XXI SMB PLANT BIOLOGY CONGRESS

4TH ASPB MEXICO SECTION MEETING

OCTOBER 20TH–24TH ■ 2025

MÉRIDA, YUCATÁN, MÉXICO

BOOK OF ABSTRACTS

Welcome to the 21st SMB Plant Biology Congress & 4th ASPB Mexico Section Meeting!

We are delighted to have you join us for this exciting event, taking place in the vibrant city of Mérida, Yucatán, from October 20–24, 2025.

This special event brings together the national Plant Biologist Congress, hosted by the Mexican Society of Biochemistry (SMB), and the 4th Mexico Section Meeting of the American Society of Plant Biologists (ASPB). We are honored to have distinguished scientists joining us from Mexico, the United States, Europe (Switzerland, Austria, England, Germany, Italy, the Netherlands), Asia (Pakistan), and across the Americas (Canada, Guatemala, and Peru).

Our organizing committee has selected an exceptional program featuring presentations from leading experts in plant biology. Attendees will explore a wide range of topics, including metabolism, plant growth, stress responses, genetics, evolution, and plant communication. This congress is designed to inspire discussions and offer invaluable networking opportunities, particularly for Mexican students and postdoctoral researchers willing to interact with leading scientists from a variety of biological disciplines.

We look forward to an inspiring, collaborative, and enriching experience with all of you!!!

ORGANIZING COMMITTEE

Patricia Coello, President	Facultad de Química, UNAM
Stewart Gillmor	Unidad de Genómica Avanzada, Cinvestav, México
Jesús Montiel	Centro de Ciencias Genómicas, UNAM
Alejandra Rougon	ENES Unidad León, UNAM
Elizabeth Córdoba	Instituto de Biotecnología, UNAM
Fulgencio Alatorre	Centro de Investigación Científica de Yucatán, A.C.
Karolina Heyduk	University of Connecticut, USA

LOGISTICS COORDINATOR Jessica Carcaño Macías

TECHNICAL SUPPORT TEAM

- Aída A. Hernández Quinto
- José Adolfo Martínez Olmedo
- Brenda Álvarez
- Verónica Ramón
- Sonia Barragán
- Luis Carlos Gutiérrez Pacheco
- Jesús R. Alatorre Cobos

Scientific Program: XXI SMB Plant Biology Congress & 4th ASPB Mexico Section Meeting

Monday October 20

09:00-18:30	Registration & Welcome
Keynote session	
18:30-19:15	**Siobhan M. Brady** (UC Davis) Environmental Integration with Root Cell Type Development
19:30-21:30	Welcome Cocktail

Tuesday October 21

·Abiotic Stress and Genetics & Genomics·

Chair, **Stewart Gillmor**

Keynote session	
09:00 - 09:45	**Alejandra Covarrubias** (IBT-UNAM) Dry Times, Tough Proteins: Roles of LEA proteins in plant responses to water scarcity
Plenary sessions	
09:45 - 10:15	**Minerva Trejo** (Institute of Science and Technology, Austria) Leveraging the evolutionary information in genomic and epigenomic databases to reconstruct DNA methylation systems
10:15 - 10:45	**Ken Birnbaum** (New York University) One Call, Many Responses: Decoding Auxin's Cell-Specific Language
10:45 - 11:15	**Damien Formey** (CCG-UNAM) Phabase: A High-Quality Genome Assembly and Platform for *Phaseolus vulgaris* cv. Negro Jamapa
11:15 - 11:45	Coffee break
11:45 - 12:15	**Dan Chitwood** (Michigan State University) Iconography and Indigenous Knowledge in the De la Cruz-Badiano Nahuatl Herbal (1552): A Text Mining and Morphometric Study of Mexican Plants
12:15 - 12:45	**Azalea Guerra** (Unidad Irapuato, Cinvestav) Genetic Diversity of Orphan Crops and Wild Relatives: A Foundation for Resilient Agroecosystems
12:45 - 13:15	**Zachary Lippman** (Cold Spring Harbor Labs) Hidden genetic complexities and phenotypic consequences of evolving plant stem cell networks in nature and agriculture
13:15 - 14:45	Lunch

Short talks	Chair, **Fulgencio Alatorre**
14:45 - 15:00	**José Alberto Valenzuela** (CIAD) A comparative genomic analysis reveals key conserved cuticular genes in fleshy fruits cuticular biosynthetic machinery.
15:00-15:15	**David Díaz** (Unidad Irapuato, Cinvestav) WIP2/NTT modulates cytokinin homeostasis
15:15-15:30	**Elsa Góngora** (Unidad Mérida, Cinvestav) What did we lose by domesticating papaya? Genome sequencing of wild and commercial varieties reveals genetic clues to resistance
15:30-15:45	**Sergio Pérez** (Penn State University) The MexMAGIC population and novel whole genome assemblies define signals of clinal adaptation in Mexican native maize
15:45-16:00	**Teresa Nava** (IBT-UNAM) Structural disorder and distinctive motifs in the C-terminal region of the MADS-domain transcription factors is conserved across diverse taxa
16:00-16:15	**Hong Ma** President of the American Society of Plant Biologists **(ASPB)**
Flash talks	
16:15-16:45	**Coral Martínez** (IBT-UNAM) Anhydrobiosis in plants: from biocondensates in seeds, to the native cellular structure of the pollen grains. **Donaji García** (Facultad de Química-UNAM) ZmVDAC and ZmHXK4 are involved in plant drought response and the modulation of cell death **Damaris Guerrero** (Facultad de Química-UNAM) PCNA association with CycB1;2/CDKB1;1 complex and its role in DNA Damage Response during maize germination. **Gerardo Contreras** (Facultad de Química-UNAM) Structural insights into the functional regulation of DPE2 during bean fruit development **Noe García** (ENES León, UNAM) Identification and classification of pathogens effectors in agronomical relevant crops using meta-transcriptomics, proteome structural data, and artificial intelligence **Jessica Segura** (CCG-UNAM) Characterization of *Botrytis cinerea* infection in *Lotus japonicus* and effect of nodulation on the defense response **Goretty Caamal** (CIBNOR) MeJA triggers *Capsicum annuum* response against *Phytophthora capsici* through defense-related genes activation in early stages of infection.

	Rocío Reyero (CCG-UNAM) The common bean mutant (*Phaseolus vulgaris*) non-nodulating mutant: nnod(2114) is affected in the infection thread development during symbiosis with rhizobia. **Elizabeth Quintana** (CIATEC) How ozone can affect pollination syndromes in strawberry plants? **Mingyar López** (Facultad de Química-UNAM) Revealing new mechanisms of maize Wee1 kinase regulation **Juan Ramos** (Unidad Irapuato, Cinvestav) Arabidopsis gynoecium development: confocal imaging of the cell cycle and cytokinin signaling **Iván López** (Leibniz Institute) Gradual domestication of root traits in ancient maize shows an adaptation to the Neolithic soils from Tehuacán
16:45-17:15	Coffee break
17:15-18:30	Poster session (even numbers)
Keynote session	
18:30-19:15	**Sue Rhee** (Michigan State University) Drug discovery for agriculture:Center for Sustainable Plant Innovation and Resilience through International Teamwork (C-SPIRIT)

Wednesday October 22

·Plant-Biotic Interactions and Metabolism·

Chair	Chair, **Jesús Montiel**
Keynote session	
	The EMBO Lecture
09:00 - 09:45	**Cyril Zipfel** (Universitat Zurich) Roles and recognition of stress-regulated secreted signaling peptides
Plenary sessions	
09:45 - 10:15	**Jeongmin Choi** (University of Cambridge) Identification of a suppressor of arbuscular mycorrhizal symbiosis in high phosphate conditions
10:15 - 10:45	**Jean Michel Ané** (University of Wisconsin) Biological Nitrogen Fixation on the Aerial Roots of Maize and Sorghum for Sustainable Agriculture
10:45 - 11:15	**Clelia de la Peña** (CICY) Integrative study of *in vitro* albinism in *Agave angustifolia*: Transcriptomic, Epigenetic, and Metabolomic evidence
11:15 - 11:45	Coffee break

11:45 - 12:15	**Robertas Ursache** (CRAG) GELP proteins as architects of essential plant biopolymers
12:15 - 12:45	**Samuel Zeeman** (ETH Zürich) Beyond Simple Storage: Revealing the Cellular Machinery and Processes Underpinning Starch Granule Biosynthesis
12:45 - 13:15	**Berkley Walker** (Michigan State University) Photorespiration as a crossroads between photosynthesis, metabolism, and leaf energetics
13:15 - 14:45	Lunch
Short talks	Chair, **Alejandra Rougon**
14:45 - 15:00	**Carmen Quinto** (IBT-UNAM) Unraveling the role of the phospholipase PvPLDα2 in the symbiotic interaction between common bean and rhizobia
15:00-15:15	**Naveenkumar Athiyannan** (Unidad Irapuato, Cinvestav) A tandem kinase fusion protein in *Aegilops tauschii* confers wheat leaf rust resistance
15:15-15:30	**Rosana Sánchez** (IBT-UNAM) The dynamics between the infection thread formation and cell division at the rhizobial-infection site in *P. vulgaris* roots
15:30-15:45	**Roxana Coreas** (University of California, Berkeley) Nano-omics enables pathogen infection in plants through stress biomarker enrichment in plants and crops within hours of infection
15:45-16:00	**Aurora Lara** (Facultad de Química-UNAM) Glucose-6-phosphate dehydrogenase is regulated via Cyclin/CDK phosphorylation during maize germination
16:00-16:15	**June Simpson** (Unidad Irapuato, Cinvestav) Inactive cell wall invertases are necessary for correct development of *A. thaliana* seedlings and affect aspects of sucrose metabolism
16:15-16:30	**Carlos Rodríguez** (Tecnológico de Monterrey) Integrating multi-omics data of Olive (*Olea europaea*) and transcriptomics of multiple Oleaceae species for tracing oleuropein biosynthesis
Flash talks	
16:30-16:45	**Alexis Cadena** (CICY) ROS dynamics among three *Agave angustifolia* Haw. Phenotypes
	Gema Hernández (CIBA-IPN) Phytochemical evaluation of *E. beecheyanum* a medicinal plant from the Purépecha Plateau
	Elizabeth Monroy (CCG-UNAM) EXPA1 and EXPA2, cell wall α-expansins, are required for symbiotic infection and nodule development in *Lotus japonicus*

	Catalina Bucarey (CCG-UNAM) Functional Roles of Pectin Methyl Esterase Isoforms in Intra- and Intercellular Symbiotic Infection of *Lotus japonicus*
	Diana Bravo (UMSNH) Fungal endophytes associated to *Asclepias curassavica* influence the oviposition and foraging in the monarch butterfly (*Danaus plexippus*)
	Sandra Adame (UMSNH) The role of volatiles emitted by the entomopathogenic fungus *Beauveria bassiana* in interaction with plants
	Pablo Suárez (Facultad de Química) Taxonomic and functional overlap of the coralloid root and seed microbiome from cycad *Dioon edule*
	Victoria Vela (Unidad Irapuato, Cinvestav) The role of mir156 and SQUAMOSA PROMOTER BINDING PROTEIN-LIKE transcription factors in Arabidopsis embryo development
16:45-17:15	Coffee break
17:15-18:30	Poster session (odd numbers)
Keynote session	
18:30-19:15	**Marisa Otegui** (University of Wisconsin Madison) Membrane remodeling by ESCRT proteins in plant cells

Thursday October 23	
	·Development and Evolution·
Chair	Chair, **Karolina Heyduk**
Keynote session	
09:00 - 09:45	**Alfredo Cruz** (Unidad de Genómica Avanzada, Cinvestav) Exploring the AINTEGUMENTA/PLETHORA–Auxin Feedback Loop: Insights from an EvoDevo Perspective on Stem Cell Niche Regulation
Plenary sessions	
09:45 - 10:15	**Robert VanBuren** (Michigan State University) Leveraging evolutionary innovations in grasses to develop climate resilient agriculture
10:15 - 10:45	**Ana Caicedo** (U Mass Amherst) Converging on competitiveness: the repeated evolution of seed shattering in weedy rice
10:45 - 11:15	**Joseph G. Dubrovsky** (IBT-UNAM) Conceptual developments and new players in lateral root initiation and morphogenesis
11:15 - 11:45	Coffee break

Plenary session

11:45 - 12:15	**Tzvetanka D. Dinkova** (Facultad de Química, UNAM) microRNA regulatory roles during de-differentiation and differentiation in maize Somatic Embryogenesis
12:15 - 12:45	**Paul Jarvis** (Oxford University) Protein import into chloroplasts and its regulation by the ubiquitin-proteasome system
12:45 - 13:15	**Zhiyong Wang** (Carnegie Institution for Science) The crosstalk between sugar and brassinosteroid regulates plant growth and acclimation
13:15 - 14:45	Lunch

Short talks

Chair, **Patricia Coello**

14:45 - 15:00	**Brenda López** (Instituto de Biología-UNAM) The unique shoot apical meristems in Cactaceae
15:00-15:15	**Renata Rivera** (CICY) Morpho-anatomical characterization of fruit dehiscence in Bixa orellana L.: implications for plant development and crop improvement
15:15-15:30	**Edgar Demesa** (Unidad Irapuato, Cinvestav) Imputation integrates single-cell and spatial gene expression data to resolve transcriptional networks in barley shoot meristem development
15:30-15:45	**Isabel Goldaracena** (Stanford University) Engineering multi-input protein logic gates for programmable gene expression control in plants
15:45-16:00	**Martin Heil** (Unidad Irapuato, Cinvestav) Resistance triggered by self-DNA: specificity and application potential of an incomprehensible immune response
16:00-16:15	**Arturo Guevara** (IBT-UNAM) Exploring the signaling pathway that controls arsenite (As [III]) responses in the root of *Arabidopsis thaliana* L.
16:15-16:30	**Beatriz Ruiz** (Unidad Irapuato, Cinvestav) Interactions Between the Transcription Factor BOL/DRNL/ESR2 and the Jasmonate Pathway
16:30-16:45	**Héctor Hugo Torres** (Stanford University) Understanding and engineering crown root developmental program in the C4 grass *Setaria viridis*
16:45-17:15	Coffee break
17:15-18:30	Business meeting

Keynote session

18:30-19:15	**Keiko Torii** (University of Texas) *Environmental control of stomatal development - From intricate modulation to innovation*
20:30-23:30	Final Banquet

Friday October 24

08:30-09:15	Departure

AMERICAN SOCIETY OF PLANT BIOLOGISTS
1924
INVENIAMVS VITALIA ARCANA PLANTARVM

CVTT Coordinación de Vinculación y Transferencia Tecnológica

SMB

CTR® SCIENTIFIC

ASOCIACIÓN MEXICANA DE SEMILLEROS, A.C.
AMSAC
Porque la vida en el campo comienza con una semilla.

SDI Secretaría de Desarrollo Institucional

Instituto de Biotecnología UNAM

Cinvestav Genómica Avanzada

Escuela Nacional de Estudios Superiores Unidad León

CICY

The Company of Biologists

CCG Centro de Ciencias Genómicas

BIOCIENTIFICA®

VISIT YUCATAN

· Table of Contents ·

KEYNOTE SESSION, October 20th

Siobhan M. Brady
University of California Davis

Environmental integration with root cell type development

University of California, Davis. Department of Plant Biology and Genome Center
1 Shields Ave. Davis, CA 95616. Email: sbrady@ucdavis.edu

A plant's roots serve as a major line of defense against environmental stress to protect the plant as a whole. Roots of diverse plant species have found ways to deal with stress by devising cell wall modifications and natural barriers to resist drought, flooding, mineral deficiencies, and other insults that impair plant growth. Many plant species have evolved unique cell wall forms composed of specialized biopolymeric metabolites that are deposited in elegant patterns in specific cell types. These walls are largely molecularly understudied although they are linked with a variety of stress responses. I will describe my group's approaches that merge classic developmental genetics with systems biology to elucidate these developmental programs in multiple plant species.

KEYNOTE SESSION, October 21st

Alejandra Covarrubias

Instituto de Biotecnología, UNAM

Dry times, tough proteins: Roles of LEA proteins in plant responses to water scarcity

Alejandra A. Covarrubias[1*], Inti Arroyo-Mosso[1], David Rendón-Luna[1], P. Sofía Romero-Pérez[1,3], Laura V. Martínez-Castro[1], Coral Martínez-Martínez[1,2], Dayana Martínez-Basulto[1], Alejandro Arroyo-Mosso[1], Francisco Campos[1], Cesar Cuevas-Velazquez[1], Alejandro Garciarrubio[1], Shahar Sukenik, Marisa S. Otegui[2], José Luis Reyes[1].

[1]Plant Molecular Biology Department, Instituto de Biotecnología, Universidad Nacional Autónoma de México. Cuernavaca, Mor. México. [2]Botanic Department, Michigan University, USA. [3]Department of Chemistry and Biochemistry, University of California at Merced, Merced, California, USA.
*Email: alejandra.covarrubias@ibt.unam.mx

During development, orthodox seeds undergo a distinctive process characterized by substantial water loss, eventually reaching a desiccated state. Despite the severity of this dehydration, this process is essential for successful seed germination. At the onset of dehydration, in the late stages of embryogenesis, these seeds accumulate a group of proteins known as Late Embryogenesis Abundant (LEA) proteins. LEA proteins also accumulate in response to water deficit in both vegetative and reproductive plant tissues. Due to their amino acid composition and physicochemical properties, LEA proteins are classified as Intrinsically Disordered Proteins (IDPs). Like other IDPs, they exhibit remarkable structural flexibility, enabing interactions with a wide variety of molecular components within the cellular environment. Our research has aimed to elucidate the roles of some of these proteins in plant responses to low water availability occurring during development or under environmental stress. We have focused on two highly conserved LEA protein families, LEA4 and LEA6, in the model plant Arabidopsis thaliana. Through genetic, biochemical, cellular and molecular approaches, we have gathered evidence that these proteins help prevent enzyme inactivation and further aggregation during in vitro dehydration. Our findings also indicate that their different physicochemical properties establish the conditions where they perform this protective effect. Proteins from both families acquire alpha helical structure under high osmolarity and/or macromolecular crowding. This structural flexibility allowed the development of a biosensor, based on a LEA4 protein, designed to detect osmotic stress effects in cells. Genetic analyses of Arabidopsis mutants deficient in these proteins revealed reduced tolerance to water deficit. Additionally, a LEA6 protein was shown to play key roles in maintaining the glassy state and longevity in seeds. Mutants in this gene are also affected in dormancy and desiccation tolerance. Using LEA proteins fused to fluorescent tags, we characterized their tissue and subcellular localization in seeds, during early post- germination development, and under water-limiting conditions. Notably, in imbibed seeds, AtLEA4 is not only abundant but also forms large aggregates that behave as biomolecular condensates. Similar condensate formation was observed in vegetative tissues under water deficit. In the cytosol, the protein exists in at least two distinct forms: as stable condensates and as a more dynamic soluble molecule. The relationship between the subcellular organization of these proteins and their functional roles will be further discussed. This work was partially supported by CONAHCyT grant CF-2023-I-503.

PLENARY SESSIONS, October 21st

Minerva Trejo Arellano
Institute of Science and Technology Austria

Leveraging the evolutionary information in genomic and epigenomic databases to reconstruct DNA methylation systems

Trejo-Arellano Minerva S.[1], Caisova Lenka[1], Hollwey Elizabeth[1], Lyons Dave[2], Zilberman Daniel[1*]

[1]Institute of Science and Technology, Am Campus 1, Klosterneuburg, 3400, Austria.
[2]Department of Cell & Developmental Biology, John Innes Centre, Norwich, NR4 7UH, UK.
[*]Email: mtrejoar@ist.ac.at; [*]Daniel.Zilberman@ist.ac.at

The basal unit of biological information is the DNA sequence. Furthermore, DNA nucleotides can be methylated, impacting genome structure and readout. DNA methylation can be epigenetically inherited over many generations, is crucial for plant and animal development, and its mis-regulation underlies several human diseases (Law & Jacobsen, 2010). Despite its importance, our understanding of DNA methylation is far from complete, partly because it is absent from the most common genetic model organisms (Ponger & Li, 2005). To overcome this challenge, we are leveraging the available genomic and epigenomic data to uncover the evolution of DNA methylation. We systematically sampled multiple databases to recover proteins bearing a functional catalytic DNA methyltransferase (DNMT) domain, particularly focusing on poorly studied branches of the tree of life. This data, combined with bisulfite sequencing of representative species has allowed to uncover ancestral patterns and configurations of eukaryotic DNA methylation, and their evolution across millions of years.

Ken Birnbaum
The Center for Genomics and Systems Biology
New York University, USA

One call, many responses: decoding auxin's cell-specific language

Auxin's role in mediating so many different responses in the plants is one of the central enigmas in plant tissue patterning. For example, in the root, auxin signaling has been implicated in the formation of the cap, root hairs, the quiescent center, and the specification of the xylem. How does one hormonal signal have a role in specifying so many different tissues? To add new tools to address this question, we have been developing high-throughput methods to dissect auxin signaling combining cell-based screening methods and new *in-silico* protein interaction approaches. For rapid cell-based screening of specific cell types, we developed a cell-based method that preserves much of a cell's endogenous identity while knocking out multiple targets using ribonuclear CAS9 complexes. The result showed that A-class ARFs have largely overlapping targets but the same ARF has vastly different downstream effects in different cell types. This supports a model in which ARFs interact with different localized transcription factors to mediate cell-type specific effects. To explore this possibility, we conducted a genome AlphaFold3 screen of the A-class ARFs against the entire genome. AlphaFold captured the known interactions with AUX/IAAs. In addition, independent DAP-seq data of genome wide transcription factor binding sites showed that top AlphaFold interactors also showed a significant over-representation of co-binding on promoters. In one case, our screen showed that ARF5 (*MONOPTEROS*, *MP*) interacted strongly with C-class ARFs 10 and 16, which have been implicated root cap maturation. Our transcriptional analysis of the ARF5 knockdown showed that mature columella identity was largely missing, in agreement with the ARF10/16 double mutant phenotype. This leads to a model in which the broadly expressed A-class ARF5 interacts in a subset of its domain with the C-class transcription factors ARF10 and 16 to specifically control columella identity. Overall, the unbiased screen shows ARFs interacts most strongly with transcription factors that are highly cell-type specific. The results suggest many potential avenues by which ARFs could mediate different processes in different cells.

Damien Formey

Center for Genomic Sciences, UNAM

Phabase: A high-quality genome assembly and platform for *Phaseolus vulgaris* cv. Negro Jamapa

Heladia Salgado Osorio[1], Emmanuel Pacheco[1], Eber Daniel Villa Rodríguez[2], Lavinia Ioana Fechete[2], Turgut Yigit Akyol[2], Jesús Montiel González[1]*, Damien Formey[1]*

[1]Center for Genomic Sciences, UNAM, Cuernavaca, Morelos, Mexico.
[2]Aarhus University, Aarhus, Denmark.
*Email: formey@ccg.unam.mx.

The common bean (Phaseolus vulgaris) is one of the most consumed legumes, distributed in two major gene pools: the Andean and Mesoamerican. Here, we present a high-quality genome assembly of the Mesoamerican cultivar Negro Jamapa, widely cultivated in Mexico. Using PacBio long-read sequencing and advanced assembly pipelines, we generated a chromosome-level assembly with improved contiguity and accuracy compared to the current Andean reference genome (G19833). Genome completeness assessments (BUSCO) revealed higher representation of conserved orthologous genes in the Negro Jamapa assembly. Structural comparisons highlighted significant variations between the Mesoamerican and Andean genomes, underscoring the genetic diversity within P. vulgaris. This new genomic resource has been incorporated into an integrated platform, named Phabase, that includes tools for visualizing genomic assemblies, exploring transcriptomic and microtranscriptomic data, performing sequence alignments, facilitating data download and enabling comprehensive comparisons between different Phaseolus genomes. Designed for continuous expansion, this platform will incorporate additional Phaseolus genomic and transcriptomic resources in the future. Together, these efforts provide a valuable foundation for advancing functional genomics and harnessing the genetic potential of common beans.

Dan Chitwood
Michigan State University

Iconography and indigenous knowledge in the De la Cruz-Badiano nahuatl herbal (1552): a text mining and morphometric study of Mexican plants

J. Noé García-Chávez[1], Mariana J. Ruíz-Amaro[1], Jimena Jazmin Hurtado-Olvera[1], Kylie DeViller[3], Lachlann Simms[2], Plants & Python class of 2023, Zoë Migicovsky[2], Robert VanBuren[4], Alejandra Rougon-Cardoso[1], and Daniel H. Chitwood[5]*

[1]Laboratory of Agrigenomic Sciences, ENES Unidad León, Universidad Nacional Autónoma de México, México. [2]Department of Biology, Acadia University, Wolfville, Nova Scotia, Canada. [3]Jodrey School of Computer Science, Acadia University, Wolfville, Nova Scotia, Canada. [4]Plant Resilience Institute, Michigan State University, East Lansing, MI, USA/Department of Plant Biology, Michigan State University, East Lansing, MI, USA. [5]Department Computational Mathematics, Science & Engineering, Michigan State University, USA.
*Email: dhchitwood@gmail.com.

From ahuacatl to chīlli, many of the world's most economically important plants trace their names and cultural significance to Nahuatl, the language of the Mexica and other Indigenous peoples of central Mexico. These words have transcended linguistic boundaries—embedding themselves not only in Spanish but in many languages worldwide—through the global spread of these plants and their culinary and medicinal traditions. This persistence of Nahuatl terminology is more than linguistic: it reflects the enduring Indigenous botanical knowledge systems that resisted and survived European colonization. One of the most remarkable documents preserving this knowledge is the Libellus de Medicinalibus Indorum Herbis, commonly known as the De la Cruz-Badiano Herbal of 1552. This manuscript provides a rare window into a pre-Hispanic classification system, offering plant names in Nahuatl alongside Latin and stylized botanical illustrations. In this study, we interpret the Herbal as both a medical manuscript and a repository of Indigenous iconographic data. We present a text mining analysis of the Nahuatl plant names and their embedded associations with English and Spanish words, as well as a morphometric analysis of leaf shapes from the manuscript's illustrations. Our findings shed light on the structure of Nahua botanical classification and how it reflects cultural, medicinal, and economic understandings of the natural world. By reinterpreting this colonial-era document through computational and visual analysis, we aim to highlight the sophistication of Indigenous botanical knowledge and contribute to ongoing conversations about decolonizing plant science. Project website: https://danchitwood.github.io/DeLaCruzBadiano1552/

PLENARY SESSIONS, October 21st

Azalea Guerra García

Cinvestav Unidad Irapuato

Genetic diversity of orphan crops and wild relatives: a foundation for resilient agroecosystems

Departamento de Biotecnología y Bioquímica, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN), Irapuato, México.
Email: azalea.guerra@cinvestav.mx

Domestication is a dynamic and non-linear process shaped by both human selective pressures and natural evolutionary forces, resulting in the rich genetic and phenotypic diversity observed in cultivated species. In centers of domestication, wild and feral populations, landraces, breeding lines, and even genetically modified varieties coexist and interact, often exchanging genetic material. Growing evidence shows that gene flow from wild relatives has played a key role in enhancing the adaptive potential of crops, contributing valuable genetic diversity. As we face global challenges such as climate change, emerging pathogens, and declining crop diversity, it is essential to explore and harness the full breadth of genetic variation within crop gene pools—including landraces and wild relatives adapted to diverse environments. In the scarlet runner bean (*Phaseolus coccineus*), a Mesoamerican orphan legume, we identified a severe genetic bottleneck associated with domestication, followed by asymmetric gene flow from wild populations that helped restore genetic diversity in cultivated forms. Traditional varieties of the year bean (*P. dumosus*) not only have a broader distribution than their wild relatives but have also significantly increased their genetic diversity, likely because of the interaction and gene flow with other domesticated *Phaseolus* species in milpas of southern and central Mexico. In lentil (*Lens culinaris*), landraces from the Middle East and South Asia exhibit strong regional differentiation, while wild lentil (*L. orientalis*) populations show no clear geographic structure. Instead, their distribution is better explained by climatic variables, suggesting local adaptation to specific environmental conditions. These findings underscore the importance of conserving and utilizing traditional varieties and wild relatives to maintain useful genetic diversity and agrobiodiversity. However, significant gaps remain in germplasm collections, and many orphan crops and their wild counterparts are still underrepresented and unexplored.

Zachary Lippman
Cold Spring Harbor Laboratory, USA

Hidden genetic complexities and phenotypic consequences of evolving plant stem cell networks in nature and agriculture

Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, USA.
Howard Hughes Medical Institute, Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, USA.
Email: lippman@cshl.edu

Decades of research on the few model systems that dominate plant studies have revealed the genetic and molecular mechanisms that govern stem cell control and the natural and agricultural traits they influence. While the core factors and their interconnected regulation are highly conserved, recent advances in pan-genomes and genome editing have led to the emergence of new model systems that uncover previously hidden complexities of stem cell control within and between lineages. I will present our recent findings on using pan-genomics and pan-genetics in the Solanaceae family to expose critical roles for gene duplications, their diversifications, and interactions among stem cell regulators controlling flowering, inflorescence architecture, and fruit size. All of these findings have implications for both the evolvability of phenotypes in nature and the engineering of traits in agriculture.

KEYNOTE SESSION, October 21st

Sue Rhee
Michigan State University

Drug discovery for agriculture: Center for Sustainable Plant Innovation and Resilience through International Teamwork (C-SPIRIT)

Department of Plant, Soil, and Microbial Sciences, Michigan State University, East Lansing, MI, USA.
Email: rheeseu6@msu.edu

The Center for Sustainable Plant Resilience and Innovation through International Teamwork (C-SPIRIT) unites experts from the U.S., Canada, Japan, Korea, and the U.K. to develop sustainable, nature-based solutions that boost crop resilience and stabilize global food production amid increasing environmental stressors. By harnessing the chemical diversity of plants and microbes, C-SPIRIT aims to create green chemistry alternatives to synthetic fertilizers and pesticides, enhancing plant and soil health while reducing reliance on non-sustainable practices. Its multidisciplinary research integrates genomics, AI, and synthetic biology to discover, synthesize, and validate bioactive metabolites that protect crops from drought, heat, and fungal pathogens. C-SPIRIT will focus on seven major crops including maize, rice, soybean, potato, tomato, pennycress, and cassava. Through collaboration with industry and agricultural stakeholders, public engagement, and international training programs, the Center ensures its innovations are scalable, accessible, and contribute to a more sustainable global bioeconomy. In this talk, I will describe our research pipeline, some preliminary results, and a future outlook.

KEYNOTE SESSION, October 22nd

Cyril Zipfel
University of Zurich

Roles and recognition of stress-regulated secreted signaling peptides

[1]Institute of Plant and Microbial Biology, Zürich-Basel Plant Science Center, University of Zürich, 8008 Zürich, Switzerland.
[2]The Sainsbury Laboratory, University of East Anglia, Norwich Research Park, Norwich, United Kingdom.
Email: cyril.zipfel@uzh.ch

Plants must constantly adapt to changing environmental conditions. Cell-surface receptor kinases play an important role in the sensing of stimuli originating from the environment itself or that are produced endogenously often in response to such external cues. I will present our recent work on stress-regulated secreted signaling peptides, their perception mechanisms, and diverse functions in adaptive stress responses.

