several specific light regimes. Our results suggest a differential paramylon production profile that is strongly dependent on illumination and our findings reinforce the fact that far-red component enhances paramylon production [2].

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Study of the function of Pet494 in Cox3 biogenesis in mitochondria of *Saccharomyces cerevisiae*

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Abstract

Cytochrome *c* oxidase or complex IV is the terminal complex of the respiratory chain. It is an integral complex localized in the mitochondrial inner membrane, with one side facing the mitochondrial matrix (MM) and the other facing the intermembrane space (IMS). It oxidizes cytochrome *c,* which is previously reduced during the Q cycle in complex III. Complex IV also reduces molecular oxygen to generate H_2O , a byproduct of cellular respiration.
In *Saccharomyces cerevisiae* complex IV is formed by twelve protein subunits, three of which are encoded in the mitochondrial DNA (Cox1, Cox2, and Cox3) and nine in the nucleus (Cox4-9, Cox12, Cox13, and Cox26).

The mitochondrial mRNAs lack the elements required for the initiation of translation commonly found in nuclear mRNA in eukaryotes and prokaryotes such as the Shine-Dalgarno and Kozak sequence, which facilitate ribosome positioning and translation initiation through the recognition of a consensus sequence. Alternatively, mitochondrial translation initiation is facilitated by translational activators.

Translational activators are proteins that act on their own or in complexes on the 5' untranslated region (5'UTR) of the mitochondrial mRNA. Each mitochondrial transcript has its own set of translation activators. Even though the molecular mechanism of translational initiation is not yet fully understood, it is believed that translational activators are key to stabilize mitochondrial mRNA and facilitate proper positioning of the mitochondrial ribosome.

One of these translational activators is Pet494, a nuclearly encoded protein which, together with Pet54 and Pet122, activates the translation of *COX3* mRNA. We have evidence in our laboratory suggesting that Pet494 might potentially have an additional function as an assembly chaperone for the Cox3 peptide. For this reason, we decide to explore this potential alternate function.

CONTRIBUTION OF SPHINGOLIPIDS TO THE REGULATION OF PLASMA MEMBRANE H^+-ATPase ACTIVITY IN *ARABIDOPSIS*

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The activity of the plasma membrane (PM) H^+-ATPase in plants is highly regulated. It has been proposed that the lipid environment can modulate its activity, either through specific lipid-protein interactions or more generally through the physical properties of the bilayer. In our research group, using *Arabidopsis* plants with modified levels of sphingolipids in the PM (*lcb2b hp / lcb2a (+)*), a nearly two-fold increase in H^+-ATPase activity was found compared to the wild-type. The objective of this work was to identify the sphingolipid species that could be involved in this increased activity, using plants that express a different sphingolipid content than the wild-type genotype. To this end, PMs were isolated from the leaves of *Arabidopsis* genotype *lcb2b hp / lcb2a (+)*, the wild-type (wt), and, as controls, the *lcb2a-1 (-)*, *lcb2a-1 (+)*, and *lcb2b hp / lcb2a (-)* genotypes. The PM sphingolipids were extracted according to Markham and Jaworski (Rapid Commun Mass Spect 2007, 21, 1304), and the separation and identification of the species were performed by HPLC/ESI-MS/MS. Based on the sphingolipid quantities, univariate and multivariate comparative analyses were conducted to determine the diversity, abundance, and predominance of the species in the plants with higher H^+-ATPase activity. The sphingolipidome analysis revealed 101 sphingolipid species (27 ceramides [Cers], 24 hydroxyceramides [hCers], 19 glucosylceramides [GlcCers], and 31 glycosylinositol phosphoceramides [GIPCs]). Principal Component Analysis (PCA) and a heat map showed that the PMs of *lcb2b hp / lcb2a (+)* plants exhibited a specific and distinct sphingolipid profile compared to the wt and control plants. The analysis of the total contents of the hydrophobic moiety of sphingolipids in the PMs of *lcb2b hp/lcb2a (+)* demonstrated that these membranes have a lower amount of unsaturated long-chain bases (LCBs) and a higher content of long-chain fatty acids (LCFAs) compared to the control and wt plants. To associate specific sphingolipid species with the increase in enzyme activity, an ANOVA and post hoc analysis was performed on the 101 detected species to find those that showed changes in their content only in the *lcb2b hp / lcb2a (+)* plants compared to the wt and controls. In the PMs of *lcb2b hp / lcb2a (+)* plants, 11 sphingolipid species were identified: 2 Cers, 2 hCers, and 7 GIPCs whose contents showed significant differences with respect to the wt and control plants. Considering their abundance, the identified Cers and hCers accounted for less than 0.4% of the total sphingolipid content. Therefore, these species could be associated with the higher enzyme activity, perhaps through specific sphingolipid-H^+-ATPase interactions. In the case of the seven GIPC species, which represented approximately 25 mol% of the total sphingolipid content, this abundance could influence the physical properties of the PM, leading to the increase in enzyme activity.

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Kinetic characterization and polypeptide composition of the dimer and monomer of the F_1F_O-ATP synthase from *Yarrowia lipolytica*

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F_1F_O-ATP synthase produces adenosine triphosphate (ATP), the universal energy currency in the cell, from adenosine diphosphate and inorganic phosphate by rotary catalysis, a process common to all forms of life (Kühlbrandt, 2019). The ATP synthase can be found in a monomer (V_1) as well as a dimer (V_2); the last one has an important role in the mitochondrial cristae architecture. The kinetic characterization of V_2 from *Ustilago maydis* and *Polytomella sp* suggests that the interface monomer-monomer has an important role in the activity of the oligomer (Esparza-Perusquía *et al.,* 2017; Villavicencio-Queijeiro *et al.,* 2015). To achieve a more comprehensive insight into the function of the V_2, we isolated the V_1 and V_2 of the F_1F_O-ATP synthase from *Yarrowia lipolytica*, a strictly aerobic yeast (Nicaud, 2012) that depends on oxidative phosphorylation for its supply of ATP. The V_2 was solubilized with digitonin and V_1 was solubilized with dodecyl maltoside, and their ATPase activity was performed as described (Esparza-Perusquía *et al.,* 2017). Interestingly, analysis of the supercomplexes by BN-PAGE showed that upper than 95% of the F_1F_O-ATP synthase amount was present as V_2, while V_1 was scarce; this suggested that the crista architecture could principally be tubular. Preliminary, our results showed a value of a V_{max} of 0.7038 ± 0.01 µmol ATP hydrolyzed·min^{-1}·mg protein^{-1} and a K_m of 363 ± 32.7 µM for V_2. In the presence of dodecyl maltoside, the V_2 showed V_{max} and K_m values of 2.57 ± 0.05 µmol ATP hydrolyzed·min^{-1}·mg protein^{-1}, and 411.2 ± 47.7 µM, respectively. The V_1 showed a V_{max} of 0.1154 µmol ATP hydrolyzed·min^{-1}·mg protein^{-1} and a K_m of 748 ± 95.9 µM. In the presence of dodecyl maltoside, the V_1 showed V_{max} and K_m values of 0.1094 µmol ATP hydrolyzed·min^{-1}·mg protein^{-1}, and 310 ± 17.8 µM, respectively. The V_2 and V_1 samples were analyzed by MS/MS tandem spectrometry. V_1 showed the complete set of F_1F_O-ATP synthase subunits; the V_2 showed the dimer-specific subunits *e*, *g* and *k*. Additionally, the mitochondrial ADP/ATP carrier and phosphate carrier (PIC) were joined to both oligomeric states. Also, ATP synthase regulation protein NCA2, and chaperones ATP11 and ATP10 were associated with both oligomers.

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Ancient respirasome: Kinetics and polypeptide composition

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Respiratory chain supercomplexes have been isolated from mammalian and yeast mitochondria, and bacterial membranes. Bacterial supercomplexes are characterized by their relatively high detergent-stability compared to yeast or mammalian supercomplexes. The mobility of substrate cytochrome c increases in the order bacterial, yeast, and mammalian respiratory chain. The Electron Transport Chain of *P. denitrificans* is constituted by the NADH-Q oxidoreductase (complex I), the succinate-Q oxidoreductase (complex II), the QH_2-cytochrome c oxidoreductase (complex III), and the cytochrome c oxidase (complex IV); all of them are embedded in its plasma membrane with their catalytic regions oriented to the cytoplasm (John *et al.,* 1977). Because these complexes are constituted by a few polypeptide chains, they are considered as a simplified version of those present in eukaryotes; however, they can use NADH or succinate as high-potential electron donors to carry out the proton electron gradient, which is used by F_1F_O-ATP synthase to produce ATP. In our laboratory we are interested in the study of supercomplexes, particularly the respirasome which is constituted by the complexes I, III_2, and IV, which oxidizes NADH and reduce oxygen. In this case, isolated the different supercomplexes from *P. denitrificans* and selected only the respiratory complexes activity upper 1600 kDa. This “megacomplex” is a stable interaction between I, III_2 and IV. The NADH dehydrogenase activity from the *P. denitrificans* respirasome was inhibited by rotenone, antimycin or cyanide, even if coenzyme Q or cytochrome c are added, indicating a functional intercommunication between them. Mass spectrometry showed the 14 canonical subunits of complex I, in addition to the canonical ones of complex III and IV (manuscript in progress).

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Mitochondrial ROS induces lysosomal dysfunction impairing autophagic flux in human cells carrying the APOE4 allele

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Background: The ApoE4 allele is a significant genetic risk factor for late-onset Alzheimer's disease (AD) and decreased longevity. Increased brain activity has been observed in asymptomatic young carriers of ApoE4, potentially linked to enhanced glycolysis. However, cells expressing ApoE4 fail to adapt their metabolic status in response to high-energy demands, suggesting a lack of metabolic plasticity. Consequently, a reduction in glycolysis later in life may initiate pathogenesis by revealing deficiencies in other metabolic pathways, such as OXPHOS. Mitochondrial dysfunction is frequently observed with autophagic/lysosomal function changes, suggesting these may be related processes. Studies using human tissue from AD-ApoE4 patients demonstrate a blockage of the autophagy flux and a decrease in the level of the lysosomal autophagy transcripts. Nevertheless, it remains unclear how autophagosome clearance is linked to mitochondrial deficiency associated with ApoE variants.

Methods: Adult human fibroblasts from ApoE4 heterozygotes, AD patients, and ApoE3 homozygotes were analyzed by RT-qPCR to evaluate changes in mRNA transcripts related to mitochondrial biogenesis. Samples were also processed for TEM to assess mitochondrial morphology. Autophagy flux was examined using cell biology and biochemical approaches, which included the mCherry-GFP LC3 sensor and WB analysis to detect LC3 and p62 proteins. Specifically, mitophagy was displayed by IF and immunoblot techniques. To determine whether ApoE4-induced autophagic flux suppresses lysosomal degradation, we performed a DQ-BSA dequenching analysis, measured LAMP2 protein levels, and examined lysosome clustering. The bioenergetic profile was determined using the Seahorse technology and a genetically encoded H_2O_2 sensor. Finally, the impact of mitochondria-targeted antioxidants was evaluated through time-resolved confocal fluorescence microscopy with MitoSOX, and the turnover of autophagy-lysosome fusion was described.

Results: In Apo4 carriers, deficiencies in the mitochondrial respiratory chain led to increased mitochondrial mass and biogenesis, as evidenced by elevated levels of the PGC-1α transcript and mitochondrial DNA copy number. Additionally, the number of mitochondria colocalizing with the punctate pattern of LC3 decreased, indicating

reduced mitophagy. This observation was confirmed by the lack of significant changes in LC3-II and TOMM20 protein levels after treating ApoE4 cells with bafilomycin. These abnormalities of mitochondria were associated with an increase in mitochondrial ROS, which could be abolished by selective inhibitors of mitochondrial superoxide production from complexes I and III, S1QEL, and S3QEL, respectively. Moreover, reducing mitochondrial ROS production significantly restored lysosomal proteolytic capacity, prevented autophagic flux blockage, and enhanced basal respiration mitochondrial respiration in ApoE4 cells. Finally, forcing fibroblasts to rely on OXPHOS while chronically inhibiting mitochondrial complexes, I and III revealed that ApoE4 cells do not depend on OXPHOS for survival. This suggests that the alkalinization of lysosomes is due to elevated levels of mitochondrial superoxide, that suppressed mitophagy may be due to low energy availability, with cells actively utilizing alternative survival pathways.

Conclusions: Our results show that mitochondrial ROS induces a lysosomal dysfunction related to the ApoE4 allele. The combined alterations in mitochondrial function and lysosomal activity outlined in our study may be critical in the development of neurodegenerative diseases. Thus, we provide novel insights into the etiology of AD associated with the ApoE gene.

The NDi1 starts the electron flux in the *Saccharomyces cerevisiae* respirasome.

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During evolution, *Saccharomyces cerevisiae* discarded mitochondrial complex I, but retained three rotenone-insensitive NADH dehydrogenases: two on the external (Nde1 and Nde2) and one on the internal (Ndi1) leaf of the inner mitochondrial membrane. Previously, we reported the presence of a supercomplex in *S. cerevisiae* constituted by the Ndi1 and complexes III_2 and IV. Here, the respirasomes from WT and NDE1Δ/NDE2Δ strains were isolated, and their activities and protein composition were characterized. Mass spectrometry of respirasomes from WT and NDE1Δ/NDE2Δ strains showed the canonical subunits of complex III, IV, and the Ndi1, showing the minimal composition of the respirasome. To confirm the respirasome function, its activity was characterized. Kinetic characterization of NADH:DBQ oxidoreductase activity from respirasomes, as well as free Ndi1, showed V_{max} values of 16 ± 0.2, 20 ± 0.4, and 14 ± 0.3 μmol NADH oxidized·min^{-1}·mg^{-1} for WT respirasome, NDE1Δ/NDE2Δ respirasome, and free Ndi1, respectively. The kinetic model for WT and NDE1Δ/NDE2Δ respirasome was a Ping Pong Bi-Bi mechanism with two different stable enzyme forms, free (E) and modified enzyme (F); while the free Ndi1 exhibited a Random Bi-Bi mechanism with the ternary complex NADH-Ndi1-ubiquinone. This suggests that the interaction of Ndi1 with complexes III_2 and IV in the respirasome modifies its kinetic mechanism. Oxygen consumption values were 6.7 ± 1.4 and 8 ± 1.6 μmol O_2·min^{-1}·mg^{-1} for WT and NDE1Δ/NDE2Δ respirasomes, respectively. The values for NADH:O_2 activity were 2.4 ± 1.4 and 2.5 ± 1.6 for WT and NDE1Δ/NDE2Δ respirasomes, respectively, suggesting that electron flux from NADH to oxygen occurs in the *S. cerevisiae* respirasome. The electron transfer from NADH to oxygen was inhibited by flavone, antimycin A, or cyanide, but the Ndi1 activity was insensitive to antimycin A or cyanide, indicating that no codependence of respirasomal-NADH:DBQ oxidoreductase activity occurs as reported in *Ustilago maydis* respirasome. This result indicates that the activity of respirasomal Ndi1 may contribute to the quinol pool with no evidence of direct substrate channeling. This is the first evidence of the Ndi1 role as the electron input in the respirasome from *S. cerevisiae*.

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The protein import machinery in the colorless plastids of the chlorophycean alga *Polytomella parva*

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Over the course of evolution, the loss of photosynthetic capacity has occurred in several lineages of plants and algae. *Polytomella parva* is a unicellular alga evolutionarily related to *Chlamydomonas reinhardtii*, that has lost the ability to carry out photosynthesis. *P. parva* is a suitable model to study the adaptation of organisms to an heterotrophic form of life[1]. *P. parva* carries remnants of an ancestral chloroplast, a colorless plastid also named amyloplast, that maintains several metabolic routes but whose main function is the synthesis and storage of starch. In addition, *P. parva* lacks a plastid genome, suggesting that amyloplast proteins cannot be synthesized inside the organelle and must be imported from the cytosol. In organelles of endosymbiont origin, such as plastids, a protein import machinery is required for their biogenesis and homeostasis. This machinery allows the internalization of proteins synthesized by cytosolic ribosomes into the organelle. In chloroplasts this import system consists mainly of the membrane complexes TOC-TIC and its associated motor Ycf2-FtsHi. TOC-TIC recognizes and translocates protein precursors, and Ycf2-FtsHi is the associated motor that enables translocation at the expense of ATP hydrolysis. In *C. reinhardtii*, the TOC-TIC and Ycf2-FtsHi complexes are made up of subunits encoded both in the plastidial and in the nuclear genome. Tic214 and Ycf2 are subunits of the TOC-TIC complex and of the motor respectively that are encoded in the chloroplast[2,3]. Based on the structure of TOC-TIC and Ycf2-FtsHi complexes in *C. reinhardtii*, orthologs of these complexes in *P. parva* were searched in the databases and compared by bioinformatic and proteomic analyses. So far we identified several putative components of TOC-TIC and Ycf2-FtsHi in *P. parva*. Our observations also suggest the absence of the Tic214 and the Ycf2 subunits in the colorless alga, two crucial proteins for the import of nucleus-encoded chloroplast components[4].

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Exploring the druggability of the binding sites of exogenous allosteric inhibitors of FOF1-ATP synthase

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In addition to its central role in mitochondria as the primary producer of ATP, F_OF_1-ATP synthase also performs several essential regulatory functions at the cell membrane. Dysregulation of this enzyme has been increasingly associated with a wide range of human disorders, including hypertension, atherosclerosis, cancer, and various neurodegenerative, autoimmune, and age-related diseases. Moreover, the inhibition of F_OF_1-ATP synthase compromises the viability of multiple bacterial pathogens of significant public health concern. As a result, this enzyme has emerged as a promising therapeutic target, both for treating human diseases and addressing the growing threat of antibiotic resistance [1,2]. In this study, we conducted a computational analysis of the binding sites for the exogenous aurovertin and polyphenol inhibitors within the bovine F1 subcomplex, which shares high sequence identity with the human enzyme [3]. Molecular dynamics simulations revealed that although these sites are largely preformed, inhibitor binding disrupts inter-subunit communication and induces long-range dynamic perturbations at the catalytic site. End-point binding free energy calculations identified key hot spot residues involved in stabilizing the recognition. These residues also contributed to stabilizing solvent-exposed regions identified via mixed-solvent molecular dynamics, which mimic inhibitor-enzyme interactions and may serve as pharmacophore constraints for virtual screening. To investigate the potential for designing species-specific inhibitors targeting the inhibitor binding sites, we performed free energy calculations using two bacterial F_1-ATP synthases with experimentally solved structures. Finally, a sequence conservation analysis was carried out to evaluate the conservation of the inhibitor binding sites among pathogen homologs. Together, our findings represent an initial step toward the structure-based design of novel allosteric inhibitors targeting exogenous binding sites in F_OF_1-ATP synthase.