Jeongmin Choi
University of Cambridge

Identification of a suppressor of arbuscular mycorrhizal symbiosis in high phosphate conditions

Saskia Birch[a], Sophie Perryman[a], Nikoline Mekjan[a], Evan Elise[a], Thomas Ralfs[a], Miles Bate-Weldon[b], Alex Williams[b], Boas Pucker, Giles Oldroyd[b], Boas Pucker[c], Emma Wallington[d], Katie Field[b], and Jeongmin Choi[a,*]
[a]Crop Science Centre, Department of Plant Sciences, University of Cambridge, Cambridge, CB3 0LE, UK.
[b]School of Biosciences, University of Sheffield, Sheffield, S10 1AE, UK.
[c]Institute of Cellular and Molecular Botanics (IZMB), University of Bonne, Boone, 53115, Germany.
[d]National Institute of Agricultural Botany, Park Farm, Villa Road, Histon, Cambridge, CB24 9NZ, UK.
*Email: jc913@cam.ac.uk

Phosphate is an essential mineral nutrient for plants. However, the majority of phosphate remains inaccessible to plants as it forms complexes with organic and inorganic matter. To mineralise these phosphate complexes beyond the root zone, approximately 80% of land plants form beneficial associations with the soil fungi arbuscular mycorrhizae (AM). AM fungi provide phosphate, along with nitrogen and many other mineral nutrients, to plants in exchange for carbon-based photosynthates used for fungal nourishment and spore production. However, AM symbiosis is a highly dynamic process influenced by many environmental factors. For example, high phosphate levels in soil (e.g., after fertiliser application) strongly suppress AM symbiosis, limiting the application of AM symbiosis in modern agriculture. Recently, the molecular mechanisms of phosphate suppression have begun to be unveiled. Internal phosphate sensors (SPXs) and the transcription factors (PHRs) that drive the Phosphate Starvation Response directly regulate AM colonisation in a phosphate-dependent manner. However, genetic perturbation of these genes did not entirely suppress AM symbiosis, suggesting the presence of additional unknown components. Here, I will present a novel negative regulator of arbuscular mycorrhizal (AM) symbiosis that is responsible for the collapse of AM symbiosis under high phosphate conditions. By taking a genetics, molecular biology, and transcriptome approach, we discovered that the loss-of-function mutant retained AM colonisation even under high phosphate treatment by maintaining the phosphate starvation response. Together with the known molecular players, our findings support the idea that plants actively regulate arbuscular mycorrhizal (AM) symbiosis by fine-tuning the phosphate-sensing and signalling pathways.

PLENARY SESSIONS, October 22nd

Jean-Michel Ané

University of Wisconsin-Madison

Biological nitrogen fixation on the aerial roots of maize and sorghum for sustainable agriculture

R.E. Venado[1], R. Diogo[1], D. Laspisa[2], V. Infante[1], J. Wilker[1], K. Gibson[1], Emily Wolf[3], Saddie Vela[3], C.I. Calderon[4], J. Wallace[2,5], W. Vermerris[3,6], and, J.M. Ané[1,4]*

[1]Department of Bacteriology, University of Wisconsin-Madison, Madison, WI, U.S.A
[2]Institute of Plant Breeding, Genetics, and Genomics, University of Georgia, Athens, Georgia, U.S.A
[3]Plant Molecular and Cellular Biology Graduate Program, University of Florida, Gainesville
[4]Department of Plant and Agroecosystem Sciences, University of Wisconsin-Madison, Madison, Wisconsin, U.S.A
[5]Department of Crop & Soil Sciences, University of Georgia, Athens, Georgia, U.S.A
[6]Department of Microbiology & Cell Science and UF Genetics Institute, University of Florida, Gainesville, Florida, USA
*Email: jeanmichel.ane@wisc.edu

Cereals, such as Maize (*Zea mays*) and Sorghum (*Sorghum bicolor*), are essential crops that sustain millions of people and serve as livestock feed. Sorghum also holds promise as a sustainable bioenergy crop due to its high biomass yield. Our research has demonstrated that particular maize landraces from southern Mexico derive 29%–82% of their nitrogen from the atmosphere by harboring nitrogen-fixing (diazotrophic) communities in a mucilage produced by their aerial roots after rainfall. More recently, we have demonstrated similar nitrogen-acquiring traits in specific sorghum accessions. Through acetylene reduction, ^{15}N gas enrichment, and ^{15}N isotope dilution assays, we have shown that the sorghum plants with this trait can acquire nitrogen from the atmosphere through diazotrophs in the mucilage, obtaining approximately 43% of their nitrogen from this process. The mucilages of maize and sorghum are similar and produced by border cells that detach from the aerial root cap, resembling the process on underground roots. However, border cells are more abundant and larger in the mucilage from aerial roots than in underground ones. We have conducted a comprehensive study of the aerial root development in various maize landraces and sorghum accessions, elucidating the influence of both genetic and environmental factors. Using QTL and GWAS approaches, we have identified genetic regions governing this trait in maize and sorghum.Contrary to our initial expectations, humidity, and soil nitrogen levels were found to have only a limited impact on aerial root development in these genotypes. Our research also isolated over 400 distinct diazotrophs from maize and sorghum mucilage, along with the development of non-fixing mutants as controls and ammonium-excreting mutants from some of these isolates.The unique symbiotic relationship between diazotrophs and the aerial roots of maize and sorghum presents an encouraging opportunity to leverage biological nitrogen fixation, thereby reducing reliance on synthetic fertilizers and fostering more sustainable and productive agricultural practices.

Clelia de la Peña

Centro de Investigación Científica de Yucatán

Integrative study of *in vitro* albinism in *Agave angustifolia*: transcriptomic, epigenetic, and metabolomic evidence

Andrade-Marcial, Mauricio, Aguilar-Méndez, Edder D., and De-la-Peña, Clelia*

Unidad de Biotecnología, Centro de Investigación Científica de Yucatán, Calle 43 No. 130 x 32 y 34. Col. Chuburná de Hidalgo, 97205 Mérida, Yucatán, México.
*Email: clelia@cicy.mx

The *in vitro* propagation of *Agave angustifolia* has enabled significant progress in large-scale plant production; however, it has also revealed the emergence of albino phenotypes as a consequence of somaclonal variation. This presentation showcases integrative findings obtained through physiological, transcriptomic, epigenetic, and metabolomic approaches aimed at elucidating the alterations in chloroplast biogenesis in albino plants. Our analyses reveal that albino phenotypes exhibit a complete loss of photosynthetic efficiency, along with a drastic reduction in pigment accumulation. At the molecular level, we observed significant deregulation of genes encoding pentatricopeptide repeat (PPR) proteins involved in plastid differentiation, as well as marked changes in DNA methylation patterns and histone modifications. Additionally, metabolic profiling indicates disruption in key pathways such as flavonoid biosynthesis, lipid metabolism, and aromatic amino acid biosynthesis. These findings point to a complex mechanism in which epigenetic and transcriptional factors—possibly induced by *in vitro* conditions—converge to impair chloroplast development. Understanding this phenomenon not only provides fundamental insight into phenotypic plasticity in *Agave*, but also offers valuable perspectives into the regulation of chloroplast biogenesis and the molecular bases of photosynthesis disruption in plants.

Robertas Ursache

Centre for Research in Agricultural Genomics

GELP proteins as architects of essential plant biopolymers

Luca Piccinini[1], Fabien Nirina Ramamonjy[1], Vanessa Reyes Loaiza[1], Olga Serra Figueras[2], and Robertas Ursache[1]*

[1]Centre for Research in Agricultural Genomics (CRAG), CSIC-IRTA-UAB-UB, Barcelona, 08193 Spain.
[2]Laboratori del Suro, Department of Biology, University of Girona, Campus Montilivi, Girona, 17003 Spain.
*Email: robertas.ursache@cragenomica.es

The seed coat and leaf epidermis develop lipid-based protective barriers composed of cutin and suberin, which are essential for seed dormancy, germination, longevity, and leaf water retention. While significant progress has been made in elucidating the suberin and cutin biosynthetic pathways, the enzymatic machinery responsible for their precise assembly and tissue-specific deposition remains poorly understood. This presentation will highlight the role of GDSL esterase/lipase (GELP) proteins in the assembly of seed coat and leaf surface biopolyesters. Using a combination of optimized clearing and staining techniques, tissue-specific transcriptomics, and metabolomics approaches, we identify GELPs as key enzymes of cutin and suberin polymerization in distinct plant organs and tissues. Understanding the molecular mechanisms underlying GELP-mediated biopolymer formation offers a framework for improving stress resilience in plants.

Samuel Zeeman
Institute of Molecular Plant Biology, ETH Zurich

Beyond simple storage: revealing the cellular machinery and processes underpinning starch granule biosynthesis

Institute of Molecular Plant Biology, ETH Zurich, Switzerland. HPT E78, Auguste-Piccard-Hof 1, 8093 Zürich, Switzerland.
Email: szeeman@ethz.ch

Starch accumulates in the plastids of green algae and plants. It is critically important, both for the plant and for human use. The value of starch crops (e.g., cereal and legume seeds, storage roots, and tubers) is clear, while the links between starch metabolism and plant growth, development, and environmental responses are still being explored. Chemically, starch is simple. Its major component, amylopectin, is a branched glucose homopolymer, similar to glycogen - the soluble storage polymer in animals, fungi, and prokaryotes. However, amylopectin is structurally more complex, due to the coordinated actions of multiple enzymes and isozymes. This produces a polymer that undergoes a phase transition, forming insoluble, semi-crystalline granules that can grow substantially in size. *Arabidopsis thaliana* serves as a powerful model for investigating starch biosynthesis. In leaf chloroplasts, starch granules form between thylakoid membranes. Using electron tomography, isotope labelling, and NanoSIMS imaging, we showed how starch granules initiate and grow. Through biochemical and genetic analyses, we identified proteins involved in granule initiation, with their modulation significantly affecting granule number. One such protein, MFP1 (MAR BINDING FILAMENT-LIKE PROTEIN 1), is anchored to thylakoid membranes and forms discrete domains. MFP1 directs the location of granule formation, and re-localizing it shifts starch deposition sites. Several biosynthetic proteins become embedded in granules during growth. Two of these, LIKE EARLY STARVATION 1 and EARLY STARVATION 1, have a conserved carbohydrate-binding surface. Functional studies in yeast and plants suggest they promote the transition of amylopectin from a soluble to a semi-crystalline phase, stabilizing granules—challenging the view that crystallization occurs spontaneously. These insights into the cell biology of starch biosynthesis reveal new gene targets to enhance starch yield and properties in crops.

Berkley Walker
Michigan State University

Photorespiration as a crossroads between photosynthesis, metabolism, and leaf energetics

Berkley Walker*, Xinyu Fu, Kelem Gashu, Kaila Smith, Anne Steensma, Ludmila Roze, and Joshua Vermaas

Michigan State University, East Lansing, Michigan, United States of America.
*Email: berkley@msu.edu

Photorespiration is a key component of plant metabolism that recycles inhibitory metabolites produced as a byproduct when the CO_2-fixing enzyme, rubisco, reacts with oxygen instead of CO_2. Because of photorespiration's sensitivity to CO_2 concentrations and temperature, it is an important driver of plant response to climate change. Photorespiration requires 30-40% of photosynthetic energy in illuminated leaves and releases carbon dioxide, greatly reducing rates of net carbon assimilation in many plant species and comprising a large portion of total metabolic flux. Photorespiration has been a perennial target for crop improvement, but many questions about its *in vivo* function remain unresolved. For example: What is the role of photorespiration in photoprotection? What is the temperature response of the photorespiratory pathway itself, and how does it adapt to changing conditions? Do photorespiratory intermediates, like glycine and serine, proceed according to the "textbook pathway" or are they diverted to support amino acid synthesis in planta? and, Does the large flux of carbon in photorespiration supply other aspects of metabolism like one-carbon synthesis? The answers to these questions are important for designing "next generation" improvement strategies for photorespiration and for understanding the fundamental responses of plants to future climates. Here I discuss our progress addressing these questions using gas exchange measurements, biophysical measures of photosynthetic energy use, metabolic flux analysis, and reaction-kinetic simulations of photorespiration.

KEYNOTE SESSION, October 22nd

Marisa Otegui
University of Wisconsin-Madison

Membrane remodeling by ESCRT proteins in plant cells

Center for Quantitative Cell Imaging and Department of Botany, University of Wisconsin-Madison.
Email: otegui@wisc.edu

The dynamic flow of proteins to and from the plasma membrane and between organelles is and crucial for cellular function, signaling, development. We study the cellular functions and biophysical properties of ESCRT proteins to uncover the fundamental principles governing membrane remodeling in plants and other organisms. ESCRT proteins are essential for endosomal trafficking of plasma membrane molecules as well as many other crucial cellular events. However, we are still far from understanding how ESCRTs mediate membrane remodeling in the context of all their many cellular functions. Using live imaging, electron tomography, genetics, biochemistry, structural biology, and computational modeling we analyze endosomal membrane vesiculation in several plant lineages and other organisms to understand the common principles that underly ESCRT membrane bending. We also have uncovered novel functions of ESCRT proteins in cell division and nuclear dynamics. These findings are helping us to elucidate basic mechanisms essential for cell function and plant development, with broader implications for cell biology and plant biotechnology.

Alfredo Cruz Ramírez
Unidad de Genómica Avanzada, Cinvestav

Exploring the AINTEGUMENTA/PLETHORA–auxin feedback loop: insights from an evoDevo perspective on stem cell niche regulation

Molecular and Developmental Complexity Group. Advanced Genomics Unit of CINVESTAV, Irapuato; México.
Email: alfredo.cruz@cinvestav.mx

AINTEGUMENTA-LIKE/PLETHORA/BABYBOOM (*APB*) genes are recognized as part of the ancient developmental toolkit shared among land plants. In *Arabidopsis thaliana*, these transcription factors are activated by auxin and predominantly expressed in actively dividing tissues, underscoring their vital role in organ formation. The auxin signaling pathway itself is an evolutionarily conserved mechanism present since the earliest land plants, where it plays a fundamental role in regulating growth and development. In *Marchantia polymorpha*, a liverwort representing one of the most basal land plant lineages, a single APB ortholog, *MpAINTEGUMENTA* (*MpANT*), is encoded in the genome. This gene was the focus of our study aimed at elucidating its function. We found that *MpANT* expression is largely confined to meristematic regions, where cell division is most active. Expression assays confirmed its sensitivity to auxin and auxin transport inhibitors, indicating that *MpANT* functions downstream of auxin signaling and Auxin Response Factors (ARFs). Functional studies of both loss- and gain-of-function *MpANT* alleles highlight its essential role in maintaining meristem identity and promoting cell proliferation. Furthermore, *MpANT* appears to act upstream of the auxin transporter *MpPIN1*, influencing auxin distribution within developing tissues. Collectively, our findings reveal a feedforward regulatory loop involving auxin, *MpANT*, and *MpPIN1* that is critical for proper development in *Marchantia*. This work underscores how key components of the auxin signaling network—including both the pathway and downstream targets such as APB genes—have been conserved since the early evolution of land plants.

PLENARY SESSIONS, October 23rd

Robert VanBuren

Michigan State University

Leveraging evolutionary innovations in grasses to develop climate resilient agriculture

Department of Plant Biology. Department of Plant, Soil, and Microbial Sciences.
Plant Resilience Institute, Michigan State University.
Email: vanbur31@msu.edu

Drought is the most pervasive issue we face in agriculture today, and water deficit is responsible for most of the global loss in yield and quality. Work in the VanBuren lab focuses on exploring the mechanisms that plants use to combat drought stress in natural and agricultural settings. We leverage large-scale, high-resolution datasets across space and time to model and predict the genetic elements, physiological signatures, and alleles controlling drought tolerance phenotypes. Our work centers on the C4 cereals maize, sorghum, teff, and various millets as well as numerous wild and model species with unique evolutionary innovations.

Joseph Dubrovsky

Universidad Nacional Autónoma de México

Conceptual developments and new players in lateral root initiation and morphogenesis

Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México (UNAM), Cuernavaca, 62210, Morelos, Mexico joseph.dubrovsky@ibt.unam.mx

Primary, adventitious, basal, and lateral roots produce new roots through a reiterative process of the formation of lateral roots (LRs) which are the building blocks of root systems involved in multiple functions important for plant life. In this plenary lecture I will outline new concepts in the field of LR formation and will discuss new players involved in LR initiation (LRI) and morphogenesis. Founder cell (FC) identity acquisition precedes LRI. We have established that LRI starts from a single FC followed by auxin-dependent recruitment of other FCs. Therefore, while FC recruitment is taking place, the LR primordium (LRP) morphogenesis is already in progress. This challenges the paradigm of separation of LRI and morphogenesis. As a single FC starts LRI, Stage 0 of LRP is recognized, resulting in the first division of FC. We established that the first cell divisions in LRP are extremely short, the shortest in plants, which is essential for lateral inhibition and acropetal pattern of LRI. We found that during LRP morphogenesis, its core and flanks show different cell proliferation capacity. In search of new factors involved in these processes, using biosensors, we addressed the role of anionic phospholipids, phosphoinositides (PI4P & PI(4,5)P_2) and phosphatidylserine (PS), in LRI and LRP morphogenesis. We established that specific spatiotemporal dynamic changes in anionic phospholipids play important role in orchestration of these processes. Particularly, PI(4,5)P_2 & PS are actively involved in graded differentiation of pericycle and LRI. Also, PI(4,5)P_2 promotes LRI and LRP morphogenesis. The latter process also depends on epigenetic regulation by an H3K4 methyltransferase ARABIDOPSIS HOMOLOG OF TRITHORAX1 (ATX1) which controls LRP dome shape through RedOx control mediated by *PEROXIDASE35*. I will outline respective mutant phenotypes and describe what morphogenetic processes are affected in *atx1* mutants. I thank all my collaborators, DGAPA-PASPA-UNAM (sabbatical support) and DGAPA-PAPIIT-UNAM (grant IN203024).

Ana Caicedo
University of Massachusetts Amherst

Converging on competitiveness: the repeated evolution of seed shattering in weedy rice

Ana Caicedo[1*], Xiang Li[1],

[1]University of Massachusetts Amherst, Amherst, MA, U.S.A., 611 N. Pleasant St., caicedo@umass.edu

Weedy rice (*Oryza* spp.), also known as red rice, is a weedy type of rice that invades cultivated rice (*Oryza sativa* L.) fields all around the world. Multiple studies have shown that weedy rice has evolved many times independently from a variety of ancestral backgrounds. However, most weedy rice origins have occurred through de-domestication of cultivated rice varieties. Seed shattering — that is, the easy release of seeds from the panicle — is one of the most important traits distinguishing weedy rice from cultivated rice, as it enables efficient seed dispersal in the weed. Weedy rice origins imply that the seed shattering trait was necessarily lost during domestication and then regained during weed evolution. Using comparative histology approaches we have shown that modifications to the abscission zone (AZ), a specialized cell layer in the rachilla, underlie both loss of shattering during domestication and regaining of shattering during weed evolution, but the nature of the modifications differs across domesticated-weedy lineages. QTL mapping using weed x crop ancestor crosses, have revealed that mutations in different loci are responsible for the convergence of the seed shattering trait in different weedy rice populations. Likewise, comparative transcriptomics of the AZ region and surrounding tissues indicate that most genes exhibiting AZ-region exclusive expression patterns do not overlap between weedy lineages. Despite lack of genetic convergence, expression data suggests that transcription factors related to the repression of lignin and secondary cell deposition have played a role in the modification of the AZ in multiple weedy lineages. Our findings suggest genetic flexibility in shaping AZ morphology, with some genetic constraints on AZ identity determination, and shed light on the evolutionary lability of this competitive agricultural weed.

Tzvetanka Dimitrova
Universidad Nacional Autónoma de México

microRNA regulatory roles during de-differentiation and differentiation in maize somatic embryogenesis

Tzvetanka D. Dinkova[1*], Eduardo Luján-Soto[1], Nahum E. Zambrano-Zepeda[1], Mayra Aquino-Luna[1], Vasti T. Juárez-González[2], Brenda A. López-Ruiz[3], Ma. Teresa de Jesús Olivera-Flores[1]

[1]Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México, Ciudad de México 04510, México. [2]Department of Plant Biology, Uppsala BioCenter, Swedish University of Agricultural Sciences and Linnean Center for Plant Biology, Uppsala, 75007, Sweden. [3]Jardín Botánico, Instituto de Biología, Universidad Nacional Autónoma de México, Ciudad de México 04510, México.
[*]Email: cesy@unam.mx

Somatic embryogenesis (SE) is an alternative plant reproduction process, where differentiated somatic cells restructure their fate to develop totipotency. High concentrations of the synthetic auxin 2,4-D are used to promote SE *in vitro*. During the past decade, “molecular omics” provided potential biomarkers for successful accomplishment of SE induction and development. Between them, microRNAs have emerged as essential regulators of gene expression necessary to fulfill developmental reprograming. We used a Mexican maize synthetic variety VS-535 from Tuxpeño landrace, to explore the regulatory role of development and stress-related miRNAs during immature embryo de-differentiation, callus embryogenic potential acquisition, and plant differentiation. Initial transcriptomic and small RNA omic comparisons revealed contrasting accumulation levels for miR156, miR167, miR390 and miR528, between the immature embryo explant and de-differentiated embryogenic callus (EC). miR156, miR167 and miR390 are land plant conserved miRNAs that regulate transcription factors involved in development and auxin responses, while miR528 is a monocot-specific miRNA involved in stress responses. Noticeably, miR528 is the most abundant miRNA in maize EC. miRNA and their target levels were analyzed at short temporal stages during embryogenic callus (EC) induction and after proliferation establishment. Their accumulation displayed contrasting patterns during dedifferentiation from embryogenic and non-embryogenic explants. When differentiation was promoted from embryogenic tissues, miR390 and miR156 showed the earlier switches associated with plant regeneration. To confirm the relevance of miRNA-mediated regulation in embryogenic potential maintenance, short tandem target mimics (STTM) were used. A closer inspection on miR528 transcription regulation revealed an interconnection of auxin transcriptional responses with upstream and downstream miRNA regulators. Such interconnection provides insights on the relevance of precise temporal and possibly local miRNA accumulation for successful dedifferentiation and differentiation processes in maize SE. Acknowledgement to financial support: PAPIIT: IN218921, IN212024; SECIHTI: CF-2023-1-115; FQ: PAIP 5000-9118.

Paul Jarvis
University of Oxford

Protein import into chloroplasts and its regulation by the ubiquitin-proteasome system

Section of Molecular Plant Biology, Department of Biology, University of Oxford, Oxford OX1 3RB, UK.
Email: paul.jarvis@biology.ox.ac.uk

The development and operation of chloroplasts (or other members of the plastid family of plant organelles) requires the participation of thousands of different organellar proteins. Most chloroplast proteins are nucleus-encoded and synthesized in the cytosol in precursor form. These preproteins pass through multiprotein import machines in the organelle's outer and inner envelope membranes – these are called the TOC and TIC translocons, respectively. Receptor components of the TOC complex exist in multiple isoforms, and genetic studies in the model plant *Arabidopsis* have indicated that the isoforms have distinct preprotein recognition specificities and function in different import pathways. Operation of such client-specific import pathways controls the organelle's proteome and functions, and plays a role in the differentiation of different plastid types (e.g., chromoplast formation in ripening tomato fruit). Our work has shown that the chloroplast protein import machinery is proteolytically regulated by direct action of the ubiquitin-proteasome system (UPS), in a process termed "chloroplast-associated protein degradation" (CHLORAD). The CHLORAD machinery has three key components: the SP1 ubiquitin E3 ligase, the SP2 β-barrel channel protein, and the CDC48 AAA+ ATPase. The SP1 and SP2 proteins form a complex in the outer envelope membrane, and respectively mediate the ubiquitination and extraction (or retrotranslocation) of TOC protein targets. The CDC48 protein, located in the cytosol, provides the energy that drives the retrotranslocation step, delivering the target proteins to the cytosolic 26S proteasome for degradation. Recent advances in our understanding of the mechanisms and functions of CHLORAD will be discussed.

Zhiyong Wang
Carnegie Institution for Science, Stanford

The crosstalk between sugar and brassinosteroid regulates plant growth and acclimation

Carnegie Institution for Science, Stanford, CA 94305, USA.
Email: zywang24@stanford.edu

Plant growth is driven by both sugar and growth-promoting hormone brassinosteorids (BR). The interplay between sugar and BR is crucial for plant growth regulation. BR promotes growth by activating the BRI1 receptor kinase, which triggers a phosphorylation cascade that leads to inactivation of the GSK3-like kinase BIN2, activation of the BZR1 family of transcription factors, and expression of BR-responsive genes. We have identified hundreds of BIN2 substrates and thousands of BZR1 target genes, illustrating a detailed BR signaling network. To understand how sugar and BR co-regulate plant growth, we study the crosstalk between sugar and BR signaling pathways. We have demonstrated that sugar signaling through the Target Of Rapamycin (TOR) pathway prevents BZR1 degradation to enhance BR-responsive gene expression and growth. Starvation increases the level of the UPL3 ubiquitin ligase, which interacts with BZR1 and promotes BZR1 degradation, thereby inhibiting growth and improving survival under stresses. To maintain sugar homeostasis, BR not only promotes sugar consumption (growth) but also promotes gluconeogenesis. Sugar availability is also monitored by protein O-GlcNAcylation (O-linked-N-acetylglucosamine, O-GlcNAc) and O-fucosylation (O-fucose). In Arabidopsis, two homologous proteins, SPINDLY (SPY) and SECRET AGENT (SEC), catalyze protein O-fucosylation and O-GlcNAcylation, respectively. Our proteomic studies have identified hundreds of nucleocytoplasmic proteins modified by O-GlcNAc and O-fucose. A significant portion of these O-glycosylated proteins are also substrates of BIN2 kinase, suggesting network-wide crosstalk between sugar-sensing O-glycosylation and BR-regulated phosphorylation. Our research unveils a complex network of sugar-BR interplay that regulates and optimizes plant growth and acclimation.

KEYNOTE SESSION, October 23rd

Keiko Torii
University of Texas at Austin

Environmental control of stomatal development - From intricate modulation to innovation

Howard Hughes Medical Institute and Department of Molecular Biosciences, The University of Texas at Austin, USA.
The Institute of Transformative Biomolecules, Nagoya University, Japan.
Email: ktorii@utexas.edu

Stomata, cellular valves on the plant epidermis, serve as critical interface between plant and atmosphere. The presence of stomata is critical for plant growth, survival and water-use efficiency, and it further impacts global carbon and water cycles. In the past two decades, molecular genetic studies in the model plant Arabidopsis unraveled the key regulators of stomata differentiation and the mechanism that ensures proper differentiation and patterning of stomata. The development of stomata occurs through a consecutive action of 'master-regulatory' basic-helix-loop-helix (bHLH) transcription factors, SPEECHLESS (SPCH), MUTE, and FAMA, all of which form a heterodimer with a partner bHLH, SCREAM (SCRM). Plants respond to diverse environmental cues and adjust the numbers and density of stomata in order to optimize their growth and survival. Therefore, understanding exactly how environmental stress signaling pathways impact the expression, activity, and function of these transcription factor modules is critical. Our genetics and chemical biology studies have revealed the importance of bHLH protein heterodimerization, and we are identifying a novel regulator mediating the inhibition of stomatal differentiation upon environmental stress. While we are learning much about the molecular mechanisms of stomatal patterning and differentiation from the model system Arabidopsis, new insights are coming from the study of diverse plant species. For example, we are expanding our study into Brassicaceae amphibious plant, *Rorippa aquatica*, to understand how this extremophyte has invented a new way to re-wire environmental/hormonal signal transduction pathways to swiftly switch to astomatous life style under water.

A comparative genomic analysis reveals key conserved cuticular genes in fleshy fruits cuticular biosynthetic machinery

[1]Valenzuela-Avilés, J.A.,[1]Silva-Córdova, F.J., [1]Contreras-Vergara, C.A., [2]Cruz-Mendívil, A., [3]CalderónVázquez, C.L., [1]Tiznado-Hernández, M.E., [4*]Hernández-Oñate, M.A.

[1]Centro de Investigación en Alimentación y Desarrollo, A.C., Hermosillo, México. [2]SECIHTI-Instituto Politécnico Nacional, Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional. Guasave, México.
[3]Instituto Politécnico Nacional, Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional. Guasave, México.
[4*]SECIHTI-Centro de Investigación en Alimentación y Desarrollo, A.C., Hermosillo, México.
*Email: miguel.hernandez@ciad.mx

The cuticle is a lipophilic layer produced by epidermal cells covering aerial organs in terrestrial plants. It plays essential functions in plants, protecting them against transpiration, radiation, pathogens, and mechanical damage. Additionally, it performs critical roles in fleshy fruits during their ontogeny and influences economically important characteristics during postharvest. The cuticle biosynthetic machinery is conserved in all land plants; however, its structure and composition vary among species, organs, and is influenced by environmental conditions, generating cuticles with specific characteristics between species. This study aims to conduct a comparative genomic analysis of the genes involved in cuticular biosynthesis, focused on fleshy fruits, to understand the differences and similarities related to the species-specific cuticular composition and behavior. To achieve this, we collected genomic data from 55 plant species, including 43 fleshy fruits from public databases, and made a quality assessment using BUSCO and QUAST. To identify the orthologues to the canonical biosynthetic pathways described in model species, we used OrthoFinder. Then, we generated a phylogenetic tree using FastTree to analyze the evolution of these gene families with CAFE5. The consensus domains of each gene family were obtained. Syntenic networks were created using SynNet, and the response elements of each family were analyzed using the MEME suite. We identified genes responsible for the cuticle biosynthesis in several fleshy fruit species and the gene families that were expanded significantly in the viridiplantae taxa. The synteny analysis shows the conservation of a core of essential cuticular genes. Moreover, the promoter analysis associates these genes with response elements related to water deficit and stress responses. Our findings show that some gene families involved in cuticular biosynthesis are increasing in gene copy number in the viridiplantae taxa. These have highly conserved domains, and their promoters contain clusters of response elements against water deficit by regulating transpiration.

WIP2/NTT modulates cytokinin homeostasis

David Díaz-Ramírez[1], Edgar Demesa-Arevalo[2,3], Yolanda Durán-Medina[1], Rosa E. Becerra-García[1,4], José E. Cruz-Valderrama[4], Beatriz E. Ruiz-Cortés[1], Humberto Herrera-Ubaldo[4], Ricardo A. Chávez Montes[4], Maurizio Di Marzo[5], Clelia de la Peña Seaman[6], Lucia Colombo[5], Martin Sagasser[7], Rüdiger Simon[2,3], Ondrej Novak[8], Stefan de Folter[4], Nayelli Marsch-Martínez[1*]

[1]Department of Biotechnology and Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav), km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, Mexico. [2]Heinrich-Heine Universität, Düsseldorf, Germany. [3]Cluster of Excellence on Plant Sciences, Heinrich Heine University, Düsseldorf, Germany. [4]Advanced Genomics Unit (UGA-Langebio), Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav), km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, Mexico. [5] Department of Biosciences, University of Milan, Italy. [6] Centro de Investigación Científica de Yucatán (CICY), Yucatán, México. [7]Center for Biotechnology, University of Bielefeld, Germany. [8]Laboratory of Growth Regulators, Centre of the Region Haná for Biotechnological and Agricultural Research, Institute of Experimental Botany, Czech Academy of Sciences, and Faculty of Science, Palacký University, Czechia.
*Email: nayelli.marsch@cinvestav.mx

Cytokinins (CKs) are key phytohormones orchestrating critical aspects of plant development, like meristem maintenance and organogenesis. Understanding the precise molecular mechanisms regulating CK levels and their spatial distribution is essential for grasping plant growth dynamics. Here, we investigate the role of the transcription factor WIP2/NO TRANSMITTING TRACT (WIP2/NTT) in modulating cytokinin homeostasis and signaling in *Arabidopsis thaliana.* Using an inducible expression system to overcome genetic redundancy, we demonstrate that NTT activation alters CK levels and signaling in a tissue-specific manner, as evidenced by a hormone quantification and the cross with the *TCSn::GFP* reporter. Transcriptomic profiling identified targets of NTT, including *IPT5* (biosynthesis), *AHP6* (signaling), and *CKX7* (degradation). Yeast one-hybrid and transactivation assays confirmed NTT's binding and regulation of these genes, which was further validated by genetic crosses with an *IPT5::GUS* reporter line. Notably, analysis of the crosses with *ckx7* mutant revealed that *CKX7* is required for the observed NTT induction phenotype. To visualize cytokinin distribution at high resolution, we optimized a whole mount immunolocalization protocol for small molecules. These experiments showed significant alterations in cytokinin distribution, both in *ntt* loss-of-function mutant and upon NTT induction. Together, our integrative approach—combining molecular genetics, transcriptomics, high-resolution imaging, and in vivo protein–DNA interaction assays—demonstrates that WIP2/NTT modulates cytokinin homeostasis through direct transcriptional regulation, consequently affecting cytokinin signaling in a tissue-specific manner. These findings advance our understanding of cytokinin regulation and provide new insights into the mechanisms that control plant development.

What did we lose by domesticating papaya? Genome sequencing of wild and commercial varieties reveals genetic clues to resistance

Diego Garza-González[1], Mayra A. Sallas-Barreto[1], Perla A. Contreras-de la Rosa[1], Adriana Quiroz-Moreno[1], Arianna C. Chan-León[1], Mirbella Cáceres-Farfán[1], Jorge M. Santamaría[1], Luisa López-Ochoa[1], Mariana Chávez-Pesqueíra[1], Aileen O'Connor-Sánchez[1], Kevin Childs[2], Elsa B. Góngora-Castillo [3*]

[1]Unidad de Biotecnología, Centro de Investigación Científica de Yucatán, Calle 43 No. 130 x 32 y 34. Col. Chuburná de Hidalgo, 97205 Mérida, Yucatán, México.
[2]Department of Plant Biology. Michigan State University. 612 Wilson Road. 48824 East Lansing, Michigan, USA.
[3]Departamento de Recursos del Mar. Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional. Km 6. Antigua carretera a Progreso. Cordemex, 97310. Mérida, Yucatán, México.
*Email: elsa.gongora@cinvestav.mx

Mexico is the world's third-largest producer and leading exporter of papaya (*Carica papaya*), making the understanding of its genetic diversity essential for crop improvement and long-term sustainability. Domestication theory suggests that wild varieties serve as valuable reservoirs of genetic variation, which can be impacted by the process of domestication. This reduction in variability can be reflected in the lower tolerance exhibited by commercial varieties to various biotic and abiotic stresses. In light of the challenges posed by climate change, which has led to an increase in extreme temperatures and the spread of diseases, it is critical to comprehend the defense mechanisms of this economically important crop. In this study, we sequenced, assembled, and annotated the genomes of a wild *C. papaya* variety and the commercial variety Maradol using Oxford Nanopore long-read sequencing technology, achieving over 100X coverage for both genomes. The assembled genomes measured approximately 280 megabases (Mb) for the wild variety and 300 Mb for the commercial variety, consistent with expected genome sizes. Using predicted protein datasets, we analyzed the gene family encoding nucleotide-binding site (NBS) domain proteins, which play a key role in plant immune responses. Our results revealed novel NBS genes in the wild variety, along with differences in domain composition and regulatory elements in their promoter regions when comparing both varieties. These findings highlight the importance of wild populations as reservoirs of genetic diversity and their potential as a source of biotechnological resources for crop improvement.

The MexMAGIC population and novel whole genome assemblies define signals of clinal adaptation in Mexican native maize

Sergio Pérez Limón[1]; Ana Laura Alonso Nieves[2]; Maria del Rosario Ramírez Flores[3]; Stewart Gillmor[4]; Ruben Rellán Álvarez[5]; Ruairidh Sawers[6*].

[1] Department of Plant Science, The Pennsylvania State University, University Park, PA 16802, USA. [2]Departamento de Fitomejoramiento, Universidad Autónoma Agraria Antonio Narro, Calzada Antonio Narro 1923, Colonia Buenavista, Saltillo, Coahuila, 25315, México. [3]Bioscience Division, Oak Ridge National Laboratory, 1 Bethel Valley Rd, Oak Ridge, TN 37830, USA. [4]Departamento de Biotecnología y Bioquímica, Centro de Investigación y de Estudios Avanzados (CINVESTAV-IPN), Irapuato, Guanajuato, 36821, México. [5]Department of Molecular and Structural Biochemistry, North Carolina State University, Raleigh, NC 27695. [6]Department of Plant Science, The Pennsylvania State University, University Park, PA 16802, USA. Email: sjp6181@psu.edu; *rjs6686@psu.edu.

Mexican Native Maize Varieties are connected populations growing over continuous environmental gradients, representing an excellent system to study the genetic architecture of adaptive traits along environmental and physiological clines, nevertheless, the study of this system is limited by the lack of full genome assemblies and presence of strong population structure. Multi-Parent Advanced Generation Inter-Cross (MAGIC) populations are powerful mapping populations that combine the power and low population structure of biparental populations with a measure of the diversity captured in diversity panels. Here we present the MexMAGIC population, generated with the genomic contribution of eight Native Maize Varieties sourced from different climatic-adaptation groups of maize cultivation in Mexico, and the novel full genome assemblies of three of its founders. We combine association and linkage mapping with founder environmental data to investigate the genetic architecture of Tassel Branch Number and (TBN) and Early Flowering (DTA), putative adaptive traits, and the role of selection acting on the observed clinal patterns along altitude. We demonstrate that the MexMAGIC population is suitable to identify functional variation and correctly estimate allelic effects at kernel pigmentation loci. We detected an oligogenic architecture for DTA, different for lowland and highland early varieties, and recovered evidence of selection acting along thermal requirement and not along altitude, highlighting the importance of farmer selection for this trait. We identified a major QTL for TBN on chromosome 7, linked to the gene *Ramosa1*, observing genome-wide and gene level signals of selection acting along an elevational cline. The analysis of the full genome assemblies identified a duplication around *Ramosa1*, unique to Palomero Toluqueño, that might cause a reduced TBN in the variety. We will discuss these results in light of complementary work that will exploit the potential of the MexMAGIC population to characterize the genetic architecture of adaptive traits in Mexican Native Maize.

Structural disorder and distinctive motifs in the C-terminal region of the MADS-domain transcription factors is conserved across diverse taxa

Teresa Beatriz Nava[1], Erandi Ramírez-Aguirre[2], Alejandra A. Covarrubias[1*] and Adriana Garay-Arroyo[2*]

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, México. [2]Laboratorio de Genética Molecular, Desarrollo y Evolución de Plantas, Depto. de Ecología Funcional, Instituto de Ecología, Universidad Nacional Autónoma de México, Ciudad de México, México.
[*]Email: agaray@iecologia.unam.mx

Plants exhibit remarkable developmental plasticity, allowing them to adapt to fluctuating environmental conditions through precise regulation of gene expression. Transcription factors (TFs) are central to these processes, integrating environmental cues and modulating developmental and stress-responsive pathways. In *Arabidopsis thaliana*, over 90% of TFs have intrinsically disordered regions (IDRs), which contribute to functional versatility by facilitating dynamic interactions, like protein-protein and protein-nucleic acids interactions. MADS-domain TFs constitute a conserved eukaryotic family involved in key developmental and stress-response functions. Through in silico analysis, we found that most *Arabidopsis* MADS-domain proteins contain IDRs (≥30 residues) in their C-terminal region, with a higher degree of global disorder in Type II compared to Type I proteins, in both types several members have propensity for have liquid-liquid phase separation (LLPS). Interestingly, orthologous MADS-domain proteins from non-plant eukaryotes (*Drosophila melanogaster, Saccharomyces cerevisiae, Homo sapiens*) also exhibit disordered C-terminal regions, with longer IDRs than those found in plants. Conserved motifs and putative activation domains were identified within the C-terminal IDRs of both Type I and Type II proteins, supporting their potential roles in co-regulator recruitment. Furthermore, Molecular Recognition Features (MoRFs) were detected in Type II proteins, reinforcing the hypothesis that IDRs facilitate specific, transient interactions essential for transcriptional regulation. These findings suggest that the presence and conservation of C-terminal IDRs in MADS-domain TFs across eukaryotic lineages are evolutionarily maintained features that contribute to their functional diversity and regulatory capacity in plant development and stress adaptation. Acknowledgements: This work was partially supported by Consejo Nacional de Humanidades, Ciencias y Tecnologías (CONAHCyT) with grants CF-2023-I-503 and CF-2019/252952 to AAC, and CBF2023-2024-1002 to AGA. The first authors wish to thank Consejo Nacional de Humanidades, Ciencias y Tecnología (CONAHCyT) for the postdoctoral scholarships granted (E.R.A., CVU number 413896; T.N.R., CVU 501149).

Unraveling the role of phospholipase PvPLDα2 in the symbiotic interaction between common beans and rhizobia

Ronal Pacheco, Noreide Nava, Ana Chávez-Martínez, Janet Palacios-Martínez, Luis Cárdenas and Carmen Quinto*

Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Universidad 2001, Colonia Chamilpa 62210, Cuernavaca, Morelos, Mexico.
*Email: carmen.quinto@ibt.unam.mx

The common bean (*Phaseolus vulgaris* L.) can establish a symbiotic relationship with soil bacteria known as rhizobia, which culminates in the formation of new organs in the roots, called nodules, in which nitrogen fixation occurs. Nodulation in legumes requires the synthesis and transport of plant-synthesized lipids for the formation of the symbiosome membrane during nodule organogenesis or for that of bacteroids during nitrogen fixation. Therefore, we are interested in evaluating the synthesis and turnover of lipids and phospholipids in this process. Physiological studies have reported the participation of phospholipases D (PLDs) in the establishment of this interaction. PLDs are phospholipid-degrading enzymes involved in diverse plant processes, for example, in defense responses against bacterial and fungal pathogens, although their role in the aforementioned symbiosis remains unexplored. Herein, the functional characterization of common bean PLD, *PvPLDα2A,* was performed in the interaction between *P. vulgaris* and *Rhizobium tropici*. An *in silico* analysis showed that the common bean genome encodes 22 PLDs and that *PvPLDα2A* transcription is induced by rhizobia in roots and nodules. The latter was confirmed by qPCR analysis. Downregulation of *PvPLDα2A* by RNAi enhanced rhizobial infection and increased nodules number in bean roots. Furthermore, overexpression of this gene decreased the number of nodules formed. These data suggest that PvPLDα2A acts as a negative regulator of the legume-rhizobial symbiosis, which is consistent with a study published in soybean (2). Our results confirm the relevance of PLDs and lipid metabolism in the regulation of the legume-rhizobium symbiosis and contribute to the understanding of the molecular mechanisms underlying this process. Acknowledgments. We appreciate the financial support of UNAM-PAPIIT (IN204024 to CQ) and SECIHTI (CF-2023-I-297 to CQ), and scholarship 749422 to RPS.

A tandem kinase fusion protein in *Aegilops tauschii* confers wheat leaf rust resistance

Naveenkumar Athiyannan[1,2*] Andrea González-Muñoz[1], Emile Cavalet-Giorsa[1], Evans S. Lagudah[3], Sambasivam K. Periyannan[3,4], Brande B. H. Wulff[1], Simon G. Krattinger[1]

[1]Plant Science Program, Biological and Environmental Science and Engineering Division (BESE), King Abdullah University of Science and Technology (KAUST), Thuwal, Saudi Arabia.
[2]Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Unidad Irapuato, Irapuato, México.
[3]Commonwealth Scientific and Industrial Research Organization (CSIRO), Agriculture and Food, Canberra, Australia
[4]Centre for Crop Health, University of Southern Queensland, Toowoomba, Queensland, Australia.
*Email: *naveenkumar@cinvestav.mx

Leaf rust, caused by the fungal pathogen *Puccinia triticina* (*Pt*), is one of three rust diseases that affect wheat. Wild relatives of wheat possess untapped sources of genetic resistance to manage leaf rust. An accession of the D-genome progenitor of bread wheat, *Aegilops tauschii* TA1675, exhibited resistance to an Australian *Pt* isolate. In this study, we employed bi-parental mapping to map the leaf rust resistance locus to the short arm of chromosome 2D and identified it as *LR39*. Concurrently, *k*-mer-based association mapping of an *Ae. tauschii* diversity panel that was phenotyped with an *Lr39*-avirulent *Pt* isolate revealed a peak at the telomeric end of chromosome arm 2DS. Long-read genome assembly of TA1675 revealed that the *k*-mers corresponded to a 2.33–2.45 Mb region in 2DS, which overlapped with markers flanking *LR39* at 1.20–2.84 Mb. The genomic region underlying the association peak contained three candidate genes in TA1675: one wheat tandem kinase (WTK) and two genes of unknown function. The biparental mapping interval harbored 16 genes, including WTK and an NBS-LRR-encoding gene. Based on functional annotations and polymorphisms compared with the susceptible *Ae. tauschii* accession AL8/78, the WTK gene emerged as the most promising candidate. Virus-induced gene silencing of the WTK candidate gene in TA1675 resulted in greater susceptibility to leaf rust. However, silencing an NLR-encoding gene located just outside the peak region did not result in increased susceptibility, indicating WTK gene is *Lr39*. The genomic sequence of the *Lr39* candidate gene is 11,699 bp with 21 exons. The corresponding 3,408-bp coding sequence encodes a protein with two N-terminal kinase domains of the LRR_8B subfamily, followed by a major sperm protein (MSP) domain and a WD40 repeat-containing domain at the C terminus. The fusion of MSP and WD40 domains in Lr39 constitutes a novel tandem kinase fusion protein.

The dynamics between the infection thread formation and cell division at the rhizobial-infection site in *P. vulgaris* roots

Rosana Sánchez-López[1,*] Elizabeth Monroy-Morales[1], Raúl Dávila-Delgado[1], Emmanuel Ayala-Guzmán[1], Alicia Gamboa-deBuen[2]

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Campus Morelos, Avenida Universidad 2001, Colonia Chamilpa, Cuernavaca 62210, México. [2]Instituto de Ecología, Universidad Nacional Autónoma de México, Circuito exterior s/n Ciudad Universitaria, Ciudad de México, 04510, México.
*Email: rosana.sanchez@ibt.unam.mx.

The legume-rhizobium symbiosis is a complex and finely regulated developmental program that leads to the *de novo* formation of an N_2-fixing nodule. It involves two well-coordinated processes: infection and organogenesis. Epidermal infection begins with the formation of a rhizobial-microcolony in a responsive and curled root hair, which facilitates the entry of rhizobia via an infection thread (IT). Concomitantly, cell division is activated in the neighboring cortical cells, forming a nodule primordium, through which the IT elongates. Last, the organogenesis of a N_2-fixing nodule where rhizobia are enclosed in the central tissue. In our model, *Phaseolus vulgaris* the initial cell division occurs in the sub-epidermal (S-E) cell. Recently, we developed a visualization strategy to explore the dynamics between the IT elongation in responsive-root hairs, its crossing into the S-E cell, and the cell-cycle progression in the S-E cell to be invaded by the IT. Here we summarize our findings using the nuclear marker NLS-Turquoise, a cell plate specific marker YFP-*Pv*KNOLLE and *Rhizobium etli* CE3-DsRed pMP604[1] excellent tools to trace the nuclear position, cytokinesis and rhizobia-containing structures, respectively. We have found that, at the microcolony stage, the nuclei in S-E cells were relocated to the center of the cell, suggesting the cell cycle activation, as well as a correlation between the IT elongation toward the base of the root hair and cell plate formation. Last, the IT invades the S-E cell when it is in a late cytokinetic status. Currently, we explore the correlation between IT and cell-cycle progression. This work was partially supported by Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica/UNAM (PAPIIT/ UNAM IN218524) to RSL.

Nano-omics enables pathogen infection in plants through stress biomarker enrichment in plants and crops within hours of infection

Roxana Coreas[1], Nikitha Sridhar[1], Teng-Jui Lin[1], Henry J. Squire[1], Sophia Tomatz[2], Elizabeth Voke[1], and Markita P. Landry[1]*

[1]Department of Chemical and Biomolecular Engineering, University of California,Berkeley, USA. [2] Department of Plant and Microbial Biology, University of California, Berkeley, USA.
Email: roxanacoreas@berkeley.edu; *landry@berkeley.edu.

Global food demand is projected to grow by 60% over the next 20 years yet stressors compromise food security by reducing agricultural productivity. Nanotechnology, which adapts nanomaterials as sensors in agriculture, can detect singular stress signalling molecules. However, 'single-signal' nano-sensors do not provide a comprehensive understanding of complex stress responses, particularly at early infection pre-symptomatic timepoints. By delivering nanomaterials into biosystems, their surfaces become spontaneously coated with biomolecules forming nanoparticle-corona complexes. In this study, we hypothesized that the intrinsic interfacial interactions between nanomaterials and plant biomolecules could be harnessed as a nano-tool for the highly sensitive enrichment of stress-induced biomarkers in plants, to capture early-onset crop infections. We exposed *A.thaliana* and *Z.mays* to pathogenic *P.syringae* (DC3000) to elicit production of stress-induced biomarkers. We collected time-dependent (0.5-, 1-, 3-, and 7-days post infection [dpi]) leaves and used 10-nm gold nanoparticles (AuNPs) with different surface charges to enrich and detect plant stress biomarkers via a 'nano-omics' approach, which leverages nanoparticle-corona formation and analysis with ultra-high performance liquid chromatography tandem mass spectrometry (UHPLC-MS/MS). Our nano-omic results show that several unique biomarkers are detectable in 'healthy-appearing' plants ≤1-dpi, prior to the onset of phenotypic responses and several days prior to infection detectable with conventional UHPLC-MS/MS. Interestingly, the sensitivity of nano-omics extended to 'distal' leaves of infected plants: we did not measure any *P.syringae* pathogen in distal leaves, but were able to capture systemic pathogen-triggered immune signalling at 0.5-dpi using nano-omics, capturing low-abundance stress proteins putatively transported to non-infected leaves through apoplastic trafficking. We compared nano-omics to standard techniques and found that our nano-omic approach was far more sensitive at detecting early-onset infections. Our work demonstrates that nanoparticle-corona complexation can be engineered for ultra-sensitive biomarker identification in infected plants, with a generic workflow that can be extended to other crops to support global sustainability goals.

Glucose-6-phosphate dehydrogenase is regulated via Cyclin/CDK phosphorylation during maize germination

Lara-Núñez A*, Labastida-Cuevas R, Guerrero-Molina ED, Rodríguez-Sotres R, Vázquez-Ramos JM

Facultad de Química-UNAM. Edificio E, Lab. 114, Ciudad Universitaria, CDMX C.P. 04510. Tel. 55 56284860.
*Email: auroraln@unam.mx.

Glucose-6-phosphate dehydrogenase (G6PDH) is the key regulatory enzyme of the oxidative pentose phosphate pathway (OPPP). During seed germination, G6PDH plays a pivotal role by supplying reducing equivalents (NADPH) and metabolic intermediates essential for macromolecule biosynthesis. DNA replication, in particular, demands high levels of NADPH and nucleotide precursors. Therefore, cell proliferation must be tightly coordinated with OPPP activity to ensure an adequate supply of reducing power and substrates—such as ribose-5-phosphate, an OPPP intermediate—required for cell cycle initiation. Cyclins (Cyc) and cyclin-dependent kinases (CDKs), which form active heterodimeric complexes, are central regulators of the cell cycle through phosphorylation of specific protein targets. This post-translational modification modulates substrate activity, initiating a cascade of events that drive cell division. In this study, we investigated whether ZmG6PDH5—the isoform exhibiting the highest catalytic activity during maize germination—interacts with ZmCycs or ZmCDKs, serves as an in vitro and in vivo substrate for Cyc/CDK-mediated phosphorylation, and how this modification affects its enzymatic function. Our results demonstrate that ZmG6PDH5 interacts with several CycD proteins, one CycB, and CDKA;1, but not with CycB1;2 or CDKB. Phosphorylation by Cyc/CDK complexes significantly enhanced G6PDH activity (approximately fivefold). Moreover, treatment of maize embryo axes with RO-3306—a specific CDK inhibitor—during germination led to reduced total G6PDH activity compared to untreated controls. Notably, reactive oxygen species (ROS) levels increased in inhibitor-treated embryo axes, showing an inverse correlation with G6PDH activity. These findings suggest that G6PDH contributes to ROS scavenging during early germination and that its regulation via CDK-mediated phosphorylation is critical for maintaining redox homeostasis and ensuring proper seedling development.
Acknowledges: PAPIIT-DGAPA IN211423, PAIP 5000-9130

Inactive cell wall invertases are necessary for correct development of *A. thaliana* seedlings and affect aspects of sucrose metabolism

Andrea Castro Mexicano, Luisa Cendejas Gutiérrez, Lucila Ortiz Castellanos and June Simpson*

[1]Department of Genetic Engineering, Cinvestav, Unidad Irapuato Km. 9.6 Libramiento Norte, Carretera Irapuato-León C.P.36824, Irapuato, Guanajuato, México.
*Email: june.simpson@cinvestav.mx.

Cell wall invertases (CWI) which irreversibly breakdown sucrose into fructose and glucose are highly regulated at the transcriptional and post-translational level. CWI are essential components of sucrose metabolism with roles in sucrose partitioning, signaling, development and stress tolerance. Most plant species have several CWI isoforms and in many species some of these isoforms are known or predicted to be inactive and incapable of breaking down sucrose. The prevalence of inactive CWI isoforms has led to the hypothesis that they may carry out alternative functions. *A. thaliana* has 2 confirmed inactive CWI (AtCWINV3 and AtCWINV6) with specific and distinct expression patterns. Knock-down lines (individual and double) show severely affected and non-viable phenotypes where seedlings fail to develop normally and only the AtCWINV6 and the AtCWINV6/AtCWINV3 knock-down lines are partially restored to normal growth by the addition of sucrose. Sucrose also induces AtCWINV6 expression but represses AtCWINV3 expression in the wild type. The AtCWINV3 knock-down lines show lower invertase activity and higher expression of the AtCif1 invertase inhibitor. Preliminary BiFC results show a probable interaction between the inactive CWI and AtCif1, suggesting that the inactive CWI could regulate AtCif1 at the post translational level by binding to the inhibitor and thereby reducing inhibition of active CWI.

Integrating multi-omics data of olive (*Olea europaea*) and transcriptomics of multiple Oleaceae species for tracing oleuropein biosynthesis

Ornella Calderini [1], Mohamed O. Kamileen [2], Yoko Nakamura [2], Sarah Heinicke [2], Ryan M. Alam [2], Benke Hong [3], Yindi Jiang [4], Alma Gutiérrez-Vences [5], Fiammetta Alagna [6], Francesco Paolocci [1], Maria Cristina Valeri [1], Edoardo Franco [1], Soraya Mousavi [1], Roberto Mariotti [1], Lorenzo Caputi [2], Sarah E. O'Connor [2], Carlos E. Rodríguez-López [5,7]*

[1]Institute of Biosciences and Bioresources, CNR, Perugia, 06128 Italy. [2]Department of Natural Product Biosynthesis, Max Planck Institute for Chemical Ecology, Jena, 07745 Germany. [3]Zhejiang Key Laboratory of Precise Synthesis of Functional Molecules, Department of Chemistry, School of Science and Research Center for Industries of the Future, Westlake University, Hangzhou, China. [4]Key Laboratory of Quantitative Synthetic Biology, Shenzhen Institute of Synthetic Biology, Shenzhen Institutes of Advanced Technology, Chinese Academy of Sciences, Shenzhen 518055, China. [5]Tecnológico de Monterrey, Escuela de Ingeniería y Ciencias, Ave. Eugenio Garza Sada 2501, Monterrey, N.L. 64700, Mexico. [6]Department of Energy Technologies and Renewable Sources, National Agency for New Technologies, Energy and Sustainable Economic Development (ENEA), Trisaia Research Centre, Rotondella (MT), 75026 Italy. [7]Tecnológico de Monterrey, The Institute for Obesity Research, Integrative Biology Unit, Ave. Eugenio Garza Sada 2501, Monterrey, N.L. 64700, Mexico.
*Email: c.e.rodriguez@tec.mx.

The dietary, economic and cultural importance of Olive cannot be overstated; along with wine and bread, olive oil forms the base triad of the traditional Mediterranean diet, shown to reduce risk of metabolic disorders among others. Oleuropein, an oleoside-type secoiridoid, is the major determinant of flavor and quality of olive oil, often attributed as the main contributor of the health benefits of virgin olive oil. Despite its agricultural and biomedical importance, oleuropein biosynthetic pathway remains uncharted. We ran MS-based metabolomics and RNA-Seq experiments on olive fruit mesocarp of six commercial olive cultivars with varying oleuropein content, during maturation and ripening. Using this data we discovered three polyphenol oxidases with oleuropein synthase (OS) activity, a novel oleoside-11-methyl ester glucosyl transferase (OMEGT) synthesizing a potential intermediate in the route, and a 7-*epi*-loganic acid O-methyltransferase (7eLAMT). Integrating transcriptomics data from 15 plant species from three iridoid-producing plant orders (Lamiales, Gentianales, and Cornales), and tissue expression panels from Arabian jasmine (*Jasminum sambac*) and European Ash (*Fraxinus excelsior*), we discovered two 2-oxoglutarate dependent dioxygenases (named 7eLAS) that synthesize 7-*epi*-loganic acid; in contrast *C. roseus* 7-deoxy-loganic acid hydroxylase (7DLH), a known bottleneck in MIA production, is a cytochrome p450. An analysis of public datasets point to a role of the genes discovered in this work in response to infection of olive to pathogens *Spilocaea oleagina* and *Verticillium dahliae*, as well as infestation by the Emerald Ash Borer in *F. excelsior*. This comparative co-expression method, which combines guilt by association and comparative transcriptomics approaches, can successfully leverage big datasets for untargeted discovery of enzymes. Given the increasing availability of expression data from species across the plant kingdom, the methods used for gene discovery used in this work can be readily applied to other untraced pathways.

The unique shoot apical meristems in Cactaceae

Brenda A. López-Ruiz, Valeria Romero-Gómez, Ivonne S. Altamirano-Hernández C. GenaroRamírez-Castro, Delil A. Chincoya, Eduardo Torres-Flores, Ulises Rosas*

Jardín Botánico, Instituto de Biología, Universidad Nacional Autónoma de México, Tercer Circuito Exterior, Ciudad Universitaria, Coyoacán, Ciudad de México 04510, México.
*Email: urosas@ib.unam.mx

Cactaceae is a large family of plants that is native to the Americas, with Mexico being home to the highest number of species. A key area of research in cacti is the development of their Shoot Apical Meristem (SAM), which is larger than the SAMs found in other plants, including model species like Arabidopsis thaliana. The mechanisms that regulate the size of the SAM in cacti remain a mystery; the genetic regulation in these plants may be similar to that in other model organisms. However, it is likely that this regulation involves a recycling and modification of genetic pathways, leading to the unique evolutionary traits observed in the Cactaceae family. Two genes, WUSCHEL (WUS) and SHOOT MERISTEMLESS (STM), are part of a genetic network that regulates meristem size. In this study, we utilized the extinct cactus species Mammillaria san-angelensis as a model system because of its short life cycle, short size, ease of reproduction, and sequenced genome. We describe seven developmental stages of the SAM, ranging from one day post-germination to four years of growth. Additionally, we compared the vegetative SAM with the floral meristem and areole, a unique structure found in Cactaceae. We identified the orthologues of WUS and STM from the genome of M. san-angelensis and obtained the corresponding gene models. Furthermore, we measured the expression levels of these genes across the developmental stages. The results provide insights into the genetic basis of gigantism in Cactaceae, an evolutionary novelty observed in these plants. This work was supported by PAPIIT DGAPA UNAM IN219725 and UNAM-DGAPA Posdoctoral Program (POSDOC) to BALR.

Morpho-anatomical characterization of fruit dehiscence in *Bixa orellana* L.: implications for plant development and crop improvement

Renata Rivera-Madrid*[a] Rocio Tamayo-García[a], José Alberto Narváez-Zapata[b], Margarita Aguilar-Espinosa[a]

[a]Unidad de Biología Integrativa. Centro de Investigación Científica de Yucatán, A.C. Calle 43 # 130, Chuburná de Hidalgo, CP 97205, Mérida, Yucatán, México.
[b]Instituto Politécnico Nacional-Centro de Biotecnología Genómica, Blvd Del Maestro esq. Elias Piña, 88710, Reynosa, Tamaulipas, México.
*Email: renata@cicy.mx

Bixa orellana L., well known as achiote in Mexico, is considered a commercially important species due to the bixin pigment in its seeds. This plant exhibits notable phenotypic variability, especially in fruit dehiscence, a trait correlated with pigment content. Understanding fruit dehiscence in *Bixa orellana* is crucial for optimizing seed dispersal and pigment yield, directly impacting agricultural productivity and breeding strategies. This study details the morphological and anatomical characterization of *B. orellana* fruits with contrasting dehiscence phenotypes: dehiscent, indehiscent, and partially dehiscent. An analysis histological analysis of the fruits was conducted 58 days after floral anthesis, revealing crucial differences in the dehiscence zone and the distribution and number of endocarp glands. Dehiscent fruits showed smaller, isodiametric cells and a clearly defined dehiscence zone with evidence of enzymatic degradation, facilitating seed release. In contrast, indehiscent fruits lacked a clear distinction in this zone, and partially dehiscent fruits exhibited intermediate characteristics. Analysis of lignin deposition using autofluorescence and phloroglucinol-HCl staining revealed distinctive patterns. Dehiscent fruits presented low lignin fluorescence in all areas. In contrast, indehiscent fruits showed precise lignin deposition in the appendix, exocarp, vascular bundle sheath, and placenta, but not in the dehiscence zone. Partially dehiscent fruits reflected an intermediate pattern. These findings suggest that lignification and enzymatic degradation in the dehiscence zone are crucial characteristics determining the fruit's opening capacity. Furthermore, variations in the presence and distribution of endocarp glands and secretory cells were observed, being more abundant in dehiscent fruits. The detailed characterization of these phenotypes significantly contributes to understanding the traits associated with dehiscence and their potential application in breeding programs to optimize bixin production in *B. orellana*. This work provides valuable insights into the developmental processes governing fruit dehiscence, advancing our knowledge of plant reproductive biology and its implications for crop improvement.

Imputation integrates single-cell and spatial gene expression data to resolve transcriptional networks in barley shoot meristem development

Edgar Demesa-Arevalo[1,7,8]*, Hannah Dörpholz[2], Isaia Vardanega[1], Jan Eric Maika[1], Itzel Pineda-Valentino[6], Stella Eggels[2], Tobias Lautwein[3], Karl Köhrer[3], Thorsten Schnurbusch[4,5], Maria von Korff[7], Björn Usadel[2,6], Rüdiger Simon[1]*

[1]Heinrich Heine University, Faculty of Mathematics and Natural Sciences, Institute for Developmental Genetics, CEPLAS, Düsseldorf, Germany
[2]Institute of Bio and Geosciences (IBG-4: Bioinformatics) & Bioeconomy Science Center (BioSC), CEPLAS, Forschungszentrum Jülich, Jülich, Germany.
[3]Heinrich Heine University, Medical Faculty, Biological and Medical Research Center (BMFZ), Genomics and Transcriptomics Laboratory (GTL), Düsseldorf, Germany.
[4]Research Group Plant Architecture, Leibniz Institute of Plant Genetics and Crop Plant Research (IPK), OT Gatersleben, 06466 Seeland, Germany.
[5]Martin Luther University Halle-Wittenberg, Faculty of Natural Sciences III, Institute of Agricultural and Nutritional Sciences, Halle, Germany.
[6]Heinrich-Heine University Düsseldorf, Faculty of Mathematics and Natural Sciences, Institute for Biological Data Science, CEPLAS, Düsseldorf, Germany
[7]Heinrich Heine University, Faculty of Mathematics and Natural Sciences, Institute for Plant Genetics, CEPLAS, Düsseldorf, Germany, Institute for Plant Genetics
[8]Genetic Engineering Department, Centre for Research and Advanced Studies (CINVESTAV-IPN) Irapuato Unit, Mexico. Km. 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato 36824, Guanajuato, México.
*Email: edgar.demesa@cinvestav.mx, ruediger.simon@hhu.de

Grass inflorescences are composite structures, featuring complex sets of meristems as stem cell niches that are initiated in a repetitive manner. Meristems differ in identity and longevity, generate branches or split to form flower meristems that finally produce seeds. Within meristems, distinct cell types are determined by positional information and the regional activity of gene regulatory networks. Understanding these local microenvironments requires precise spatio-temporal information on gene expression profiles, which current technology cannot achieve. Here we investigate transcriptional changes during barley development, from the specification of meristem and organ founder cells to the initiation of distinct floral organs, based on an imputation approach integrating deep single-cell RNA sequencing with spatial gene expression data. The expression profiles of more than 40.000 genes can be analysed at cellular resolution in multiple barley tissues using the web-based graphical interface *BARVISTA*, which enables precise virtual microdissection to analyse any sub-ensemble of cells. Our study pinpoints previously inaccessible key transcriptional events in founder cells during primordia initiation and specification, characterises complex branching mutant phenotypes by barcoding gene expression profiles, and defines spatio-temporal trajectories during flower development. We thus uncover the genetic basis of complex developmental processes, providing novel opportunities for precisely targeted manipulation of barley traits.

Engineering multi-input protein logic gates for programmable gene expression control in plants

Isabel Goldaracena, Jennifer A. N. Brophy*

Bioengineering Department at Stanford University, Stanford, California, USA. *Email: jbrophy@stanford.edu.