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Gene expression modulation of SERCA3 in MCF-7 cells treated with the extract of *Capsicum annuum* L. var. Fascinato

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Calcium ions (Ca^{2+}) serve as essential second messengers in maintaining cellular homeostasis, orchestrating a myriad of physiological processes. Intracellular Ca^{2+} concentration is tightly regulated through a complex network of transporters, binding proteins, and enzymes that modulate its release, uptake, or storage across cellular compartments. Disruption of this finely tuned balance is associated with metabolic disorders, endoplasmic reticulum (ER) stress, dysregulated apoptosis, and cancer progression, as evidenced by numerous studies (Berridge et al., 2003; Monteith et al., 2007). Since 2009, our laboratory has focused on the spatial characterization of ER-resident Ca^{2+}-regulating proteins in MCF-7 breast cancer cells. Early investigations revealed a homogeneous distribution of IP_3 and ryanodine receptors across the ER, whereas SERCA, the only canonical ATPase responsible for Ca^{2+} reuptake into the ER, displayed a polarized localization. This asymmetry suggests a spatially restricted regulation of Ca^{2+} signaling, potentially impacting cellular proliferation and survival pathways, processes central to tumorigenesis. To explore novel therapeutic strategies, viability assays were performed using ethanolic extracts from three *Capsicum annuum L.* vars.: Baselga, Fascinato, and Orangela. Half Maximal Inhibitory Concentration (IC_{50}) values were determined to assess antiproliferative efficacy in MCF-7 cells. Fascinato exhibited the most promising profile, prompting further analysis of its mechanisms of action. Intracellular Ca^{2+} measurements demonstrated that the extracts induced Ca^{2+} mobilization in a dose-dependent manner, suggesting interference with ER Ca^{2+} dynamics. Transcriptomic profiling of Fascinato-treated MCF-7 cells revealed upregulation of *ATP2A3* (SERCA3), and differential expressions of several genes related to calcium handling, signal transduction, and transcriptional regulation. Notably, the affected pathways encompassed both direct Ca^{2+} regulators and secondary effectors involved in cell cycle and apoptosis. This systems-level approach underscores the therapeutic potential of plant-derived bioactives in modulating calcium homeostasis. Our findings support the hypothesis that targeted disruption of Ca^{2+} equilibrium via natural agents such as Fascinato may selectively compromise cancer cell viability. The modulation of SERCA3 and associated signaling networks highlights new avenues for developing adjuvant or alternative oncologic interventions.

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Cysteine Oxidation as a Regulator of the A Subunit of the Vacuolar ATPase in *Saccharomyces cerevisiae*

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The vacuolar ATPase (V-ATPase) is responsible for generating electrochemical and proton gradients across the membranes of the vacuolar system. The *VMA1* gene encodes the A subunit of V-ATPase, which contains a cysteine residue (C261) that is involved in the catalytic activity of the complex.

A connection has been established between the vacuole and the degradation of cysteine for the production of hydrogen sulfide (H_2S). Deletion of any of the subunits comprising the V_1 subcomplex of the V-ATPase leads to decreased H_2S production from cysteine. However, the exact mechanism by which V-ATPase regulates this process remains unclear. Oxidative stress can trigger post-translational modifications (PTMs) that oxidize specific amino acid residues such as cysteines. These modifications may alter protein structure and function, impacting their roles in various cellular processes. H_2S can counteract these effects through persulfidation, which reduces oxidized cysteines and restores protein function.

The main objective of this study is to elucidate how redox conditions regulate the activity of the vacuolar ATPase, and to determine whether H_2S production mediated by *CYS4* plays a role in this regulation. Through the purification of vacuoles from the *wt* and *Δcys4* strains, the activity of the V-ATPase was assessed, and it was found that the addition of cysteine and H_2O_2 induces changes in its activity. Additionally, labeling of the vacuolar proteome revealed the addition of H_2O_2 and cysteine to PTMox modifications in vacuolar proteins. Additionally, *Δcys4* strains present early vacuole acidification and biogenesis, suggesting a role for H_2S in vacuolar function. Using CRISPR–Cas9-mediated genome editing for scarless and marker-free DNA integration into yeast, we introduced a point mutation in cysteine residue C261 of the catalytic A subunit of the cytosolic V-ATPase complex. This allows us to evaluate the behavior of the cysteine residue under reduced and oxidized conditions.

Our findings may reveal a novel redox-dependent regulatory mechanism affecting vacuolar function and metabolic adaptation in yeast.

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Kinetic characterization of respirasomes and free complex I from *Yarrowia lipolytica*

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Mitochondria are highly dynamic organelles that adapt to cellular energy demands through structural and functional remodeling. A key feature of this adaptability is the supramolecular assembly of respiratory complexes I, III_2, and IV into structures known as respirasomes, which enhance protein density in the inner mitochondrial membrane and support efficient electron transfer and ATP production.

In this study, we characterized the subunit composition and enzymatic activity of digitonin solubilized respirasomes and free complex I from *Yarrowia lipolytica*, focusing on their roles in reactive oxygen species (ROS) generation. NADH:DBQ oxidoreductase activity was comparable between the two forms, with rates of 1376 ± 110 and 1632 ± 105 nmol NADH oxidized/min·mg complex I for respirasomes and free complex I, respectively. Respirasome-mediated respiration was sensitive to inhibition by rotenone, antimycin A, and cyanide, coinciding with elevated ROS production. A NADH:O_2 ratio of 1.6 ± 0.2 was determined, consistent with a functional coupling of electron transfer and oxygen consumption.

ROS production was quantified under conditions optimized for maximal enzymatic activity: 400 µM DBQ, 150 µM NADH and 5 mM cytochrome c. Basal (only with endogenous substrate) ROS levels were low in both assemblies: 17.5 ± 6.9 and 9.4 ± 3.9 pmol H_2O_2/min·mg complex I for respirasomes and free complex I, respectively. Upon substrate stimulation, ROS generation in respirasomes increased significantly to 44.5 ± 18.4 pmol H_2O_2/min·mg, approximately fivefold higher than in the free complex I (5.9 ± 0.84 pmol H_2O_2/min·mg). Under inhibitor-treated conditions respirasomes exhibited markedly elevated ROS levels 425 ± 111 and 357 ± 118 pmol H_2O_2/min·mg complex I in the presence of rotenone (100 µM) and antimycin A (5mM), respectively while free complex I showed no detectable ROS increase.

These results underscore a distinct functional role for supramolecular organization in the regulation of mitochondrial ROS production. Our findings suggest that respirasome architecture enhances the propensity for ROS generation under stress conditions, revealing a potential mechanism for redox imbalance during mitochondrial dysfunction.

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DESIGN AND MOVEMENT: SPHINGOLIPID INFLUENCE ON MEMBRANE FLUIDITY

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Sphingolipids are membrane amphiphiles which constitute one third of the total lipids from the plasma membrane in the plant *Arabidopsis thaliana*. The diversity of these lipids is extraordinary, and about 100 of sphingolipid species have been identified, among the most abundant belonging to two classes: the glycosylinositolphosphoceramides (GIPCs) and the glucosylceramides (GlcCers). These species have considerable polar heads integrated by carbohydrates and phosphate groups, while their hydrophobic region contains one fatty acid (FA) and a long chain sphingoid base (LCB). These two acyl chains give a rigid, compact and straight structural frame, features that are based on the abundant very long chain of the FA, the presence of few double bonds that are in *trans* configuration both in the FA and the LCB, and the presence of OH groups at the neck of the sphingolipid molecule. Our question was to determine the contribution of the different sphingolipid chemical groups and species to the membrane fluidity. The strategy was to use plasma membranes from mutants with different sphingolipid composition and then to study their fluidity. The latter was determined by fluorescence polarization from the membrane probes 1,6-diphenyl-1,3-hexatriene (DPH) and *cis* parinaric acid (PA). Sphingolipid composition was performed by HPLC/ESI-MS/MS analyses. Both sets of results were processed by correlation analysis using Pearson correlation (r) and Welch's t-test for comparing sample means within technical and biological replicates. The Shapiro-Wilk test, Spearman correlation and t-test were applied to the acyl chain length (FA), OH position (LCB) and type of polar head. So far, our results indicate that VLCFA promote membrane fluidity, while LCFA decrease membrane fluidity and therefore, the VLCFA/LCFA ratio is fundamental to modulate membrane fluidity, yet it is not through an interdigitation mechanism.

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The plasma membrane of beetroots submitted to waterlogging

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In plants, roots are responsible for providing water and mineral nutrients to shoots. They obtain the ATP needed for the metabolism of their cells mostly by respiration. They are thus very sensitive to decreases in oxygen content in the soil, such as those caused by waterlogging. This condition can produce programmed cell death in beetroots, especially when other stress condition (i.e. heat) is added. However, in the absence of other stresses, beetroots can tolerate waterlogging up to 17 days and recover if the soil is drained. In this study, we focused mainly on the changes in the proteins of the plasma membrane, especially on H^+-transporting ATPases.

Plasma membranes were prepared from beetroots flooded for 1, 3, 5 or 15 days and after 15 days of recovery in well-drained soil. Proteins in these membranes were analyzed by mass spectrometry. About 280 plasma membrane proteins were quantified and grouped according to their changes in each condition. Some proteins showed early and transient (at 1 or 3 days) increases or decreases, while others presented changes in amount in all days of waterlogging. Most proteins recovered to near control levels after 15 days of recovery in well-drained soil.

Among the proteins whose amount changed the less, were the plasma membrane H^+-ATPases PMA1 and PMA4. These small changes were also observed in immunoblots. However, the activity of the H^+-ATPases (presumed to be especially sensitive to ATP levels) presented larger changes, in line with the (immunologically measured) activating phosphorylation of their penultimate Thr residues. These results indicate that the activities of plasma membrane H^+-ATPases are controlled mainly by post-translational modifications in response to environmental changes.

Kinetic characterization of *Bos taurus* respirasome

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Mitochondrial respirasomes formed by the association of respiratory complexes I, III_2, and IV, are proposed to optimize electron transfer and the protonelectrochemical gradient generation (Reyes-Galindo *et al.,* 2019). Complex I (NADH:ubiquinone oxidoreductase) plays a central role as the entry point for electrons into the respiratory chain, coupling NADH oxidation to proton translocation across the inner membrane ultimately driving ATP synthesis (Zickermann *et al.,* 2015; Laube *et al.,* 2022). Supercomplexes and individual complexes from Bos taurus heart mitochondria have been widely used in structural studies, however, their comparative kinetic analyses remain scarce.

Here, we report the successful isolation of mitochondrial supercomplexes and complex I from Bos taurus heart mitochondria. Mitochondrial supercomplexes and free complexes were solubilized with an optimal digitonin/protein ratio (2:1) and isolated by sucrose continuous gradient centrifugation (16%–42%) as described by Reyes-Galindo *et. al.,* (2019); lead to distinct fractions consistent with the composition of the respirasome and the free complex I.

These results confirm the viability of obtaining high-purity preparations. Ongoing work will focus on kinetic characterization (Km,Vmax) using NADH, decylubiquinone, and cytochrome c to evaluate potential differences in catalytic efficiency between the respirasome and the free complex I.

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Epigenetic regulation of *ATP2A2* and *ATP2A3* genes and their potential role in hepatocellular carcinoma progression

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ATP2A2 and *ATP2A3* genes encode for Sarco/Endoplasmic Reticulum Calcium-ATPase 2 and 3, SERCA2 and SERCA3. This family of enzymes, with multiple isoforms, pumps Ca^{2+} from the cytoplasm into the endoplasmic reticulum, playing a central role in intracellular calcium homeostasis and signal transduction.[1] It has been shown that SERCA2 is downregulated in human oral cancer, and mice lacking a copy of this gene develop squamous cell carcinoma in the oral and gastric tracts.[2,3] Similarly, SERCA3 is downregulated and almost absent in several types of cancer and cancer cell lines, such as breast, lung, colon, gastric, and choroid plexus, suggesting an important role of this gene in cancer.[4-8] Previous findings demonstrated that histone deacetylase inhibitors (HDACi) increase *ATP2A3* expression in gastric, colon, lung, breast, and HCC cancer cells.[6-10] We have shown that sodium butyrate and trichostatin A, two HDACi, induce *ATP2A3* expression by increasing H3K9 and H3K27 acetylation in human breast cancer and rat HCC cells.[9,10] In this study, we investigated whether LBH589, a clinically relevant HDACi, and 5-Azacitydine, a DNA hypomethylating molecule, regulate *ATP2A2 and ATP2A3* expression in HepG2 HCC cells, as well as whether overexpression of these genes regulates proliferation and migration of these cells. Our results show that LBH589 induced *ATP2A3* expression through acetylation of lysines 9 and 27 of histone 3, suggesting that these modifications regulate this gene expression. Treatment of HepG2 cells with 5-Azacytidine induced *ATP2A3* expression; however, the CpG sites evaluated at its promoter region remained methylated, even after 5-Aza treatment. In contrast, the *ATP2A2* gene showed no changes in expression after treatment with these inhibitors, suggesting that these epigenetic mechanisms do not regulate its expression. Overexpression of *ATP2A3,* but not *ATP2A2*, inhibited colony formation and migration of HepG2 cells, suggesting that *ATP2A3* might be an important regulator of proliferation and migration in HCC. Our findings provide new evidence on the epigenetic regulation of the *ATP2A3* gene and its potential role in HCC progression.

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EFFECT OF CANCER CELLS-DERIVED CONDITIONED MEDIUM ON CARDIOMYOCYTE ENERGY METABOLISM

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Recent reports reveal a close relationship between heart damage and cancer, where cancer cells may negatively affect cardiac function by releasing factors (cytokines and metabolites), as has been observed in experimental murine models. Our research group evaluated the effect of hepatoma AS-30D on heart function. After seven days of cell implantation, an adverse effect on heart function was observed. 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 cells. Due to the limited experimental evidence on this topic, we are interested in analyzing the modifications induced by a conditioned medium from cancer cells on fluxes of energy pathways, enzyme activity, and protein content (oxidative phosphorylation and glycolysis) in cardiomyocytes.

In this study, the experimental models were rat C6 glioma cells and H9c2 rat cardiomyocytes. Both cell lines, at 100% confluence, were incubated in DMEM without fetal bovine serum (FBS) in both normoxic and hypoxic conditions for 24 hours to obtain the conditioned medium (CM). Afterward, H9c2 cells at 80% confluence were incubated with CM for 24 hours. Fresh medium (FM) without FBS was used as the control. The oxygen consumption associated with ATP synthesis (OxPhos) decreased by 50%, and glycolytic flux increased by 56% in H9c2 cells exposed to CM generated in hypoxia (CM-C6-H) compared to control cells with FM. The activity of mitochondrial enzymes (CS, IDH) showed a downward trend compared to the FM. PFK-1, the controlling step in glycolysis, increased its activity by approximately 30% in H9c2 cells exposed to CM-C6-H, which would explain the increased pathway flux in this condition.

HIF-1α, a transcriptional regulator of glycolytic enzymes, was observed to be active in cells treated with CM generated under hypoxic conditions, which may contribute to the increased activity of glycolytic enzymes such as phosphofructokinase I and, consequently, to the enhanced glycolytic flux. The stabilization of HIF would occur through the accumulation of lactate, which in the CM-C6-H was found at a concentration of 30 mM, significantly higher than in the condition with FM. When H9c2 cells in FM were exposed to 30 mM lactate, glycolytic flux increased by 45% compared to the control without exogenous lactate. These results suggest that CM-C6-H induces metabolic remodeling in H9c2 cells through the activation of HIF-1α.

Exercise combined with metformin and tert-butyl hydroquinone improves hepatic mitochondrial bioenergetics and redox status in middle-aged obese female Wistar rats.

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The world's population continuous to shift towards older, less active and more sedentary lifestyles especially during middle age. In addition consumption of high-caloric diets, increases the risk of metabolic and cardiovascular afflictions. Developing clinical strategies to mitigate those health complications represent a difficult challenge. Our group has previously shown that combining metformin (MTF) and tert-butyl hydroquinone (tBHQ) treatments, in addition to exercise, partially prevents liver damage associated with obesity. Hence, we evaluated the role of exercise in combination with MTF and tBHQ (triple-treatment) to counteract mitochondrial damage in the liver from obese middle-aged female rats. Animals were fed a high-fat diet (HFD) starting at 21 days till 15 months of age. The treated groups performed a Fartlek-type exercise 5 days/week for 30 min/session. MTF and tBHQ were administered at a dose of 250 mg/kg/day, and 10 mg/kg/day, respectively, for 7 days/month from 10 to 15 months of age. The triple-treatment therapeutic approach promoted animal survival, and increased AMPK and PGC1α expression. The treatments increased mitochondrial ATP synthesis and OXPHOS complexes activities; they also recovered the membrane potential and the redox state by decreasing oxidative damage and increasing SOD2, CAT, and GPx content and enzymatic activities. In summary, exercise in combination with intermittent tBHQ and MTF treatments proved to be an excellent intervention to prevent mitochondrial damage caused by HFD.