Plants must balance normal growth and development with stress responses when facing environmental challenges. Recent advances in genetic engineering allow us to introduce genes from other organisms or plant species that help plants defend against pathogens or modify plant structure to better survive harsh conditions. However, when these protective genes are always active (constitutive expression), they can interfere with normal plant development. Therefore, we need smarter gene control systems that can sense multiple environmental signals and activate protective responses only when and where they are truly needed. In this project, I developed a toolkit that uses split transcription factors to create precise gene control systems. Each fragment of the transcription factor responds to a different signal - such as an environmental cue, a specific cell type, or a developmental stage. The target gene only activates when all required conditions are present simultaneously. I tested this approach using transient expression in *Nicotiana benthamiana* to develop a 3-input system and characterized its performance under various conditions. I also demonstrated a 2-input system in stable *Arabidopsis thaliana* transformations using cell-type specific promoters. This technology provides much tighter control over gene expression and should help maintain normal plant development. To make this approach more accessible, I developed computational tools to guide the selection of split sites and explored machine learning methods to automate the engineering process. I am currently expanding this system by testing additional developmental and cell-type specific promoters in stable plant lines and identifying practical applications for the technology.

Resistance triggered by self-DNA: specificity and application potential of an incomprehensible immune response

Martin Heil[1]*

Centro de Investigación y de Estudios Avanzados (CINVESTAV) – Unidad Irapuato, Irapuato, México.
*Email: martin.heil@cinvestav.mx.

Fragments of self-DNA are damage-associated molecular patterns (DAMPs) that activate an immune response. Since the first reports, DNA-triggered resistance to pests and pathogens was reported for various plant species. Own work showed that self-DNA treatment can increase yield of common bean (*Phaseolus vulgaris*) more than 2-fold and the biomass of lettuce (*Lactuca sativa*) 1.3-fold. *Arabidopsis thaliana* and *P. vulgaris* responded to fragmented self-DNA or non-self-DNA with a differential induction of H_2O_2 and jasmonic acid (JA) but a non-specific induction of salicylic acid (SA). Correspondingly, only self-DNA induced resistance to chewing caterpillars (*Spodoptera frugiperda*) while self- and non-self-DNA induced resistance to bacterial and fungal pathogens to similar degrees. This self/non-self-specificity remains an enigma. Human immune cells hardly distinguish bacterial or viral DNA from eukaryotic DNA and respond more strongly to non-self-DNA, since immune responses to self-DNA can lead to autoimmune diseases (Lupus, Psoriasis) or fatal outcomes of viral infections (HIV-1, COVID-19). Understanding these differences is difficult without knowing how plants perceive extracellular DNA. Recent work linked the DNA damage response with the immune response and identified CpG methylation as a relevant factor, but no receptor has been identified so far. The quest for the plant DNA receptor is open.

Exploring the signaling pathway that controls arsenite (As [III]) responses in the root of *Arabidopsis thaliana* L

Andreina Ocampo-Santana[1], Salvador Barrera-Ortiz[1a], Luz Breton-Deval[1,2], Gabriel Guillén[1], and Ángel Arturo Guevara-García[1*]

[1]Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Universidad #2001, colonia Chamilpa, C. P. 62210, Cuernavaca, Morelos, México.
[2]Consejo Nacional de Humanidades, Ciencia y Tecnología, Avenida Insurgentes Sur 1582, colonia Crédito Constructor, Ciudad de México, México.
*Email: atrturo.guevara@ibt.unam.mx.

Rising metal concentrations in the environment are one of the most serious pollution problems today. Due to their sessile nature, this problem is particularly important for plants that complete their life cycle exposed to contaminants present in the soils where their seeds germinate. Despite interest in studying their toxic effects and how to control them, very little is known about the signaling mechanisms by which plants respond to exposure to heavy metals and metalloids. Arsenic (As) is a metalloid known to affect plant development, notably of the root, which is the organ responsible for the uptake of water and nutrients from the soil. As [III] is the most toxic form of inorganic As, and the elucidation of the mechanism by which plant roots respond to its exposure is just beginning. Here, using a pharmacogenetic strategy, it is shown that As [III] causes cell damage and inhibits the growth of the primary root of *A. thaliana*, arresting cell division and elongation. Furthermore, thanks to the use of inhibitors (N-1-naphthylphthalamic acid/NPA) and mutants (*pin2*) that affect auxin transport, and to the analysis of the expression of an auxin-inducible gene (*IAA17*), it is proposed that auxins participate in the response. Finally, using reporter lines for the *MPK6* and *MPK3* genes, the participation of a MAP kinase module in the signaling pathway activated by As [III] is suggested.

Interactions between the transcription factor BOL/DRNL/ESR2 and the jasmonate pathway

Beatriz E. Ruiz-Cortés[1], Yolanda Durán-Medina[1], C. Cecilia Ramos-Tamayo[1,2], Herenia Guerrero-Largo[1], Ma. Isabel Cristina Elizarraraz-Anaya[1], Omar Fabián Hernández-Zepeda[3], Enrique Ramírez-Chávez[1], Michiel Lammers[4], Ruud A. de Maagd[4], Jorge Molina-Torres[1], Stefan de Folter[2] and Nayelli Marsch-Martínez[1]*

[1]Irapuato Unit, Biotechnology and Biochemistry Department, Cinvestav, Irapuato C.P. 36824, Guanajuato, México. [2]Advanced Genomics Unit, Cinvestav, Irapuato C.P. 36824, Guanajuato, México. [3]Genómica Alimentaria, Universidad de la Ciénega del Estado de Michoacán de Ocampo, Sahuayo C.P. 59103, Michoacan, Mexico. [4]Wageningen University & Research, P.O. Box 16, 6700 AA Wageningen, The Netherlands.
*Email: nayelli.marsch@cinvestav.mx.

BOL/DRNL/ESR2, an AP2/ERF transcription factor, regulates early organ development in Arabidopsis (*Arabidopsis thaliana*). Its loss of function causes flower organ defects, while its overexpression induces green callus formation in roots without the addition of hormones. Jasmonates (JA), plant hormones known as major players in stress responses, also regulate some aspects of organ development like stamen development and plant growth. In this work, we studied the interaction between BOL and the JA pathway revealing bidirectional regulation with developmental implications. We found that exogenous application of methyl jasmonate (MeJA) partially rescued the stamen defects in CRISPR-generated *bol-cr* mutants, suggesting JA deficiency contributes to these phenotypes and linking BOL and JA mediated stamen development. Conversely, MeJA treatments in wild-type plants partially mimicked some *bol-D* mutant phenotypes like reduced rosette and root size, while JA inhibition restored wild-type leaf curvature, suggesting an alteration in JA homeostasis in the gain-of-function mutant. Quantitative analyses show *BOL* overexpression elevates JA levels, whereas *bol* loss-of-function reduces them. Furthermore, inducible BOL activity led to downregulation of a JA responsive marker. Finally, JA biosynthesis inhibition affected BOL induced root callus formation and led to an expansion of the *BOL* expression domain in roots. Together, these results position BOL as a modulator of JA pathway, regulating JA accumulation, signaling sensitivity, and developmental responses, while JA reciprocally fine-tunes *BOL* expression. Our work uncovers a previously unrecognized feedback loop integrating BOL mediated development with JA homeostasis, offering new insights into how transcriptional regulators coordinate hormonal and morphogenetic programs.

Understanding and engineering crown root developmental program in the C4 grass *Setaria viridis*

Héctor H. Torres-Martínez[1], Shiqing Yao-Lu[1], Janina Tamborski[2], William G. Viana[1], Sunil Kenchanmane-Raju[3], Kenneth D. Birnbaum[4], Jennifer A. N. Brophy[2], José R. Dinneny[1*]

[1]Department of Biology, Stanford University, Stanford, CA 94305, USA.
[2]Department of Bioengineering, Stanford University, Stanford, CA 94305, USA. [3]Department of Botany and Plant Sciences, University of California, Riverside, CA 92521, USA.
[4]Center for Genomics and Systems Biology, New York University, New York, NY 10003, USA.
*Email: dinneny@stanford.edu.

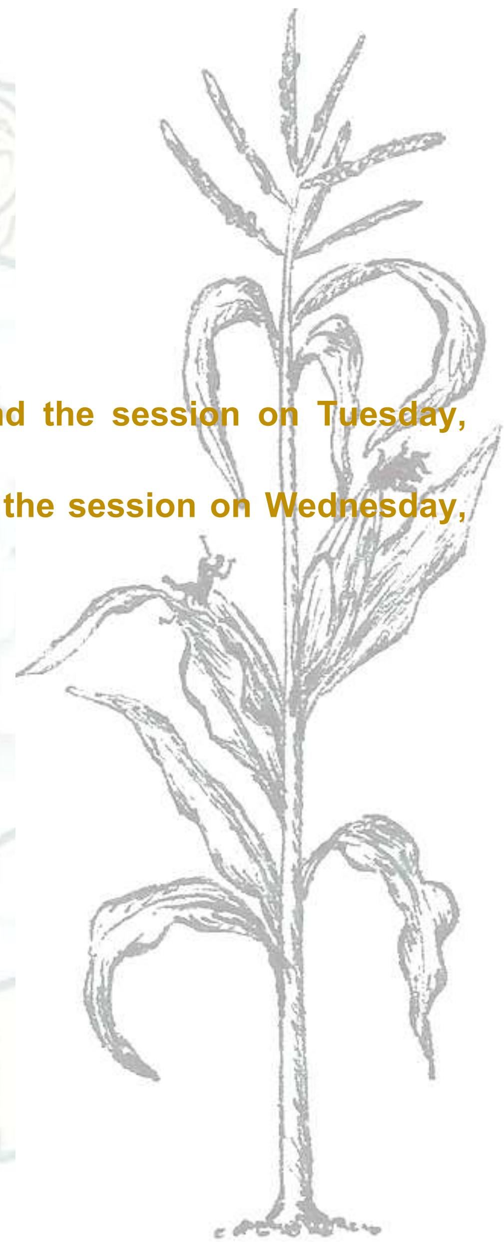
Despite grasses being among the world's major crops, fundamental aspects of their developmental programs remain poorly understood. Grasses develop a fibrous root system predominantly composed of branching adventitious roots, including crown roots, the stem basal roots that emerge near the soil surface. This project aims to elucidate the cellular and genetic basis of crown root primordium development and to explore synthetic genetic circuits for engineering root system architecture in the C_4 model grass *Setaria viridis*. Unlike *Arabidopsis thaliana*, where lateral roots arise from a single parental tissue, grass lateral roots originate from multiple tissue layers. Whether a similar ontogeny underlies crown root development remains unclear. To address this, we have optimized high-resolution imaging and in situ hybridization chain reaction (HCR) protocols to examine gene expression patterns during the early stages of crown root development. In parallel, we are conducting tissue-specific transcriptomic analyses to identify promoters that enable precise dissection of tissue-specific contributions to crown root initiation. Preliminary analysis of a CRISPR/Cas9-generated *crown root defective* (*crd*) mutant revealed impaired primordium morphogenesis, particularly affecting the root cap lineage. Given that the endodermis contributes to primordial root cap formation in grass lateral roots, we hypothesize that CRD plays a key role in specifying the primordial root cap from endodermal-like derivatives. Further experiments are underway to test this hypothesis. Because *crd* loss-of-function mutants exhibit a significantly reduced number of crown roots, we are fine-tuning *CRD* expression using synthetic biology tools to modulate root system architecture. Ongoing characterization of transgenic lines carrying buffer gates with varying CRD expression levels will help determine whether *CRD* enables precise control over crown root number. Altogether, this research demonstrates the potential of integrating developmental biology and synthetic biology to engineer grass root systems, ultimately contributing to the development of crops with improved resilience under environmental stress.

Abiotic stress

Posters

Participants with even numbers will attend the session on Tuesday, October 21.

Participants with odd numbers will attend the session on Wednesday, October 22.



Poster 1. PCNA association with CycB1;2/CDKB1;1 complex and its role in DNA damage response during maize germination

Estefany Damaris Guerrero Molina[1], Mingyar Neftalí López Hernández[1], Ulises Martínez Ortega[2], Elpidio García Ramírez[1], Jorge Manuel Vázquez Ramos[1]*

[1]Facultad de Química, Departamento de Bioquímica, Universidad Nacional Autónoma de México Ciudad de México, 04510, México.
[2]Facultad de Química, Departamento de Farmacia, Universidad Nacional Autónoma de México Ciudad de México, 04510, México.
*Email: jorman@unam.mx.

Genome stability in seed cells is crucial for crop productivity and thus DNA damage by genotoxic agents and metabolic processes can reduce stability. To counteract this damage, plant cells have developed DNA repair mechanisms similar to those found in mammals. Cyc/CDK protein complexes are essential for cell cycle advancement but are inhibited in response to DNA damage, particularly double-strand breaks (DSB), however, CycB1/CDKB1 complexes participate in repairing DSB through homologous recombination. On the other hand, the Proliferation Cell Nuclear Antigen (PCNA), an essential protein for DNA replication and cell cycle regulation, also importantly participates in repairing DSB through homologous recombination (HR) and this involves RAD51 recombinase. In this project, studies on maize demonstrated the effect of gamma radiation-induced DNA damage in reducing germination and seedling establishment, with the concomitant observation of differential changes in the expression and protein abundance of PCNA, CycB1;2, CDKB1;1, and RAD51 after radiation. Furthermore, interaction simulations were developed between PCNA and the aforementioned proteins. Predictions indicate a potential macro-complex formation between these proteins and DNA. Phosphorylation assays showed that PCNA enhanced CDKB1;1 kinase activity over RAD51 in radiated seeds, suggesting their involvement in the regulation of DNA damage repair.

Poster 2. Vector for stable protein expression in *Capsicum annuum* with only native genetic elements. A biosafe alternative

Lilian Gabriela Valencia-Turcotte, Felipe Leon-Garcilazo, María Teresa Olivera-Flores and Rogelio Rodríguez-Sotres*

Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México, Av. Universidad 3000, Col. Cd. Universitaria, Alc. Coyoacán, c.p. 04510. Cd. México.
*Email: sotres@unam.mx.

Transgenic plants have long been the subject of public concern, mainly because of the potential contamination of natural crop species pangenomes with foreign genes. In this study, a pCAMBIA1300 plasmid was refurbished with intragenic elements to drive protein expression, transiently in onion epidermis, and constitutively in *Capsicum annuum* callus. The bacterial kanamycin resistance gene was replaced with a native chili pepper transporter gene as selection marker. The intragenic expression cassette has the *C. annuum* promoter of fructose-1,6-bisphosphate aldolase controlling one chimeric open reading frame (ORF) for one phosphate-starvation stress response protein; either a soluble inorganic pyrophosphatase (PPa), a membrane-bound inorganic pyrophosphatase (PPv), or a member of the haloacid dehalogenase superfamily (PS2); which was in-frame with the fluorescent reporter iLOV (light, oxygen, or voltage flavoprotein domain from *C. annum* phototropin) and compassed by the *C. annuum* aspartic protease terminator (ASPt). The results demonstrate constitutive expression driven by the strong promoter, as visualized by confocal microscopy of a sharp iLOV fluorescent signal. Compared to commonly used animal fluorescent proteins, iLOV is smaller, has a shorter emission delay, and shows quick spontaneous photobleaching recovery in living cells. Transient expression was observed in onion epidermis within 24 hours and constitutive expression was detected in *C. annuum* callus at 30, 60, and more than 370 days post-transformation. The intensity and spatial distribution of iLOV fluorescence varied depending on the fused protein, supporting the expected vacuolar localization for AVP and the cytoplasmic localization for PPa and PS2. ASPt was found to produce the most stable expression. Based on these findings, this vector is proposed as a non-toxic intragenic alternative for genetic stable transformation of plants using only native elements. Combined with iLOV, it also proves suitable for subcellular localization studies in *Capsicum* species and potentially in other Solanaceae crops. Funding: CONAHCyT CB 218-A1-S-8650, PAPIIT-DGAPA IN219518, PAIP-FQ 5000-9122.

Poster 3. ZmVDAC and ZmHXK4 are involved in plant drought response and the modulation of cell death

García-Ortiz D., Burgos-Palacios A. and Sánchez-Nieto S.*

Laboratorio de Transporte y Percepción de Azúcares en Plantas, Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México, 04510, Mexico City, Mexico.
*Email: sobeida@unam.mx.

Voltage-dependent anion channels (VDACs) are the most abundant proteins in the mitochondrial outer membrane and have essential roles in metabolite and high-energy molecule exchange, as well as in processes such as programmed cell death, contributing significantly to cytoplasmic and mitochondrial signaling. In plants, the study of VDAC is limited, what is known is that they are multigene families that code for several isoforms, which have been shown to participate in different types of biotic and abiotic stresses. In *Zea mays*, an important agronomic species, nine putative VDAC genes have been identified through *in silico* analyses , all of which exhibit high potential to encode functional proteins.ZmVDAC1b and ZmVDAC4b isoforms have shown a potential association with a mitochondrial hexokinase from maize (ZmHXK4), based on pull-down assays using the soluble version of the hexokinase (ZmHXK4Δ30) as bait. In this study, we analyzed the physiological effects resulting from the individual and combined expression of ZmVDAC1b or ZmVDAC4b and ZmHXK4 in plants.The genes encoding *ZmVDAC1b* and *ZmVDAC4b* were cloned, and both proteins were shown to be translated and localized to the mitochondria. Transient expression of these isoforms in *Nicotiana benthamiana* leaves induced tissue damage, which was exacerbated under drought stress conditions but was alleviated by co-expression of ZmHXK4 (either in its full-length or soluble form). However, ROS detection revealed greater histological damage in leaves expressing ZmVDACs alone or in combination with ZmHXK4Δ30, compared to those co-expressing ZmVDACs with the full-length ZmHXK4. These results suggest a functional interaction between ZmVDAC and ZmHXK4, potentially involved in modulating programmed cell death in plants. We are currently exploring the physical interaction between ZmVDAC and ZmHXK4 through molecular dynamics simulations and experimental approaches.

Poster 4. Anhydrobiosis in plants: from biocondensates in seeds, to the native cellular structure of the pollen grains

Coral Martínez Martínez[1,2], Alejandra A. Covarrubias Robles[1], and Marisa Otegui[2]*

[1]Instituto de Biotecnología, Departamento de Biología Molecular de Plantas, Universidad Nacional Autónoma de México. Cuernavaca, Morelos, México. [2]University of Wisconsin, Department of Botany. Madison, Wisconsin, United States.
Email: otegui@wisc.edu

Water is essential for all life forms on Earth. However, certain organisms, or specific structures within organisms, can survive prolonged desiccation by entering a state with no detectable metabolic activity, yet retain the ability to resume life upon rehydration. This phenomenon is known as anhydrobiosis: a biological state in which an organism can lose up to ~95% of its cellular water and later restore normal metabolic functions once rehydrated. In plants, anhydrobiosis is particularly evident in resurrection plants, which can tolerate near-complete desiccation and revive when water becomes available again. It also occurs in specialized plant structures such as seeds and pollen grains of certain species. During desiccation, plants employ diverse protective strategies—biochemical, structural, and molecular—to survive extreme dehydration and preserve viability. Among these, the accumulation of protective proteins is crucial, for example, Late Embryogenesis Abundant (LEA) proteins play key roles in preventing protein aggregation and preserving cellular structures under water stress. In addition, desiccation-tolerant tissues undergo structural rearrangements in components such as the plasma membrane, cell wall, organelles, and cytoplasm. These adaptations are critical for maintaining cellular integrity in a desiccated but viable state over extended periods. In this work, we explore the behavior of a Group 4 LEA protein from *Arabidopsis thaliana*. Our findings show that this protein forms condensates in the cytosol of seeds, a phenomenon likely promoted by the intracellular environment unique to this anhydrobiotic structure. In contrast, under water-deficit conditions in vegetative tissues, the same protein displays a diffuse localization in both the cytoplasm and nucleus. Additionally, we will investigate the potential role of this protein in pollen grains, another anhydrobiotic structure in the plant. As part of our study, we will present the native ultrastructure of *Arabidopsis* pollen obtained using Cryo-Electron Tomography (Cryo-ET), a technique that enables us to study structure-function relationships at molecular and cellular resolution in anhydrobiotic systems. This work is supported by Consejo Nacional de Ciencia y Tecnología-México (CONACyT F-1615 221448) and by Water & Life Interface Institute (WALII).

Poster 5. ROS dynamics among three *Agave angustifolia* Haw. phenotypes

Alexis I. Cadena-Ramos and Clelia De-la-Peña*

Unidad de Biotecnología, Centro de Investigación Científica de Yucatán, Calle 43 No. 130 x 32 y 34 Col. Chuburná de Hidalgo, 97205 Mérida, Yucatán, México.
*Email: clelia@cicy.mx.

Reactive oxygen species (ROS) are highly reactive molecules generated as metabolic byproducts in living organisms, playing dual roles as cellular damage agents and signaling molecules. In plants, ROS are primarily produced in organelles such as chloroplasts, peroxisomes, and mitochondria, with additional contributions from the cell wall and plasma membrane, especially under stress conditions. This thesis investigates ROS dynamics in Agave angustifolia Haw., focusing on green, albino, and variegated phenotypes. Transcriptome analysis revealed strong upregulation of antioxidant enzyme genes in albino plants, indicating enhanced ROS-responsive mechanisms. Conversely, proteomic analysis showed fewer differentially accumulated proteins in albino plants, though key enzymes—including cytoplasmic ascorbate peroxidase (APX), catalase (CAT), and superoxide dismutase (SOD)—were elevated. Histochemical localization using nitroblue tetrazolium (NBT) revealed intense superoxide accumulation in albino and non-green sectors of variegated plants, while fluorescent probing with 2',7'-dichlorofluorescein diacetate demonstrated ROS accumulation in the apoplast and stomatal regions of albino phenotypes, whereas in variegated tissues, ROS were predominantly found in the cytoplasm of non-photosynthetic areas. Notably, the absence of functional chloroplasts in albino plantlets suggests that ROS originate mainly from non-chloroplastic sources such as the plasma membrane (via NADPH oxidases), peroxisomes, and the apoplast, rather than the photosynthetic electron transport chain. This is supported by the observed apoplastic and stomatal ROS localization, as well as the downregulation of mitochondrial and Krebs cycle proteins, indicating altered metabolic and redox pathways. In variegated plants, ROS production is compartmentalized, with cytoplasmic accumulation restricted to non-green regions, reflecting the interplay between chloroplast-dependent and independent ROS sources. Enzymatic analysis confirmed higher SOD activity in albino and variegated tissues compared to green tissues, supporting molecular findings. These results reveal distinct ROS production and antioxidant strategies among phenotypes, with albino plantlets exhibiting unique adaptations to cope with light-induced oxidative stress.

Poster 6. The absence of HXK reduces the survival of *Arabidopsis thaliana* subjected to freezing challenge

King-Díaz B[1], Mendieta-Juárez AL[2], Burgos-Palacios A[1], Sánchez-Nieto S[1*]

[1]Sugar Transport and Perception Laboratory in Plants, Department of Biochemistry, Faculty of Chemistry, National Autonomous University of Mexico, 04510, Mexico City, Mexico. 2 Instituto Tecnológico de Milpa Alta. Independencia Sur No. 36, Colonia San Salvador Cuauhtenco, Alcaldía Milpa Alta, C.P. 12300, CdMx.
*Email: sobeida@unam.mx.

Hexokinases are best known for their hexose phosphorylating activity, but they also exhibit various functions, including acting as a glucose sensor, a protein kinase, and an RNA processing protein. In *Malus domestica,* HXK1 is involved in protecting the plant from salt stress by phosphorylating two proteins: one transcription factor and the vacuolar Na^+/H^+ antiporter. Overexpression of CsHXK4 in *Camelia sinensis* made the plants resistant to cold temperatures by an unknown mechanism. The goal of this work was to investigate the participation of the HXK in the resistance to freezing challenge in *Arabidopsis thaliana*. To this end, four lots of plants were grown for six weeks at 22°C, then challenged at subzero temperatures for 2 h, and then kept for 2 weeks at 22°C for recovery. The plants used were: 1) wild type, 2) wild-type treated for two weeks with *N*-acetyl-D-glucosamine (an HXK inhibitor), 3) the null HXK1 mutant, gin2-1, and 4) the gin2-1 complemented with the HXK4 from maize. The results show that the wild-type plants and the gin2-1 plants complemented with ZmHXK4 survive the freezing temperatures. However, gin2-1 plants and the wild-type plants treated with the HXK inhibitor did not survive the challenge. The results suggest that the HXK is a key in the survival of the plants to the freezing challenge.

Poster 7. The m6A reader YTH factor ECT8 contributes to seed germination in response to abscisic acid in Arabidopsis

Ana Gabriela López-Pérez[1], José Martín-Rodríguez[1], Claudia Díaz Camino[1], Yuniet Hernández[1], Cécile Bousquet-Antonelli[2], Alejandra A Covarrubias[1], José L Reyes[1]*

[1]Instituto de Biotecnología, Universidad Nacional Autónoma de México. Av. Universidad 2001, Col. Chamilpa, Cuernavaca Mor. CP 62210 MEXICO..
[2]Institut de Biologie Moléculaire des Plantes, CNRS, Université de Strasbourg, 12 rue du Général Zimmer, 67084 Strasbourg, France.
*Email: jose.reyes@ibt.unam.mx

Methylation of adenosine at the N6 position (m^6A) is the most common internal modification found in eukaryotic mRNA and affects the fate and function of the modified transcripts, thus regulating fundamental biological processes. In plants, proteins containing a highly conserved domain known as YTH recognize m^6A in modified mRNAs to regulate their fate by inducing their translation, promoting degradation or stabilizing the transcripts. In Arabidopsis, we show that the transcript encoding for ECT8, a protein containing the YTH domain, increases its accumulation under different conditions associated with water deficit or abscisic acid (ABA) treatment. In germination tests, we observed that three different *ect8* mutant lines showed reduced sensitivity to ABA. Using RNA-seq, we identified genes involved in ABA signaling that were affected during germination in one of these mutants. Among them, we found that the transcript encoding PYL8, an ABA receptor is m6A-modified. Using a complementation line expressing pECT8:ECT8:GFP, we performed co-immunoprecipitation assays and confirmed that PYL8 mRNA is directly recognized by ECT8. Using the same line and a transient expression system, we also confirmed that ECT8 localizes to cytoplasmic bio-condensates and colocalizes with RBP47, a stress granule marker. Finally, LARP1a, another RNA binding protein that affects mRNA half-lives, was suggested to interact with ECT8 based on two hybrid assays. We later confirmed that both genes interact genetically in response to ABA during germination, and that the two proteins physically interact *in vivo*. These results show that ECT8 regulates ABA responses during germination in Arabidopsis by recognizing m6A-modified transcripts. We suggest that LARP1 contributes to the regulation these mRNAs within stress response pathways regulated by mRNA methylation. This work is supported by DGAPA-PAPIIT project IN-212124 to JLR, and ECOS-Nord project 315630 to JLR and CBA.

Poster 8. **Differential responses of grain amaranths to waterlogging stress**

Padilla Escamilla Jazmín Patricia[1], García Ortega Luis Fernando[2], Ríos Castro Emmanuel[3], González de la Vara Luis Eugenio[1], Délano Frier John Paul[1*]

[1]Departmento de Biotecnología y Bioquímica, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Unidad Irapuato, Irapuato, Guanajuato, México
[2]Departmento de Ingeniería Genética, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Unidad Irapuato, Irapuato, Guanajuato, México
[3]Unidad de Genómica, Proteómica y Metabolómica (UGPM), LaNSE, Cinvestav-IPN, Ciudad de México, México
[*]Email: john.delano@cinvestav.mx

Grain amaranth stands out for its high nutritional value and its ability to grow in extreme environments. They are also known to have a high tolerance to salinity, drought, defoliation and other (a)biotic stresses. However, their response to waterlogging-induced stress has been barely explored. This project describes the response of two grain amaranth species (*Amaranthus cruentus* and *A. caudatus*) subjected to different waterlogging treatments under greenhouse conditions, as well as the impact that waterlogging stress had on seed yield and quality. In both species, the phenotypic responses to this stress condition included the development of adventitious roots, complete loss of the primary root, disrupted development and alterations in the plant's life cycle. However, *A. cruentus* produced adventitious roots earlier and more abundantly than *A. caudatus*, which most probably allowed *A. cruentus* to survive longer in waterlogged soils. Seed yield and viability produced by surviving stressed plants were evaluated. *A. cruentus* was identified as the more tolerant species, since no significant differences with control, unstressed plants, were observed in these parameters. In contrast, the fewer *A. caudatus* plants that recovered from waterlogging stress exhibited a marked reduction in seed production, in addition to delayed germination and lower seed viability. Seed protein composition was analyzed by mass spectrometry. This proteomic analysis revealed 137 differentially expressed proteins in *A. cruentus* under treatment conditions, 84 in *A. caudatus*, and 186 differential proteins between the two species ($p < 0.05$). These changes occurred predominantly in seed storage proteins and starch/oil synthesis- and stress-related proteins The results obtained in this comparative study contribute to further understand the mechanisms employed by grain amaranths to tolerate stressful ambient conditions. Also, they support the proposal that *Amaranthus cruentus* represents a promising model for studying the mechanisms of waterlogging stress tolerance in plants.

Poster 9. Molecular basis of the protective activity of LEA6 proteins during *in vitro* dehydration

Dayana Martínez Basulto, Inti Arroyo Mosso, Francisco Campos, Alejandro Arroyo Mosso, Iván Arenas Sosa, Elda F. Morales Velarde, Alejandro Garciarrubio, and Alejandra A. Covarrubias*

[1]Plant Molecular Biology Department, Instituto de Biotecnología, Universidad Nacional Autónoma de México. Cuernavaca, Mor. México.
*Email: alejandra.covarrubias@ibt.unam.mx

Abiotic factors such as water scarcity, salinity, and extreme temperatures are environmental conditions that significantly affect plant growth, development, and productivity. To cope with these conditions, plants have evolved various physiological, biochemical, and molecular responses that allow them to minimize cellular damage and maintain their functionality. Among these responses is the accumulation of Late Embryogenesis Abundant (LEA) proteins, which are intrinsically disordered proteins commonly accumulated under water-deficit. *In vitro* evidence indicates that these proteins have protective capabilities toward macromolecules and cellular structures during dehydration processes. In this study, we focused on the family the LEA protein family 6, originally identified in our laboratory, with the aim of understanding the molecular mechanism involved in the protection these proteins exert on target proteins *in vitro* under progressive dehydration. TheAtLEA6-2.1 protein from Arabidopsis was used as model. From the wild-type protein, mutants were generated in conserved residues; these were expressed in bacteria and purified to evaluate them in in vitro progressive dehydration assays. Lactate dehydrogenase (LDH) enzyme was used as a model target protein. Analysis of the different mutants showed that those with mutations in the conserved LEDYK motif had significantly lower protective capacity that the wild-type protein. Evaluation of additional mutants in this motif suggested the involvement of a possible helix formed in this region. With the support of bioinformatic structural models, we propose a mechanism of action. These findings will allow to advance our understanding of the structural elements responsible for the protective activity of LEA6 proteins in plants. Project supported by CONAHCyT CF-2023-I-503.

Poster 10. The AtLEA6-2.1 protein plays a key role in desiccation tolerance, longevity, and maintenance of the vitreous state in seeds

Inti A Arroyo-Mosso[1], U G V S S Kumara[2], Alejandro Garciarrubio[3], Thomas C Boothby[2] and Alejandra A Covarrubias*[1]

1Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, Mexico.
[2]Department of Molecular Biology, University of Wyoming, Laramie, Wyoming, USA.
[3]Departamento de Ingeniería Celular y Biocatálisis, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, México.
*Email: alejandra.covarrubias@ibt.unam.mx

Plants have evolved a wide range of adaptive and adjustment mechanisms to deal with different levels of dehydration. Among these are those involving LEA (Late Embryogenesis Abundant) proteins, which accumulate abundantly during seed maturation and in vegetative tissues under water-deficit conditions. Due to their ubiquity and conservation across the plant kingdom, as well as their close association with low water availability, understanding their function is essential to reveal how plants endure adverse environmental conditions while continuing their development. In this study, we analyzed the AtLEA6-2.1 protein, a member of Group 6 (LEA6) of the LEA proteins in *Arabidopsis thaliana*. Phylogenetic analyses indicated that this family emerged with seed plants, suggesting a specialized function in seed viability. Using mutant lines, we demonstrated that AtLEA6-2.1 is crucial for the acquisition of desiccation tolerance in seeds. Furthermore, we found that this protein is essential for seed longevity, as its absence leads to reduced germination after both natural and accelerated aging. Interestingly, this phenotype was associated with increased fragility of the vitreous state and lower cellular viscosity in the seed—parameters that correlate with seed viability. So far, AtLEA6-2.1 is the only protein for which these properties have been reported. Finally, we evaluated its protective capacity *in vitro* under progressive dehydration, and found that the acidic nature of these proteins requires specific conditions to exhibit their function. Altogether, our results position AtLEA6-2.1 as a key component in the plant response to water scarcity, as well as in seed longevity and the maintenance of the vitreous state. Project supported by CONAHCyT CF-2023-I-503.

Poster 11. **The role of XAL2, AGL24, and SOC1 in the cross-effect of root growth and osmotic stress**

Ramírez-Aguirre E.[1], Sánchez M. P.[1], Álvarez-Buylla E.[1], Garay-Arroyo A[1*].

[1] Laboratorio de Genética Molecular, Desarrollo y Evolución de Plantas, Depto. de Ecología Funcional, Instituto de Ecología, Universidad Nacional Autónoma de México, Ciudad de México, México.
*Email: agaray@iecologia.unam.mx

One of the most common environmental challenges for plants is osmotic disequilibrium, which induces Ca_2^+ and ROS signaling, affecting the auxin pathway and eliciting mechanisms to prevent the loss of water. When osmotic stress occurs during primary root development, root growth is limited, affecting cell-wall production and cell elongation. Transcription factors mediate the response to stress, maintaining the developmental pathways even during stressful conditions. Previous data suggest that *XAL2*, *AGL24*, and *SOC1* MADS-box transcription factors have antagonistic effects on root growth. *XAL2* is a growth promoter, but *AGL24* and *SOC1* repress *XAL2*, limiting root growth. However, under osmotic stress conditions, only *XAL2* mutants' root growth is less affected than that of *SOC1* and *AGL24* mutants. Double mutants *xal2-2 agl24-4* and *xal2-2 soc1-6* show a slightly shorter primary root than WT, and the root of *agl24-4 soc1-6* does not differ from WT. We also analyzed the functional roles of three *XAL2*, *AGL24*, and *SOC1* in root development with a combinatorial bulk RNA-seq among six mutant lines and the WT line (Col-0) under control and osmotic stress conditions. *XAL2*, *AGL24*, and *SOC1* respond differently to osmotic stress. *XAL2* functions are related to oxidative stress and cell-wall production, *SOC1* is associated with flavonoid and phenylpropanoid biosynthesis, and AGL24 is associated with pyrophosphatase activity. Under stress conditions, XAL2 is associated with cell-wall production and response to deprivation of water; SOC1 is associated to response to osmotic stress and different phosphatases activity, and AGL24 is associated to water deprivation and cold, among others.These results suggest that both *XAL2* and *SOC1* are involved in two different pathways elicited by osmotic stress, and that *AGL24* may affect this response indirectly.

Poster 12. **Functional characterization of transcription factors involved in root penetration of *Arabidopsis thaliana* in compacted soils**

Lugo-Rodríguez Aurelia Andrea[1], González-Morales Sandra Isabel[1], Oropeza-Aburto Araceli[1], Bello-Bello Elohim[2], Herrera-Estrella Luis Rafael[1,3*]

[1]Center for Research and Advanced Studies of the IPN, Advanced Genomics Unit, Irapuato, Gto Libramiento Norte Carretera Irapuato-León 36821.
[2]Salk Institute for Biological Studies, La Jolla, CA, United States 10010 N Torrey Pines Rd, 92037
[3]Texas Tech University, Institute of Genomics for Crop Abiotic Stress Tolerance, Lubbock, TX, 1006 Canton Ave, 79409.
*Email: lherrerae@cinvestav.mx

Soil compaction is a phenomenon difficult to solve. Plant roots growing on compacted soils are usually exposed to a multi-stress environment, experiencing high mechanical impedance, low oxygen levels, water stress, and nutrient privation. During evolution, plants acquired different adaptive mechanisms, which include a broad range of anatomical, morpho-physiological, and molecular responses, to cope with the edaphic complexity of soil compaction. To better understand the genetic regulation of root responses to this type of stress, we focused on transcription factors (TFs) with potential regulatory roles. *A. thaliana* is a suitable model to study the molecular mechanisms underlying root penetration under controlled conditions. Therefore, the aim of this study was the characterization of transcription factors related to root penetration using transcriptomes from comparisons between Ler-0 and Sha accessions. For this purpose, we developed a biphasic agar system based on the Root Penetration Index (RPI) assay (Bello-Bello *et al*., 2022) where a 2% agar concentration successfully simulated increased mechanical impedance (>2 MPa), providing a reliable *in vitro* model for studying root penetration in *A. thaliana*. Also, we supplemented in one treatment the upper agar with 10% PEG 8000 to simulate water stress, this combined-stress setup provided a reliable *in vitro* model for studying root penetration in *A. thaliana* under osmotic and physical constraints. Genotyping confirmed six homozygous T-DNA insertion mutants for transcription factors potentially involved in root response to compaction. Penetrability tests of mutant lines under both single (mechanical) and combined (mechanical + water) stress showed the expected phenotype under mechanical stress alone, characterized by low penetration into compacted soil and altered root architecture. Furthermore, these results confirmed that plants employ various morphological and molecular mechanisms that act synergistically to improve their penetration capacity in soils with high mechanical resistance.

Poster 13. Soybean improvement through gene editing for Mexican producers

Adriana Vargas-Almendra, Roberto Ruiz-Medrano, Berenice Calderón-Pérez y Beatriz Xoconostle- Cázares*

Departamento de Biotecnología y Bioingeniería, Centro de Investigación y de Estudios Avanzados (CINVESTAV), Ciudad de México 07360, México.
*Email: bxoconos@cinvestav.mx

Soybean is one of the most important crops worldwide due to its high seed protein and oil content, which are widely used in both food and industrial applications. However, soybean productivity is constrained by several biotic and abiotic factors, with drought being one of the most severe, capable of causing yield losses exceeding 80%. Therefore, the development of new varieties with improved tolerance to water deficit is of great importance. In this study, we targeted the TREH and ARGAH genes in soybean using the CRISPR/Cas9 system to generate drought-tolerant plants. Genetic transformation of the cultivar Huasteca 400 mediated by *Agrobacterium tumefaciens* was standardized using the RUBY:35S reporter gene. Key parameters during co-cultivation, including temperature, bacterial strain, incubation time, and photoperiod, were optimized, resulting in multiple explants exhibiting reporter gene expression, visible as a reddish coloration. Transformation efficiency was highest when using the *A. tumefaciens* C58C1 strain under co-cultivation for five days at 23 °C with a 16/8 h light/dark photoperiod. Based on these optimized conditions, transformations with the TREH and ARGAH constructs were performed, achieving an efficiency of 8%. Surviving explants under selection with ammonium glufosinate were validated by both the ImmunoStrip® assay and PCR amplification. Although *in vitro* regeneration of the Huasteca 400 variety is limited during the shoot elongation phase, we found that adjusting herbicide concentration and exposure time improved plantlet development at this critical stage, enabling the successful induction and elongation of multiple shoots.

Poster 14. Proteomic insights into the role of eIF4E homologous protein (4EHP) during heat stress response in *Arabidopsis thaliana*

[1]Karen Quiroz Nuñez, [2]Jorge Herrera Díaz, [1]Tzvetanka D. Dinkova

[1]Departamento de Bioquímica.
[2]Unidad de Servicios de Apoyo a la Investigación y la Industria, Facultad de Química, Universidad Nacional Autónoma de México, 04510, CDMX, Mexico.
Email: kanquisoy@gmail.com

4EHP is a non-canonical class II isoform of the eIF4E family, which plays a key role in translation regulation by recognizing the 5'cap of mRNAs. In *Arabidopsis thaliana*, the absence of 4EHP results in delayed flowering and increased production of rosette leaves, inflorescences, and siliques. Furthermore, under heat stress 4EHP knockout mutants exhibit enhanced heat tolerance compared to the wild-type plants. To investigate the role of 4EHP in heat stress response, here we analyzed proteomic changes, by shotgun and two-dimensional gel electrophoresis (2DGE), in wild type and *4ehp* knockout *A. thaliana* plants exposed to acclimation and heat stress. Shotgun proteomic analysis revealed a higher accumulation of heat shock proteins (HSPs) including HSP101, HSP70.1, HSP81.2, HSP60, HSP20, and HSP17.6 in the mutant line under heat stress. Additionally, through the 2DGE gel electrophoresis patterns we identified differentially accumulated protein spots, some of which were represented by stress-related proteins including HSP20 and Glutathione S-transferase, among others, and were more abundant in the mutant. A Gene Ontology (GO) functional classification of shotgun identified proteins revealed overrepresentation of proteins involved in heat stress response, recovery, regulatory processes, and cell growth in the mutant line compared to wild type. These results suggest that 4EHP may function as a regulator of protein synthesis during heat stress, possibly through its role in stress granule dynamics. Acknowledgement to financial support: PAPIIT: IN218921, IN212024; SECIHTI: CF-2023-1-115; FQ: PAIP 5000-9118.

Poster 15. **Enhancing drought tolerance in malting and forage barley through mutagenesis**

Cruz-Muñoz, D.C., Rodríguez-Gandarilla, M. G[1]., Avila-Perches, M.A., Urrea-López, R., Massange-Sánchez, J. A.[1]*

[1]Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C. (CIATEJ), Unidad de biotecnología vegetal, Zapopan, México. Camino Arenero 1227, El Bajío, 45019, Zapopan, Jalisco.
[2]Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP), Campo experimental Bajío, Mexico
*Email: jmassange@ciatej.mx

Barley (*Hordeum vulgare* L. ssp. *vulgare*) is the world's fourth most important cereal, primarily used for animal feed and malting. Various barley varieties have been developed to meet these distinct uses and adapt to diverse environments. However, increasing climate change pressures underscore the need to enhance crop resilience. Induced mutagenesis using ethyl methanesulfonate (EMS) is a well-established strategy for increasing genetic variability and selecting for key traits. This study aimed to determine the optimal EMS treatment in malting and forage barley for the selection of drought-resistant mutants. Seeds underwent EMS treatments at varying concentrations (0.1%–1.0% v/v) and exposure times (1–16 h), assessing germination, survival, shoot and root length, biomass, and seedling vigor. The optimal EMS treatment was 0.5% for 2.7 h in the forage variety and 2.6 h in the malting variety. A set of mutants was subsequently subjected to drought stress; ten forage mutants survived, whereas none from the malting group did. Further selection based on leaf water loss identified three promising forage mutants (For-7, For-8, and For-10), highlighting their potential for breeding drought-tolerant barley. These findings advance our understanding of mutagenesis in barley and support future efforts to enhance stress tolerance in this vital crop.

Poster 16. Regulation of a long non-coding RNA in response to heat stress in *Arabidopsis thaliana*

Vargas Susana, Nieto Jesús and Arenas-Huertero Catalina

Laboratorio del metabolismo del RNA, Facultad de Ciencias, Universidad Autónoma de San Luis Potosí, San Luis Potosí.

The agricultural industry has been profoundly affected by the drastic increase in global temperatures, which has resulted in significant losses in the production of essential food crops like soybeans, maize, and wheat. In response to this challenge, various genes associated with heat stress have been identified, including members of the *HSF* (Heat Shock Factor) family, which are considered key elements in plant defense against this type of stress. Studying these genes is crucial for understanding the mechanisms of heat adaptation and for identifying tools that help elucidate this process. Not only have protein-coding genes been identified, but also non-coding RNAs that are positively regulated under heat stress conditions. In *Arabidopsis thaliana*, a long non-coding RNA (lncRNA) named *lincEIN2* was found to be expressed under heat stress. This lncRNA is encoded downstream of the gene that codes for the *EIN2* (ETHYLENE INSENSITIVE 2) protein, a key component of the ethylene perception pathway. Using the reporter line promlincEIN2: GUS, it was confirmed that *lincEIN2* is expressed under heat stress in true leaves, and its expression depends on the temperature to which the plant is exposed. Furthermore, silencing lines of *lincEIN2* revealed its involvement in thermotolerance and in the ethylene perception pathway through interaction with the carboxyl-terminal domain of EIN2.

Poster 17. Multi-omics analyses on Maya-land tomatoes shed light on plant strategies to thrive in high temperatures

Luis Fernando Maceda-López[1,2], Annie Espinal-Centeno[2], Jose Juan Ordaz-Ortiz[1]* and Alfredo Cruz-Ramírez[2]*

[1]Metabolomics and Mass Spectrometry Laboratory, Unidad de Genómica Avanzada, Centro de Investigación y Estudios Avanzados, Instituto Politécnico Nacional, Irapuato 36824, Guanajuato, México.
[2]Molecular and Developmental Complexity Group, Unidad de Genómica Avanzada, Centro de Investigación y Estudios Avanzados, Instituto Politécnico Nacional, Irapuato 36824, Guanajuato, México. Presenting and Corresponding Authors' *Emails: alfredo.cruz@cinvestav.mx; jose.ordaz.ortiz@cinvestav.mx

Heat stress significantly limits tomato yield and fruit quality, making it essential to identify genes and metabolites linked to heat tolerance. In this study, we analyzed the transcriptomes and metabolomes of roots and shoots under heat stress in a Maya-land ecotype of heirloom tomato (Calkiní), which grows in lowlands and elevated temperatures. These profiles were compared to those of another heirloom ecotype (Acaxochitlán) that grows in highlands and lower temperatures. Our omics approach revealed that several transcripts encode enzymes involved in various biosynthetic pathways, leading to the differential accumulation of metabolites. While both ecotypes activate heat stress response mechanisms, the roots of the Calkiní ecotype exhibited a greater accumulation of diverse metabolites and up-regulation of key genes linked to molecular and physiological strategies for coping with heat stress. A major finding of this study is the increased accumulation of diverse metabolites and the up-regulation of key genes involved in phenylpropanoid and suberin biosynthesis in the heat-tolerant Calkiní ecotype. These molecular changes are consistent with significant suberin deposition in the exodermis and endodermis of Calkiní roots, along with elevated gibberellin levels in the meristematic zone under heat stress conditions. Additionally, we observed differential expression and metabolite accumulation related to the PA-GAPC-NFY heat stress tolerance pathway between the two ecotypes. Our findings highlight the importance of studying native Mexican tomato varieties to uncover traits that enable plants to withstand diverse biotic and abiotic stresses, offering valuable insights into the genetic and metabolic pathways underlying heat tolerance across ecotypes and tissues.

Poster 18. **Selection of reference genes and identification of marker genes for drought stress and senescence in forage and malting barley**

Rodríguez-Gandarilla, M. G., Massange-Sánchez, J. A.*

Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C. (CIATEJ), Unidad de biotecnología vegetal, Zapopan, México. Camino Arenero 1227, El Bajío, 45019, Zapopan, Jalisco.
*Email: jmassange@ciatej.mx

Gene expression analysis is a key tool for understanding the molecular mechanisms that regulate plant development and stress responses. While RNA-seq enables global transcriptome profiling, RT-qPCR remains the most accessible and reliable method for targeted expression studies, provided that stable reference genes are used. Barley (*Hordeum vulgare* L. ssp. *vulgare*) is the fourth most important cereal crop globally, primarily cultivated for forage (~65%) and malting (~30%). Varieties developed for these purposes differ in growth cycle, cultivation conditions, and climatic adaptation. In this study, 14 candidate reference genes previously reported in the literature were evaluated in developmental tissues (young leaf, flag leaf, senescent leaf, and grain) and under drought stress (young leaf) in forage and malting barley varieties. Additionally, 14 marker genes associated with drought stress and senescence were analyzed. All reference genes showed amplification in endpoint PCR; however, only seven (TUBα, EF1, UPL, EIF5A2, MALD, HSP70 and qRefB) yielded specific bands on agarose gel and a single melting curve peak. Out of the 14 marker genes evaluated, eight were successfully amplified and exhibited high specificity. These included P5CS2, Hsdr4, HSP17, and WRKY1, associated with drought response, and RBCS, GS2, SAG12, and AGTX, associated with senescence. Stability analyses using BestKeeper, NormFinder, and GeNorm are currently in progress to identify a robust set of reference genes and validate stress-responsive markers for use in barley genetic improvement under defined environmental conditions. Preliminary results underscore the importance of validating reference genes under specific experimental conditions, as literature-based selections may not always yield optimal performance.

Poster 19. Effects of heat stress on carbohydrate synthesis in tolerant and sensitive wheat genotypes

Encinas-Montes, AL., Assaf-Torres, JA., Huerta-Ocampo JA., Vazquez-Moreno L[1]., Hernandez-Leon, SG*

Centro de Investigación en Alimentación y Desarrollo A.C., Gustavo Enrique Astiazarán Rosas No. 46 Col. La Victoria, Hermosillo, Sonora, México.
Email: sergio.hernandez@ciad.mx

Abiotic stress represents a major challenge for agriculture. Heat stress (HS) has become a leading cause of crop losses in various crops, such as wheat, significantly affecting production and yield. Wheat (*Triticum aestivum*) is one of the most important staple foods, ranking third in cereal production worldwide. The HS caused by climate change influences growth and development at all plant stages, causing alterations that negatively impact their productivity. Therefore, it is important to study various physiological and biochemical processes to find genotypes tolerant to climate change. Herein, two experiments were performed: control and HS during the vegetative stage using four bread wheat genotypes (two sensitive and two tolerant to HS, classified according to their grain yield reduction under HS at the reproductive stage in field conditions) in a greenhouse at Centro de Investigación en Alimentación y Desarrollo (CIAD) in Hermosillo, Sonora, Mexico. The content of chlorophyll, carotenoids, and starch, as well as RuBisCO and Sucrose Phosphate Synthase (SPS) activity were determined. Genotype 6 (tolerant) showed increased susceptibility, reflected in reduced total chlorophyll, carotenoid, starch content, and RuBisCO activity. In contrast, genotype 24 (sensitive) showed a more favorable response by maintaining RuBisCO activity, conserving chlorophyll and carotenoid levels, and increasing starch content, suggesting greater efficiency in metabolic adaptation under heat conditions at the vegetative stage. Genotypes 18 (sensitive) and 26 (tolerant) increased SPS activity, which could be associated with compensatory mechanisms to maintain carbon flow. Due to heat stress, carbohydrate synthesis is affected in a genotype-dependent manner. However, genotype 24 managed to generate a certain tolerance to continue carbohydrate synthesis and have the resources to face the following stages under heat stress conditions. Understanding these differential responses is essential to identify physiological and biochemical characteristics associated with heat tolerance.

Poster 20. Role of XAL2 in the response to salt stress in *Arabidopsis thaliana*

Manuel Lucas Mateo[1,2], M. Elena Álvarez-Buylla Roces[1], María de la Paz Sánchez Jiménez[1], Adriana Garay Arroyo[1*]

[1]Laboratorio de Genética Molecular, Epigenética, Desarrollo y Evolución, Instituto de Ecología, Universidad Nacional Autónoma de México, CDMX, México.
[2]Posgrado en Ciencias Bioquímicas, Universidad Nacional Autónoma de México, CDMX, México.
*Email: agaray@iecologia.unam.mx

Salinity is an important factor limiting plant growth and development. Plants respond to excess salt through complex genetic regulatory networks involving numerous genes and transcription factors, including members of the MADS-box family. *XANTAAL2* (*XAL2*/*AGL14*), a type II MADS-box gene, is predominantly expressed in the primary root, where it promotes root development. Recently, XAL2 has been associated with the regulation of salt-responsive genes. However, the role of XAL2 in response to salt stress remains to be identified. Therefore, we characterized the function of XAL2 in response to salt stress. We found that XAL2 acts as a negative regulator of primary root growth under salt and osmotic stress conditions because loss-of-function mutants exhibit increased salinity resistance, while overexpressing lines are more sensitive. In contrast, during germination under salt stress, XAL2 acts as a positive regulator, as overexpression lines had a higher germination rate, while loss-of-function lines showed the opposite phenotype. Under control conditions, XAL2 accumulates in the primary root (vascular bundle), lateral roots, and their primordia. Interestingly, we observed that under salt stress, XAL2 accumulation increases in the vascular bundle of the primary root, lateral roots, and their primordia. We also examined the expression pattern using a reporter line that included the long intron of *XAL2* fused to GUS, without a minimal promoter. We found that the long intron of XAL2 is sufficient to drive a similar expression pattern, localized in the vascular bundle and lateral roots, but not in the primordia of lateral roots. This expression is enhanced under saline conditions. Overall, our results suggest that XAL2 is an important component in the response to salt stress.

Poster 21. Role of miR396 and growth regulating factors (GRF) in *Phaseolus vulgaris* leaf development during water deficit

María Beatriz Pérez-Morales, Cecilia Contreras-Cubas, Alejandra A. Covarrubias and José L. Reyes*

Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, UNAM. C.P. 62210. Cuernavaca, Morelos, México.
*Email: jose.reyes@ibt.unam.mx

Plants are sessile organisms constantly challenged by adverse environmental conditions such as drought, which strongly limits growth and productivity. MicroRNAs (miRNAs) are 21–22 nt small RNAs that post-transcriptionally regulate gene expression and play central roles in plant development and stress responses. The conserved miR396 family targets Growth-Regulating Factors (GRFs), a plant-specific transcription factor family that controls cell proliferation. In *Arabidopsis thaliana*, miR396 accumulates in the distal region of developing leaves, restricting GRF expression proximally and thereby attenuating cell proliferation. Given that inhibition of leaf growth is an early response to water deficit, we investigated the role of the miR396–GRF regulatory module in *Phaseolus vulgaris* under drought stress. We observed that common bean plants significantly reduce leaf growth rate and leaf number per plant in response to water deficit, while maintaining positive allometry under both control and stress conditions. Genome-wide analysis identified 11 *P. vulgaris GRF* genes, 9 of which contain a functional recognition site for miR396, suggesting direct regulation. In leaves subjected to water deficit, the *GRF* genes showed a complex pattern of mRNA accumulation while miR396 reduced its accumulation in the distal region under well-watered conditions but not under water deficit. In particular, *PvGRF9* decreased its accumulation, suggesting it is involved in the regulation of growth arrest under water deficit conditions.The functional characterization of the miR396–GRF module in *P. vulgaris* under drought stress provides new insights into the molecular mechanisms that modulate leaf development under adverse conditions. This work was partially supported by a grant from PAPIIT-DGAPA (IN212124) to JLR.

Poster 22. **Identification and characterization of ECT8 interacting proteins during stress response in *Arabidopsis thaliana***

Castro-Bustos S[1], Diaz-Camino C[1], Pérez-Morales B[1], Reyes-Taboada J.L[1*]

[1]Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Universidad 2001,62210, Cuernavaca, México.
[*]Email: jose.reyes@ibt.unam.mx

Adenosine methylation at the N6 position (m6A) is the most common internal modification present in eukaryotic mRNAs. Different protein complexes participate in this dynamic and reversible post-transcriptional modification pathway. Besides proteins capable of adding and removing the m6A, there are other proteins responsible for its recognition. So-called reader proteins recognize m6A transcripts and modulate their lifetime and intracellular destination. In plants, proteins with a YTH domain can specifically recognize m6A and particularly in *Arabidopsis thaliana* 13 proteins have been identified. Among these, ECT8 participates in the response to water deficit and forms stress granules in response to ABA and NaCl treatments. Furthermore, interaction with mRNA decapping protein 5 (DCP5) has been described. In addition, database search revealed seven possible interacting proteins, which are related to the response to stress and ABA. Herein, we are interested in exploring the possible interaction of ECT8 with these proteins to define its molecular function, to elucidate the pathways in which it participates, and to determine its contribution to stress responses. For this, we are performing water deficit stress experiments in Arabidopsis lines expressing an ECT8-GFP fusion protein to perform Co-immunoprecipitation Mass Spectrometry (Co-IP/MS) assays. To confirm this interaction, ECT8-candidates will be analyzed by Bimolecular Fluorescence Complementation (BiFC) using an Agrobacterium-mediated transient transformation system in *Nicotiana* leaves. Live cell images will be visualized under a confocal microscope. Because ECT8 has been observed within biocondensates in response to stress, we will determine the cellular localization of the protein-protein interactions identified here. Identification of the ECT8 interactors will contribute to our understanding of the mechanisms employed by plants to cope with adverse conditions. This work is supported by a grant from PAPIIT-DGAPA, UNAM (IN212124) to JLR.

Poster 23. **Spatio-temporal expression analysis of *Physcomitrium patens* *PpDREB-A3-like* genes under abiotic and energetic nutritional stress**

Alejandro Serratos-Ramírez[1], Angélica Concepción Martínez-Navarro[1], Alejandra Chamorro-Flores[1], Miguel Angel Villalobos-López[1], Patricia León-Mejía[2], Analilia Arroyo-Becerra[1*]

[1]Centro de Investigación en Biotecnología Aplicada, Instituto Politécnico Nacional, Ex-Hacienda San Juan Molino Carretera Estatal, Km 1.5, 90700 Santa Inés Tecuexcomac, Tlaxcala, México.
[2]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Av. Universidad 2001, Col. Chamilpa, Cuernavaca, Morelos, 62210 México.
*Email: alarroyo@ipn.mx

Transcriptional factors of the DREB-A3 type belong to the AP2/EREBP family and are specific to plants. In most vascular plants, this group is represented by single member, ABI4, which acts as a key regulator in the abscisic acid (ABA) signaling pathway, sugar level perception and plays central roles during early stages of germination and abiotic stress responses. While ABI4 has been extensively studied in vascular plants, knowledge of this transcription factor in non-vascular plants remains minimal. In our research group, we identified two genes homologous to *Arabidopsis thaliana ABI4* in the moss *Physcomitrium patens*. To investigate the regulation of *PpDREB-A3-like1* and *PpDREB-A3-like2,* we analyzed their putative *cis*-regulatory elements associated with abiotic stress. Expression analyses showed that both genes respond to high-carbon conditions, ABA, osmotic stress, salinity, heat and dehydration. To further characterize their regulation, we generated transgenic plants carrying transcriptional fusion of *PpDREB-A3-like1* with GFP. The *PpDREB-A3-like1::Δabi4*GFP(2) line exhibit basal promoter activity at the juvenile protonemal stage, with high activity under abiotic stress. At gametophore (under dehydration stress) and sporophyte stages, the *PpDREB-A3-like1* promoter also displayed basal activity. Additionally, we found that *PpDREBA3-like1* contributes to brachyocyte development under extreme abiotic stress conditions, including cold, freezing, heat and dehydration. We thank Instituto Politécnico Nacional, SIP, BEIFI, and SECIHTI for financial support.

Poster 24. The exocyst and autophagy: their interrelationship during the response to osmotic stress and endoplasmic reticulum stress.

Ausencio Galindo, Salvador Barrera-Ortiz, Luis Castillo-Olamendi, and Helena Porta*

[1]Departamento de Biología Molecular de Plantas. Instituto de Biotecnología, Universidad Nacional Autónoma de México. Av. Universidad 2001, Cuernavaca, Morelos 62210, México.
*E-mail: helena.porta@ibt.unam.mx.

Autophagy is a conserved catabolic process in most eukaryotic cells that helps break down and recycle unwanted or dysfunctional cytosolic material. This includes proteins, protein complexes, nucleic acid aggregates, and even entire organelles. These components are enclosed in double-membrane vesicles called autophagosomes, which are later degraded after fusing with the vacuole. The exocyst is a heteroctameric protein complex involved in targeting and anchoring secretory vesicles to the plasma membrane during normal exocytosis. The exocyst consists of eight subunits (Sec3, Sec5, Sec6, Sec8, Sec10, Sec15, EXO70, and EXO84). In plants, the exocyst plays key roles such as polarized growth, cell wall formation, and responses to pathogens. Furthermore, the increasing number of genes encoding EXO70 suggests functional specialization across different tissues and environmental conditions. An example is EXO70B1, which is believed to be important during autophagy. In this study, Bimolecular Fluorescence Complementation (BiFC) assays in N. benthamiana showed interaction between ATG8i and EXO70B1 in the cytoplasm and the vacuole. Additionally, after inducing endoplasmic reticulum stress, this interaction intensified in the cytoplasm. To further explore the exocyst's role in autophagy, a construct was made to express GUS under the control of the EXO70B1 promoter (pMDC163-EXO70B1p), along with one for EXO70B1-GFP (pB7FWG0-EXO70B1p:EXO70B1). The significance of EXO70B1 during hydrotropic curvature was also examined using the exo70b1 mutant (GK-156G02). We aim to investigate the role of the exocyst at different stages of autophagosome formation, including autophagosome closure, fusion with the vacuole, and the potential for specific degradation of exocyst proteins through autophagy.

Poster 25. Atmospheric pollutants: toxic effects of benzo[*ghi*]perylene on *Physcomitrium patens*

Oyarvide-Rodríguez, E[1]. Arenas-Huertero, C[1].* & Arenas-Huertero, F.*

[1]Facultad de Ciencias de la Universidad Autónoma de San Luis Potosí, S.L.P, México, Av. Parque Chapultepec 1570, Privadas del Pedregal, C. P. 78295.
[2]Hospital Infantil de México Federico Gómez, Ciudad de México, México, Avenida Dr. Márquez 162, Colonia Doctores, Cuauhtémoc, C. P. 06720.
*Email: catalina.arenas@uaslp.mx; farenashuertero@yahoo.com.mx

Polycyclic aromatic hydrocarbons (PAHs) are organic pollutants released from human activities, particularly from vehicle and industrial emissions. The absorption of PAHs by plants depends on the structure of their benzene rings. Low molecular weight (LMW) PAHs, which contain 2 to 3 benzene rings, can enter plants through the stomata during foliar uptake. In contrast, high molecular weight (HMW) PAHs, with 4 to 7 benzene rings, can penetrate the cuticle and be absorbed by the roots, using both active and passive transport processes. Atmospheric studies conducted in Mexico City in 2006 and 2016 identified benzo[*ghi*]perylene as the predominant PAH. Although it is classified as non-carcinogenic to humans, the limited research on its toxic effects in plants raises concerns. Previous studies have shown that PAHs have a strong lipid affinity, enabling them to bioaccumulate and be transported throughout the food chain. This poses potential risks to economically important species and human health. Therefore, it is essential to assess the toxicity of benzo[*ghi*]perylene in *Physcomitrium patens*, a model moss with a haploid/diploid life cycle and protonemal tissue composed of chloronema and caulonema. This research is significant as it can provide valuable insights into plant cell development and differentiation, which could be applicable to a broad range of plant species.

Poster 26. A comparative study of adaptive stress tolerance in the Brassicaceae family

Andrea Ramirez[1], Prashanth Ramachandran[1], Evelyn Alferez[1], José Dinneny[1*]

Stanford University, Biology Department, Palo Alto, USA.
[*]Email: dinneny@stanford.edu

Due to their sessile lifestyle, plants often face adverse environmental conditions such as drought, flooding, and soil salinity. Salt stress is particularly damaging, representing one of the most severe global threats to crop productivity. Roots are the primary site of stress perception, where anatomical and physiological adjustments enable selective uptake of essential nutrients while restricting the accumulation of toxic ions. Yet, much of our current understanding comes from the salt-sensitive model *Arabidopsis thaliana*, limiting the discovery of adaptive traits that confer high levels of salt tolerance. The Brassicaceae family provides a powerful framework to address this gap, as it includes both *A. thaliana* and extremophiles such as *Eutrema salsugineum* and *Schrenkiella parvula*, which naturally thrive in saline environments. In this study, we employ a comparative phenomics approach that integrates root anatomical analyses with physiological and gene expression data to investigate mechanisms of stress adaptation. We identify striking differences in endodermal organization that likely influence solute retention and exclusion. These findings uncover key adaptive traits and offer new strategies for breeding or engineering stress-resilient crops.