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4H-benzo[d][1,3]oxazines inhibits Proliferation, Migration, and Invasion Cervical Cell Lines

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A family of 4H-benzo[d][1,3]oxazines were obtained from a group of N-(2-alkynyl)aryl benzamides precursors via gold(I) catalyzed chemoselective 6-exo-dig C-O cyclization. The precursors and oxazines obtained were studied in breast cancer cell lines MCF-7, CAMA-1, HCC1954 and SKBR-3 with differential biological activity showing various degrees of inhibition with a notable effect for those that had an aryl substituted at C-2 of the molecules. 4H-benzo[d][1,3]oxazines showed an IC50 rating from 0.30 to 157.4 µM in MCF-7, 0.16 to 139 in CAMA-1, 0.09 to 93.08 in SKBR-3, and 0.51 to 157.2 in HCC1954 cells. We observed that etoposide is similar to benzoxazines while taxol effect is more potent. Four cell lines responded to benzoxazines while SKBR-3 cell line responded to precursors and benzoxazines. Compounds 16, 24, 25 and 26 have the potent effect in cell proliferation inhibition in the 4 cell lines tested and correlated with oxidant activity suggesting a possible mechanism by ROS generation. These compounds represent possible drug candidates for the treatment of breast cancer. However, further trials are needed to elucidate its full effect on cellular and molecular features of cancer.

Estrogen Deficiency Aggravates Mitochondrial and Cardiac Dysfunction in a Female Mouse Model of Cardiometabolic Injury Through Impaired Phospholamban Signaling

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Introduction: Postmenopausal women face increased cardiovascular risk due to estrogen decline, yet female-specific mechanisms of cardiac dysfunction remain understudied. Cardiac function relies on excitation-contraction-bioenergetics coupling where Phospholamban (PLN), a key regulator of Ca^{2+} reuptake via SERCA, may be dysregulated by estrogen deficiency. Impaired PLN phosphorylation can exacerbate Ca^{2+} mishandling, mitochondrial dysfunction, and contractile impairment. This study evaluated how the lack of estrogen influences PLN signaling and cardiac function in a female mouse model of cardiometabolic injury (CMI).
Methods: Four groups of 6-week-old female C57BL/6 mice were studied: (1) SHAM (control surgery), (2) SHAM-CMI (high-fat diet + L-NAME), (3) OVX, and (4) OVX-CMI. Weight, blood pressure, glucose, and cardiac function were recorded over 18 weeks. Cardiomyocytes were analyzed by confocal microscopy, and cardiac tissue was analyzed by qPCR and western blot. Data were analyzed with ANOVA and Tukey’s post hoc test ($p<0.05$), presented as mean ± SEM.
Results: At 18 weeks, OVX-CMI mice showed increased body weight (SHAM 7.6±0.7 g vs. OVX-CMI 18.3±5.4 g, p=0.04) and adiposity (SHAM 8.8±1.2% vs. OVX-CMI 18.3±3.1%, p=0.03). Peak fasting glucose occurred at 12 weeks (OVX-CMI 141.2±6.3 mg/dL, vs. SHAM 99.3±7.6 mg/dL, OVX 106.6±7.7 mg/dL and SHAM-CMI 111.3±5.8 mg/dL $p<0.05$). Diastolic dysfunction was exacerbated in OVX-CMI group (E/e’ OVX-CMI 19.1±5.4 vs. SHAM 4.4±2.6, p=0.001; vs. OVX 5.9±3.0, p=0.01). TMRE fluorescence was significantly reduced (fold change SHAM .99±0.05 vs. OVX-CMI 0.67±0.08, p=0.003), indicating mitochondrial depolarization. pPLN-Thr17 expression was reduced (SHAM 2.2±0.3 vs. OVX-CMI 0.38±0.003), suggesting impaired phosphorylation of PLN, which may enhance SERCA inhibition and impair relaxation. Ca^{2+} transient amplitude increased with CMI (SHAM 1.9±0.1 $\Delta F/F_0$ vs. SHAM-CMI 3.4±0.1, p=0.0008; vs. OVX-CMI 3.1±0.1, p=0.01), independent of hormonal status.
Conclusion: Estrogen deficiency amplifies the deleterious effects of cardiometabolic stress by disrupting PLN–SERCA signaling and impairing mitochondrial function, leading to pronounced diastolic dysfunction. These findings underscore the importance of including female-specific models in cardiovascular research and support targeting PLN pathways in future therapies.

Upregulation of oxidative metabolism through HIF-1α is related to the high cytokine and chemokine levels in peripheral blood mononuclear cells from patients with pulmonary arterial hypertension

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Introduction: Pulmonary Arterial Hypertension (PAH) is a progressive disease of the pulmonary vascular system that alters clinical parameters and the site of involvement, while innate immune activation has been linked to the development of PAH, the relationship between this immune system and its metabolic processes remains poorly understood and, little is known about the changes that occur in the periphery under this condition.

Methods: In this retrospective study, we worked with peripheral blood mononuclear cells (PBMCs) and plasma from 20 PAH patients and 8 healthy subjects. PBMCs were isolated from blood samples, and mRNA was extracted to analyze by RT-PCR gene expression of AMPKα2, HIF-1α, GLUT1, PFK2, PDHp, MTCO1, CO4, and MTATP6. Then, protein levels of HIF-1α, AMPK, subunits from OXPHOS complexes were analyzed by western blots. In addition, mitochondrial DNA (mtDNA) from PBMCs was quantified. Finally, inflammatory circulating cytokines also were quantified by flow cytometry, and correlation analysis were performed. We carried out the shapiro test in R studio to determine that the data followed a non-parametric distribution hence Mann-Whitney test were performed to analyze gene and protein expression and Spearman correlation for the correlation analysis.

Results: Gene expression showed significant trends of change in the HIF-1a gene and a significant difference in the expression of subunit 1 of ETC complex IV ($p<0.05$). Patients with PAH showed significantly elevated protein levels of HIF-1α ($p<0.005$), AMPK activity ($p<0.005$), and subunits of OXPHOS complexes: CIII ($p<0.005$), CIV ($p<0.05$), and CV ($p<0.05$). Correlation analysis of cytokine levels with those proteins showed a positive correlation with protein levels; interestingly, HIF-1α and AMPK protein levels positively correlated with levels of OXPHOS complexes.

Conclusions: A significant elevation of protein levels of HIF-1α and OXPHOS complexes, as well as an upregulation in AMPK activity in patients with PAH, indicates an adaptive response mechanism in mononuclear cells that, in turn, modifies the systemic inflammatory response.

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Cannabidiol modulates via PPARγ endocrine activity of white-like adipocytes

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Cannabidiol (CBD) activates the master regulator of adipogenesis (PPARγ) which regulates lipid metabolism in adipocytes. 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 ana 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 being 3.9 ± 0.8-folds ay day 10, and 1.6 ± 0.02-folds at day 14. Also, CBD increased 1.81 ± 0.5-fold of triglycerides at day 10 and 1.97 ± 0.45-fold at day 14. 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 relative expression of PPARγ (1.87-fold), PGC-1α (1.2-fold) and adiponectin (2.6-fold) at day 10, which additionally correlates with high adiponectin release in supernatant. Also, it decreased the relative expression of pref-1 (0.18-fold), suggesting major differentiation. Preliminary data demonstrates that CBD promotes surface markers associated with browning adipocytes. Finally, CBD increased the expression and release of adiponectin (anti-inflammatory), while it decreased expression and release of leptin (pro-inflammatory) on adipocytes treated with CBD, demonstrating its ability to modulate the endocrine function of adipocytes. Overall, our data demonstrated that CBD enhances adipogenesis in adipocytes via PPAR-γ, promoting a white adipocyte molecular phenotype and regulating endocrine activity toward an anti-inflammatory balance. These findings open the door to studying the use of CBD in obesity context.

Lysine hyperacetylation impairs the mitochondrial ATP synthase complex in the cardiometabolic HFpEF heart

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Introduction. Heart failure remains a leading cause of death worldwide. In particular, cardiometabolic heart failure with preserved ejection fraction (HFpEF) can be viewed as the heart response to a systemic metabolic disruption, typically secondary to obesity. In the current study, we sought to elucidate the mechanisms of energetic impairment in the heart of a preclinical model of HFpEF. **Methods.** HFpEF mice were subjected to an eight-week regimen of high-fat diet and L-NAME. Metabolism of ventricular cardiomyocytes was explored via high-resolution respirometry. Gene expression profile of the heart was assessed by NGS-based bulk RNA-seq. In-gel activity assays followed BN-PAGE of solubilized heart mitochondria. Mitochondrial complexome profiling as well as identification of acetylated lysine residues were determined by mass-spectrometry of isolated mitochondria samples. Acetylated lysine was also approached by immunoblot. ATP synthase activity was measured by a NADH-coupled spectrophotometric assay in isolated mitochondria and ventricular myocytes acutely exposed to sirtuin 3 activator honokiol (HKL). **Results.** HFpEF animals were characterized by visceral adiposity, elevated blood pressure, pulmonary congestion and overt diastolic dysfunction. Respirometric analysis of ventricular myocytes showed a compromised spare respiratory capacity (CTRL 2.3±0.23, HFpEF 1.6±0.12) along with a 1.5-fold increase of leak respiration in HFpEF. Next, RNA-seq revealed 1535 differentially-expressed genes (DEGs; FC>2, FDR<0.05). Notably, 146 DEGs (142 down-regulated in HFpEF) were contained in the GO-CC “mitochondrion” term, particularly encoding subunits of respiratory complexes I, IV, and V. Both in-gel and NADH-coupled assays showed a diminished ATP synthase activity in HFpEF mitochondria. Mitochondrial complexome profiling yielded no changes in abundance of respiratory complex subunits at the protein level, however, mass spectrometry did identify increased acetylation of lysine residues at both F1 and F0 regions of the ATP synthase complex (e.g. Atp5a1 K230 log2FC=5.80, FDR=7.8E-03). Lysine acetylation negatively correlated with ATP synthase activity in both isolated mitochondria and ventricular myocytes. Of note, HKL 10 µM brought acetylated lysine profiles of HFpEF myocytes back to CTRL levels and restored ATP synthase activity. Finally, we found a depressed ATP synthase activity in biopsies from HF human patients in which hyper-acetylation was previously found to be a feature. **Conclusion.** The present findings suggest impaired mitochondrial bioenergetics in the heart of a mouse model of cardiometabolic HFpEF, probably due to increased acetylation of lysine residues in the F1/F0 ATP synthase complex.

Effect of *killer* conjugated Ag nanoparticles over biological systems

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Silver nanoparticles (AgNPs) are widely used as antimicrobial agents but with high precipitation in aqueous solutions, to prevent this, several stabilizers can be used like polyvinyl pyrrolidone (PVP), PEG or proteins [1]. The present study aims to conjugate a fresh protein fraction of *killer* yeast supernatant containing the *K*1 toxin, to a chemically synthesized Ag nanoparticle. The protein fraction was obtained by ultrafiltration with 10 kDa columns, visualized on SDS-PAGE, and quantified by Bradford method. For the AgNPs synthesis, optimal conditions were achieved testing several concentrations of $AgNO_3$ (0.2 - 2 mM), and D-(+)-Glucose (0.5 - 300 mM). The freshly prepared NPs were conjugated with the *killer* fraction by passive adsorption for 24 hrs. at room temperature. Both, conjugated and free AgNPs were characterized with spectroscopy UV-Vis, FTIR and TEM. The antimicrobial effect of the nanocomposites was evaluated by agar well diffusion assays against *S. cerevisiae* 5x47 (sensitive strain to *K*1), *S. cerevisiae* 42300 (*K*1 producer), *B. subtillis*, and *P.* aeruginosa. Additionally, the toxicity, mitochondria viability, and DAPI internalization were evaluated on HEK-293 cells. As expected, the chemical synthesized AgNPs shown a uniform circular morphology and diameters around 20 nm with high aggregation. In comparison, those NPs conjugated with the *killer* fraction are much more scatter with similar size and shape. The spectroscopy characterization shows absorption peaks at 380-400 nm on UV-Vis and differential signals at 1075, 480-410 cm-1 on FTIR, which demonstrates the presence of AgNPs. On the other hand, both preparations demonstrate antimicrobial effect, however, the *killer* conjugated AgNPs shows higher activity against all the microorganisms tested ($P<0.05$). On mammal cells, the nanoparticles are capable of interact with the cell membrane, increasing DAPI internalization; for longer exposure times (24 - 48 hrs.), the interaction with the nanoparticles results lethal for cells.

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Cannabidiol prevents pathological cardiac hypertrophy via activation of PPARs and preservation of mitochondrial function.

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Background: Pathological cardiac hypertrophy is a precursor to heart failure, characterized by mitochondrial dysfunction, calcium mishandling, and oxidative stress. Cannabidiol (CBD), a non-psychoactive phytocannabinoid, modulates metabolic regulators, including peroxisome proliferator-activated receptors (PPARs), which are involved in cardiac remodeling. This study evaluates the PPAR-dependent cardioprotective effects of CBD in an *in vitro* hypertrophy model.

Methods: Rat H9c2 cardiomyoblasts were treated with Angiotensin II (1 μM, 48 h) to induce hypertrophy and exposed to CBD (0.1 μM). Hypertrophy biomarkers (cell size, BNP, and Col1a) were assessed by confocal microscopy and qPCR. The role of PPARγ and PPARα was evaluated using specific antagonists (GW9662, GW6471) and agonists (rosiglitazone, fenofibrate); gene and protein expression were quantified by qPCR and Western blot. Mitochondrial ROS and Ca^{2+} levels were analyzed using mitoSOX and Calcium Green.

Results: CBD prevented the Ang II-induced 60%±7 increase in cell size and reduced BNP and Col1a expression by 80%±5. Mitochondrial ROS levels decreased by 75%±6, and MCU expression was downregulated by 90%±10. CBD treatment doubled PPARγ and PPARα expression, and the inhibition of either receptor abolished these effects, restoring hypertrophic parameters and mitochondrial dysfunction. Complementary analysis of cardiac tissue samples from a murine model of Ang II-induced hypertrophy (L-NAME/high-salt, 0.7 mg/kg/day, 28 days) revealed reduced cardiomyocyte area and fibrosis (60%±10 and 35%±5, respectively), along with improvements in mitochondrial membrane potential and respiration in CBD-treated animals.

Conclusion:
These findings demonstrate that CBD exerts strong anti-hypertrophic effects through PPARγ and PPARα activation and mitochondrial protection, highlighting its potential to prevent cardiac hypertrophy.

Muscle-type–specific mitochondrial stress responses reveal dissociation between function and structure in a murine cardiometabolic HFpEF model

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Objective: Exercise intolerance is a hallmark of heart failure with preserved ejection fraction (HFpEF), largely attributed to peripheral, non-cardiac factors. However, the extent and nature of muscle-type–specific mitochondrial alterations in the context of HFpEF remain unclear. This study aimed to evaluate mitochondrial function, structure, and stress response pathways in two functionally distinct skeletal muscles in a well-characterized murine model of cardiometabolic HFpEF with confirmed exercise intolerance.
Methods: Male C57BL/6J mice were fed a 60% high-fat diet and L-NAME (0.5 g/L) for 12 weeks to induce a cardiometabolic HFpEF phenotype (n=32); controls received standard chow and water (n=31). Cardiac diastolic dysfunction and reduced exercise performance were confirmed. In manually isolated fibers from gastrocnemius and soleus, we evaluated triglyceride accumulation, lipid peroxidation, mitochondrial oxygen flux (complex I- and II-supported, and ADP-stimulated respiration), membrane potential (ΔΨm), and mitochondrial network structure. Gene expression analysis included markers of mitochondrial biogenesis, dynamics, mitophagy, and metabolic stress responses.
Results: HFpEF mice exhibited increased body mass (+73.4%) and impaired exercise performance (~30% reduction in distance, ~13% decline in relative power). Gastrocnemius muscle showed a marked (~300%) increase in triglycerides and elevated lipid peroxidation, unlike soleus. Complex I-supported respiration was significantly reduced in both muscles (−42% in gastrocnemius, −25% in soleus), with preserved complex II- and maximal ADP-stimulated flux. ΔΨm declined (~15% in gastrocnemius, ~9% in soleus) despite no changes in mitochondrial network organization or biogenesis/dynamics gene expression. Notably, Parkin expression was differentially regulated (−67% in gastrocnemius, +72% in soleus), alongside selective upregulation of HIF-1α and AMPK in gastrocnemius.
Conclusion: Our findings reveal a dissociation between mitochondrial function and structure in skeletal muscle during cardiometabolic HFpEF, with distinct alterations in lipid metabolism, mitophagy, and stress signaling between glycolytic and oxidative muscle types. This muscle-type–specific mitochondrial stress phenotype highlights an underexplored peripheral mechanism contributing to exercise intolerance in HFpEF, providing new insight into potential metabolic targets beyond the heart.

Does the δ-subunit of F_1-ATPase from *Polytomella parva* has a role in regulating the hydrolytic activity of this enzyme?

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Mitochondrial ATP synthase is an enzyme complex that carries out ATP synthesis from ADP and Pi. It is present in the plasma membrane of bacteria, in the thylakoid membrane of chloroplasts, and in mitochondrial inner membranes. This enzyme uses the energy of an electrochemical gradient to couple proton pumping with ATP synthesis (1). However, when the electrochemical gradient is compromised, either by the absence of oxygen or by the presence of uncouplers, this complex can carry out the opposite reaction by hydrolyzing ATP to reestablish the membrane potential (2). On the other hand, due to the risk of exacerbated ATP consumption, there are several mechanisms aimed at controlling the hydrolytic activity of this enzyme. The presence of small peptides that function as natural inhibitors of ATP hydrolysis have been described, for example the ε subunit in *E. coli*, the ζ subunit in *P. denitrificans*, the mitochondrial IF1 peptide or the ATPθ subunit in cyanobacteria (3). ATP synthase from the colorless alga *Polytomella parva* has attracted interest due to its atypical subunit composition, of the proteins involved in the dimerization of the enzyme and those forming the peripheral arm, which functions as a stator preventing the movement of the F_1 region during rotational catalysis of the enzyme. In addition to having 10 extra subunits (Asa1-10) not found in other ATP synthases, the subunits forming the catalytic core of the enzyme (α and β) have amino acid extensions in the N-terminal and C-terminal regions, respectively (4). There is also no report (biochemical or structural) of the presence of a natural inhibitor for *Polytomella parva* mitochondrial ATP synthase. In this context the δ subunit (homologous to the bacterial ε subunit) that participates in the binding of the hydrophilic part of the enzyme with the hydrophobic part, also exhibits an atypical extension in the N-terminal region. Bioinformatics and molecular dynamics approaches suggest that this N-terminal region is the most flexible or dynamic area of the protein, so it was hypothesized that this region could be prone to undergo conformational changes that allow it to interact with the subunits of the catalytic core of the enzyme (α or β) inhibiting ATP hydrolysis. Here, we explored the role of the δ subunit in the control of the hydrolytic activity of the mitochondrial complex V of the alga *Polytomella parva.*

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Identification of oxidative phosphorylation complexes of *Paramecium multimicronocleatum*

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For decades, the study of mitochondrial bioenergetics has been fundamental to describe and understand the function of oxidative phosphorylation complexes (OXPHOS) [1]. However, this research has focused on classical biological models such as yeasts and animals, leaving out other organisms of ecological importance such as ciliates [2], cosmopolitan microorganisms fundamental in the trophic chain of aquifer ecosystems and in some cases human parasites [3]. Due to this bias in the knowledge available to date, we sought to identify the OXPHOS machinery of the ciliate *Paramecium multimicronucleatum.*

Large scale *P. multimicronucleatum* culture conditions were standardized using minimum mineral medium of Tris-phosphate supplemented with vitamins and yeasts. After cell lysis, membranes were enriched by differential centrifugation and solubilized with non-ionic detergents: n-dodecyl-β-D-maltoside (β-DDM) and glyco-diosgenin (GDN101). OXPHOS complexes were separated by blue- and clear-electrophoresis in polyacrylamide gels in native conditions and further identified by *in-gel* zymography. We identified the activity of complexes I, IV and V. Additionally, a putative CI + $CIII_2$ supercomplex was identified which was not previously described for this species.