Poster 27. Silicon and H^+-ATPase activity: implications in K^+ loss in barley under salt stress

Luis Enrique Flores-Pantoja[1,2], Cesar Leobardo Aguirre-Mancilla[1], Manuel Martínez-Estévez[3], Juan Gabriel Ramírez-Pimientel[1], Ahuitzolt de Jesús Joaquín-Ramos[1] and Isaac Zepeda-Jazo[2]*

[1]Doctorado en Ciencias en Producción Agroalimentaria, Tecnológico Nacional de México/IT de Roque. Carretera Celaya-Juventino Rosas, km.8, C.P. 38110.
[2]Licenciatura en Genómica Alimentaria, Universidad de La Ciénega del Estado de Michoacán de Ocampo. Avenida Universidad 3000, Col. Lomas de la Universidad, C.P. 59103.
[3]Centro de Investigación Científica de Yucatán, A. C. Calle 43 No. 130 x 32 y 34 Chuburná de Hidalgo. CP 97205.
*Email: z_isaac@hotmail.com

Silicon is considered a beneficial element for plants exposed to stress conditions. It has been reported that silicon induces a reduction in sodium (Na^+) uptake and maintains adequate levels of cytosolic potassium (K^+) in tissues in plants under saline environments. K^+/Na^+ homeostasis is essential for plant adaptation to salt stress. It has been proposed that silicon enhances H^+-ATPase activity and contributes to Na^+ exclusion and K^+ uptake. K^+ loss in plants exposed to NaCl is triggered by plasma membrane depolarization provoked by augmented intracellular Na^+ concentration, which is counteracted by increased H^+-ATPase activity. The objective of this study was to evaluate the effects of silicon on K^+ loss and net K^+ and H^+ fluxes in roots of barley seedlings (*Hordeum vulgare* var. Esperanza) under saline conditions. Seeds were germinated hydroponically in darkness (0.5 mM KCl and 0.1 mM $CaCl_2$) with and without silicon (0, 1 and 2 mM Na_2SiO_3, pH 5.7). Four-day-old seedlings were evaluated at three salinity levels (80, 160 and 320 mM NaCl). K^+ loss was determined by flame photometry after exposing seedling roots to the NaCl concentrations for 24 h. The net fluxes of K^+ and H^+ were measured *in vivo*, *in situ* and in real time using the MIFE® technique. The saline treatments were applied after five minutes of the record. Salt induced K^+ loss in the roots of control seedlings was inhibited by silicon, and this effect was dose-dependent under all salt conditions tested. Similarly, the K^+ and H^+ efflux was induced by salinity, silicon at both dosages inhibited outward K^+ transport while increasing H^+ release. These findings suggest that silicon modulates K^+ transport in barley roots under salt stress, which may be associated with reduced plasma membrane depolarization due to increased H^+-ATPase activity.

Poster 28. Mechanosensory signaling pathways underlying natural variation in root penetration responses in *Arabidopsis thaliana*

González-Morales Sandra I[1], Bello-Bello Elohim[3], Oropeza-Aburto Araceli[1], Herrera-Estrella Luis*[1,2]

[1]Laboratorio Nacional de Genómica para la Biodiversidad/Unidad de Genómica Avanzada, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Irapuato, Gto 36821, México.
[2]Department of Plant and Soil Science, Institute of Genomics for Crop Abiotic Stress Tolerance, Texas Tech University, Lubbock, TX 79409, United States.
[3]Plant Molecular and Cellular Biology Laboratory and Integrative Biology Laboratory, Salk Institute for Biological Studies, La Jolla, CA, USA.
*Email: lherrera@cinvestav.mx

Root system architecture plays a central role in plant adaptation to challenging soil environments. Soil compaction generates mechanical impedance that restricts root elongation and limits access to deep water and nutrient reservoirs, ultimately reducing crop productivity. Despite its agricultural relevance, the molecular basis of root mechanosensory responses remains poorly understood. Here, we combined time-resolved RNA-seq, gene regulatory network analysis, and functional categorization to dissect the mechanisms underlying natural variation in primary root penetrability among Arabidopsis thaliana accessions. Our results reveal the existence of a coordinated signaling route specifically activated by mechanical impedance during early root–soil interactions. This pathway integrates stress perception with transcriptional reprogramming and downstream metabolic adjustments, enabling tolerant accessions to sustain penetration under compacted conditions. Network-level analyses identified key regulatory nodes that orchestrate gene expression waves across successive stages of root penetration, pointing to a previously uncharacterized layer of mechanosensory signaling in plants. Collectively, these findings provide novel insights into the molecular determinants of root penetration capacity and open new avenues for engineering crops with enhanced resilience to compacted soils and improved capacity for deep carbon sequestration.

Poster 29. **Functional analysis of TELOMERASE ACTIVATOR 1 (*TAC1*), a telomerase regulator, in drought tolerance in *Arabidopsis thaliana***

Juan Fernando Gutiérrez-García[2*], Daniel Sierra-Cacho[1], Lisset Herrera-Isidrón[2]
Plinio Guzmán[1*]

[1]Departamento de Ingeniería Genética, Centro de Investigación y de Estudios Avanzados del IPN, Unidad Irapuato, Irapuato, Guanajuato 36824, México.
[2]UPIIG, Instituto Politécnico Nacional, Silao de la Victoria, Guanajuato 36275, México
[*]Email plinio.guzman@cinvestav.mx

Telomeres are essential DNA sequences that protect chromosome ends and ensure genomic stability through mechanisms conserved in eukaryotes, which include maintenance by the reverse transcriptase enzyme telomerase. Alterations in telomere dynamics trigger cellular senescence and are associated with aging processes, oxidative stress, and cancer. This study explores the relationship between dehydration—a type of abiotic stress that negatively impacts plant development—and telomere dynamics. Previous research showed a differential expression of telomere metabolism genes under water stress in wild-type (wt) *Arabidopsis thaliana* plants compared to the *atl78* mutant, which encodes for a ubiquitin ligase involved in dehydration tolerance. Among the differentially expressed genes, the TELOMERASE ACTIVATOR1 (*TAC1*)—a C2H2-type zinc finger transcription factor that promotes telomerase expression in *A. thaliana*—showed enhanced expression after one day of rehydration, following a prolonged period of dehydration. We are currently functionally characterizing *TAC1* and evaluating mutant lines, including knockout and ectopic expression variants, with conditional expression in response to dehydration. To this end, we have implemented a genetic system in *A. thaliana* inducible by drought through the use of a water deficit-sensitive promoter.

Poster 30. When phosphate deficiency meets realistic soil iron chemistry: roots grow, not arrest

Javier Mora-Macias[1,2,*], Alma Armenta-Medina[1,3], Carlos Erazo[1], Huajin Zheng[1], Mutsutomo Tokizawa[1], Rahul Chandnani[1], Hanh Nguyen[1], Connor Burbridge[1], Jon Shaff[4], Raju Datla[1], William J. Lucas[5,*], Leon V. Kochian[1*]

[1]Global Institute for Food Security, University of Saskatchewan, Saskatoon, Canada S7N 4J8
[2]Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C., Unidad Sureste, Parque Científico y Tecnológico de Yucatán, Tablaje Catastral 31264, Km 5.5 Carretera Sierra Papacal–Chuburná Puerto, C.P. 97302, Mérida, Yucatán, México
[3]Centro Nacional de Recursos Genéticos (CNRG), INIFAP, Jalisco, México
[4]Robert W. Holley Center for Agriculture and Health, USDA-ARS, Cornell University, Ithaca, NY 14853, USA
[5]Department of Plant Biology, College of Biological Sciences, University of California, Davis, CA 95616, USA
*Email: leon.kochian@gifs.ca; wjlucas@ucdavis.edu; jmora@ciatej.mx

For years, phosphate (Pi) deficiency in *Arabidopsis* has been described as a stress that arrests root meristems through iron (Fe) toxicity. This model, based on experiments with Fe supplied as Fe-EDTA, proposed that excess free Fe^{3+} inhibits root growth via specific malate transport and redox pathways. However, Fe-EDTA is a weak chelate that releases unusually high levels of soluble Fe^{3+} compared to what plants typically encounter in aerated agricultural soils. We asked what would happen if Pi deficiency were studied under conditions that better reflect realistic soil Fe chemistry. Using strong Fe chelates such as Fe-EDDHA and Fe-HBED, which maintain much lower levels of free Fe^{3+}, we observed the opposite of the classical view: Pi deficiency stimulated both primary and lateral root growth. Remarkably, this stimulation still required malate efflux and redox pathways and was accompanied by enhanced auxin activity in expanded root caps. Moreover, an auxin response regulator also contributed to this effect, suggesting a coordinated Fe-dependent mechanism that transforms Pi deficiency into an opportunity to expand the root system. Our findings overturn the long-standing model of Pi-induced root growth inhibition, showing that the outcome critically depends on Fe chemistry. Under soil-like Fe conditions, *Arabidopsis* roots do not shut down; they grow deeper and wider—just as we also observed in crop species—potentially improving P foraging.

Poster 31. **Uncovering the GST family in mango: from genes to heat stress adaptation**

Contreras-Vergara C. A. [1*], Paredes-Coronado N.[2]

[1]Laboratorio de Genética y Biología Molecular de Plantas y Hongos, Centro de Investigación en Alimentación y Desarrollo, A. C. Hermosillo, Sonora, Carr. Gustavo Enrique Astiazarán Rosas No. 46, 83304 México.
[2]Universidad de Sonora, Facultad Interdisciplinaria de Ciencias Biológicas y de la Salud, Hermosillo, Sonora, Luis Donaldo Colosio s/n, 83000 México.
*Email: contreras@ciad.mx

Glutathione S-transferases (GSTs) constitute a large multigene family of multifunctional enzymes that play essential roles in plant physiology. Beyond their canonical activity catalyzing the conjugation of diverse substrates with glutathione, GSTs contribute to fundamental processes such as growth, development, detoxification of xenobiotics, and protection against oxidative stress. Their functional diversity is attributed to the existence of distinct GST classes, which enable plants to respond to a wide range of biotic and abiotic stressors. Comparative studies across species as rice, orange, *A. thaliana*, among others, have highlighted the evolutionary expansion and diversification of GST genes, underscoring their importance in adaptive responses, and maintaining cellular homeostasis. To investigate the composition of the GSTs family in mango fruit, the mango genome was analyzed using bioinformatics approaches. A total of 67 GST genes were identified and classified into ten groups (Tau, Phi, Lambda, Theta, Zeta, DHAR, TCHQD, metaxins, PARB, and microsomal mPGEs classes), according to their evolutionary connection and the existence of conserved structural domains and motifs, exhibiting diverse physicochemical properties. Subcellular localization prediction revealed that GST were located at the cytoplasm, mitochondria, and chloroplasts, and some isoforms were associated with membranes (microsomal GSTs). Analysis using the available transcriptome data from mango, demonstrated notable expression patterns for tau class GSTs in response to heat stress, suggesting their potential role in stress resistance. These findings provide new insights into the molecular basis of GSTs function in mango, and stablish a foundation for functional studies aimed to improve stress tolerance in this commercially important fruit.

Poster 32. Biochemical and physiological responses of four wheat genotypes with differing heat-stress tolerance in the Yaqui Valley, Sonora

Edgar Robles-Rodriguez [1], Huizar L. Díaz-Ceniceros[2], Elisa M. Valenzuela-Soto[1†], Sergio G. Hernandez-Leon[1*]

[1]Centro de Investigación en Alimentación y Desarrollo, A.C. Carretera G.E. Astiazarán Rosas No. 46, Col. La Victoria, C.P. 83304, Hermosillo, Sonora. México.
[2]Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias, Carretera Norman E. Bourlaug SN, Cajeme 85000, Sonora, México
[†]In memoriam; We gratefully acknowledge the contributions of Dr. Elisa M. Valenzuela-Soto and dedicate this work to her memory.
[*]Email: sergio.hernandez@ciad.mx

Heat stress during the reproductive stage threatens wheat productivity in northwestern Mexico, especially in Sonora, the country's main wheat-producing state. To support breeding, we evaluated simple, integrated markers of tolerance. In previous field trials under optimal and heat stress (HS) conditions, diverse CIMMYT and INIFAP genotypes were classified by yield loss as tolerant or sensitive to HS. From these, four genotypes: two tolerant (G02, G13), one intermediate (G09), and one sensitive (G21), were selected for detailed physiological and biochemical analyses. Plants were grown under control and HS at the Norman Borlaug Experimental Field in the Yaqui Valley, Sonora. We measured chlorophyll a, b, total and carotenoids, chlorophyll fluorescence with a Handy PEA Plus, osmolytes proline and glycine betaine, antioxidant enzymes activity (catalase and ascorbate peroxidase, APX), and yield-related traits (spike length and weight, seed number and weight, and tillering). HS triggered genotype-specific responses. G02 and G13 maintained total chlorophyll and carotenoids content, while G09 and G21 showed marked pigment declines. Nonetheless, Fv/Fm remained stable (0.80-0.83) across genotypes, indicating that the maximum efficiency of photosynthesis was preserved. Proline content decreased under HS, mainly in G13 and G21, while glycine betaine increased in all genotypes, suggesting its central role in heat protection. Catalase activity decreased and APX increased in all genotypes, highlighting APX as the primary pathway for hydrogen peroxide detoxification under HS. Yield-related traits were reduced under HS, with the greatest losses in G09 and G21 and the smallest in G02. Overall, pigment stability, higher glycine betaine accumulation, enhanced APX activity and stable Fv/Fm emerge as practical potential markers for selecting wheat lines with improved tolerance to HS.

Poster 33. Analysis of the factor(s) responsible for the antifungal activity of *Burkholderia* spp.

Maria Goretti Rivas-Guardado[1], John Paul Délano Frier[1]*, Guadalupe Coyolxauhqui Barrera-Galicia[2], Norma Martínez-Gallardo[1]

[1]Centro de Investigación y de Estudios Avanzados del IPN, Unidad Irapuato, Irapuato, Guanajuato, México, Libramiento Norte Carretera Irapuato-León Kilómetro 9.6, Carretera Panamericana Irapuato-León, 36821. [maria.rivas@cinvestav.mx], [john.delano@cinvestav.mx]*
[2]Universidad Autónoma de Zacatecas, Facultad de Ciencias Químicas. Carretera Zacatecas-Guadalajara Km. 6, ejido "La Escondida", C.P. 98160. Ciudad Universitaria campus Siglo XXI, Zacatecas, Zacatecas [coyolba@uaz.edu.mx]

Guaranteeing worldwide food safety has lately become a pressing problem due to several factors, some of which involve an increased severity of biotic and abiotic factors that seriously affect crop production. One of the main biotic factors involves increasingly damaging plant diseases caused by fungi, which are mostly controlled through the use of fungicides. Currently, the use of antagonistic microorganisms has become a popular alternative to fungicides in integrated disease management programs considering that their use is a sustainable alternative for managing plant diseases that can lead to a reduced dependence on chemical means of control.
Interactions between beneficial microorganisms represent a promising strategy for the biological control of phytopathogens. In this study, the antifungal capacity various genera of *Burkholderia* bacteria was evaluated against four commercially important fungal phytopathogens: *Alternaria alternata*, *Rhizoctonia solani*, *Colletotrichum gloesporioides*, and *Neopestalotiopsis rosae* through direct confrontation tests in PDA medium. The *in vitro* confrontation assays allowed the observation of significant mycelial growth inhibition, suggesting that compounds having antifungal activity were being secreted by the bacteria. To explore the nature of the compounds, extracts were collected from the culture medium after the interaction period and subjected to a preliminary analysis by gas and liquid chromatographic separations. Differentiated metabolic profiles were observed in the confrontation zones, supporting the hypothesis of an induced response leading to the production of bioactive secondary metabolites. These results indicate a possible mechanism of chemical competition, reinforcing the potential use of these bacterial strains as biocontrol agents.
Future analyses using mass spectrometry will allow the identification of specific metabolites involved in antifungal activity and their *in vivo* effect to be confirmed. This study will contribute to the understanding of trophic interactions between microorganisms in competitive environments and to the discovery of bioactive compounds of interest to agriculture.

*Plant Biotic Interactions Poster

Poster 34. *In silico* characterization of the promoter of the GmTRE1 gene of soy (*Glycine max* L. Merr) and analysis of functionality in tobacco (*Nicotiana tabacum* L.)

Axel Fernando Acosta Charles [1*] and Deyanira Quistián Martínez [1]

Universidad Autónoma de Nuevo León, Facultad de Ciencias Biológicas, Laboratorio de Botánica Experimental; Av. Pedro de Alba y Manuel L. Barragán s/n, Ciudad Universitaria, San Nicolás de los Garza, Nuevo León, México. C.P. 66455
[*] Email: axelferac@gmail.com

Plants possess tolerance mechanisms that allow them to withstand different types of stress, including water stress, since genes such as Trehalase (TRE1) have been identified as factors influencing drought resistance in various plant species, including soybean (*Glycine max* L. Merr.). In this study, the promoter of the soybean GmTRE1 gene (PGmTRE1) was isolated by PCR from seeds of the cultivars (cv.) Huasteca 400 (H400), Tamesí (H500), and Huasteca 02 (H02), with the aim of sequencing them and performing an *in silico* characterization. This analysis identified 43 binding sites for 38 different transcription factors (TFs), grouped into 11 known TF families and shared among the three cultivars. Notably, 10 binding sites for 9 TFs involved in stress responses, belonging to the AP2, Dof, HD-ZIP, MYB, and WRKY families, were also found in the Huasteca 100 (H100) cultivar. Additionally, a binary vector was constructed by fusing the PGmTRE1 from cv. H100 (PH100GmTRE1) with the reporter genes EGFP:GUS, and used to transform tobacco leaves (*Nicotiana tabacum* cv. Xhanti) via agroinfiltration. Finally, histochemical GUS staining was performed, and its expression area and intensity were quantified from images under no stress control conditions and in plants subjected to 100 μM ABA treatment and 20 h desiccation. A greater GUS expression area and mean GUS intensity were observed in plants exposed to ABA stress (0.33) compared to those subjected to desiccation (0.037) or to no stress (0.076), suggesting that PH100GmTRE1 is induced by the ABA signaling pathway, and may contribute to this mechanism of resistance to water stress.

Poster 35. Transcriptomic and genomic analysis of chili (*Capsicum annuum* L) response to drought and salt stress

[1]Ruiz-Ortega, H.A*., [1]Asaff-Torres, A.J., [2]Calderón-Vázquez, C.L., [1]Campillo-Badilla, D., [1]Durazo-Arvizu, E.E., [1]Huerta-Ocampo, J.A., [3]Villalobos-Escobedo, J.M., [1]Sánchez-Estrada, A., [4]Hernández-Oñate, M.A.*

[1]Centro de Investigación en Alimentación y Desarrollo, A.C. Carretera Gustavo Astiazarán No. 46. Hermosillo, Sonora, México. Sede Hermosillo.
[2]Instituto Politécnico Nacional, Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional CIIDIR Unidad Sinaloa, Guasave, Sinaloa, México.
[3]Tecnológico de Monterrey, Instituto de Investigación sobre Obesidad. Eugenio Garza Sada 2501. Monterrey, Nuevo León, México.
[4]SECIHTI - Centro de Investigación en Alimentación y Desarrollo, A.C. Coordinación de Ciencia de los Alimentos. Carretera Gustavo Astiazarán No. 46. Hermosillo, Sonora, México. Sede Hermosillo.
*Email: ruizo.hector@gmail.com; miguel.hernandez@ciad.mx

Throughout plant domestication, selective breeding for desirable traits such as ease of handling or a major source of food has resulted in a significant reduction of genetic diversity, particularly within regions of the genome related to transcriptional regulation. This results in domesticated plants with reduced tolerance to abiotic stressors such as drought and salinity, thereby diminishing crop yield. Climate change will exacerbate these effects, posing a significant challenge to the food security of an ever-growing global population. Therefore, to address this issue, it is necessary to perform studies aimed at identifying differences at the genomic and transcriptomic levels between wild and domesticated chilli varieties during drought and salt stress scenarios. In this context, the goal of the current study is to identify and characterize differences and similarities in gene structure, expression patterns, and molecular mechanisms involved in the response of wild and domesticated chili varieties to drought and salt stress. We created a database containing the genomes of wild and cultivated varieties of chilli, as well as transcriptomes in response to drought and salt stress. We have constructed a comprehensive database containing 14 chili pepper genomes (13 domesticated and one wild) and integrated transcriptomic data from nine independent studies on chili response to drought and salinity. In our current analyses, we focus on identifying relevant genes that respond to both drought and salt stress. Subsequent analyses will involve comparative analysis of gene expression, structure, and regulatory networks between wild and cultivated chilli varieties. This study will generate relevant insights into the understanding of domestication effects on chilli tolerance to drought and salt stress, which will help develop novel strategies to enhance chilli tolerance to abiotic stress in the current climate change scenario in the future.

Poster 36. Expression Analysis of *HvNAC* Genes Associated with Senescence and Grain Filling in Mexican Barley Varieties

Hernández-Baca, M.F.[1], Rodríguez-Gandarilla, M.[1], Massange-Sánchez, J.A.[1*]

[1]Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C. (CIATEJ), Unidad de biotecnología vegetal, Zapopan, México. Camino Arenero 1227, El Bajío, 45019, Zapopan, Jalisco.
fehernandez_al@ciatej.edu.mx; jmassange@ciatej.mx

Barley (*Hordeum vulgare*) is the fourth most important cereal crop globally, with major uses in beer production and forage. In Mexico, breeding programs have released 17 malting and 5 forage varieties, where 'Esperanza' (malting) and 'Maravilla' (forage) are two of the most prominent. These varieties differ in end-use, growth conditions, and physiological maturity, suggesting distinct gene regulatory mechanisms, especially those involving transcription factors associated with senescence and grain filling. The NAC gene family has been implicated in regulating both processes in cereals. This study aims to characterize the expression patterns of selected *HvNAC* genes in 'Esperanza' and 'Maravilla' using RT-qPCR at three key developmental stages: anthesis, milk development, and dough development. Samples include flag leaves, senescent leaves, and developing grains collected at 5 days before anthesis (DBA), and 15 and 30 days after anthesis (DAP). Thirteen *HvNAC* genes, including seven linked to grain development and six to senescence, were selected based on prior transcriptomic data and functional studies. To date, all primer pairs have been validated via endpoint PCR and melting curve analysis. RNA extraction from grain tissue is being optimized, and plants have reached the target phenological stages. Preliminary data indicate that grain-related *HvNACs* peak at the milk stage, while senescence-associated *HvNACs* increase at later stages of grain development. These findings will inform the selection of candidate *HvNAC* genes for functional validation and support breeding strategies aimed at enhancing grain yield and stress adaptation in Mexican barley.

*Plant Genetics & Genomics poster

Poster 37. Exploring long-term CO_2 exposure as a driver of molecular and physiological adaptation in urban plants

Pasquale Luca Curci[1*], Francesco Punzi[1,2], Benedetta Leonetti[3], Gabriella Sonnante[1], Marta Beccaluva[4,7], Tommaso Gabbriellini[5], Luciana Renna[6], Stefano Mancuso[6,7], Giovanni Stefano[5] and Federico Vita[3]

[1]Institute of Biosciences and Bioresources, National Research Council, Bari, Italy
[2]Department of Soil, Plant and Food Sciences, University of Bari “Aldo Moro”, Bari, Italy
[3]Department of Biology, University of Bari "Aldo Moro", Bari, Italy
[4]Department of Architecture, University of Florence, Florence, Italy
[5]Department of Biology, University of Florence, Florence, Italy
[6]Department of Agricultural, Food, Environmental and Forestry Sciences and Technologies (DAGRI), University of Florence, Sesto fiorentino (FI), Italy
[7]Fondazione per il Futuro delle Città (FFC), Florence, Italy
*Email: pasqualeluca.curci@cnr.it

Urban environments are characterized by elevated CO_2 concentrations, increased temperatures, air pollution, and disrupted hydrological cycles, posing a complex array of abiotic stresses to plant life. Among these, chronic CO_2 enrichment represents a persistent and often underestimated factor influencing plant adaptation. In this study, we investigate the impact of long-term exposure to urban CO_2 levels on plant physiology and molecular responses by comparing Populus individuals from urban Florence with those from nearby non-urban green areas. Using a multidisciplinary approach, we assess RuBisCO enzymatic activity through high-throughput assays and examine alterations in chloroplast morphology, cuticle structure, and photosynthetic tissues using confocal and transmission electron microscopy. Enzymatic activity of key redox-related enzymes (CAT, APX, SOD, peroxiredoxin, and glutathione reductase) is also measured to determine redox balance shifts associated with urban CO_2 stress. In parallel, chloroplast genomes are analyzed for structural changes and mutations potentially linked to photosynthetic efficiency. To broaden the scope of the study, we perform a meta-analysis of publicly available RNA-seq datasets from *Arabidopsis thaliana*, Populus, and other species exposed to CO_2-related stress. This allows identification of differentially expressed genes, organ-specific expression patterns, and co-expression modules related to CO_2 responses. Furthermore, we integrate these findings with metabarcoding analysis of fungal and bacterial endophytes to explore the role of plant-associated microbiota in modulating stress responses. Conducted within the URbEVOLUTION project (PRIN MUR, 2022RYTHE3, Italy), this work contributes to the identification of molecular markers and physiological traits linked to CO_2 resilience, supporting the development of climate-adaptive strategies for urban green infrastructure.

Poster 38. Zinc oxide nanoparticles modulate bioaccumulation, metabolism, and bacteriome in *Capsicum annuum* L.

Luis Alberto García[1], Rosa Sánchez[2], Gilberto Velázquez[1]*

[1]University of Guadalajara, Chemistry Department. Advance Biochemistry Lab. Guadalajara, Jalisco, México
[2]National Laboratory for Agri-Food and Forestry Research and Services (LANISAF), Chapingo Autonomous University, Mexico.
*Email: gilberto.velazquez@academicos.udg.mx

Nanotechnology has emerged as a promising field in agriculture, particularly through the application of metal oxide nanoparticles to improve plant growth and resistance. This study evaluated the effect of zinc oxide nanoparticles (ZnONPs) on *Capsicum annuum L.* by examining bioaccumulation, metabolic profile, and phyllosphere bacteriome responses. The experimental design involved controlled exposure of seedlings to varying concentrations of ZnONPs, followed by assessment of zinc accumulation, antioxidant activity, volatile organic compound (VOC) profiles, and metagenomic analysis of associated microbial communities. Results demonstrated that zinc content in plant tissues increased proportionally with the concentration of ZnONPs applied to the soil. Antioxidant defense systems were significantly altered, as indicated by increased ABTS, DPPH, FRAP values, total polyphenols, and modifications in enzymatic activities of guaiacol peroxidase, ascorbate peroxidase, catalase, and superoxide dismutase. Using SPME-GC-MS, approximately 49 volatile compounds were detected across treatments, of which 23 were identified as green leaf volatiles, aldehydes, benzene derivatives, and terpenoids. Notably, ZnONP treatments were correlated with the reduction or inhibition of key green leaf volatiles such as hexanol, hexyl acetate, (Z)-3-hexenyl acetate, (E)-2-hexenol, and (E)-2-hexenyl acetate. Metagenomic analysis revealed that ZnONP exposure had limited effects at the phylum level; however, shifts in abundance were observed in Firmicutes, Actinobacteria, and Aquificae. Higher concentrations of ZnO favored colonization of genera such as *Tumebacillus*, suggesting adaptive microbial responses. Overall, the findings indicate that ZnONPs influence zinc bioaccumulation, modulate antioxidant defense, alter volatile metabolic pathways, and shape the phyllosphere microbiome of *C. annuum L.* These results contribute to understanding the environmental and agricultural implications of nanoparticle use, highlighting both potential applications and ecological considerations.

Poster 39. Beyond ABA: Exploring Physiological Drought Tolerance Mechanisms in Wild *Carica papaya*

Arroyo-Álvarez, Erick[1], Fuentes-Ortiz, G.[2], and Santamaría, J.M.[1]*

[1]Centro de Investigación Científica de Yucatán, A.C. (CICY), Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo, C.P. 97205, Mérida, Yucatán, México
[2]Independent Researcher, Mérida, Yucatán, México
*Corresponding author: jorgesm@cicy.mx

Wild accessions of *Carica papaya* exhibit distinctive physiological strategies to withstand drought, contrasting with the ABA-dependent responses commonly observed in commercial cultivars. In this study, we characterized the drought response of the wild genotype Yuc2 by evaluating endogenous ABA levels, expression of the ABA biosynthetic gene CpNCED3, and key water relations parameters, including leaf water potential, osmotic potential, and turgor pressure, throughout a progressive drought and recovery cycle.

Despite prolonged exposure to water deficit, Yuc2 maintained relatively stable ABA concentrations, with only minor increases detected in leaves and roots, and consistently low expression of CpNCED3 compared to the commercial cultivar Maradol. These patterns suggest that Yuc2 does not rely on a strong ABA-mediated response to regulate stomatal closure or osmotic adjustment. Instead, physiological data indicate that this genotype sustains higher leaf water potential and exhibits a more moderate decline in osmotic potential, allowing it to maintain turgor pressure and delay dehydration symptoms, which contributes to more efficient recovery upon rehydration.

The dissociation between CpNCED3 expression, ABA accumulation, and physiological adjustments in Yuc2 points to a partially ABA-independent drought tolerance strategy. However, it is likely that this response involves a broader combination of adaptive mechanisms that have not yet been fully explored. Further research is needed to investigate the potential role of alternative hormonal signaling pathways, the accumulation of compatible solutes, and antioxidant responses, among other possible strategies present in wild papaya relatives.

Plant genetics and genomics

Posters

Participants with even numbers will attend the session on Tuesday, October 21.

Participants with odd numbers will attend the session on Wednesday, October 22.

Poster 40. Elucidating the transfer of symbiotic plasmid from *R. etli* CFN42 into *M. truncatula* root nodules

Luis Alfredo Bañuelos-Vazquez[1*], Susana Brom[2], David Romero[2], Laura Cervantes[2] and Kimberly Gallagher[1*]

[1]University of Pennsylvania, Penn Arts & Sciences, Department of Biology, Philadelphia, Pennsylvania, USA. [2]Programa de Ingeniería Genómica. Centro de Ciencias Genómicas. UNAM. Cuernavaca, Morelos. México.
[*]Email: luisv@sas.upenn.edu.

Rhizobium etli CFN42 has great agricultural importance due to its symbiotic association with *Phaseolus vulgaris* (common bean), through the formation of determinate nodules. The genome of *R. etli* contains one chromosome and six large plasmids (pRet42a-pRet42f). Most of the genetic information involved in the establishment of the symbiosis is located on plasmid pRet42d, named as the symbiotic plasmid (pSym). This plasmid is able to perform conjugation, using pSym-encoded transfer genes controlled by the RctA/RctB system. It has been shown that bacterial conjugation occurs inside root nodules, a niche previously unrecognized for genetic exchange. Furthermore, how the pSym can be transferred to endophytic bacteria that are within the root nodules. These findings strengthen the proposal that nodules constitute a perfect environment for the exchange of genetic information among bacteria, in addition to their function as structures where nitrogen fixation and assimilation take place. This project aims to examine the effects of acquiring the pSym of CFN42 on particular strains of *Ensifer meliloti* that form indeterminate nodules in *M. truncatula* and to accurately assess the symbiotic capacity of both plant species. Also, this research aims to enhance our understanding of plant-microbe interactions and to identify potential strategies for improving legume productivity in agricultural systems.

Poster 41. Improving maize regeneration through the modulation of calcium signaling

Nicole Gómez Capetillo[1], Marcela Hernández Coronado[2], Carlos Humberto Ortiz Ramírez[1*]

[1]UGA Laboratorio Nacional de Genómica para la Biodiversidad, CINVESTAV Irapuato, Guanajuato 36821, México.
[2]CINVESTAV Unidad Irapuato, Guanajuato 36821, México.
*Email: carlos.ortiz@cinvestav.mx.

Improving the efficiency of genetic transformation in *Zea mays* remains a major challenge. The high recalcitrance exhibited by some genotypes has historically hindered genetic engineering and functional characterization efforts in this staple crop. Hence, we aim to enhance callogenesis and plant regeneration by modulating calcium signaling through the application of the ion gadolinium (Gd^{3+}), a known inhibitor of calcium channels. Mature and immature embryos from two contrasting maize genotypes (A188, low recalcitrance; B73, high recalcitrance) were exposed to varying concentrations and durations of Gd^{3+} and the effects of these treatments were evaluated. We identified a protocol that significantly boosted embryogenic callus induction in mature embryos for both genotypes. This was particularly evident in the highly recalcitrant B73 line, which saw a remarkable 80% increase in callus induction (n=40, pval=0.00001). In addition the application of Gd^{3+} improved plant regeneration in both mature and immature embryos. In the A188 genotype, a regeneration efficiency of 54% (n=33) was achieved in immature embryos. Compared to the control, the Gd^{3+}-based treatment led to a 10% increase in regeneration efficiency, underscoring the potential of calcium channel modulation to enhance transformation outcomes across different developmental stages. Finally, we employed CRISPR/Cas9 gene editing mediated by *Agrobacterium tumefaciens* to functionally characterize candidate genes putatively involved in Kranz anatomy development. Our findings contribute to the development of a protocol that improves transformation even in recalcitrant maize genotypes facilitating functional characterization and genetic engineering approaches.