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Bioenergetics in aging yeast: the interplay between sirtuin2 and caloric restriction

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Aging is characterized by a progressive decline in cellular function, with mitochondria playing a central role. In *Saccharomyces cerevisiae*, chronological aging, defined as the loss of viability in non-dividing, stationary-phase cells, provides a useful model for post-mitotic senescence. Under caloric restriction (CR), yeast upregulate mitochondrial biogenesis and cell respiratory activity, while attenuating ROS production through ill-defined mechanisms. NAD^+-dependent deacetylase Sirtuin 2 (Sir2) has been identified as a key regulator, linking nutrient sensing to mitochondrial metabolism and genomic stability. Sir2 activity is modulated by the NAD^+/NADH ratio and regulates the expression of genes involved in stress resistance and metabolic remodeling. Together, CR-induced respiratory enhancement and Sir2-mediated epigenetic control constitute a reciprocal regulatory circuit: elevated mitochondrial respiration sustains a favorable NAD^+/NADH ratio to activate Sir2, while Sir2-driven transcriptional programs reinforce mitochondrial integrity and stress resilience. This extends chronological lifespan in yeast and highlights mitochondria as both sensors and effectors of longevity programs. In *S. cerevisiae* the molecular crosstalk between caloric intake, sirtuin activity, and mitochondrial bioenergetics was explored in order to analyze strategies preserving cellular function during senescence.

Kinetic characterization of dimeric F_1F_O-ATP synthase

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F_1F_O ATP synthases, found in bacterial, thylakoid and inner mitochondria membranes, are multiproteic molecular machines that carry out the production of ATP by a mechanism of rotational catalysis[1] using the proton motive force[2]. In mitochondria, this complex (denominated complex V, CV) is found in distinct oligomeric forms, mainly as a dimer (CV_2). The core structure of F_1F_O-ATP synthase, which comprehends the *c* ring, the central stalk and the catalytic domain is highly conserved among organisms. With the description of the high-resolution structures of mitochondrial complexes of novel eukaryotic study models it is now clear that mitochondrial ATP synthases pose highly divergent structures among evolutive distant eukaryotic supergroups. The major differences found in CV_2 are related to the peripheral stalk and the region embedded in the membrane, which directly impacts the stability of the complex and the ultrastructure of mitochondrial cristae [3].

Throughout decades, most kinetic studies of ATP hydrolysis by mitochondrial CV have been carried out in the monomeric enzyme from classical study models such as *Saccharomyces cerevisiae* and *Bos taurus*, both within Opisthonkonta supergroup. Therefore, we seek to compare the catalytic activity of CV_2 ATP hydrolysis of distant eukaryotic lineages which present structural divergence. *Saccharomyces cerevisiae* (Opisthokonta), *Polytomella parva* (Chlorophyceae), *Euglena gracilis* (Discoba), *Paramecium multimicronucleatum* (Alveolata), and *Phaeodactylum tricornutum* (Stramenopila) were cultured and further lysated. Mitochondria enriched preparations were obtained by differential centrifugation and membranes solubilized by the non-ionic detergent n-dodecyl-β-D-maltoside and glyco-diosgenin. Stable CV_2 was purified by ionic exchange liquid chromatography followed by exclusion chromatography. Finally, kinetic activity was evaluated by Pullman's coupled assay of ATP hydrolysis in isolated dimers. Preliminary results indicate that CV_2 present distinct kinetic parameters for ATP hydrolysis among the studied species.

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MCU inhibition via AAV9 transfection confers cardiac protection by maintaining mitochondrial health

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Introduction: Mitochondrial calcium (Ca^{2+}) overload through the mitochondrial calcium uniporter (MCU), a protein that transports Ca^{2+} into to the mitochondrial matrix, have been related to disease progression and development in various cardiovascular pathologies. Pharmacological treatment with inhibitors, such as Ru360, have proven useful in ameliorating or preventing cardiovascular diseases in animal models. Nonetheless, pharmacological inhibition is also subject to unforeseen off-targets that could be contributing to the protective effect. To verify these claims, we decided to use an *in vivo* transfection approach using AAV9, a virus that has tropism for cardiac tissue, to inhibit MCU expression in an acute model of catecholamine overload.
Objectives: Evaluate the cardioprotective effect of MCU inhibition via AAV9 transfection in a murine model of catecholamine overload.
Materials and methods: AAV9 were synthesized by transfecting HEK293T cells with a plasmid containing reporter protein tdTomato and 3 short hairpin sequences against MCU and collected using Iodixanol gradient ultracentrifugation. $5x10^{11}$ v.p. of AAV9 or normal saline solution was administered to 8-week-old C57bl/6 male mice via retro orbital injection. After 4 weeks of the administration, transfection confirmation was assessed by the presence of the reporter protein tdTomato in cardiomyocytes and MCU expression. These mice were subjected to a catecholamine overload model that consists in Isoproterenol (ISO, 400mg/kg) administration subcutaneously after a baseline ECG recording. Afterwards, cardiomyocytes and mitochondria were isolated for characterization studies.
Results: AAV9 synthesis yielded virus with properly packaged sh-MCU. 4 weeks after AAV9 administration, fluorescence from the reporter protein tdTomato was present in cardiomyocytes isolated from hearts administered with AAV9 but no fluorescence was detected in control cardiomyocytes. MCU protein expression was also concomitantly reduced by 49% (±10%) from control levels. After catecholamine overload, mitochondria from AAV9 hearts showed mitochondrial preservation as evidenced by a higher mitochondrial membrane potential (-180.3 ±8.9 mV vs -166.4 ±9 mV) when compared to ISO hearts without AAV9 administration.
Conclusions: Mitochondrial Ca^{2+} overload prevention through MCU inhibition is a promising new strategy for cardiac protection. AAV9 seems to confirm that the effects seen are indeed due to MCU inhibition and not an off-target effect of pharmacological inhibition. This also raises the possibility of using a AAV9 strategy of protein inhibition in cardiac tissue as a new treatment for cardiovascular diseases or to elucidate the role of specific proteins in the context of various cardiovascular pathologies.

Distribution of the F_1F_O-ATP synthase regulatory ζ subunit in alphaproteobacteria

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The F_1F_O-ATP synthase is the enzyme that condenses ADP + Pi into ATP [1]. This enzyme is present in most living organisms; in bacteria it's located in the inner cytoplasmic membrane and in mitochondria in the inner mitochondrial membrane. The F_1F_O-ATP synthases hydrolyze ATP when no terminal electron acceptor is available for the electron transport chain [2]. This hydrolytic activity can consume the ATP pool, which would lead to cellular death. To prevent the complete hydrolysis of intracellular ATP, there are regulatory mechanisms that can inhibit the ATPase activity [3]. One of these inhibitory mechanisms is the presence of endogenous regulatory subunits: in most bacteria, it is ε; in mitochondria, it is IF_1; and in alphaproteobacteria, it is ζ. The ζ subunit is the most recently discovered regulatory subunit; because of this, its origin within the alphaproteobacteria is still unknown. Unlike the canonical α, β, γ, δ, and ε, subunits of the F_1 which are clustered together in the same operon, the ζ subunit is not located in the same operon, because of this situation, its distribution pattern can be different than that of the other F_1 subunits. Here, we study the distribution of this ζ subunit among alphaproteobacteria and compare its distribution with that of the other subunits from the F_1-ATPase complex. To achieve this, we made a phylogenetic analysis of the ζ subunit and the α, β, γ, δ, and ε, subunits and compared their distribution in the alphaproteobacterial class. We compared their distribution with that of the 16s phylogenetic tree of life [4,5].

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Characterization of Mitochondrial Oxidative Phosphorylation Complexes from *Auxenochlorella protothecoides*

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Green microalgae perform two coordinated, coexisting bioenergetic processes: respiration in the mitochondria and photosynthesis in the chloroplast [1]. Both organelles have an endosymbiotic origin, a process that enabled them to acquire significant genetic variability, leading to the vast diversity of eukaryotes [2]. In the final stage of respiration, oxidative phosphorylation (OXPHOS) occurs, with its machinery composed of five multiprotein complexes (CI-CV) embedded in the inner mitochondrial membrane. The activities of CI-IV generate a proton gradient that results in the synthesis of ATP through the activity of CV [2]. It is expected that, because of their endosymbiotic origin, genetic variability would be reflected in the structural variability of the OXPHOS machinery. However, high structural conservation has been observed among complexes from different eukaryotes. A comparative study of OXPHOS complex structures shows that this high conservation has been overestimated, as the described OXPHOS complexes are from classic eukaryotes like plants, fungi, and animals. Few studies have been conducted across different members, hiding the structural diversity of OXPHOS complexes (Rico-Luna, A. *in preparation*). This is especially true for the *Chloroplastida* supergroup, where plants (*Streptophyta*) are the focus of research. In contrast, there are no described members of the *Trebouxiophyceae* class, even though the photosynthetic genus *Chlorella*, which is widely studied for its biotechnological importance, and the non-photosynthetic genus *Prototheca*, containing some infectious species that cause human protothecosis, belong to this class. We are interested in studying the species *Auxenochlorella protothecoides* because it is the intersection between these genera, showing a greater similarity to the pathogenic algae *Prototheca wickerhamii* than to other members of its genus [3]. The characterization of the OXPHOS complexes from *A. protothecoides*, used as a non-pathogenic model in this work, will enable the identification of structural differences to find therapeutic inhibitors as a more effective alternative for treating *P. wickerhamii* protothecosis.
To begin this study, the dark culture conditions for *A. protothecoides* were standardized and scaled up for biomass production. This was used to standardize cell disruption and membrane purification, which were then utilized to extract complexes with different types of mild detergents (β-DDM and GDN) at various concentrations. The extracts were subjected to native blue electrophoretic separation (BN-PAGE) and zymography to identify CI, CIV, and CV. The conditions for growing *A. protothecoides*, scaling up, and biomass production, as well as the conditions for cell disruption, membrane extraction, and solubilization, were established. So far, we have identified complexes I and V.

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The role of the ubiquitin Ligase Gzl in apical-basal transcytosis during the development of *Drosophila*.

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The respiratory system of *Drosophila* melanogaster is a network of branched tubes of epithelial cells. Terminal cells supply the respiratory surface for gas exchange. To fulfill their function, they branch extensively their basement membrane and extend a tubular lumen of apical membrane filled with air within each branch. Their growth is guided by FGF secreted by surrounding tissues. An important aspect is that initially the entire membrane and integral proteins are secreted to the apical domain, later they are moved by transcytosis to the basal domain. In another Drosophila epithelium, ubiquitination of SNARE Synaptobrevin by the ubiquitin ligase Godzilla has been shown to be necessary for transcytosis to occur.

Here we explore whether Godzilla's function on Synaptobrevin is necessary for the development of Drosophila terminal cells

Quantifying the photoprotective effect of qE NPQ in Chlamydomonas

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Photoinhibition, or light-induced damage to Photosystem II (PSII), irreversibly reduces PSII activity and linear electron flow. In the presence of chloroplast protein synthesis inhibitors, photoinhibition rates are proportional to irradiance. However, variations in light absorption, pigment composition, or cell culture density make it difficult to compare between cultivation modes or algal strains. In addition, photoprotective mechanisms such as thermal energy dissipation via the LHCSR3 antenna protein in Chlamydomonas (qE) are expected to decrease the probability of photodamage.

We propose using the PSII excitation rate as a functional parameter allowing normalization of the photoinhibition rate. *Chlamydomonas reinhardtii stt7* mutant cells (unable to perform state transitions) were grown under different light intensities and trophic modes. Cells were then exposed to photoinhibitory conditions, and functional measurements of photosynthesis were undertaken. To account for differences in functional architecture of the photosynthetic apparatus, we estimated PSII excitation rates by analyzing chlorophyll fluorescence rise in the presence of the PSII inhibitor DCMU. Electrochromic shift signals were also used to measure PSII charge separation rates for cross-validation.

Using PSII excitation rate values, we calculated the number of charge separations during photoinhibition. Cells grown under low light and mixotrophic conditions showed similar fractions of PSII activity leading to photodamage. In contrast, the photoprotective capacity of qE led to a lower photoinhibition rate per PSII charge separation. We discuss the feasibility of this and alternative methods to assess PSII excitation in the presence of qE.

Ammonia detoxication via photosynthetic reactions: a tale about a natural solar powered ammonia scrubbing system in salamander eggs.

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The spotted salamander (*Ambystoma maculatum*) and the green alga *Oophila amblystomatis* represent one of the few known examples of a vertebrate–photosynthetic symbiosis. During early development, salamander egg capsules are colonized by the algae. While previous research has emphasized the mutual exchange of oxygen and carbon compounds between host and alga, our study highlights a remarkable additional function of the algae in nitrogen cycling. We observed that toxic ammonia, a common metabolic waste product produced by developing embryos, was significantly reduced in egg capsules exposed to light. Our results show that *O. amblystomatis* assimilates this ammonia, effectively detoxifying the microenvironment by lowering concentrations to undetectable levels (<0 ppm), even without water changes.

Photosynthetic activity became detectable five days post-fertilization and remain high during development. Eggs exposed to medium to high light exhibited faster developmental progression than those kept in darkness over the course of the 30–40-day experiment. Medium light exposed eggs displayed higher speeds of embryonic growth, higher levels photosynthetic growth and Photosystem II (PSII) efficiency in the algae. Interestingly, eggs exposed to low light conditions displayed higher Photosystem II efficiency and algal growth when compared to those exposed to high light but showed slower embryonic developmental speed.

We hypothesize that increased light levels drive higher rates of oxygen evolution, which are essential for rapid embryogenesis in the salamander. However, the uninterrupted development of embryos in darkness, along with unsuccessful attempts to establish algal cultures in Basal Bold- (a commonly used medium for *Oophila* cultivation) and Sueoka-media (rich in ammonia) from green eggs, suggest that nitrogen exchange may not be the primary driver underpinning this symbiotic interaction. The implications of this biochemical cooperation for symbiotic nitrogen processing, and its potential applications in ammonia management within aquatic and engineered systems, will be discussed.

Posters Abstracts

Relieve del Edificio de los Danzantes de Monte Albán

Evaluation of L-glutaminase activity in the extremophile yeast *Rhodotorula mucilaginosa*.

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In order to grow, organisms need access to nutrients necessary to form their different structures. Some molecules are synthesized by the organism itself, while others are absorbed from the environment. In addition, organisms can detect the concentration of each nutrient and adjust their metabolic activity. Carbohydrates, fats and proteins may be used as carbon sources. Yeasts are unicellular eukaryotic organisms that are extremely useful in biochemical studies due to a metabolism resembling multicellular organisms. In addition, these grow in a few hours, require little space and can be genetically manipulated. It was decided to evaluate whether yeasts can grow using the amino acid glutamine (Gln) as the sole carbon source, thus resembling mammalian tumor cells. It was found that the yeast *Rhodotorula mucilaginosa*, which is highly resistant to adverse conditions, can grow using Gln as the only carbon source. Subsequently, the glutaminase covalent inhibitor 6-diazo-5-oxo-L-nor-leucine (DON), used as a chemotherapeutic agent, was tested. It is proposed that *R. mucilaginosa* is a good model to assessing sensitivity to glutaminase inhibitors, aiming to using them for anti-tumour therapy.

"Bioenergetic Architecture of a Decoupled Aquaponic System: Metagenomic Characterization of Microbial Flux and Nutrient Cycles"

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The bioenergetic efficiency of decoupled aquaponic systems depends on microbial communities that drive key biogeochemical cycles [1]. This study characterizes the microbial community structure and flux within such a system, composed of a fish tank (FT), a high-surface area bead filter for nitrification, and a floating raft root bed (RB). Microbial communities from the FT (source community) and the RB (sink community) are profiled via 16S rRNA gene amplicon sequencing and correlated with key physicochemical parameters. In an aquaponic ecosystem, there are two different types of analysis that can be done to help understand the microbial community structure and function within it. Alpha diversity analysis measures the microbial diversity within a single sample (like a water sample or a biofilter). In contrast, beta diversity analysis compares the diversity between different samples or compartments (like comparing biofilters to plant roots). Our beta diversity analysis shows no statistically significant difference between the FT and RB communities, confirming the FT acts as the primary microbial reservoir seeding the rhizosphere. Our alpha diversity indices, however, are consistently lower in the RB, indicating strong niche selection pressure in the plant root zone. The taxonomic profiling identifies a dynamic community. Key genera include *Carboxydoclivirga* and *Clostridium* as drivers of organic matter mineralization and fermentation; *Acinetobacter*, with its known versatility in the nitrogen cycle [2]; and *Sulfuricurvum*, revealing the sulfur cycle as another key bioenergetic pathway within the system. Furthermore, the photosynthetic cyanobacteria *Cyanothece aeruginosa* contributes to primary production, introducing new energy into the ecosystem via carbon fixation. This work deciphers a functioning metacommunity dynamic and identifies the key bioenergetic players, from heterotrophic recycling to chemosynthesis and photosynthesis, that define the resilience and efficiency of this ecosystem.

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Mitochondrial dynamics and mtROS regulate B cell differentiation in response to non-bilayer phospholipid arrangements

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Abstract:

Mitochondria play a crucial role in B cell responses against lipidic antigens through metabolic changes associated with differences in mitochondrial mass, membrane potential (ΔΨm) and mtROS production. Cell membrane lipids can acquire antigenic properties when they form stable non-bilayer phospholipid arrangements (NPAs) induced by drugs such as chlorpromazine. Stable NPAs induce germinal center responses and the production of anti-NPA IgG antibodies, which are present in autoimmune diseases such as Systemic Lupus Erythematosus. In this study we analyzed by flow cytometry the association between B cell response and mitochondrial dynamics, ΔΨm and mtROS production in a murine model that resembles human lupus induced by the administration of liposomes bearing NPAs. We found that B cells mainly respond through germinal center pathway and differentiated into plasma cells and memory B cells. Mitochondria from germinal center B cells increased their ΔΨm and decreased mtROS production, consistent with mitochondrial fusion, which favored their differentiation into memory B cells. In contrast, mitochondria from IgG^+ plasma cells showed decreased ΔΨm and elevated mtROS, associated with mitochondrial fission. These events result from changes in mtROS production; however, little is known about how mtROS contributes to B cell activation and differentiation to enable an effective immune response. Our findings establish a strong relationship between the *in vivo* B cell response against lipidic antigens and mitochondrial function, demonstrating that mtROS contribute to the modulation of mitochondrial dynamics and ΔΨm, with mitochondrial fission driving plasma cell differentiation.

Insights in activation of *COX1* mRNA translation in yeast mitochondria

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Mitochondrial translation represents a crucial regulatory step in the expression of the mitochondrial genome. In *Saccharomyces cerevisiae*, translational activators act on the 5′ untranslated regions (5′UTRs) of their target mRNAs to promote translation, presumably by guiding the mitochondrial ribosome to the start codon. The mitochondrial mRNA encoding subunit I of Cytochrome *c* Oxidase (*COX1*) is regulated by two known translational activators: Pet309 and Mss51. Pet309 directly interacts with the *COX1* mRNA via its pentatricopeptide repeat (PPR) motifs. In contrast, Mss51 lacks recognizable RNA-binding domains, and no direct interaction between Mss51 and *COX1* mRNA has been observed within mitochondria. Despite their established roles, the precise mechanisms by which *COX1* mRNA translation start remain poorly understood. To gain deeper insight into this process, we investigated the association of Pet309 and Mss51 with the mitochondrial ribosome. Both proteins were found to interact with the mitoribosome independently of the presence of *COX1* mRNA and of each other. The interaction of Pet309 with both the ribosome and *COX1* mRNA is mediated by its first six PPR motifs, located near the N-terminus of the protein. Notably, Pet309 appears to be entirely ribosome-associated, as no unbound protein was detected. In contrast, Mss51 primarily exists in lower molecular weight complexes, with only a small fraction co-sedimenting with the ribosome. These findings indicate that Pet309 and Mss51 stably and constitutively associate with the mitochondrial ribosome in a manner that does not depend on ongoing *COX1* mRNA translation. This supports the hypothesis that specialized mitoribosome populations exist within mitochondria, potentially dedicated to the translation of specific mRNAs.

Adenosine derivative compound IFC-305 reverses epithelial-mesenchymal transition induced by palmitic acid and TGF-β1 in HepG2 cells.

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Hepatocellular carcinoma (HCC) is the most common liver cancer and one of the leading causes of death in Mexico. In HCC, cancer cells exhibit metabolic reprogramming, increasing glycolysis to produce lactate. Lactate affects cell behavior and the cell environment to induce epithelial-mesenchymal transition (EMT). EMT plays an important role in providing changes in gene expression that promote cellular migration, tumor progression, and therapy resistance. EMT is characterized by cells with polarity loss, migration, and invasiveness capabilities, similar to those of mesenchymal cells. In HCC, hepatocyte transformation begins with E-cadherin loss, and N-cadherin and vimentin expression are induced by Snail, zeb1, and zeb2 transcription factors.

EMT is influenced by various factors, including fatty acids and growth factors, such as palmitic acid (PA) and transforming growth factor beta (TGF-β).

Dysregulation of fatty acid metabolism and the activation of profibrogenic pathways act as inducers of oncogenic signaling pathways, posing challenges for EMT reversal treatment. Thus, the search for new therapeutic targets and effective treatments is indispensable.

IFC-305 is an aspartate salt of adenosine that has therapeutic effects. These include improved mitochondrial bioenergetics, inducing antioxidant enzymes, autophagy induction in the HepG2 cell line, preventing alterations observed in animals with metabolic syndrome, and suppressing fibrotic pathways in both *in vivo* and *in vitro* models.

In this research, we aimed to develop a new EMT in vitro model using PA and TGF-β as inducers in HepG2 cells to evaluate IFC-305 in EMT transition. Preliminary results suggest that PA and TGF-β1 treatment transform hepatocytes into a mesenchymal phenotype, upregulate Snail, Zeb1, Zeb2, and increase protein levels of N-cadherin and vimentin. Furthermore, IFC-305 prevents cellular migration, decreases mesenchymal markers. We suggest that IFC-305 can revert transformed hepatocytes to an epithelial phenotype and inhibit metabolic reprogramming; therefore, the ability of IFC-305 to decrease lactate production is being evaluated.

Bioenergetic disruptions in neutrophil-like differentiated HL-60 cells caused by high glucose culture

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Type 2 diabetes is characterized by chronic hyperglycemia, which affects neutrophils functions leading to reduced pathogen clearing and increased morbidity. It is unknown how increased glucose affects neutrophils response, and their study is hampered by their short lifespan. Herein, we use a myeloid progenitor cell line widely used to study neutrophils differentiation *in vitro*. We induce neutrophil-like differentiation of HL-60 cells over 7 days using 1.3% DMSO in a standard medium with either 25mM (control) or 50mM glucose (hyperglycemia modeling) to study the effect of hyperglycemia during neutrophil production. We confirmed the neutrophilic phenotype by investigating CD15 and CD11b surface expression using flow cytometry, and we used Mito Tracker Green to assess mitochondrial mass. We used high-resolution respirometry to measure routine respiration and evaluated bioenergetic changes in response to relevant inhibitors during neutrophilic-like differentiation. We evaluated citrate synthase (CS) and CPT1 activity in protein extracts. After 7 days of differentiation, we obtained a similar percentage of $CD11b^{+}CD15^{+}$ cells for both treatments (93.35% for control and 94% for hyperglycemic modeling). We measured an increase in CD11b in hyperglycemic modeling compared to the control. During differentiation, we observed a decrease in routine respiration, following the decrease in mitochondrial mass at days 5 and 7 compared to HL-60 cells for both glucose concentrations. We did not see any change of non-mitochondrial respiration during differentiation. In high glucose, mitochondrial electron transfer capacity showed a significant decrease at day 5 and 7 compared to day 2 and 4. Furthermore, we measured a decrease in CS activity in high glucose compared to control on days 5 and 7. CPT1 activity decreased from day 4 onwards in high glucose compared to the control. Overall, high glucose in neutrophilic-like HL-60 cells differentiation could mimic some reported features of hyperglycemic patients such as the increase in CD11b. The change in glucose concentration showed a metabolic dysregulation, especially in the CS and CPT1 downregulation regarding mitochondrial mass. This demonstrates that HL-60 cells under high glucose differentiation could be used to model hyperglycemic neutrophils.

Keywords: Hyperglycemia, Neutrophils, T2D

Cannabidiol as an Anti-inflammatory and Mitochondrial Function-Protective Therapy in Cardiorenal Syndrome

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Renal dysfunction (RD) and heart failure (HF) often coexist, forming cardiorenal syndrome (CRS), which increases morbidity and mortality. This bidirectional relationship is driven by hemodynamic, neurohormonal, and inflammatory mechanisms, worsening both organs. Hypertension (HTN) is a key factor, increasing cardiac load and promoting hypertrophy, fibrosis, and endothelial dysfunction. Additionally, HTN impairs renal perfusion, accelerating RD and CRS. About 40–50% of HF patients develop RD, elevating mortality risk. Oxidative stress and mitochondrial dysfunction play a critical role, as excess reactive oxygen species (ROS) from HTN and HF induce cellular damage, inflammation, and fibrosis, altering the renin-angiotensin-aldosterone system (RAAS) and worsening RD. Preclinical studies show cannabidiol (CBD) reduces oxidative stress and inflammation in HF models, improving function and reducing fibrosis, but its effects in CRS remain unknown. This study evaluates CBD's protective effects in a CRS model. Male C57BL/6 mice (12 weeks) received a subcutaneous angiotensin II pump for 4 weeks plus L-NAME (0.1 g/L) + 1% NaCl in drinking water (CRS group). CBD was administered subcutaneously (1 mg/kg and 10 mg/kg) every third day. CRS increased blood pressure, which CBD did not reduce. However, low-dose CBD decreased plasma creatinine and proteinuria. Histology revealed glomerular hypertrophy and mesangial expansion, which CBD attenuated. CBD also reduced inflammatory markers (IL-6, TNF-α, MCP-1) and oxidative stress (protein carbonylation, thiol levels, TBARS, aconitase activity). Additionally, CBD improved mitochondrial function, enhancing respiratory activity and energy efficiency. These findings suggest CBD has reno-protective effects in HTN-induced CRS, reducing glomerular hypertrophy, inflammation, and oxidative stress. Further research is needed to explore molecular mechanisms and the therapeutic potential of CBD in CRS and renal diseases.

Key words: CBD, heart failure, renal dysfunction

Wolbachia metabolism after incubation in an axenic medium

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Wolbachia are obligate endosymbiotic Gram-negative bacteria infecting more than half of all insect species as well as filarial nematodes, crustaceans and arachnids. *Wolbachia* shows a diverse variety of symbiotic associations with its hosts, ranging from obligate mutualism in filarial nematodes to commensal, parasitic, or pathogenic associations in insects and other arthropod hosts. Wolbachia seems to contribute with biosynthetic pathways for haem, nucleotides, riboflavin, and/or FAD. In return seems to need help due to the lack of two glycolytic pathway enzymes, namely hexokinase and 6-phosphofructokinase. To test the glycolytic pathway in *Wolbachia* we incubated *Wolbachia* for 48 h in an axenic medium. Then evaluated if isolated bacteria use different glycolysis substrates (glucose, glucose 6-phosphate, fructose 1,6-Bisphosphate, glyceraldehyde 3-phosphate and phosphoenolpyruvate). Wolbachia was able to use only phosphate-containing substrates. It did not use glucose. Nonetheless, it was ableto synthesize ATP and it did release phosphate.

Effect of the unsaponifiable fraction of avocado oil on electron transport chain function in liver and kidney mitochondria of rats fed a high-fat and fructose diet

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Increased consumption of ultra-processed foods containing high fat and high fructose (HF-HFr) increases the risk of metabolic syndrome, which causes damage to organs such as the liver and kidneys. HF-HFr diets cause oxidative stress and mitochondrial dysfunction by impairing electron transport chain (ETC) complex activity. Previous studies have shown that avocado oil improves ETC function in the liver and kidney mitochondria of rats fed an HF-HFr diet, thereby improving liver and kidney function. However, it is unclear whether the antioxidant molecules in the unsaponifiable fraction of avocado oil (UFAO) are responsible for avocado oil's effects on the ETC. In this study, we investigated whether UFAO can prevent impaired ETC function in the liver and kidneys of rats fed an HF-HFr diet. The rats were divided into three groups: a control group consuming a standard rodent diet, an HF-HFr group consuming an HF-HFr diet for 12 weeks, and an HF-HFr+UFAO group consuming an HF-HFr diet plus 10 mg/kg of UFAO for 12 weeks. Liver and kidney mitochondria were isolated by differential centrifugation. ETC activities were assayed spectrophotometrically using substrates and inhibitors specific to each complex. In liver mitochondria, the activities of complexes I, II-III, and IV were 50% or less of those of the control group, while the activity of complex II remained unchanged. Complex I activity increased twofold in the HF-HFr+UFAO group compared to the HF-HFr group. The activity of the complex II-complex III segment in the HF-HFr+UFAO group returned to levels observed in the control group. No changes in the activity of complex IV in the HF-HFr+UFAO group were observed compared to the HF-HFr group. In kidney mitochondria, the activity of all ETC complexes decreased by more than 50% in the HF-HFr group compared to the control group. The activities of complexes II and II-III in the HF-HFr+UFAO group were similar to those in the control group. For complexes I and IV, however, the activity in the HF-HFr+UFAO group was twice that in the HF-HFr group. These results suggest that avocado oil prevents the inhibition of ETC complexes in rats consuming a HF-HFr diet due to its antioxidant components.

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

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Cervical cancer (CeCa) is the second most common cancer in women in Mexico and still a public health problem. Cancer cells possess characteristics that normal cells do not have. One of these characteristics is the metabolism reprogramming specifically to the mitochondria. Our research group has shown JAK/STAT pathway is active in CeCa cell lines, and recently prove it is involved in the metabolism regulation, since upregulation genes from glycolysis and the down regulation of genes from the electron transport chain (ETC) in CeCa cell lines.

In this work we analysed the role of STAT5 on the expression of mitochondrial genes involved in ETC. In cervical cancer cell lines, we demonstrate that STAT5 is localised in mitochondria in a basal form and stimulation with 10 UI/mL of IL-2 increases activation and the translocation of STAT5 to mitochondria. Consequently, the IL-2 treatment decreases the transcription of ETC-related genes, specifically UQCRC1, NDUFV1 and ATP5FB1. Furthermore, the oxygen consume from oxidative phosphorylation (OXPHOS) and mitochondria respiration decrease with also 10 UI/mL at 24 hours, supporting and favouring the metabolic reprogramming.

Additionally, transient transfection with RNA interference (RNAi) to knock down STAT5 demonstrated that IL-2 stimulation did not decrease the transcription of this ETC-related genes compared to wild-type CeCa cells.

The results of STAT5 translocation to the mitochondria, the regulation of the gene expression of the proteins that include the complexes of ETC and OXPHOS regulation suggest that STAT5 is an important regulator of mitochondria metabolic and contributes to the metabolic reprogramming in cancer cells.

Lipid Antigen-Driven Activation of T γδ Cells via NPA-Containing Liposomes in a Murine Model of Lupus

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Systemic lupus erythematosus (SLE) is a complex autoimmune disease marked by a breakdown of self-tolerance and the production of pathogenic autoantibodies. Although protein antigens have been extensively researched, the immunogenic capabilities of lipid-based structures remain underappreciated. In this research, we utilized liposomes bearing stable non-bilayer phospholipid arrangements (NPA) to induce a murine lupus model. These liposomes act as both a disease trigger and a novel platform for presenting lipid antigens that mimic membrane alterations associated with autoimmunity. BALB/c mice immunized with NPA-bearing liposomes exhibited lupus-like features, including the formation of autoantibodies. We focused on T γδ cells, which are a subset capable of recognizing non-peptide antigens, especially lipids. We utilized flow cytometry to examine T γδ cells from the spleen and mesenteric lymph nodes, evaluating their activation, proliferation, cell cycle status, mitochondrial dynamics, and cytokine production. Lupus-induced mice showed a significant increase in T γδ cell numbers and activation compared to controls. These cells actively proliferated (mainly in the S and G2/M phases, and exhibited signs of mitochondrial fission, indicating metabolic reprogramming associated with effector functions. Notably, they secreted IL-4 and IFNγ, which are key cytokines for B cell activation and IgG class switching.

Our findings highlight NPA-bearing liposomes as valuable tools for modeling lupus and studying lipid-driven immune responses. In this study, T γδ cells seem to connect innate lipid recognition with adaptive immunity. Our results suggest that T γδ cells are vital in both the initiation and maintenance of the adaptive immune response in SLE, and they might be directly involved in producing high-affinity antibodies against NPA. This model opens new pathways for targeting lipid-reactive mechanisms in autoimmune diseases and enhances our understanding of the role non-conventional T cells play in SLE pathogenesis.

Interaction of Amphotericin B with Lipid Bilayers via Molecular Dynamics

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Abstract:
Amphotericin B is a polyene antibiotic widely used in the treatment of systemic fungal infections. Its mechanism of action is associated with pore formation in ergosterol-rich membranes, although its interactions with other lipid components are not yet fully understood. In this work, molecular dynamics simulations were performed to analyze the behavior of amphotericin B in lipid bilayer systems composed of POPC–ergosterol and POPC–cholesterol. The aggregation of the molecule, its specific interactions with sterols, and its potential insertion into the membrane were evaluated. The results reveal differences in the molecular association patterns depending on the sterol type, as well as in the stability of the aggregates formed. These findings contribute to a better understanding of amphotericin B selectivity toward fungal membranes and may guide the design of derivatives with improved efficacy and reduced toxicity.

Keywords: Amphotericin B, molecular dynamics, lipid bilayer, ergosterol, cholesterol.

New real-time method (Nar-JJ) to measure the activity of membrane nitrate reductase (Nar) of denitrifying bacteria.