Poster 42. The function of the *VIM5* gene in seed development of Arabidopsis

José David Velázquez-Fierro, Karina Orozco-Natividad, C. Stewart Gillmor*

Unidad de Genómica Avanzada, Centro de Investigación y Estudios Avanzados del Instituto Politécnico Nacional (Cinvestav), Irapuato, Guanajuato, México.
*Email: stewart.gillmor@cinvestav.mx

Seed development is a hallmark process in the life cycle of higher plants, with a plethora of genetic pathways driving the proper development of the embryo, endosperm, and seed coat. Although the endosperm is also a product of double fertilization, its transcriptional landscape differs from that of the embryo. This is the result of different epigenetic modifications established mostly during gametogenesis. The different epigenetic profiles between the maternal diploid central cell and the paternal haploid sperm cell render the endosperm a unique genomic system, a triploid genome with a 2:1 maternal to paternal ratio. Although many genes show triploid expression, others show imprinted expression that is biased toward the maternal or paternal allele. Imprinted expression has been fine-tuned by evolution and deviation from the 2:1 maternal:paternal ratio can result in seed malformation or even seed abortion. The *VARIANT IN METHYLATION 5* (*VIM5)* gene is a paternally expressed gene that encodes a ubiquitin E3 ligase only present during development of the endosperm. VIM5 regulates endosperm gene methylation via the ubiquitination and degradation of the CG methyltransferase MET1 and the CHH and CHG methyltransferase CMT3. In this work we demonstrate how loss of *VIM5* affects chalazal endosperm size and hinders embryo development in Arabidopsis seeds, casting a light on the importance of paternally expressed genes and paternal contribution during early seed development.

Poster 43. **Genetic bases of photosynthetic plasticity in maize varieties adapted to diverse climates**

Elizabeth Hernández Jaimes[1], Carlos Humberto Ortiz Ramírez[1], Marcela Hernández Coronado[2*]

[1]Developmental Genetics and Genomics Laboratory. Advanced Genomics Unit. CINVESTAV-Irapuato. Libramiento Norte León Km 9.6, 36821 Irapuato, Gto., Mexico.
[2]Laboratory of Regulation of Totipotency and Regeneration. CINVESTAV-Irapuato. Libramiento Norte León Km 9.6, 36821 Irapuato, Gto., Mexico.
*Email: marcela.hernandez@cinvestav.mx

Phenotypic plasticity is the ability of a single genotype to produce different phenotypes in response to changes in the environment. This is especially relevant for plants that, being sessile organisms, rely on adapting to their surroundings. However, there is limited information on the genetic basis of plasticity. Native maize has been domesticated and adapted to contrasting environments, and it also has a broad genotypic and phenotypic diversity. Therefore, it is an excellent model for studying the mechanisms underlying plasticity. Previous studies in our lab have determined that there are changes in leaf vein density, depending on the environment and the native maize variety. This is relevant due to the impact of vein density on photosynthetic efficiency, which directly affects crop yield. Therefore, we used vein patterning as a model to identify the genetic bases that modulate phenotypic plasticity in eight native maize varieties from Mexico. We collected leaf primordia tissue at the exact stage in which veins start developing under different environmental conditions and conducted an RNA-seq study. The obtained data was analyzed to identify differentially expressed genes and co-expression profiles associated with the most plastic varieties. Our results revealed that plasticity in vein density could be a coordinated response that starts with the induction of chaperone proteins like HSP90, consistently expressed in the varieties with the highest plasticity, and is followed by the downstream expression of a set of transcription factors known for controlling leaf development. In conclusion, this study provides a deeper understanding of the genes and processes that regulate phenotypic plasticity in the vein density of native maize. The acquired knowledge could have important implications for crop improvement to increase their adaptability to changing environmental conditions.

Poster 44. Whole genome sequencing reveals genomic variations and selection signals in ancient Mexican grapevines

Sara Covarrubias[1], Cristina Dominguez-Castro[2], Alejandra Rougon-Cardoso[1], Rufina Hernández-Martínez[3], Luis Delaye[4] & Antonio Hernández-López[1*]

[1]Laboratorio de Investigación Interdisciplinaria, Escuela Nacional de Estudios Superiores Unidad León, Universidad Nacional Autónoma de México, Blvd. UNAM 2011, Predio El Saucillo y Comunidad de los Tepetates, El Potrero, 37684 León, Guanajuato, México. [2]Facultad de Enología y Gastronomía, Universidad Autónoma de Baja California, Carretera Transpeninsular Ensenada-Tijuana 3917, Col. Playitas, Ensenada Baja California 22860, México. [3]Departamento de Microbiología, Centro de Investigación Científica y de Educación Superior de Ensenada, CICESE, Carr. Tijuana-Ensenada 3918, Zona Playitas, Ensenada Baja California 22860, México. [4]Departamento de Ingeniería Genética, Centro de Investigación y de Estudios Avanzados del IPN, Unidad Irapuato (CINVESTAV), Libramiento Norte Carretera Irapuato León, 36821 Irapuato, México.
*Email: ahernandez@enes.unam.mx

The Mission grape (*Vitis vinifera ssp. vinifera*), known in Mexico as “Misión,” is one of the earliest grape varieties introduced to the Americas, with records of its use in winemaking dating back to the colonial period. While these wines were initially valued, the grape’s reputation declined over time, and it was long dismissed as producing characterless table wines. Despite this, the Misión grape persisted in scattered vineyards thanks to its vigorous growth, heavy yields, and remarkable drought resistance. Today, as climate change intensifies water scarcity, there is renewed interest in this historic variety, especially in Baja California, where extreme heat and limited water challenge conventional viticulture. To explore the genetic basis of these advantageous traits, we performed whole-genome analyses of Misión grape individuals from Baja California and the Bajío region. By identifying genomic regions under strong selection, we aim to uncover alleles and gene networks linked to drought adaptation, yield stability, and other agronomically important features. Preliminary results reveal distinct signals of selection in regions potentially associated with stress response and water-use efficiency, offering valuable targets for future breeding and conservation efforts. This integrative approach, combining historical narratives with modern genomics, underscores the Misión grape’s cultural importance and its potential as a model for sustainable viticulture in arid environments. Ultimately, our study not only revives appreciation for an often-overlooked variety but also provides critical genomic insights that can inform grapevine improvement strategies worldwide.

Poster 45. Modeling the molecular competition between *FT* and *TFL1* during flower transition in *Arabidopsis thaliana*

Mariana Yuste, Eugenio Azpeitia*

Centro de Ciencias Matemáticas, Universidad Nacional Autónoma de México, Morelia, Mexico.
*Email: eazpeitia@matmor.unam.mx

The correct production of flowers depends on the transition from a vegetative meristem to an inflorescence meristem, and ultimately to a flower meristem. This transition is regulated by multiple environmental and endogenous signals that converge on flowering time integrators, among which *FLOWERING LOCUS T* (*FT*) plays a central role, as it responds to all major flowering induction pathways. Interestingly, *FT* and its homologue *TERMINAL FLOWER 1* (*TFL1*) both need to bind to *FLOWERING LOCUS D* (*FD*) in order to achieve their regulatory activity. However, the FT-FD and TFL1-FD complexes have opposite functions: while FT-FD promotes flowering, TFL1-FD maintains inflorescence identity and represses flower initiation. As a result, FT and TFL1 compete for binding to FD. Yet, how this molecular competition is orchestrated during flower transition, especially how it influences the expression of flowering genes in the meristem, remains poorly understood. This work uses a mathematical model of ordinary differential equations to study how FT and TFL1 compete for binding to FD and, through the formation of their complexes, control the expression of flowering and inflorescence identity genes in the model plant *Arabidopsis thaliana*. To do this, a Gene Regulatory Network (GRN) was constructed based on prior studies, integrating the key molecular actors involved in flower transition—along with FT, FD, and the FT–FD and TFL1–FD complexes. The network's dynamics were analyzed by identifying equilibrium points that reproduce the genetic expressions observed during flower transition in both wild-type and mutant *A. thaliana*. Based on the parameters found, our results suggest that the dynamics of the FT-FD and TFL1-FD complex formation may depend on distinct affinity and timing requirements, influenced by FD abundance and its phosphorylation.

Poster 46. Connecting transcription and DNA supercoiling in the chloroplast genome

V. Miguel Palomar

Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México. Mexico City, Mexico.
*Email: vmiguelpalomar@quimica.unam.mx

Cells of photosynthetic organisms contain genetic information not only in the nucleus but also in mitochondria and chloroplasts. Organellar genomes are remnants of endosymbiosis and their unique properties have been shaped by over a billion years of co-evolution with the nucleus. A particularly mysterious property of the chloroplast genome is its packaging into a chromatin-like structure known as the nucleoid. Recently, we described how transcription shapes the chloroplast nucleoid structure by directing its interaction with the thylakoid membrane in *Arabidopsis thaliana*. If transcription is a determinant force shaping nucleoid topology, its effects should also contribute to the overall organization of this structure. In this project, we used next generation sequencing-based methods to study DNA supercoiling in the chloroplast genome. Using heterologous expression of GapR, a protein found in the bacteria *Caulobacter crescentus*, in the chloroplast of *Arabidopsis thaliana*, we found a complex pattern of positive supercoiling across different regions in the genome. Positive supercoiling is predominant on the end of actively transcribed genes and operons but is especially enriched on convergent transcriptional units. Chemical reduction of Gyrase A activity leads to a drastic change in supercoiling. Finally, by using conditions that modify chloroplast transcription we found a connection between this process and DNA supercoiling, suggesting both are required for proper expression of genes in the chloroplast and possibly shaping the nucleoid structure. This knowledge will help us understand fundamental questions about the diversity of chromatin compaction mechanisms in nature.

Poster 47. Functional analysis of putative target genes of the transcription factor GROUNDED/RKD4 in embryo development in *Arabidopsis thaliana*

Eduardo Moncada[1], Jaime Alaniz-Fabián[1], Magdy Alabady[2], Wolfgang Lukowitz[2] and Stewart Gillmor[1]*

[1]Unidad de Genómica Avanzada, Cinvestav, Irapuato, México.
[2]Department of Plant Biology, University of Georgia, Athens, Georgia, USA.
*Email: stewart.gillmor@cinvestav.mx.

The transcription factor GROUNDED/RKD4 (GRD), a member of the RWP-RK family, has been identified to have a role in promoting suspensor identity during early embryogenesis in *Arabidopsis thaliana*, yet its downstream genetic targets remain uncharacterized. Transcriptomic analysis of *grd-3* mutants at 30 hours after pollination revealed 11 genes whose transcripts decrease significantly. We are currently examining the function of these 11 genes, using indexed T-DNA insertions and ethyl methane sulfonate (ems) alleles. Our hypothesis is that mutants in at least some of these genes will recapitulate aspects of the *grd* phenotype in early embryogenesis. For four out of eleven genes, close gene homologs with expression in the embryo were found, so double and triple mutants will be analyzed to test for potential genetic redundancy. We are also designing YFP promoter-reporter constructions to determine the expression pattern of these genes and to assay for differences of expression wild type vs *grd* embryos. Our findings will help us to understand how putative GRD targets contribute to embryo development and how *GRD* functions within a broader regulatory network critical for early embryogenesis in *A. thaliana*.

Poster 48. **Unraveling meristem regulation in maize and its wild relatives**

Diego Armando Flores Bautista, Marcela Hernández Coronado*

CINVESTAV Unidad Irapuato, Carretera Irapuato-León Km. 9.6, 36821, Guanajuato. México.
*Email: marcela.hernandezco@cinvestav.mx

Meristems are pools of undifferentiated, totipotent cells with the potential for continuous cell division. In plants, gene expression programs at these stem cell niches are integrated to regulate their activity and ensure balanced growth. While significant strides have been made in understanding meristem genetics, particularly in annual model species like *Arabidopsis* (e.g., the identification of WUSCHEL and CLAVATA pathway in the shoot apical meristem, our knowledge is still limited. Intriguingly, research in maize has revealed alternative pathways, such as a KN1-mediated pathway involved in maintaining cell undifferentiation, and distinct expression patterns for WUSCHEL homologs. This suggests a diversity of mechanisms controlling meristematic activity even among annual plants, highlighting the need to broaden our investigation to other species, including perennials. Perennial plants possess the remarkable ability to retain dormant meristems that can transition to a proliferative state in response to stress removal or physiological cues. This process likely involves unique gene expression and regulation pathways that are currently poorly understood. In this project we aim to extend our knowledge in gene regulation of totipotent cell populations. For this, vegetative meristems from maize (*Zea mays*) and teosinte species (*Zea mexicana*, *Zea luxurians*, and the perennial *Zea diploperennis*), are being collected and profiled through RNA sequencing. By analyzing transcriptomic data from these four closely related species, we aim to identify divergent regulatory mechanisms, thereby contributing to a deeper understanding of meristem regulation and its evolution.

Poster 49. **Transcriptome analysis of somatic embryogenesis induction varying the inorganic nitrogen sources ratio in *Coffea canephora***

Brigitte V. Vargas-Morales, Rosa María Galaz-Avalos, Hugo A. Méndez-Hernández, Ana O. Quintana-Escobar and Víctor M. Loyola- Vargas*

Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, Calle 43, 130, Colonia Chuburná de Hidalgo, Mérida, Yucatán, C.P. 97200.
*Email: vmloyola@cicy.mx.

Coffea canephora is an economically important species that plays a valuable role in the coffee industry. Biological processes such as somatic embryogenesis (SE) can provide a means for mass plant propagation, genetic improvement, and the study of embryo formation and development. Somatic cell reprogramming to acquire embryogenic capacity involves changes in gene expression and complex signaling networks, as well as the participation of specific regulatory pathways. A change in the nitrate (NO_3^-) and ammonium (NH_4^+) ratios of the culture medium affects the embryogenic response of the SE system; nonetheless, the underlying molecular mechanisms are unknown. To study the participation of N in SE, we analyzed RNA-sequencing (RNA-seq) data of four different time points during the SE process corresponding to pretreatment (0 d) and induction (28, 42, and 56 d) in four induction treatments that vary in the inorganic N ratio as follows: A (15 mM NO_3^-:0 mM NH_4^+), C (10 mM NO_3^-:5 mM NH_4^+), D (5 mM NO_3^-:10 mM NH_4^+), and E (7.5 mM NO_3^-:7.5 mM NH_4^+). We analyzed differential gene expression in 21 pairwise comparisons (A28_C00, A42_C00, A56_C00, C28_C00, C42_C00, C56_C00, D28_C00, D42_C00, D56_C00, E28_C00, E42_C00, E56_C00, A28_C28, A42_C42, A56_C56, D28_C28, D42_C42, D56_C56, E28_C28, E42_C42, E56_C56). Furthermore, we identify several genes related to cytokinin (CK) metabolism. This study provides a step forward in understanding the molecular mechanisms by which the N source participates in the SE of *C. canephora*.

Poster 50. Chromosomal inversions evolution in sunflowers

Eric González[1], Marco Todesco[1], Mojtaba Jahani[1], Stephane Munos[2], Nicolas Langlade Loren H. Rieseberg[1*]

[1]Department of Botany, University of British Columbia, Vancouver, Canada, 6270 University Blvd.
[2]Institut national de recherche pour l'agriculture, l'alimentation et l'environnement, Toulouse, France.
*Email: loren.rieseberg@botany.ubc.ca

There is growing evidence for the importance of chromosomal inversions in local adaptation processes, especially when these occur in the presence of gene flow between populations adapted to different environments. Chromosomal inversions reduce recombination between genotypes carrying contrasting alleles. These low-recombination regions are often referred to as supergenes, as they can preserve advantageous allele combinations by limiting genetic exchange with non-adapted haplotypes. But what kinds of genes do they contain? How many of these are shared or unique between alternative haplotypes? How do these regions evolve over time? How divergent are they? As part of my postdoctoral research, I explore these questions using wild sunflowers as a model. Their genomes are mosaics of regions with reduced recombination between alternate haplotypes that are involved in adaptation and speciation. In this talk, I will introduce the genomic resources we have developed to study these regions, including haplotype-resolved reference genomes representing both maternal and paternal chromosomes. I will also present the chromosomal inversions we have identified, along with estimates of their age, divergence patterns, and gene content.

Poster 51. **Identification of Nuclear-encoded RNA polymerase (NEP) binding sites on the *Arabidopsis thaliana* chloroplast genome using ptChIP-seq**

López-Morales JJ[1], Palomar VM[2*]

[1]Undergraduate program in Chemical Pharmacobiology, Facultad de Química, Universidad Nacional Autónoma de México. Mexico City. Mexico.
[2]Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México. Mexico City, Mexico.
*Email: vmiguelpalomar@quimica.unam.mx

Mitochondria and chloroplasts evolved through the process of endosymbiosis and retain their own genome, coexisting with the nuclear counterpart. In chloroplasts, the DNA is organized in a structure similar to nuclear chromatin known as the nucleoid, which structure and regulation are still not fully understood. In *Arabidopsis thaliana* and other vascular plants, transcription of the chloroplast genome is carried out by two RNA polymerases: the Plastid-Encoded Polymerase (PEP) and the Nuclear-Encoded Polymerase (NEP). Although the function, structure, and binding sites of PEP have been widely studied, there is little knowledge about the binding sites of NEP as well as its contribution to the transcription of the chloroplast genome. In this work, using plastid chromatin immunoprecipitation coupled with high-throughput sequencing (ptChIP-seq) targeting NEP, the binding pattern of this protein in the chloroplast genome is analyzed. This study allows us to describe the role of the nuclear-encoded transcriptional machinery in the organization of the chloroplast genome, contributing to a better understanding of genetic regulation in these organelles.

Poster 52. Going back in history: Looking for the molecular components of *Mimosa pudica* sp. circadian clock

José Ambrosio Chimal-Itzincab[1], Itzel López-Rosas[2] and Alfredo Sánchez-Villarreal[1]*

[1]Colegio de Postgraduados campus Campeche, Champotón, Campeche, México. [2]SECIHTI- Instituto Tecnológico de Chiná, TecNM, Chiná, Campeche, México. *Email: asanchezv@colpos.mx.

Circadian clocks are a pervasive mechanism consisting of transcriptional and translational feedback loops that provide fitness by timing physiological processes to the external environment. In *Arabidopsis thaliana*, the circadian clock was described as comprised of *CIRCADIAN CLOCK ASSOCIATED 1* (*CCA1*), *LATE ELONGATED HYPOCOTYL* (*LHY*) and *TIMING OF CAB EXPRESSION* (*TOC1*). Through microarray analysis, it was demonstrated that the circadian clock controls a plethora of genes, thereby exerting genome-wide transcriptional control. Further description of genes and the oscillator machinery has shown its ubiquitous roles in plant development, abiotic and biotic stress, and metabolic and signalling pathways. Currently, the circadian clock has been shown to be intertwined not only with flowering time, but also with ecological interactions. In plants of agronomic interest, the circadian clock has been a key feature in domestication and adaptation to local environments. Ironically, *Mimosa pudica*, the plant in which astronomer Jean Jaques d´Ortous de Mairan made the first documentation of leaf movement and thus the first evidence of an internal mechanism keeping track of time, has not been further studied in this regard. *M. pudica* nyctinastic leaf movement (closing of leaves toward touch) has been extensively studied, but not the ~24 hour period of its leaf movement. Therefore, here we present the first approach to uncover the circadian clock related genes in the available *Mimosa pudica* sp. genome. Through BLASTn and BLASTx searches using both *A. thaliana* and legumes circadian clock genes, we have found the putative homologous of *CCA1*, *LHY, TOC1, GIGANTEA* (*GI*), *EARLY FLOWERING 3 (ELF3)*, *ELF4, TIME FOR COFFEE* (*TIC*), *ZEITLUPE* (*ZTL*), and *LUX ARRYTHMO* (*LUX*). Ongoing experimentation is focused in discerning between the *M. pudica* scaffolds as gene duplication is a common feature in Fabaceae. Our research may provide molecular tools for a neglected group of plants as Caesalpinioideae.

Poster 53. Differential expression of transcripts in the mistletoe *Psittacanthus calyculatus* throughout the infective process in mesquite trees

Cota-Pineda, M.R.[1], Mendoza-Hernández, C.S.[2], Lozoya-Pérez, N.[2], Quintana-Rodríguez, E.[2], Huerta-Ocampo, J.A.[1],Contreras-Vergara, C.A.[1], Villalobos-Escobedo, J.M.[3], Orona-Tamayo D.[2*], Hernández-Oñate, M.A.[4*]

[1]Centro de Investigación en Alimentación y Desarrollo A.C. (CIAD). Hermosillo, Sonora, México.
[2]CIATEC, A.C., Medio Ambiente y Biotecnología, León, Guanajuato, México.
[3]Tecnológico de Monterrey, Institute de Investigación sobre Obesidad. Monterrey, Nuevo León, Mexico.
[4]SECIHTI-Centro de Investigación en Alimentación y Desarrollo A.C. (CIAD). Hermosillo, Sonora, México.
*Email: dorona@ciatec.mx; miguel.hernandez@ciad.mx.

Mistletoes are hemiparasitic plants that connect to the host xylem through the parasitic organ “haustorium.” This connection is essential for the mistletoe to acquire the water and nutrients necessary for its development. In Mexico, mistletoes are highly prevalent; it has been reported that 65 % of the urban trees in León, Guanajuato, are infected. A severe infection in a host can lead to the tree’s death due to hydric stress caused by the parasitism. Mistletoe infections have been reduced through pruning, fire, and herbicides. However, these methods are inefficient and non-specific, causing damage to the host tree. Our study aims to identify key genes and pathways involved in the infection process using a transcriptomic analysis with RNA-seq across four infective stages of the mistletoe while it parasitizes a mesquite tree. Total RNA samples from these stages have been sequenced in paired-end 150 bp reads. The reads were filtered for quality and assembled *de novo* using Trinity. The assembled transcriptome was filtered based on contig quality, redundancy, and contaminant removal. The transcriptome’s quality was assessed using BUSCO, Transrate, and Trinity statistics. Read mapping and quantification were performed using RSEM, and the differential analysis was conducted using the edgeR package in RStudio. Differentially expressed transcripts were identified in all the infective stages, revealing potential key genes and pathways involved in the infection process. Transcripts up-regulated and down-regulated throughout the entire infection process, were identified, indicating a potentially significant role for these genes in mistletoe infection.

Poster 54. **Identification of *PLETHORA* genes in *Mammillaria san-angelensis* (Cactaceae): advances towards elucidating their role in root development**

Luis Fernando Cisneros-Lemus[1], Eduardo Torres-Flores[2], Viola Willemsen[3], GustavoRodríguez-Alonso[4], Svetlana Shishkova[5], Javier Andrés Juárez-Díaz[6], Ulises Rosas[7]*

[1]Posgrado en Ciencias Biológicas, Universidad Nacional Autónoma de México, CDMX, México, Ciudad Universitaria, 04510 Coyoacán.
[2]Posgrado en Ciencias Biológicas, Universidad Nacional Autónoma de México, CDMX, México, Ciudad Universitaria, 04510 Coyoacán.
[3]Laboratory of Cell Biology, Wageningen University & Research, Droevendaalsesteeg 1, Wageningen 6708PB, Netherlands.
[4]Centro de investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas. Universidad Autónoma del Estado de Morelos. Cuernavaca, Morelos, México.
[5]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Mor., 62210, México.
[6]Facultad de Ciencias, Universidad Nacional Autónoma de México, CDMX, México, Ciudad Universitaria, 04510 Coyoacán.
[7]Jardín Botánico del Instituto de Biología, Universidad Nacional Autónoma de México, CDMX, México, Tercer circuito exterior S/N, Ciudad Universitaria, 04510 Coyoacán.
*Email: urosas@ib.unam.mx

Mammillaria san-angelensis, a cactus of biocultural significance, short life cycle, ease of reproduction, and part of a highly diverse genus, is being used in our laboratory as a model system to study root development. *Mammillaria* belongs to the Cactoideae subfamily, whose members exhibit determinate growth of primary and lateral roots as a consequence of the root apical meristem (RAM) exhaustion and subsequent differentiation of all root-apex cells. The first step in understanding the mechanisms controlling determinate root growth in *M. san-angelensis* was identifying putative orthologs of the *PLT* genes using bioinformatic and molecular methods. Six PLT transcription factors have been described in *Arabidopsis thaliana*; these are crucial for root development processes including RAM maintenance. We sequenced the whole genome of *M. san-angelensis* using the PacBio platform and established a bioinformatic pipeline to isolate candidate sequences. Using those sequences, as well as the putative *PLT*s orthologs of the *A. thaliana*, we used a molecular phylogeny approach and identified the *M. san-angelensis* homologues in the *PLT1/2, PLT3/7, PLT4* and *PLT5* subclades*.* Further homology analysis revealed the relationship of the genes within the *PLT3/7* clade. Using primers designed for these sequences we validated by PCR four *PLT* genes from *M. san-angelensis* gDNA, and amplified 12 PCR fragments from cDNA of various *Mammillaria* species. Our results suggest that we have experimentally isolated the putative *AtPLT7* ortholog of *M. san-angelensis*. Finally, to perform complementation tests in the *Arabidopsis thaliana plt3 plt7* double mutant, we are currently obtaining *35S::MsaPLT3.2* and *35S::MsaPLT3.2::GFP* fusions, thus establishing tools for the functional study of the role of *PLT* genes in *M. san-angelensis* root development.

Poster 55. Seed germination of wild and cultivable amaranth seeds

Alberto Antonio Lomelí Bernal[1], Ana Paulina Barba de la Rosa[1]* & Alfredo Herrera Estrella[2]

[1]Instituto Potosino de Investigación Científica y Tecnológica (IPICYT), Camino a la Presa de San José 2055, Lomas 4ta Secc, San Luis Potosí, S.L.P., México CP 78216. Tel. (444) 8342000 Ext. 2082/2084.
[2]Unidad de Genómica Avanzada-Langebio del Centro de Investigación y de Estudios Avanzados, Libramiento Norte León Km 9.6, Irapuato, Gto., 36824, México.
*Email: apbarba@ipicyt.edu.mx

Seed germination is a critical process for the survival of most plant species, beginning with water uptake by the seed and culminating in radicle emergence. Despite its importance in the plant life cycle, the molecular mechanisms underlying seed germination remain poorly understood. Amaranth (*Amaranthus* spp.) can germinate in arid, semi-arid, saline, and low- water soils; however, the molecular mechanisms that enable it to grow under these conditions have not been extensively studied. This study aimed to analyze metabolites and miRNA changes during seed germination in arable species *Amaranthus hypochondriacus* cultivars and *A. cruentus*, as well as their potential wild ancestor, A. hybridus. Arable species exhibited higher water uptake and faster germination than the wild species, with all species displaying rapid water absorption during the first hour of imbibition (Phase I), followed by a slower uptake over the next hours (Phase II). Metabolite changes revealed that phosphatidic acid and abscisic acid play key regulatory roles in the germination of the wild species. Additionally, miRNAs analysis of amaranth seeds is underway to elucidate further the molecular pathways associated with seed germination.

Poster 56. Comparative analysis of NBS gene family in wild and commercial papaya genomes to understand implications in plant defense

Mayra Alejandra Sallas Barreto [1], Diego Garza González [1], Kevin Childs [2], Jorge H. Ramírez Prado[1], Luisa A. López Ochoa[1], José Ramon Pacheco Arjona[3], Mariana Chávez Pesqueira[1], Elsa Góngora Castillo[4]*.

[1]Unidad de Biotecnología, Centro de Investigación Científica de Yucatán, Calle 43 No. 130 x 32 y 34. Col. Chuburná de Hidalgo, 97205 Mérida, Yucatán, México.
[2]Department of Plant Biology. Michigan State University. 612 Wilson Road. 48824 East Lansing, Michigan, USA.
[3]Facultad de Medicina Veterinaria y Zootecnia. Universidad Autónoma de Yucatán. Carretera Mérida-Xmatkuil Km. 15.5, Mérida, Yucatán, México.
[4]Departamento de Recursos del Mar. Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional. Km 6. Antigua carretera a Progreso. Cordemex, 97310. Mérida, Yucatán, México.
*Email: elsa.gongora@cinvestav.mx

Mexico is the third largest producer and main exporter of papaya (*Carica papaya* L.) worldwide, so understanding its genetic diversity is key to its improvement and sustainability. The theory of domestication indicates that wild varieties are a reservoir of genetic diversity, and that it may have been affected by the domestication process. The reduction or loss of genetic variability of the species could affect the plant's ability to respond to biotic and abiotic stress. In order to understand the genetic changes in the defense response, the genomes of wild papaya and the commercial variety Maradol were analyzed to identify the differences between the genes that encode the proteins of the "nucleotide-binding site-leucine-rich repeat" (NBS-LRR) family, which are fundamental in the immunological defense of plants. Our results showed variations in the number of genes encoding these proteins, in the presence of conserved domains, and in the regulatory elements present in the promoter regions of the genes. Three-dimensional analyzes of the proteins showed a high degree of conservation of their structure, so the regulation of these genes could be key to improving the crop in the face of the environmental pressures of climate change.

Poster 57. **Analysis of the effects of auxins and cytokinins on the formation of protein-protein interactions during gynoecium development**

Karen Monserrat García Valle[1] & Stefan de Folter[2]*

[1]Unidad de Genómica Avanzada (UGA-Langebio), Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (Cinvestav), Irapuato CP 36824, Guanajuato, México. karen.garcia@cinvestav.mx
[2]Unidad de Genómica Avanzada (UGA-Langebio), Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (Cinvestav), Irapuato CP 36824, Guanajuato, México.
*Email: stefan.defolter@cinvestav.mx

The gynoecium is a complex structure whose development involves various cellular and physiological processes, which are in turn mediated by both internal and external factors such as transcriptional regulation, hormone concentration, and protein–protein interactions, among others. Transcription factors are proteins responsible for gene regulation through direct binding to DNA, and they mediate diverse developmental processes within the gynoecium, either individually or by forming protein complexes. These physical interactions between proteins can themselves be regulated by the presence of hormones such as auxins and cytokinins, which are key regulators of various plant developmental processes and stimulate cell division and differentiation. Furthermore, some auxin- and cytokinin-responsive factors interact with transcription factors from different families that are also involved in gynoecium development. In this study, we conducted bimolecular fluorescence complementation (BiFC) assays in *Arabidopsis thaliana* leaf protoplasts to test the different protein–protein interactions between the transcription factors INDEHISCENT (IND) and CRABS CLAW (CRC), which belong to the bHLH and YABBY families, respectively. We also examined interactions with the auxin-responsive factors ARF17 and ARF19, and the cytokinin-responsive factors ARR12 and ARR14, all of which are part of a previously proposed interaction network based on yeast two-hybrid assays. Additionally, we confirmed that some of these interactions are affected by the exogenous application of auxins and cytokinins. The most notable effects observed were that indole-3-acetic acid (IAA) enhances the interaction between ARF17 and IND, as well as between ARR14 and IND, whereas benzylaminopurine (BAP) suppresses most of these interactions. These findings demonstrate how auxins and cytokinins modulate the dynamics of protein–protein interactions within the gynoecium.