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Abstract. The transmembrane nitrate reductase (Nar) is the first enzyme of the anaerobic nitrate respiratory chain in all denitrifying bacteria. To date, there is no real-time method to determine its specific enzymatic activity embedded in its native inner membrane, but only alternate discontinuous or non-endogenous methods measuring the Nar activity by estimating the residual nitrate or the produced nitrite, or carried out with non-physiological electron donors or substrates. Given the lack of such a useful method to determine the endogenous Nar specific activity, in this work we standardized a new simple and useful method to measure with high precision the activity of the nitrate reductase enzyme (Nar) in real time, coupled in anaerobiosis to the NADH oxydation by Complex I in inverted vesicles of *Paracoccus denitrificans* (Sub-Bacterial Particles or SBP). This is carried out in anaerobic sealed reaction cells by firstly achieving anaerobiosis by preincubation with succinate to consume the residual dissolved oxygen, followed by malonate inhibition of further succinate respiration. Anaerobiosis is confirmed by the subsequent almost zero basal NADH oxidation, and by Oroboros confirmation of zero residual oxygen before nitrate addition. After anaerobiosis is confirmed, nitrate is added to start the Nar nitrate reductase reaction. The real-time NADH oxidation coupled to nitrate reduction is determined spectrophotometrically by subtracting the almost-zero basal NADH oxidation slope, without nitrate, from the NADH decay after nitrate addition. Controls confirmed the specific Nar activity determined by this new method by the total inhibition of the Nar reaction by sodium azide and also by cyanide, two well-known inhibitors of the Nar activity, but the former do not inhibit the Nap or water-soluble nitrate reductase. Estimation of the Michaelis-Menten affinity of Nar for NO_3^- using this so-called Nar-JJ assay gave a K_m of 70.44 ± 24.7μM [1], which is consistent with a value in the 40-75 μM range determined previously using natural substrates of the Nar enzyme [2]. This new Nar-JJ assay is a simple, low-cost, and suitable method to determine in real-time the endogenous Nar activity not only in *P. denitrificans*, but in other denitrifying bacteria such as *Brucella canis*. This Nar-JJ method is potentially useful in other pathogenic denitrifying bacteria such as *Salmonella* and pathogenic *Escherichia coli,* among others, to look for new antimicrobials targeting the essential Nar enzyme of these enteric pathogens. Funding: UNAM DGAPA-PAPIIT IN217520.

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Predicting the entry routes of nuclear-encoded mitochondrial proteins into yeast mitochondria.

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Like most eukaryotes, *Saccharomyces cerevisiae* mitochondria have their own genome (~86 kb), which encodes 8 proteins—7 of them essential for oxidative phosphorylation—along with 2 rRNAs, 24 tRNAs, and several intron-encoded maturases. However, over 99% of mitochondrial proteins are nucleus encoded, synthesized in the cytosol, and imported into the organelle. These precursors often carry a Mitochondrial Targeting Sequence (MTS) and are translocated by the TIM-TOM machinery, formed by the TOM complex in the outer membrane that serves as a general entry gate, and the TIM22 and TIM23 translocators in the inner mitochondrial membrane (MIM). Alpha-helical and beta-barrel outer membrane proteins are inserted via the *Mitochondrial Import Complex* and *SAM* pathways, respectively. Proteins targeted at the intermembrane space (IMS) follow either the Mia40/Erv1 oxidative folding pathway or a stop-transfer mechanism via TIM23. Soluble matrix-targeted proteins, with or without an MTS, are imported by TIM23. Inner membrane proteins encoded in the nucleus follow four known insertion routes: TIM23, TIM22, Oxa1-mediated conservative sorting, and the Bcs1-dependent pathway (Kizmaz et al., 2024).

Here, by compiling multiple datasets of previous proteomic studies (i.e., Vögtle et al., 2009; Morgenstern et al., 2017), we assembled a list of 1,220 proteins. After curating the data set and eliminating non-mitochondrial proteins and uncharacterized proteins, we confirmed 713 proteins which we believe represent the minimal yeast mitochondrial proteome. We analyzed this set to predict which proteins are membrane-bound, the number of transmembrane domains they contain, carry MTS, or have conserved cysteine motifs—information used to infer their likely internalization and sorting routes.

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The Unsaponifiable Fraction of Avocado Oil Improves Nonalcoholic Fatty Liver Disease, Insulin Resistance, and Mitochondrial Dysfunction in Rats Fed a High-Fat, High-Fructose Diet.

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The Western diet has been linked to an increased prevalence of non-alcoholic fatty liver disease (NAFLD), which is defined by insulin resistance, steatosis, inflammation, and mitochondrial dysfunction. Reactive oxygen species (ROS), which are produced in the mitochondrial electron transport chain (ETC), promote the progression of steatosis to steatohepatitis. Avocado oil contains unsaponifiable bioactive molecules that may reduce oxidative stress and improve mitochondrial function. This study investigated whether the unsaponifiable fraction of avocado oil (UFAO) improves NAFLD, insulin resistance and mitochondrial function in the livers of rats fed a high-fat, high-fructose (HFHFr) diet. Male Wistar rats were distributed into three groups: a control group fed a standard rodent diet, a NAFLD group fed an HFHFr diet, and a NAFLD+UFAO group fed an HFHFr diet plus 100 mg/kg b.w. of UFAO. The HFHFr diet was administered for 20 weeks. UFAO was administered after the fourth week, and streptozotocin was administered after the 16th week. The UFAO was characterized using GC-MS/MS and ESI to identify bioactive compounds. Insulin resistance was assessed using the HOMA index and an oral glucose tolerance test. Liver histologies were analyzed using hematoxylin and eosin staining. Liver mitochondria were isolated by differential centrifugation. Respiration and respiratory control ratios (RCR) were assayed polarographically. Mitochondrial membrane potential ($\Delta\Psi$) and reactive oxygen species (ROS) generation were evaluated spectrofluorometrically. Hydrocarbons, carotenoids, and xanthophylls were identified in the UFAO. Improved insulin resistance was observed in the NAFLD+UFAO group compared to the NAFLD group. The NAFLD group showed a 50% decrease in RCR compared to the CTRL group. RCR recovered to CTRL levels in the mitochondria of the NAFLD+UFAO group. $\Delta\psi$ decreased in the NAFLD and NAFLD+UFAO groups compared to the CTRL group. ROS increased in both the NAFLD and NAFLD+UFAO groups. Regarding liver histology, macro- and microsteatosis, as well as inflammation, were observed in the NAFLD group. These alterations were reduced in the NAFLD+UFAO group. In conclusion, UFAO improved liver damage, insulin resistance, and mitochondrial function in the livers of rats fed a HFHFr diet. This suggests the pharmacological potential of UFAO for treating NAFLD and its comorbidities. Further research is warranted to identify the molecules responsible for these effects.

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Evaluation of Phaseolus vulgaris L. extracts in breast cancer cells.

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Breast cancer is one of the leading causes of death among women worldwide. Although conventional treatments have improved survival rates, they still face significant limitations, such as toxicity, side effects, and tumor resistance. As a result, there has been growing interest in the use of natural compounds with anticancer potential, such as those found in the common bean (Phaseolus vulgaris L.), which is rich in phenolic compounds. This study evaluated the antiproliferative effect of hydroalcoholic extracts from the leaves and seeds of Phaseolus vulgaris L. (variety Dalia) on breast cancer cell lines (MCF-7, HCC1954, SKBR-3, and CAMA-1), as well as the use of plant-derived exosomes as delivery systems for these compounds. The results showed that leaf extracts—particularly those obtained from plants grown under temporary irrigation—had a higher concentration of phenols and a significant antiproliferative effect, unlike the seed extracts, which did not show any activity against the breast cancer cell lines. The optimal time point for evaluating these effects was determined to be 72 hours post-treatment.
These findings support the potential of bioactive compounds from Dalia bean leaves as natural anticancer agents in breast cancer treatment.

Intermittent fasting improves cardiorenal function in a murine model of heart failure

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Introduction: Heart failure with reduced ejection fraction (HFrEF) is characterized by impaired mitochondrial bioenergetics, increased oxidative stress, and inflammation, contributing to progressive cardiac and renal dysfunction. Intermittent fasting (IF) has emerged as a promising non-pharmacological intervention capable of modulating metabolic pathways, cellular stress responses, and mitochondrial homeostasis. Although its cardioprotective effects have been suggested in other models, its impact on mitochondrial function in HFrEF in a non-preventive treatment regimen remains unexplored. **Methods:** To investigate the bioenergetic and functional effects of IF in a murine model of HFrEF, male C57BL/6 mice (10 weeks old) were randomly assigned to four groups: control (CTRL), control with intermittent fasting (CTRL+IF), angiotensin II-induced HFrEF (HF), and HF with intermittent fasting (HF+IF). IF: 8-hour feeding restriction during the light phase. Echocardiography was used to assess cardiac function. Gene expression of inflammatory, mitochondrial and stress markers was quantified by qPCR. Cardiac remodeling was evaluated using hematoxylin-eosin and Masson's trichrome staining. Renal injury markers uSERPIN A3K, uKIM-1, creatinine and proteinuria levels were measured. High-Resolution respirometry and calcium handling markers analysis are currently in progress. Data distribution test (Shapiro-Wilk) and group comparisons (ANOVA) were performed. **Results:** No significant differences were observed between CTRL and CTRL+IF groups. HF mice exhibited ventricular dysfunction and renal injury. IF significantly improved left ventricular ejection fraction (HF: 37.4±4.4% vs. HF+IF: 46.4±6.6%) and cardiac output (HF: 8.8±1.4 mL/min vs. HF+IF: 23.3±3.8 mL/min). Despite persistent structural remodeling, IF reduced cardiac expression of NPPB, IL-1β and IL6, attenuated renal biomarker elevation, and improved renal function. **Conclusion:** IF improves cardiorenal performance in a murine HFrEF model. The observed functional benefits, alongside modulation of inflammation and renal injury markers, suggest that IF confers mitochondrial protection without prevention of pathological tissue remodeling. **References:** Kazmirczak F, et al. JACC Basic Transl Sci.2023;8(3):239-254., García-Rivas G, et al. JACC Basic Transl Sci. 2025;10(6):800-821., Méndez-Fernández A, et al. J Physiol. 2024, JP286410. **Keywords:** Intermittent Fasting, Mitochondria, Heart Failure, Inflammation.

Effects of IFC-305 on mitochondrial function in the B-cell precursor acute lymphoblastic leukemia (pre-B ALL) cell line NALM-6

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Precursor B-cell acute lymphoblastic leukemia (pre-B ALL) is characterized by blocked differentiation of lymphoid precursor cells, their uncontrolled proliferation, and their accumulation in bone marrow, blood, and lymphoid organs. It is the most common cancer in children and has a poor prognosis in adult patients. Therefore, new effective treatments with fewer adverse effects and that allow better recovery are necessary.

An adenosine derivate, IFC-305, has been studied in models of cirrosis and hepatocellular carcinoma, where it has been demonstrated its chemopreventive properties and it has been shown to regulate energy metabolism and mitochondrial function. Then, it could be considered as a new alternative cancer treatment. Recently, IFC-305 was found to have a cytotoxic effect on the B-cell acute lymphoblastic leukemia NALM-6 cell line at an IC_{50} of 2.4 mM. We decided to study its effect on the energy metabolism of these leukemic cells. Levels of pAMPK Thr172 increase in a time-dependent manner in treated cells at 2, 4, 6, 8 h with IFC-305 at 2.4 mM. However levels of pAMPK decrease at 24 h treatment. We also evaluated levels of ATP, ADP, and AMP in NALM-6 cells at 24 h with IFC-305 at 2.4 mM, and we found that the energy charge of NALM-6 cells increased. The membrane potential will be assessed to determine whether the IFC is capable of restoring it under conditions of metabolic dysfunction.

These results suggest that IFC-305 regulates leukemic NALM-6 cell line metabolic function, and decreases their pAMPK, preventing the activation of signaling pathways related to cell survival or repair of cell damage, thus facilitating cell death. Furthermore, the increase in ATP could suggest that cell activating energy saving and repair mechanisms, making the cells vulnerable to the effect of IFC-305 since they are unable to proliferate or remain viable in the long term.

Methanolic Extracts of *Capsicum annuum* var. *Fascinatum* (MECaF) Disrupt Calcium Homeostasis in MCF-7 Breast Cancer Cells

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Cancer results from multiple alterations in cellular mechanisms, among which calcium (Ca^{2+}) regulation plays a crucial role (1,2). Under normal physiological conditions, Ca^{2+} regulates key processes, including metabolism, cell proliferation, and apoptosis. Dysregulation of Ca^{2+} homeostasis contributes to neoplastic transformation by altering cell division, resistance to apoptosis, gene expression, and the function of organelles such as mitochondria and the endoplasmic reticulum (ER), thereby promoting a cancerous phenotype (3,4 and 5).

In this study, we conducted a transcriptomic analysis of MCF-7 cells treated with 2.3 mg/mL of MECaF for 24 hours. The results revealed changes in the expression levels of ATP2A3 (Sarcoplasmic/Endoplasmic Reticulum Ca^{2+}-ATPase 3), TRPC6 (Transient Receptor Potential Cation Channel Subfamily C Member 6), and ORAI1 (Calcium Release-Activated Calcium Modulator 1). Functional assays demonstrated that untreated MCF-7 cells exhibit robust Store-Operated Calcium Entry (SOCE), indicating active Ca^{2+} signaling and ER involvement. However, SOCE was significantly reduced in MECaF-treated cells, likely due to the coordinated upregulation of ATP2A3, along with the downregulation of TRPC6 and other Ca^{2+}-handling proteins. To validate these findings, qPCR analysis will be performed to confirm the gene expression profiles following MECaF exposure. Additionally, pharmacological modulators will be employed to assess the functional contribution of other proteins involved in Ca^{2+} influx pathways.

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Effect of IFC-305 on F_1F_O-ATPase dimerization and fibrosis in acute myocardial infarction remodeling in rats

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Cardiovascular diseases, primarily acute myocardial infarction (AMI), are the leading cause of death in Mexico and worldwide. Adenosine is known to have cardioprotective effects, as it exhibits antihypertrophic and antiadrenergic effects in the context of heart failure. A therapeutic alternative for the treatment of heart failure is the use of a new adenosine-derived drug called IFC-305, which has previously been shown to prevent infarction and modulate the activity of complexes I and II of the electron transport chain. To evaluate the compound's effect, we worked with an experimental model of myocardial infarction induced by isoproterenol, which acts directly on β-adrenergic receptors, causing functional ischemia and consequently cell death. Because mitochondrial function is diminished after isoproterenol-induced infarction, and given the fact that ATP synthase dimer arrays are a prerequisite for the formation of inner membrane cristae (Davies et al., 2012).

Then, we observed monomers and dimers in an acute infarction model induced by isoproterenol, using the technique of digitonin extraction from membrane proteins established by Dr. Marietta Tuena, and subsequently observing the enzyme's hydrolytic activity on native blue gradient gels. The following were obtained: the control rat heart sample presents activity in monomer and dimer, the heart sample with isoproterenol also presents activity in monomer and dimer but less than in the control, the heart sample that was treated with isoproterenol plus IFC-305, activity is observed in the monomer and dimer, the activity of the latter equal to the control and even a little more than in the control, which allows us to infer from these observations that the compound IFC-305 helps in the restoration of the dimers of ATP-synthase and thereby improve the energy demand of the cell. We also evaluated the Col1a1 and Col III genes. It was observed that the control group and the IFC group are similar, indicating that IFC-305 does not alter the genes studied, while those treated with isoproterenol show a higher expression of Col1a1 and Col III. The control group and the one treated with IFC-305 showed quite similar and low levels in the expression of the genes studied, which suggests that IFC-305 modifies the accumulation and has an antifibotic effect.

The Methylome Transcriptional Regulatory Network Activated by Copper: Impact on Basal Metabolism and Energy Production Genes

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Introduction: Transcriptional regulation is typically mediated by transcription factors, but an additional and crucial layer involves DNA methylation, where chemical modifications to DNA bases influence the binding affinity of the transcriptional machinery. In this study, we used *Enterococcus faecalis* as a model organism to integrate copper-induced methylome data into a transcription factor regulatory network (TRN). This approach aimed to elucidate the combined regulatory architecture of transcription factors and DNA methylation in response to copper exposure.
Materials and Methods: *Enterococcus faecalis* OG1RF strain (Id 474186). Copper treatment: 0.5 mM $CuSO_4$, 3 hours). Sequencing: Methylome (SMRT and bisulfite), Transcriptome (RNAseq). Bioinformatics: Bowtie2 mapper, SMRT Analysis tool, intersectBed tool, MEME tool, BEDTools, Cytoscape, TRN from Latore et al. 2024 (PMID: 24382465).
Results: We identified a total of 918 methylation patterns across the promoter regions of *E. faecalis*, covering approximately 55% of the genome. Notably, 20% of these methylation patterns were altered in response to copper exposure, indicating a significant epigenetic response induced by the metal. By integrating this information into the *E. faecalis* transcriptional regulatory network, we constructed a combined model of regulation involving both transcription factors and promoter methylation. The model includes 59 activated genes and 17 regulators, prominently featuring transcripts involved in energy production (ATP generation) and basal metabolism (amino acid and nucleotide biosynthesis). We identified two types of regulatory modules: a highly connected core module encompassing over 70% of the network components, and four independent modules associated with copper homeostasis (*cop* operon), as well as the regulation of phosphate, sugars, and nucleotides.
Discussion: The integration of different transcriptional regulatory mechanisms into an integrated model opens a new perspective on global gene regulation induced by metals in bacterial species.
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Administration of the aqueous extract of the aerial part of *Eryngium carlinae* and its combination with silver nanoparticles improves mitochondrial function in a model of type 2 diabetes

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Abstract

Type 2 diabetes mellitus (T2DM) is the most common type of diabetes, accounting for approximately 90 % of all diabetes cases. One of the complications of T2DM is diabetic encephalopathy (DE), which results in physiological, structural and neurochemical alterations in the central nervous system. Two of the main factors that trigger the development of DE are oxidative stress and mitochondrial dysfunction. The excess of substrates generated by chronic hyperglycemia in non-insulin-dependent tissues such as the brain, causes an increase in the generation of reactive oxygen species (ROS), leading to a state of oxidative stress and finally all of the above results in oxidative damage to mitochondrial components and the respiratory chain complexes, mitochondrial membranes, enzymes of the antioxidant system and mitochondrial DNA, ultimately resulting in mitochondrial dysfunction. This work aims to evaluate the activity of the extract-silver nanoparticles (AgNPs) combination and compare it with the extract of *E. carlinae* and commercial AgNPs on mitochondrial function and the activity of the antioxidant system in the brain mitochondria of rats with type 2 diabetes.

In this work, a T2DM model was induced by combining a high-fat diet (HFD) with a single dose of streptozotocin (STZ) (35 mg/kg body weight). The results obtained showed a significant increase in glucose levels in the diabetic group following administration of STZ. Furthermore, a significant decrease in the activity of respiratory chain complexes, mitochondrial membrane potential ($\Delta\psi_m$), respiratory control ratio (RCR) as well as the activity of the antioxidant enzymes superoxide dismutase 2 (SOD2) and glutathione peroxidase (GPx) and a significant increase in oxygen consumption in oligomycin-induced state 4 (4_o) and ROS generation were observed in the diabetic group compared to control group. However, the groups administered with the extract and the extract-AgNPs combination significantly increased the activity of the respiratory chain complexes and the antioxidant enzymes SOD2 and GPx, as well as an increase in $\Delta\psi_m$ and RCR compared to diabetic group.

Our results suggest that administration of the extract and the extract-AgNPs combination decreases oxidative stress and improves mitochondrial function in a rat model of T2DM.

Evaluation of cellular changes and cell membrane-associated proteins (metalloproteinases and ABCB1) in etoposide and paclitaxel resistant cells.

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Breast cancer is the most frequent neoplasm in women worldwide and is characterized by its biological and molecular complexity. Although treatments such as Paclitaxel and Etoposide have been effective in stopping the proliferation of tumor cells, resistance to these agents remains one of the major challenges for the treatment of this disease. Therefore, Paclitaxel- and Etoposide-resistant cell lines were generated to investigate the differences in their cellular and molecular processes compared to parental tumor cells. Proliferation assays were performed to determine the effective doses in resistant versus parental cells. The number of nucleoli, the expression and activity of MMP-2 and MMP-9, key enzymes in the degradation of the extracellular matrix and the expression of ABCB1, a protein responsible of the extrusion of compounds through cell membrane were evaluated. Paclitaxel- and Etoposide-resistant cells showed increased number of nucleoli, augmented expression of ABCB1 and activity of MMP-2 and MMP-9 compared with parental cells. Comparison between resistant and parental cells may provide a better understanding of the mechanisms that facilitate resistance, allowing for the identification of new therapeutic strategies in the future.

Mitochondrial Dysfunction in Menopause: A Journey Through the Liver, Kidney, Muscle, Heart, and Brain

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Menopause, the physiological stage that marks the cessation of ovarian function, is associated with decreased estrogen levels and systemic metabolic alterations that increase the risk of diseases such as obesity, dyslipidemia, insulin resistance, and type 2 diabetes. In postmenopausal women, the prevalence of obesity can exceed 40%, exacerbating the adverse effects of estrogen deficiency on metabolic health. Since estrogen is involved in the regulation of energy metabolism, its deficiency promotes ectopic lipid accumulation, lipotoxicity and mitochondrial dysfunction.

Furthermore, a diet high in fats and refined sugars contributes to energy imbalance, chronic inflammation, and mitochondrial dysfunction. Therefore, this study evaluated the impact of a high-fat diet (HFD) on mitochondrial bioenergetics in a rat model of menopause.

Eighteen-month-old Wistar rats (n=6 per group) were divided into four experimental groups: Sham on a standard diet (SD), ovariectomized (OVX) on a SD, Sham on HFD, and OVX on HFD. Ovariectomy or sham surgery was performed at 18 months of age, and the animals were immediately assigned to their respective diets for 2 months. At 20 months, all animals were euthanized for analysis of key organs involved in metabolic homeostasis: liver, kidney, skeletal muscle, heart, and brain.

Oxygen consumption, activity of complexes I, II, and IV of the electron transport chain (ETC), OXPHOS protein expression, and carbonyl levels as a marker of oxidative damage were quantified. The results revealed that ovariectomized rats OVX+HFD showed a significant decrease in mitochondrial complex activity and respiratory capacity compared to the Sham+HFD or SD groups, demonstrating a negative synergy between estrogen deficiency and excess fat on multi-organ mitochondrial function.

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An Unexpected Alliance: Riboflavin Production by Wolbachia in Saccharomyces cerevisiae

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Wolbachia are α-proteobacterial endosymbionts that infect 50–60% of invertebrate species, displaying a spectrum of interactions ranging from parasitism to strict mutualism. These interactions have co-evolved with their hosts, often manipulating reproduction to favor maternal transmission while occasionally providing essential metabolic benefits. Among these, riboflavin (vitamin B2), a precursor to vital flavin cofactors FMN and FAD, is notably conserved within *Wolbachia* metabolic pathways and plays a pivotal role in host survival and fitness.

Given the challenge of cultivating *Wolbachia* ex vivo, *Saccharomyces cerevisiae* was used as a heterologous model to investigate host-symbiont metabolic interactions. Two different riboflavin-auxotrophic yeast mutants, ΔRIB1 and ΔRIB4 were tested. We observed that *Wolbachia pipientis* wAlbB established an infection of the yeast host, reverting the growth defficiency in both riboflavin biosynthesis deficient strains. Our results suggest a functional riboflavin transfer from the symbiont to the eukaryotic host, offering a novel platform to dissect metabolic contributions in mutualistic endosymbiosis.

Distinct roles of NAC and RAC complexes in mitochondrial protein import and cytosolic proteostasis

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Around 99% of mitochondrial proteins are synthesized in cytosolic ribosomes and delivered to mitochondria through a process called mitochondrial import. The Nascent Polypeptide-Associated Complex (NAC) contributes to the early stages of this process by associating with the ribosomal exit tunnel. In *Saccharomyces cerevisiae*, gene duplication has led to two β-NAC subunits, enabling the formation of distinct $\alpha\beta_1$- and $\alpha\beta_2$-NAC heterodimers that interact with mitochondrial outer membrane proteins and modulate the import of specific substrates. Although NAC contributes to the fidelity of the import process, its deletion in yeast results in minimal phenotypic consequences, suggesting the presence of compensatory mechanisms. One such mechanism may involve the Ribosome-Associated Complex (RAC), comprising Hsp70 family chaperones (Ssz1, Zuo1, Ssb1, Ssb2), which could cooperate with NAC to support mitochondrial import, potentially through interactions with TPR-domain-containing receptors such as Tom20, Tom70, and Tom71.

Our findings indicate that the disruption of components within the NAC or RAC complexes exerts distinct effects on cellular physiology. Specifically, the absence of the NAC complex leads to activation of the mitochondrial retrograde signalling pathway, whereas the deletion of the Ssb1/2 chaperones, components of the RAC complex, appears to trigger general proteostasis responses mediated by the TORC1 signalling pathway. Furthermore, these mutants display differential sensitivity to endoplasmic reticulum stress and altered capacity to metabolize respiratory carbon sources such as ethanol. We also detected distinct effects on the import efficiency of mitochondrial proteins such as Sod2. Finally, using an Hsp104-GFP as a reporter for protein aggregation, we observed that NAC-deficient mutants accumulate protein aggregates under standard growth conditions. In contrast, RAC-deficient mutants exhibit low and diffuse Hsp104-GFP expression throughout the cytoplasm. Collectively, these results underscore the non-redundant roles of NAC and RAC in maintaining proteostasis.

"Gene context and location of the ζ subunit from the F_1F_O-ATP synthase of the different alphaproteobacterial"

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F_1F_O-ATP synthases condense >90 % cellular ATP through phosphorylative oxidation. These enzymes have a rotating mechanism that harnesses the energy stored in the membrane potential. This membrane potential is a proton gradient generated by the electron transport chain during cellular respiration [1]. The F_1F_O-ATP synthase allows protons to flow through its c-ring, and as the c subunits are protonated, a rotation of this c-ring occurs, then this rotation is passed to the γ subunit, which is bound on its base to the upper part of the c-ring. As γ rotates, it produces conformational changes in the α and β subunits, which then condense the ADP+Pi interface into ATP. This enzyme, in the absence of final electron acceptors, can do the inverse reaction and hydrolyze ATP. Throughout time, three different inhibitory subunits have evolved to inhibit this F_1F_O-ATPase activity. In mitochondria, the inhibitory subunit is IF_1 [2]. In bacteria, there are two known inhibitory subunits; for most bacteria, it is the ε subunit [3], and for alphaproteobacteria, the ζ subunit [1]. The ε subunit is located within the F_1-ATP operon, as this subunit is part of the canonical α, β, γ, δ, ε subunits that form the F_1 portion of the enzyme, and also as it is a structural subunit of the enzyme [4]. However, the ζ subunit, since it is not a structural subunit, is a supernumerary subunit, and it is not coded in the F_1 operon. Because the alphaproteobacteria class is composed of many different bacteria with a set of wide metabolisms and lifestyles, such as photosynthesis, nitrogen reduction or fixation, symbiotic interactions with plants and insects, as well as intracellular parasites, etc, it is our goal to analyze what genes are surrounding the ζ subunit in order to understand how this subunit is regulated in the different genomes from this class.

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MERCs and regulation of sulforaphane-modulated ER stress in cardiomyocytes subjected to chemical hypoxia.

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Introduction: Mitochondria-Endoplasmic Reticulum contact sites (MERCs) are regulators of several processes, such as ER stress (ERE), calcium signaling, and mitochondrial dynamics, among other[1], factors involved in the functioning and development of cardiovascular pathologies. Ischemia/hypoxia injury begins after deficient blood and oxygen supply to the tissue, inducing loss of cellular homeostasis and structural changes in the heart[2]; therefore, it is essential to seek strategies that help reduce this damage. Sulforaphane (SFN) is a compound present in cruciferous vegetables with different beneficial effects, such as antioxidant, anti-inflammatory, and anti-apoptotic[3]. Recently, SFN has been reported to promote a reduced mitochondria-RE association, which, in turn, increases ERE at non-toxic concentrations without *in vitro* damage stimulus[4]. Different experimental strategies have been implemented to emulate the hypoxic environment *in vitro*, including cobalt chloride ($CoCl_2$), which stabilizes hypoxia inducible factor (HIF-1α) and activates different damage responses, such as the generation of reactive oxygen species, loss of mitochondrial membrane potential, and cell death[5]. ***Aim***: To explore whether pre-treatment with SFN prevents ERE by regulating the communication of MERCs in H9c2 cells exposed to $CoCl_2$. ***Methodology***: Four experimental groups were established: non-treated cells (NT), cells treated with SFN (2.5 μM/1 h), cells exposed to $CoCl_2$ (100 μM/24 h), and the pretreated group (SFN+$CoCl_2$). Cell viability, oxidative stress, immunofluorescence, western blot, and transmission electron microscopy analysis were performed under experimental conditions. ***Results***: Cells treated with $CoCl_2$ showed decreased viability and increased oxidative stress, which were prevented by SFN treatment. Mitochondria-RE colocalization was higher when SFN induced a smaller distance between both organelles. Although cells treated with cobalt did not differ from the NT group, the distance of MERCs was significantly higher in the SFN+$CoCl_2$ *vs.* $CoCl_2$ group. In the SFN pretreated group, the expression of IRE1α increased, but its protein levels were lower than those in the NT group. In addition, HO-1 and NQO1 levels were lower in the SFN+$CoCl_2$ group compared than in the $CoCl_2$ group. ***Conclusion***: Under damage stimulus, the protective effect of SFN is associated with a lower ERE, mediated by increased communication of MERCs. ***References***: **1**. Jiang T et al. *Front Cell Dev Biol*. 2022, 10: 1036225; **2**. Chen Y et al. *Arq Bras Cardiol*. 2021, 6:1134-1144; **3**. Zhang J et al. *Biochem Biophys Res Commun*. 2022, 605: 119-126; **4**. Silva-Palacios A et al. *Chem Biol Interact*. 2023, 382: 110616; **5**. Orozco-Ibarra M et al. *Biol Res*. 2016, 49:7.

Neonatal neglect increases the risk of the cardiometabolic disorder in adulthood: a pre-clinical model

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Background: Adverse childhood experiences (ACEs), such as neglect and physical or sexual abuse, are potentially traumatic events that occur during childhood and can have long-lasting effects on physiological function into adulthood. ACEs are known risk factors for developing obesity, cardiovascular disease, and depression; however, they remain underrecognized in cardiometabolic disease pathways. This project evaluated the effect of maternal separation as a model of neglect neonatal (NN) on the cardio-metabolic phenotype in mice, using a combined model of NN and hypertensive-obesity in adulthood. **Methods:** All animal procedures were approved by the Tec de Monterrey-CICUAL. C57BL/6 male and female mice (8 weeks old) were used for breeding. The offspring male and female pups-underwent maternal separation (postnatal days 2-21, 3h/day). Mice were fed either a standard chow or high-fat diet (HFD) and HFD groups received L-NAME supplemented drinking water for 12 weeks. At 10 weeks old, mice were assigned to one of four experimental groups: 1.Control+Chow, 2.Control+HFD, 3.NN+Chow, 4.NN+HFD.. **Results:** Weekly body weight gain was significantly higher in male mice in the NN+HFD group compared to the Control+HFD from week 6 onward. In contrast female mice did not exhibit this effect. Additionally, male mice in the NN+HFD-group showed increased body fat percentage and hepatic steatosis compared to the Control+HFD group. Cardiac function was assessed by echocardiography. Systolic function parameters remained unchanged in both male and female mice across diets; however, diastolic function was impaired exclusively in male subjected to NN and HFD, as evidenced by alterations in deceleration time, isovolumic relaxation time and the E/A ratio. Signs of hypertrophy, pathological remodeling and mitochondria alterations in the left ventricle showed a trend toward an increase in male mice in the NN groups vs both the Controls group. **Conclusions:** These results suggest that the male mice exposed to early-life stress through neonatal neglect experience exacerbated cardio-metabolic alterations in adulthood when subjected to hypertensive-obesity model.

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Key-words: (up to 5): Adverse Childhood Experiences (ACEs), Neglect, Maternal Separation, Cardiometabolic disorder

Role of sodium-hydrogen exchanger 1 in cardiometabolic injury vs. pressure overload pure model

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Background: Hypertension (Hy) and metabolic disorders are major risk factors for cardiac disease, with heart failure (HF) as the final consequence when these conditions are not properly managed. While Hy typically leads to HF with reduced ejection fraction (HFrEF), the addition of metabolic dysfunction results in a more complex pathophysiology that commonly leads to HF with preserved ejection fraction (HFpEF). Sodium-glucose co-transporter type 2 (SGLT2) inhibitors have emerged as cardioprotective agents in both HF phenotypes, despite the absence of SGLT2 expression in adult cardiomyocytes. One proposed off-target of these drugs is the sodium-hydrogen exchanger isoform 1 (NHE1), a key regulator of intracellular pH and ionic balance. This study aimed to investigate NHE1 expression and activity in murine models of pressure overload (PO) and cardiometabolic injury (CMI), and to evaluate its potential as a therapeutic target of SGLT2 inhibition.
Methods: Male C57Bl/6 mice were divided into three groups: control (CTRL), CMI model, and PO model. The CMI model was induced using a high-fat diet (60%) and L-NAME (0.5%) for 12 weeks. The PO model was established by angiotensin II infusion, L-NAME (0.1%), and 1% high-sodium drinking water for 5 weeks. Cardiac function was assessed by echocardiography. NHE1 activity was measured in isolated cardiomyocytes using the ammonium pulse technique. Protein and gene expression were evaluated by Western blot and qPCR. One-way ANOVA followed by post hoc analysis was used for statistical comparisons. Data are presented as Mean ± SEM.
Results: Echocardiography revealed preserved ejection fraction in the CMI model and reduced ejection fraction in the PO model. NHE1 activity was significantly reduced in both models compared to CTRL (PO: 0.39 ± 0.06 vs. CTRL: 1.00 ± 0.26; $p = 0.01$; CMI: 0.60 ± 0.13 vs. CTRL: 1.00 ± 0.27; $p = 0.04$). Despite this, SLC9A1 (NHE1 gene) expression was significantly upregulated in the PO model (CTRL vs. PO: fold change 1.86 ± 0.32; $p = 0.04$), while it remained unchanged in CMI. Surprisingly, NHE1 protein expression was significantly reduced only in the PO group. Furthermore, cardiomyocyte cross-sectional area (CSA) and BNP levels were significantly higher in PO compared to CMI (CSA: PO 1.13 ± 0.08 vs. CMI 0.85 ± 0.05, $p = 0.01$; BNP: PO 1.92 ± 0.21 vs. CTRL 1.00 ± 0.18, $p = 0.01$), suggesting more pronounced structural remodeling and cardiac hypertrophy in the PO context.
Conclusions: These findings suggest that NHE1 activity is not uniquely altered according to HF phenotype, indicating it may not represent a selective therapeutic target for SGLT2 inhibitors. The results point toward the involvement of additional off-target mechanisms underlying the cardioprotective effects of SGLT2 inhibition.

Impact of respirasome organization on species-specific inhibition of complex III in human parasites

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Cellular respiration is carried out in the mitochondrial cristae by respiratory complexes (RCs) embedded in the mitochondrial inner membrane (MIM). These RCs can associate with each other, giving rise to supramolecular structures known as respirasomes. RCs from different eukaryotic supergroups exhibit great structural diversity, which impacts respirasome architecture. In this project, using comparative structural biology and molecular machines approaches, we analyzed the high-resolution structures of eukaryotic respirasomes available to date. The goal was to elucidate conserved assembly pathways, their contribution to MIM folding, and analyze the species-specific inhibition of drugs on mitochondrial complex III (CIII).

Our results show that respirasomes contain at least one $CIII_2$ in a central position relative to complex I, the largest of all the complexes involved. Therefore, it is proposed that $CIII_2$ acts as a molecular scaffold on which the respirasome assembles. The analysis also revealed that I/III_2 associations contribute substantially to the curvature of the MIM in most species.

Finally, in the largest human N-Respirasome, the entry cavity for the CIII-specific inhibitors antimycin A, myxothiazole, and stigmatellin A has free lateral access in the membrane, whereas in *Euglena gracilis*, the same cavities are impeded by the spatial arrangement of the CIV in the respirasome. A deeper analysis at the atomic level reveals that *E. gracilis* and *Trypanosoma brucei* have lost a phenylalanine residue involved in the π–π stacking of myxothiazole, which explains the resistance to this drug in these species. This work lays the groundwork for understanding differences in sensitivity to inhibitors and the search for potential species-specific therapeutic targets.

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Role of STAT3 on the regulation of mitochondrial activity in cervical cancer cells after IL-2 treatment

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Worldwide, cervical cancer is the fourth leading cause of cancer death in women. Cervical cancer is associated with human papillomavirus infection, which alters cells to acquire transformed characteristics such as uncontrolled proliferation, reprogramming of their energy metabolism and altered mitochondrial activity, all of which enable them to maintain rapid and uncontrolled growth. It is important to note that the JAK/STAT pathway is involved in the proliferation of cervical cancer cells, so activating the STAT3 and STAT5 proteins through tyrosine and serine phosphorylation is crucial to initiate their function. Therefore, in this work, we evaluated the role of STAT3 in the expression of critical genes crucial for the correct function of the electron transport chain in tumour cells and the effect of IL-2 on the oxidative phosphorylation of cervical cancer cell lines.

We demonstrate that treatment with 10 IU/mL of IL-2 increases the activation of STAT3 by phosphorylation on Y705 and S727 as well as cell proliferation. On the other hand, we also observed that mitochondrial genes UQCRC1 and NDUFV1 decrease after treatment with 10 IU/mL of IL-2; meanwhile, treatment with HO-3867, a specific STAT3 phosphorylation inhibitor, increases the expression of both genes, suggesting that STAT3 can function as a negative transcriptional regulator. On the other hand, we observed that treatment with 10 IU/mL of IL-2 decreases oxygen consumption, while the HO-3867 treatment increases oxygen consumption in both cervical cancer cell lines.

Altogether, these results suggest the fundamental role of STAT3 as a transcription factor in regulating critical genes essential for the correct function of the electron transport chain and, therefore, the mitochondrial activity. Furthermore, the decrease in UQCRC1 and NDUFV1 gene expression, as well as the decrease in oxygen consumption, demonstrate the importance of these nuclear-coded mitochondrial proteins for mitochondrial activity. The information generated in this project suggests that STAT3 may be a target for treating cervical cancer.

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ZmVDAC and ZmHXK4 are involved in the modulation of cell death during drought stress.

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Voltage-dependent anion channels (VDACs) are the most abundant proteins in the outer mitochondrial membrane. VDACs are essential for the exchange of ions, metabolites, and high-energy molecules (1), and they also serve as important scaffolds for the binding of various apoptosis-signaling proteins (2). In mammals, the interaction between VDAC and hexokinase II enhances cell survival, making this mitochondrial hexokinase an anti-apoptotic protein (3).

In plants, evidence for the involvement of VDAC and hexokinase (HXK) in the regulation of cell death is limited. In this study, we analyzed the physiological effects of the individual expression of two maize VDAC isoforms, ZmVDAC1b and ZmVDAC4b, as well as their co-expression with the mitochondrial hexokinase ZmHXK4, under both normal and drought stress conditions. Because abiotic stresses can trigger cell death responses (4), it is of particular interest to explore whether these proteins are involved in such processes.

The genes encoding ZmVDAC1b and ZmVDAC4b were cloned, and the proteins were transiently expressed in *Nicotiana benthamiana* leaves, where they localized to the mitochondria. Expression of ZmVDAC4b induced localized cell death and increased membrane leakage in the leaves, effects that were exacerbated under drought conditions. In contrast, co-expression with ZmHXK4 mitigated the physiological damage caused by ZmVDAC expression.

These findings suggest a functional interaction between ZmVDAC and ZmHXK4, potentially involved in modulating programmed cell death in plants. We are currently investigating their physical interaction through molecular dynamics simulations and experimental approaches.

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The use of Thioflavin T to estimate the Plasma Membrane Potential (PMP) in different yeast strains and the effect of pH.

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The use of the cationic dye thioflavin T (ThT) to evaluate the electrical plasma membrane potential difference (PMP) in baker's yeast has been described by our group. Besides evaluating fluorescence changes, we considered correction factors due to the binding of the dye to internal cell components to obtain actual PMP values in millivolts (mV). We deemed it important to investigate the feasibility of this approach with other yeast strains and to consider alternative methods, such as flow cytometry and multiwell plate readers for PMP estimation. The study included a couple of *Saccharomyces cerevisiae* strains (W303-1A and FY833) and several non-conventional yeasts, such as *Debaryomyces hansenii*, *Candida albicans*, *Meyerozyma guilliermondii*, and *Rhodotorula mucilaginosa*. Fluorescence-based PMP estimation yielded consistent results under different conditions with all strains and was further validated by measurements in mutants of the major monovalent transporters, confirming the efficacy of ThT in PMP monitoring.

Similarly, estimating yeast PMP responses to different pH values and K^+ concentrations required evaluating fluorescence changes and ThT accumulation. Qualitative observations at pH 4.0 revealed slightly lower PMP levels than at pH 6.0 and 7.0. The Nernst equation was applied with the ThT concentrations inside and outside of cells, allowing the measurement of PMP at different pH values, which aligned with the fluorescence-based observations. Given that yeast are exposed to low pH environments during fermentation, maintaining a robust PMP is crucial for survival, making the results trustworthy. We also considered the probable contribution of fermentation-derived bicarbonate to PMP establishment. These experiments reiterated the effectiveness of ThT-based methods for PMP analysis.

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Identification of F_1F_O ATP synthase from *Phaeodactylum tricornutum*

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The F_1F_O-ATP synthase is a multiprotein complex key to the synthesis of ATP, located in the mitochondrial cristae of eukaryotes [1]. Mitochondria are thought to share a single origin in all eukaryotes, resulting from primary endosymbiosis between a bacterium and an archaeon. Later secondary and tertiary endosymbiosis [2], along adaptation to the environment, and niche exploitation lead to the remarkable diversity of eukaryotic organisms observed today. As a result, the structure of mitochondrial ATP synthase varies across the different eukaryotic lineages described to date [3]. Diatoms are photosynthetic eukaryotic organisms that are cosmopolitan and predominantly marine, they represent the most abundant phytoplankton and are responsible for up to 40% of global primary production. Diatoms originated through two secondary endosymbiosis, beginning with a green alga and later involving a red alga. *Phaeodactylum tricornutum* is a model diatom and possibly the most extensively studied species. Its nuclear and mitochondrial genome, published in 2008, includes partial assemblies and annotations for genes and proteins [4]. However, there are no studies focused on mitochondrial ATP synthase, and its structure remains unknown.
Complexome profiling is a next-generation approach for identifying the abundance and arrangement of multiprotein complexes in biological samples, where the separation of native proteins is combined with tandem mass spectrometry [5]. This study aims to identify the mitochondrial ATP synthase of *P. tricornutum* using complexome profiling analysis. Using homology base methods, we identified previously unannotated subunits and compared them with known ATP synthase subunits from other eukaryotes. This comparative analysis allowed us to define a conserved core of subunits, which has been further analyzed for their structure and functional domains.

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Electrochemical Gradient and K^+ Transporters Shape *Killer* Sensitivity in *Saccharomyces cerevisiae.*

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Some *Saccharomyces cerevisiae* strains can inhibit other strain growth, due to the production of a toxin (*killer, K*1) and its interaction with its molecular target, the two-pore domain outward rectifying K^+ selective ion channel, TOK1; increasing its open probability, leading to the dysregulation of K^+ homeostasis and, in consequence, sensitive cells death. In this study, we test whether the controlled increase of extracellular K^+ modifies *Killer* effect, as predicted by Nernst Equation, annulling the electrochemical gradient, stopping K^+ efflux on sensitive cells and, thus, inhibiting *Killer* effect. We also explore the role of K^+ uniporters, Trk1 and Trk2, as compensatory mechanisms against *Killer* effect, by the influx of K^+. Experiments were made on YPD culture medium, supplemented with different [K^+] (0-400mM), where a lawn with different *S. cerevisiae* strains was made (5x47*;* BY4741*;* BY4741 *Δtok;* FY833*;* FY833 *trk1Δ;* FY833 *trk2Δ;* and FY833 *trk1, trk2Δ*, and cell spots with *K*1 producer strain, 42300. RT-PCR was performed to measure the expression of Trk1 and Trk2, so that we can determine the contribution of this proteins on *Killer* effect. We also blocked a mechanosensitive ion channel using TEA or lanthanum cations to determine its contribution to the *Killer* system. Our results suggest the electrochemical gradient is annulled at 300mM K^+, followed by an increase of *Killer* effect, probably due to volume regulation caused by an osmotic effect. Finally, we observed that *Killer* effect can appear on BY4741 *Δtok,* indicating other mechanisms independent of the interaction with TOK1, as stated by Molina-Vera (2024), via the activation of caspase Yca1p, acting like an ionophore, or by inducing apoptosis; and that other membrane proteins, including Trk uniporters, are involved on a “saving” mechanism against *Killer* effect.

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Avocado Oil Ameliorates Liver Damage and Insulin Resistance by Improving Mitochondrial Dysfunction, Diacylglycerol and ROS Levels, in Rats with Non-Alcoholic Fatty Liver Disease

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Non-alcoholic fatty liver disease (NAFLD) is a condition that ranges from simple steatosis to non-alcoholic steatohepatitis, cirrhosis, and hepatocellular carcinoma. Excessive consumption of high-fat and high-fructose diets promotes NAFLD, mitochondrial dysfunction, and insulin resistance (IR) via the accumulation of lipids, such as diacylglycerol (DAG), due to defective fatty acid β-oxidation. Previously, we demonstrated that avocado oil, a source of antioxidants, improves NAFLD, reduces hyperglycemia, and restores mitochondrial function. However, it is unclear whether avocado oil improves NAFLD and hyperglycemia by decreasing DAG levels through increased fatty acid β-oxidation and improved IR. To address this issue, male Wistar rats were divided into three groups: control, NAFLD, and NAFLD plus avocado oil (NAFLD+AO). NAFLD was induced by a high-fat, high-fructose diet for 20 weeks. AO was administered daily at 1 mL/250 g body weight (b.w.) starting on the fourth week. Streptozotocin was administered once in the 16th week to induce hyperglycemia. Hepatic damage was assessed via hematoxylin and eosin staining. Insulin resistance (IR) was determined using the HOMA-IR method. Hepatic DAG levels were assayed using an ELISA kit. Mitochondrial function was evaluated by assessing respiratory control ratios (RCR). ROS production was estimated spectrofluorometrically. The livers of the NAFLD group exhibited steatosis, inflammation, and elevated DAG levels, consistent with the increased IR observed in this group. Furthermore, NAFLD was associated with mitochondrial dysfunction and increased ROS levels. However, the mitochondria of the NAFLD group exhibited enhanced fatty acid β-oxidation. Avocado oil counteracts these alterations; the NAFLD+AO group exhibited reduced hepatic steatosis and inflammation, improved insulin resistance (IR), and decreased diacylglycerol (DAG) levels. Furthermore, avocado oil enhanced mitochondrial function and decreased ROS levels without affecting fatty acid β-oxidation. These results suggest that avocado oil improves NAFLD and hyperglycemia by decreasing DAG and mitochondrial ROS production, both of which are known to affect insulin signaling. This occurs via a mechanism independent of improvement in mitochondrial fatty acid β-oxidation.

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FKBP51 disrupts the insulin signaling pathway and impairs mitochondrial bioenergetics in HepG2 cells

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Abstract

FKBP51 reportedly antagonizes insulin signaling by reducing Akt activity, thus contributing to insulin resistance and weight gain. This has been well established in *FKBP51* knock-out mice. However, the effects of FKBP51 overexpression and its associated mechanisms have not been explored in liver cells. Here, we addressed this issue with a focus on mitochondrial bioenergetics. We overexpressed FKBP51 in the hepatocarcinoma cell line HepG2 and analyzed the changes in insulin signaling and mitochondrial function. FKBP51 overexpression impaired insulin-induced Akt and FOXO1 phosphorylation; however, downstream glycogen synthesis and gluconeogenesis inhibition were exacerbated. At the mitochondrial level, both insulin and FKBP51 overexpression decreased respiration, transmembrane potential, and ATP production, which explains why FKBP51 fails to prevent insulin metabolic response. Mechanistically, reduced ER-to-mitochondria Ca^{2+} transfer explains the drop in mitochondrial bioenergetics. Altogether, our results suggest that FKBP51 has a dual role in insulin responsiveness: it promotes insulin resistance proximal to the insulin receptor but has a negative impact on mitochondrial function, resulting in an accentuated effect on carbohydrate anabolism.

Área: Regulación del metabolismo energético

Avocado Oil Improves Renal Damage, Mitochondrial Dysfunction, and mPTP Opening in rats with Type 2 Diabetes.

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Hyperglycemia is a major contributor to diabetic nephropathy, a leading cause of renal failure worldwide. This condition involves glomerular damage, proteinuria, and mitochondrial dysfunction, including the excessive opening of the mitochondrial permeability transition pore (mPTP), which disrupts bioenergetics and increases reactive oxygen species (ROS) production. We have previously demonstrated that avocado oil improves oxidative phosphorylation, electron transport chain (ETC) function, ROS production, and kidney damage in rats with type 1 diabetes. However, it is unclear whether avocado oil can protect against kidney damage and mitochondrial dysfunction in rats with type 2 diabetes, and if its protective effects are related to improved modulation of the mPTP. To demonstrate this, male Wistar rats were distributed into control (CTRL), type 2 diabetes (T2D), and type 2 diabetes plus AO (T2D+AO) groups. The rats were fed a high-fat, high-fructose diet for 20 weeks. AO was administered daily at 1 mL/250 g body weight (b.w.) starting on the fourth week. A single administration of streptozotocin was given on the 16th week. Proteinuria was assessed on the 20th week by measuring 24-hour proteinuria. The rats were euthanized, and the mitochondria were isolated by differential centrifugation, and mPTP opening was evaluated spectrophotometrically. ROS levels were assessed spectrofluorometrically. Respiratory control ratios were calculated from mitochondrial respiration rates. Compared to the CTRL group, the T2D rats exhibited proteinuria and glomerular damage accompanied by increased mPTP opening, higher ROS production, and decreased RCR. Avocado oil prevented all these impairments; the T2D+AO group exhibited reduced proteinuria, lower mPTP opening, improved RCR, and lower ROS levels compared to the T2D group. These results show that avocado oil has nephroprotective effects in type 2 diabetes, possibly due to improved modulation of the mPTP and enhanced mitochondrial function.

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Allotopic expression of the *COX2* gene lacking the sequence encoding the leader peptide

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In most eukaryotic organisms the mitochondrial genome contains the *cox2* gene, encoding subunit II of cytochrome *c* oxidase (CcO). Nevertheless, this gene can be experimentally transferred to the nuclear genome and expressed from the nucleus. Natural nuclear relocation of the *COX2* gene has occurred in the chlorophyte algae *Polytomella parva* and *Chlamydomonas reinhardtii*, where it is split into two nuclear genes, *COX2a* and *COX2b*. During this migration the gene acquired polyadenylation signals, a mitochondrial-targeting sequence (MTS), switched from the mitochondrial to the universal nuclear genetic code, and lowered the hydrophobicity of its transmembrane segments so the encoded protein could be imported into mitochondria and assembled into cytochrome-c oxidase (CcO) (1). Inspired by this natural precedent, we recreated nuclear expression of COX2 in the yeast *Saccharomyces cerevisiae*, a strategy termed allotopic expression. Random mutagenesis produced a functional nuclear allele that encodes Cox2 fused to the Oxa1 MTS, an inner-membrane leader peptide (LP), two hydrophobic transmembrane segments (TMS1 and TMS2), and a long hydrophilic C-terminus (2). A single point mutation, W56R in TMS1, proved critical for Cox2 internalization: it reduces hydrophobicity just enough for the precursor to traverse the mitochondrial import machinery. Once inside, the Cox2(W56R) precursor is proteolytically matured, inserted into the inner mitochondrial membrane, and assembled into CcO, restoring up to 60 % of the respiratory growth of a Δcox2 mutant on non-fermentable carbon sources (3). Here, we tested whether Cox2(W56R) synthesized in the cytosol without its LP—whose gene was expressed either from a single-copy chromosomal insertion or a multicopy 2 μm plasmid— can still integrate into the inner membrane and rescue a null-Δcox2 mutant growth in non-fermentable media. This work was supported by a grant from PAPIIT (IN207023) from DGAPA-UNAM

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Disrupted mitochondrial calcium handling impairs pancreatic β-cell function under palmitate-induced lipotoxic stress.

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Introduction: Mitochondrial calcium dynamics in pancreatic β -cells, regulated by the Mitochondrial Calcium Uniporter (MCU), has shown a significant role in insulin secretion. This study investigates how a lipotoxicity-induced obesogenic environment affects MCU activity, impacting the β -cell function and cell death.

Methodology: To establish an *in vitro* lipotoxicity model, *RIN5F* cells (rat beta-cell line) were exposed to different palmitate concentrations. Cell viability and vitality were analyzed by Alamar blue and trypan blue staining, respectively. MCU activity and mitochondrial calcium dynamics were assessed by fluorometry, while reactive oxygen species (ROS) levels were measured by flow cytometry. Insulin secretion was quantified via ELISA.

Results: A dose-response curve confirmed palmitate-induced lipotoxicity. After 24 hours of exposure to 700 µM palmitate, β-cell viability decreased by 9%, while metabolic activity dropped by 47%. Mitochondrial calcium (mCa^{2+}) dynamics were also altered, with a 37% reduction in mCa^{2+} influx rate and a 41% decrease in mCa^{2+} retention capacity. Furthermore, this condition led to a 1.5-fold increase in mitochondrial ROS, while cytosolic ROS levels remained unchanged. These mitochondrial alterations were accompanied by a marked functional impairment in β-cells, evidenced by a 15-fold reduction in total insulin content relative to untreated controls. Notably, glucose-stimulated insulin secretion remained unaffected, suggesting an early stage of β-cell dysfunction in which insulin synthesis or storage is compromised before secretory failure.

Conclusion: These results suggest that MCU-mediated calcium mishandling contributes to lipotoxicity-induced beta-cell dysfunction and death, which may constitute a pathophysiological mechanism in obesity and Type 2 diabetes (T2DM). This study aims to assess whether MCU could serve as a potential therapeutic target for preventing or mitigating the progression of obesity-related T2DM.