Poster 58. **Characterization of alkaloid transporters in *Argemone mexicana* L.**

Ramiro Pérez González, Lloyd Loza Muller, Armando Muñoz Sánchez, Felipe Vázquez Flota*

Centro de Investigación Científica de Yucatán, Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México
*Email: felipe@cicy.mx

Argemone mexicana is a medicinal plant that produces benzylisoquinoline alkaloids. There is evidence suggesting the operation of transport mechanisms mediated by proteins. This work is aimed to the isolation and partial characterization of selected candidates. A total of 1012 transporter candidates frome the ABC, MATE, and PUP families, were identified by bioinformatics. Four of them were selected, all from the MATE family named AmMATE2 to AmMATE5. Specific primers designed for the isolation of them from different tissues retrieved 505 and 816 bp fragments for AmMATE2 and AmMATE3 from seedlings and cell suspensions, respectively, as well as the complete ORF for AmMATE4 and AmMATE5, both from roots. Additionally, AmMATE5 was also isolated from endosperm. Identity of the isolated candidates was confirmed by sequencing. Tissue distribution of the putative transporters was analyzed by PCR and showed that both AmMATE2 and AmMATE3 were abundant in seedlings and cell suspensions, with AmMATE3 also being abundant in leaves, roots and mature capsules. Contrastingly, expression of AmMATE4 was low in all tissues evaluated, and AmMATE5 was observed mainly in radicles, cell suspensions, and mature capsules. AmMATE4 and AmMATE5 were examined against berberine and sanguinarine using the CB-Dock2 package and found that they are very likely to interact. Amino acid domains R/KXS with a high probability to be phosphorylated is embedded in the conserved RVSNELGA site for all plant MATE proteins. AmMATE2 could be operating at certain developmental stages and not continuously; AmMATE3 results suggest that it could be a transporter widely used in this plant at all stages of development; AmMATE4 has practically no expression, which may suggest that this transporter is inducible under certain conditions, whereas AmMATE5 appears to be mainly involved in substrate mobilization in roots, both in seedlings and mature plants, in mature capsule and in cell suspensions. Supported by SECIHTI CBF-2023-2024-1879

Poster 59. Large-scale and simultaneous analysis of transcription factors binding to specific promoters in *Arabidopsis thaliana*

López Araiza Esparza K.P.[1] Marsch Martinez N.,*[1,2] de Folter S.,*[1,2]

[1]Centro de Investigación y de Estudios Avanzados del IPN, Unidad Irapuato (Cinvestav), México.
[2]Unidad de Genómica Avanzada – LANGEBIO Cinvestav Irapuato, México.
*Email: nayelli.marsch@cinvestav.mx; stefan.defolter@cinvestav.mx

Interactions between transcription factors and specific promoter regions precisely regulate gene expression in plants. However, in *Arabidopsis thaliana*, due to the abundance and diversity of these regulators, identifying the transcription factors that regulate key promoters involved in processes such as floral development remains a challenge. This lack of knowledge limits our understanding of the transcriptional networks fundamental to gynoecium biology. One of the most important cases is the *NO TRANSMITTING TRACT* (*NTT*) factor, which is responsible for the correct differentiation and development of the transmitting tract, which is necessary for correct ovule fertilization and seed development in *Arabidopsis*. To this day, it remains a mystery which transcription factors regulate *NTT* expression, despite it being essential for gynoecium development. To address these limitations, we performed *in silico* analysis of the *NTT* promoter to identify potential transcription factor binding sites in gynoecium development. With these predictions in mind, we will develop a yeast-one-hybrid (Y1H) interaction system using a customized, automated protocol on the OT-2 robot. This system allows for efficient and reproducible exploration of the multiple interactions observed *in silico* analysis between specific promoters, such as *NTT*, and the set of transcription factors involved in floral development. This approach facilitates the elucidation of regulatory networks for specific genes and opens the possibility of designing specific strategies to improve agronomic traits of interest, such as productivity or crop adaptation.

Poster 60. Application of iPBS molecular markers to assess genetic variability of Mexican sour Lime (*Citrus sp.*) from Tixtla, Guerrero

Alfredo Santana Pastor [1], Kelly Maribel Monja Mio K. M.[1], Lorenzo Felipe Sánchez Teyer [2], Antonio Rescalvo-Morales [1*]

[1]Sede CICY-Acapulco, Centro de Estudios e Investigación en Agroecología, Ambiente y Salud. Av. Bora Bora, Lote 10B, colonia La Poza, Acapulco de Juárez, Gro. C.P. 39906.
[2]Unidad de Biotecnología, Centro de Investigación Científica de Yucatán, A. C., Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo; C.P. 97205, Mérida, Yucatán, México
*Email: antonio.rescalvo@cicy.mx

Mexican sour lime (Citrus sp.), locally known as *Lima agria* is a citrus crop of high cultural and gastronomic importance in Mexico, particularly in the states of Guerrero and Yucatán, valued for its intense aroma and acidic flavor. In communities such as Tixtla, it also represents a significant element of cultural identity. However, widespread phytopathogen propagation has drastically reduced sour lime plantations, raising concerns about their genetic diversity. Retrotransposon-based inter-primer binding site (iPBS) markers have proven efficient for genetic analysis in various species, offering a simple and effective alternative for sour lime studies. This study aimed to evaluate the effectiveness of iPBS markers for assessing genetic diversity in three communities of Tixtla, Guerrero (Omeapa, Estacada, and Centro). Genomic DNA was extracted from fresh leaves and ten iPBS primers were screened. The results showed that only two of them exhibited high polymorphism, iPBS2232 and iPBS2374, generating 57 scorable bands, of which 49 (85.96%) were polymorphic. Both primers successfully grouped samples by community. Centro showed the highest genetic diversity, whereas Estacada exhibited the lowest. Overall, the analysis revealed low genetic variability, likely associated with reduced plantation size. This work is the first report on the application of the iPBS marker system in Mexican sour lime. Our findings demonstrate that iPBS markers are effective for evaluating genetic differences, providing valuable information for the development of effective conservation, propagation, and genetic improvement programs in Mexico.

Poster 61. **Establishment of a protocol for polyploidy induction on *Agave cupreata***

Tabatha N. Rios-Inurreta[1], Gabriel de Jesús-Ojeda[2], Kelly M. Monja-Mio[3], L. Felipe Sánchez-Teyer[1]*

[1]Unidad de Biotecnología, Centro de Investigación Científica de Yucatán, CICY, A.C., Mérida, México, C. 43 No. 130-x 32 y 34 Col, Chuburná de Hidalgo, Pinzón II, 97205.
[2]Dirección de Gestión Tecnológica, Centro de Investigación Científica de Yucatán, CICY, A.C., Mérida, México, C. 43 No. 130-x 32 y 34 Col, Chuburná de Hidalgo, Pinzón II, 97205
[3]Sede CICY-Acapulco, Centro de Estudios e Investigación en Agroecología, Ambiente y Salud, Av. Bora Bora, Lote 10B, colonia La Poza, Acapulco de Juárez, Gro. C.P. 39906.
*Email: santey@cicy.mx

Agave cupreata, endemic to Mexico, plays a vital role in the production of ancestral mezcal, a traditional spirit recognized under Mexican Official Standards for its artisanal and cultural value. This species is particularly important in rural communities of Guerrero, where mezcal is not only a source of income but also a symbol of cultural identity. However, *A. cupreata* propagation is severely limited by its slow growth, reproduction dependent on seeds and bulbils, and widespread harvesting of inflorescences, which prevents sexual reproduction. These challenges have contributed to its classification as “endangered” by the International Union for Conservation of Nature (IUCN). To address both conservation and agronomic needs, this study explores polyploid induction as a non-transgenic strategy which may improve characteristics such as plant vigor, biomass, and sugar content, key traits for mezcal production. Meristematic tissues from mother plants were cultured in vitro and exposed to oryzalin at two concentrations (2.5 µM and 5 µM) and two exposure durations (30 and 60 minutes), in all possible combinations. Cultures were established on modified Murashige and Skoog medium (MSb; Robert et al., 2006) optimized for *agave* species. Preliminary cytogenetic analysis confirmed that both untreated and one treated line individuals remained diploid (2n), with the mother plant karyotype showing 10 large and 50 small chromosomes (n=30). Explant survival rates decreased with longer exposure times, suggesting a cytotoxic threshold. Although polyploidy apparently was not achieved under current conditions, these results provide a baseline for future optimization. The integration of *in vitro* propagation and polyploid induction offers a promising pathway to develop improved *A. cupreata* lines, aligning biotechnology with the preservation of traditional practices. This approach supports both the conservation of a culturally significant species and the sustainable development of ancestral mezcal production systems in Mexico.

Poster 62. Study of perivascular fiber formation in *Agave fourcroydes* through a transcriptomic approach

Roberto Cruz-Balam[1], Enrique Ibarra-Laclette[2], Emanuel Villafán[2], Fulgencio Alatorre-Cobos[1]*

[1]Centro de Investigación Científica de Yucatán, Unidad de Biología Integrativa, Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo, 97205, Mérida, Yucatán, Mexico.
[2]Red de Estudios Moleculares Avanzados, Instituto de Ecología A.C., Carretera Antigua a Coatepec 351, El Haya, 91070, Xalapa, Veracruz, Mexico.
*Email: fulgencio.alatorre@cicy.mx

Although molecular mechanisms controlling the xylem development have been widely explored in several plant models, the formation of other supporting plant tissues like sclerenchyma (fiber) is scarcely known. Here, we used *Agave fourcroydes*, one of the main sources of hard fiber worldwide, to unravel the molecular events underlying ontogeny and development of perivascular fibers using a transcriptomic approach. Three different development stages of agave leaves were sampled to collect fibers and mesophyll tissues, and know their transcriptomic landscapes by RNAseq. The contrast between fiber and mesophyll tissue was more informative, with 3470 genes differentially expressed throughout the three developmental ages analyzed. Interestingly, the biological categories that were more enriched were those related to cell wall organization or biogenesis, cellulose, beta-glucan, and phenylpropanoid metabolism. Genes encoding peroxidases (PRX) and laccases (LAC), two key enzymes involved in the lignin monolignol polymerization, were identified in agave for the first time. L*AC4, LAC7, LAC11, PRX1, PRX25*, and *PRX52* were upregulated in the youngest leaf stage. Enzymatic assays using DAB revealed enhanced PRX and LAC activities in fibers compared to parenchyma cells, consistent with the RNAseq data. Ongoing analyses are focused on the genome-level identification of *PRX* and *LAC* families in *A. fourcroydes* and *A. tequilana*.

Poster 63. Identification of parental lines of maize hybrids using monomorphic molecular markers

Luna-Rodriguez P[1,2*], Burgueño J.[2], Nair S.K.[3], Gajanan S.[3]

[1]Faculty of Agronomy, Universidad Nacional Agraria La Molina, Lima, Peru, Av. La Molina s/n.
[2]International Maize and Wheat Improvement Center, Texcoco, Mexico, Carretera México-Veracruz Km. 45
[3]International Maize and Wheat Improvement Center, Hyderabad, India, 502324 Hyderabad.
[*]Email: 20210579@lamolina.edu.pe

Hybrid maize varieties are widely used because of their hybrid vigor, but the development of homozygous parental lines is time-consuming and resource-intensive. However, in different contexts, genetic material is used by third parties to develop new hybrids that are launched on the market. This situation generates the need to identify the parents used in obtaining hybrids by means of genomic analysis tools. This identification is important for protecting intellectual property, assessing the impact of parental line production programs, and regenerating hybrids with unknown genealogy. This study aims to improve the method of monomorphic profile of hybrids (UPOV, BMT/17/16 Add.), which considers only the proportion of similarities between the hybrid and the possible parental line. While the method focuses on conserved regions from one parent, it does not consider linkage disequilibrium or SNP heterozygosity. To assess this, 1223 parental lines and 20 hybrids, obtained from some of these lines, from International Maize and Wheat Improvement Center were used. Seeds were germinated, DNA was extracted, and samples were then sent to Diversity Array Technologies Pty Ltd for genotyping. The genetic diversity of the parental lines was analyzed by multidimensional scaling and linkage disequilibrium analysis. Furthermore, 6 scoring methods were developed to evaluate the similarity between hybrids and potential parents, with each method differing in how it handles matches, mismatches, missing data, heterozygous SNPs, marker correlation, allele specificity, and Bayesian probabilities. Results showed that 92.5% of actual parental lines scored within the top 30% across all methods. However, due to genetic similarities among lines and differences in missing or heterozygous SNPs, true parents did not always receive the highest scores. The scoring method that considered allele specificity performed best, identifying 25% of real parents among the top 20 scores. To improve accuracy, the study recommends methodological refinement and the use of more markers.

Poster 64. Transcriptomic analysis of homeologous genes in the allotetraploid *Coffea arabica*

Marcos David Couoh Cauich[1], Elsa Beatriz Góngora Castillo[2], Ramon Pacheco-Arjona[3], Enrique Ibarra Laclette[4], Víctor M. Loyola Vargas[1]*

[1]Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, Calle 43, No. 130 x 32 y 34, Mérida 97205, Yucatán.
[2]Departamento de Recursos del Mar, Cinvestav Mérida, Carr. Mérida - Progreso, Loma Bonita, 97205 Mérida, Yucatán.
[3]Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma de Yucatán, Carretera Mérida-Xmatkuil Km. 15.5 Tizapán, 97100 Mérida, Yucatán.
[4]Red de Estudios Moleculares Avanzados, Instituto de Ecología, A.C. (INECOL), Xalapa 91073, Veracruz.
*Email: vmloyola@cicy.mx

C. arabica is an allotetraploid species widely cultivated for the high quality of its beans. It originated through natural hybridization between *C. canephora* and *C. eugenioides*, and although it contains both subgenomes, it exhibits lower resistance to abiotic and biotic stress factors compared to *C. canephora*. Numerous transcriptomic studies on *C. arabica* have used only the *C. canephora* genome as a reference for sequence mapping, which has made it impossible to distinguish the specific contribution of each subgenome. The current availability of high-quality genomes for *C. arabica* and its parent species enables these data to be reinterpreted using a subgenome-specific, expression-targeted approach. The objective of this work is to identify homeologous genes shared between both subgenomes and to characterize the relative contribution of *C. canephora* and *C. eugenioides* derivatives in different tissues and stress conditions, using differential expression analysis based on publicly available RNA-seq data from multiple studies. In addition to global transcriptomic analysis, special attention will be paid to genes involved in auxin and cytokinin metabolism, due to their key roles in development and response to environmental stimuli. This study aims to determine the differential expression patterns between subgenomes and identify the specific contribution of each progenitor to the adaptation and development of *C. arabica*.

Poster 65. **Strategies for the genetic transformation of Mexican barley (*Hordeum vulgare* L.) cultivars based on anther culture and PDS-CRISPR/Cas9**

Pedraza-Mejía, M., Rodríguez-Gandarilla, M. G., Massange-Sánchez, J. A.*

Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco, A.C. (CIATEJ), Unidad de biotecnología vegetal, Zapopan, México. Camino Arenero 1227, El Bajío, 45019, Zapopan, Jalisco.
*Email: jmassange@ciatej.mx

Barley (*Hordeum vulgare* L.) is the fourth most important cereal crop worldwide, with significant applications in beer production and forage. In Mexico, its relevance is linked to the brewing industry, as the country is projected to remain the world's top beer exporter by 2030. Modern tools such as transgenesis and CRISPR/Cas9 offer promising alternatives to accelerate genetic improvement. However, barley is highly genotype-dependent and recalcitrant to transformation. A recent approach reported by Yong Han (2021) demonstrated the use of Agrobacterium-mediated transformation via anther culture, which regenerates different barley varieties from microspores due to their totipotent nature. This work aims to develop a genetic transformation protocol for Mexican barley cultivars using anther culture as a regeneration system and the CRISPR/Cas9 system targeting the PDS gene. Currently, we are standardizing the optimal stage for spike collection across cultivars by evaluating developmental time (8–12 weeks after sowing), booting thickness, and inter-ligule distance. Anther cultures are being tested with different growth regulators (2,4-D, Dicamba, Picloram) to optimize callus induction. In parallel, the PDS gene is being cloned and sequenced to design specific single-guide RNAs (sgRNAs). For vector construction, we follow the Golden Gate protocol. The combination of anther culture with genome editing tools could overcome current limitations in barley transformation, providing a functional platform for gene validation and trait improvement in Mexican cultivars.

Poster 66. Comparative genomics of historic grapevine varieties in Mexico

Beatriz Adriana Domínguez Flores[1], Sara Covarrubias[1], Cristina Dominguez-Castro[2], Alejandra Rougon-Cardoso[1], Rufina Hernández-Martínez[3], Luis Delaye[4] & Antonio Hernández-López[1*]

[1]Laboratorio de Investigación Interdisciplinaria, Escuela Nacional de Estudios Superiores Unidad León, Universidad Nacional Autónoma de México, Blvd. UNAM 2011, Predio El Saucillo y Comunidad de los Tepetates, El Potrero, 37684 León, Guanajuato, México.
[2]Facultad de Enología y Gastronomía, Universidad Autónoma de Baja California, Carretera Transpeninsular Ensenada-Tijuana 3917, Col. Playitas, Ensenada Baja California 22860, México.
[3]Departamento de Microbiología, Centro de Investigación Científica y de Educación Superior de Ensenada, CICESE, Carr. Tijuana-Ensenada 3918, Zona Playitas, Ensenada Baja California 22860, México.
[4]Departamento de Ingeniería Genética, Centro de Investigación y de Estudios Avanzados del IPN, Unidad Irapuato (CINVESTAV), Libramiento Norte Carretera Irapuato León, 36821 Irapuato, México.
[*]Email: ahernandez@enes.unam.mx

Climate change presents serious challenges to viticulture worldwide, with rising temperatures and increasing drought stress threatening grape production. In Mexico, the loss of historical grapevine varieties is particularly concerning, as these genotypes are often better adapted to local environmental conditions. Historic cultivars such as Rosa del Perú and Listán Prieto require significantly less care and maintenance than modern varieties while still achieving good yields and exhibiting superior tolerance to heat and drought—traits shaped by centuries of natural selection and traditional agricultural practices. This study aims to compare the genomes of Rosa del Perú and Listán Prieto from Baja California against the PN40024 (T2T) reference genome to identify genomic variants potentially linked to their stress-resilient traits. Whole-genome comparisons focused on single nucleotide polymorphisms (SNPs) and insertions/deletions (InDels), followed by variant effect prediction and contextual gene analysis to assess functional impacts on protein-coding regions. Preliminary results show that 94% of sequences map uniquely to the reference genome, and variant analysis revealed a substantial number of high-impact changes, including coding region disruptions and frameshift mutations caused by InDels. These findings provide critical insights into the genomic basis of heat and drought tolerance, disease susceptibility, and other agronomically relevant traits. The study contributes to the preservation of genetic diversity in viticulture and offers valuable information for the development of climate-resilient grapevine cultivars in the face of increasing environmental stress.

Poster 67. In-situ localization of CCDs: CCD1-1 and CCD4-4 potentially related to bixin synthesis

Lorena López-Valladolid, Margarita Aguilar-Espinosa and and Renata Rivera-Madrid*

Unidad de Biología Integrativa, Centro de Investigación Científico de Yucatán A.C., Mérida, Yucatán, México.
*Email: renata@cicy.mx

Bixa orellana L., an important tropical plant in Yucatán, is valued for its seeds, which contain bixin, a natural pigment with applications in the food, pharmaceutical, cosmetic industries, and traditional medicine. The first stage of bixin biosynthesis is mediated by a CCD-type enzyme, which uses lycopene as a substrate. Transcriptome analyses of annatto have identified BoCCD1-1 and BoCCD4-4 as potentially involved in the bixin biosynthetic pathway. Research on the cellular localization of these enzymes has found that BoCCD1-1 is located in the cytoplasm, while BoCCD4-4 is located in plastids. The high expression of the mRNAs of these enzymes in bixin storage cells (BSCs), as well as their strong expression in seeds, suggests that bixin synthesis may be occurring in both plastids and the cytosol, possibly through alternative pathways. The *in situ* PCR technique for localizing the mRNAs of the mentioned enzymes in immature seed tissues will provide further evidence about their behavior, localization, and expression.

Poster 68. Genetic regulation of haustoria formation in the hemiparasite plant *Castilleja tenuiflora* during interaction with its host

Roselia Vázquez Brito[1], Aida Araceli Rodríguez Hernández[2], Gabriela Trejo Tapia[1]*
[1]Centro de Desarrollo de Productos Bióticos, Instituto Politécnico Nacional (CEPROBI-IPN), Morelos, México.
[2]Centro de Desarrollo de Productos Bióticos, Instituto Politécnico Nacional IPN, Morelos, México, Secretaría de Ciencia, Humanidades, Tecnología e Innovación SECIHTI-CEPROBI/IPN.
*Email: gttapia@ipn.mx

Hemiparasitic plants, such as *Castilleja tenuiflora*, can establish a connection with a host plant when necessary or when the opportunity arises. These plants possess a functional photosynthetic system, which generally enables them to meet their biological requirements. The association between a parasite and its host is established through the formation of haustoria, which are specialized, multicellular structures that form a xylem bridge, allowing the bidirectional flow of molecules, such as water and nutrients. Haustoria formation begins when the parasitic plant perceives chemical signals released by the host plant. These signals, known as haustoria-inducing factors (HIFs), are perceived by the parasitic plant through transmembrane proteins and transcription factors. Activation of these signals promotes cell division, expansion, and differentiation, leading to the formation of haustoria. No genes involved in plant-plant interactions have been identified in *C. tenuiflora* to date. Therefore, the objective of this study was to identify, isolate, and evaluate the expression of genes associated with haustoria formation of *C. tenuiflora* during its interaction with the host plant *Baccharis conferta*. To this end, the genes of interest were isolated and identified using an in vitro parasite-host co-culture system. Subsequently, their expression levels were evaluated by RT-qPCR following treatments with DMBQ and syringic acid. The results demonstrated that two genes may be involved in the signaling pathways activated during haustorium formation in *C. tenuiflora*.

Poster 69. Analysis of the expression of *glx-I* in maize coleoptils infected by *Aspergillus parasiticus*

Angeles Regina García-Gutiérrez[1], Carlos Francisco Varapizuela-Sánchez[1]*, María del Socorro Pina-Canseco[2], Marco Antonio Sánchez-Medina[1]

[1]Tecnológico Nacional de México-Instituto Tecnológico de Oaxaca, Avenida Ing. Víctor Bravo Ahuja No. 125, esquina Calzada Tecnológico, C.P. 68030, Oax, Méx.
[2]Centro de Investigación, Facultad de Medicina (UNAM-UABJO), Av. San Felipe del Agua (2,69 km), C.P. 68020, Oax, Méx.
*Email: carlos.varapizuela@itoaxaca.edu.mx

In Mexico, *Aspergillus parasiticus* infects maize in the field and during storage, producing aflatoxins through the interaction between the fungus, host, and environment. The objective of this research was to evaluate the expression of the stress response gene *glx-I* in coleoptils of Bolita maize (MRBI) infected by *A. parasiticus*, in comparison with two pure lines, CML 442I and CML 495I, donated by CIMMYT. For the trials, seeds were inoculated with *A. parasiticus* ATCC 16992 ($4x10^6$spores mL^{-1}) and non-inoculated controls, incubated at 28 ± 2 °C for 7 days. Total RNA was extracted with TRIzolTM and cDNA was synthesized using the RevertAid kit with 500 ng of RNA as a template. Amplification of the *glx-I* gene was performed in 30 cycles by PCR with GoTaq® Green Master Mix, using specific oligonucleotides and *MacI* actin as an internal control. The products were analyzed by electrophoresis on a 1.5% agarose gel and quantified by densitometry with ImageJ®. The *glx-I* gene shows differences in expression levels in response to pathogen stress between genotypes. In CML 442I, expression increases significantly by 57%. On the other hand, in MRBI and CML 495I, expression decreases to 39% and 44%, respectively. In addition, *MacI* in the control group shows isoforms, possibly related to specific functions and differential regulations in cell development.

Poster 70. Dual VIGS vector: a rapid and effective tool for gene silencing in plant biotechnology

Rubio-Rodríguez E[1], Jimenez-Romero D[2], Sepúlveda-García EB[3], Ramos-Valdivia AC[2], Trejo-Tapia G[1*]

[1]Instituto Politécnico Nacional, Centro de Desarrollo de Productos Bióticos, Departamento de Biotecnología, Laboratorio de Productos Naturales, Yautepec, Morelos, 62730 México.
[2]Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Departamento de Biotecnología y Bioingeniería, Ciudad de México, 07360 México
[3]Laboratorio de Biotecnología Vegetal, Instituto de Biotecnología, División de Estudios de Posgrado, Universidad del Papaloapan, San Juan de Tuxtepec, Oaxaca 68301, Mexico
[*]Email: gttapia@ipn.mx

Virus-induced gene silencing (VIGS) is a powerful tool in plant biotechnology for studying the function of biosynthetic genes involved in the production of specialized metabolites. While the VIGS assay provides rapid qualitative and quantitative results, it often exhibits low reproducibility and high variability due to localized and transient infections that fail to impact the entire plant system. Traditional vector construction methods are constrained by the need to select restriction sites and require multiple digestion and ligation steps, resulting in increased reagent use and processing time. To obtain rapid results and study the biosynthetic pathway of specialized metabolites in non-model medicinal plants, such as *Castilleja tenuiflora*, a method for vector construction, called dual fusion vectors, was designed for use in the VIGS system. Efficient transgene construction was achieved through an overlap PCR system, which was used to fuse two genes of interest. For the insertion of the fused genes into the destination vectors, the Invitrogen Gateway recombination cloning technology was used. Five fused dual vectors were constructed, using protoporphyrin magnesium chelatase subunit H (*Cte-chlH*: ID OR458580) as a marker gene and five genes involved in the specialized metabolism of *C. tenuiflora*. Three genes are involved in the biosynthesis of phenolic compounds: phenylalanine ammonia-lyase (*Cte-PAL*: ID MN517910), cinnamate-4-hydroxylase (*Cte-C4H*: ID OR473003), pinoresinol-lariciresinol reductase (*Cte-PRL*: ID OR473002), while two are involved in the biosynthesis of terpene compounds: 1-deoxy-D-xylulose-5-phosphate synthase (*Cte-DXS*: ID MN517909) and geraniol 10-hydroxylase (*Cte-G10H*: ID MN517912). The construction of fused dual vectors yields greater efficiency in the transgene construction process, enabling visible, rapid, efficient, and successful gene silencing in medicinal plants of pharmacological interest.

Poster 71. Genome-wide identification of miRNA biogenesis genes in mango (*Mangifera indica L.*) and their response to heat stress

Mitzuko Dautt-Castro*, Andrés G. López-Virgen,
Lourdes K. Ulloa-Llanes and Maria A. Islas-Osuna*

Centro de Investigación en Alimentación y Desarrollo, A.C. Hermosillo, Sonora, México. CP. 83304.
*Email: islasosu@ciad.mx

MicroRNAs (miRNAs) are central regulators of plant responses to abiotic stress, yet their biogenesis and regulation remain poorly characterized in many fruit crops. Mango (*Mangifera indica L.*), a major tropical fruit, undergoes quarantine hot water treatment (QHWT) for postharvest disinfestation, a process that causes heat stress and accelerates fruit softening. To investigate the molecular basis of mango's stress response, we performed a genome-wide identification of gene families involved in miRNA biogenesis and assessed their expression under QHWT. We identified five *DICER-LIKE* (*MiDCL*), thirteen *ARGONAUTE* (*MiAGO*), six *HYPONASTIC LEAVES1* (*MiHYL1)*, two *SERRATE* (*MiSE*), one *HUA ENHANCER1* (*MiHEN1)*, one *HASTY* (*MiHST*), and five *HEAT-SHOCK PROTEIN 90* (*MiHSP90)* genes. Domain analysis confirmed the presence of conserved motifs in all protein sequences, which supports their putative functionality. Phylogenetic reconstruction clustered mango sequences with orthologs from Arabidopsis, *Solanum lycopersicum*, and *Citrus sinensis*, indicating strong conservation across angiosperms. *Cis*-regulatory element analysis revealed an enrichment of stress-responsive motifs (MYB, ABRE, GARE, and MeJA), suggesting potential regulation under abiotic stress. Expression profiling of selected genes in mesocarp tissue exposed to QHWT (115 °F for 75 min) showed significant transcriptional modulation. Most genes were rapidly induced between 3 and 6 hours post-treatment, consistent with an early role of miRNA pathways in stress adaptation. For instance, *MiDCL1* and *MiAGO1b* peaked at 3 h, while *MiDCL3* and *MiAGO6* were induced later, at 6 h. Notably, *MiHSP90.3* exhibited an expression pattern aligned with miRNA biogenesis genes, highlighting its dual role in heat response and RISC assembly. Our findings provide a comprehensive catalog of miRNA biogenesis genes in mango and demonstrate their dynamic regulation under heat stress. These results establish a foundation for functional studies on small RNA-mediated pathways in mango and may contribute to the development of postharvest strategies aimed at preserving fruit quality under quarantine treatments.

Poster 72. **Toward understanding WOX5 function in cacti**

Sofía Esteban-Hernández[1,2], Elizabeth Cordoba-Martinez[1], Ramsés Albarrán-Hernández[1], Svetlana Shishkova[1*]

[1]Instituto de Biotecnología, UNAM campus Morelos, Cuernavaca, Morelos, México
[2]Centro de Investigación en Dinámica Celular, UAEM, Cuernavaca, Morelos, México
[*]Email: svetlana.shishkova@ibt.unam.mx

The *WUSCHEL HOMEOBOX 5* (*WOX5*) gene plays an important role in maintaining the identity and function of root stem cell niches. *WOX5* expression is almost specific for the quiescent center (QC) of the root apical meristem (RAM) in several plant species. The presence of the QC is important for the RAM maintenance. RAM exhaustion and consequent determinate root growth has been reported for several *Arabidopsis thaliana* mutants with compromised QC identity. Also, the activation of cell divisions in the *A. thaliana* QC converges on the WOX5 transcription factor. However, little is known about the role of this transcription factor in non-model plants like cacti. In many Cactaceae species, RAM exhaustion and determinate growth of primary and lateral roots have been reported as an adaptive mechanism to cope with water deficit. Although this phenomenon was described almost 30 years ago, its molecular basis remains poorly understood. Characterizing the cacti orthologues of *AtWOX5* could help elucidate the genetic regulation of the determinate root growth and the potential role of WOX5 in RAM exhaustion. In our research group, we identified two *WOX5* paralogues, both of which are putative *AtWOX5* orthologues, in two cactus species: *Carnegiea gigantea* and *Selenicereus undatus*. In this project, the promoter region of both *WOX5* paralogues from *Carnegiea gigantea* were amplified and cloned with the aim of analyzing the expression pattern of the cacti *WOX5*. As part of this work, the tolerance of untransformed cactus calli to hygromycin was also evaluated. The future functional study of *WOX5* genes in cacti, of which this project represents the initial phase, will contribute to a better understanding of the mechanisms underlying RAM exhaustion in cacti. It could also be helpful to explore the QC functionality in cacti roots. This work was partially funded by DGAPA-PAPIIT-UNAM project IN208824.

Poster 73. **Beyond the established self-incompatibility models: a new biochemical system in *Sambucus ebulus***

Emilio García-Caffarel[1], Bruce A. McClure[2], Yuridia Cruz-Zamora[1], Felipe Cruz-García[1*]

[1]Departamento de Bioquímica, Facultad de Química, Universidad Nacional Autónoma de México. Cd. Mx, 04510, Mexico.
[2]Department of Biochemistry, University of Missouri, Life Sciences Center, Columbia, Missouri 65211.
[*]Email: fcg@unam.mx

Self-incompatibility (SI) is a genetically encoded mechanism that prevents self-fertilization in hermaphroditic flowering plants (~ 90% of the species). SI is controlled by the *S*-locus, which contains the two tightly linked pollen- and pistil-expressed *S*-determinants (pollen-*S* and pistil-*S*, respectively). The *S*-locus is polymorphic, resulting in a variety of different haplotypes. Rejection of self-pollen occurs when the pollen-*S* haplotype matches the pistil-*S* haplotype; alternatively, fertilization occurs when there is no match between the pollen- and pistil *S* haplotypes. Nowadays, both *S*-determinants have been identified in the three best-characterized SI systems. In the S-RNase based SI system, *pollen-* and *pistil-S* code for an F-box protein (SLF) and a ribonuclease (S-RNase), respectively. In the Brassicaceae SI system, *pollen-S* codes for a secreted ligand (SP11/SCR) and *pistil-S* codes for a receptor kinase (SRK). Finally, in the Papaveraceae SI system, *pollen-S* codes for a receptor (PrpS) and *pistil-S* codes for a ligand (PrsS). We are currently studying *Sambucus ebulus* (Dipsacales), which has been shown to possess gametophytic SI –meaning the pollen's compatibility phenotype is determined by its own haploid genotype–. By observing pollen tube growth within the pistil, we have evaluated the spatial and temporal SI response in this species. In addition, while studying the cross outcome, we have determined that, although fruits are sometimes formed after self-pollination, seeds are never viable, probably because pollen tubes never reach the ovary. To identify the underlying *S*-determinants in *S. ebulus,* we followed an RNAseq-based approach. Our findings suggest that the SI system in *S. ebulus* diverges from the three canonical models, as the putative *pollen-* and *pistil-S* genes differ from those described previously. Ongoing work is focused on characterizing their biochemical properties, assessing allele-specific interactions, and determining their subcellular localization, with the broader goal of elucidating the novel molecular pathway underlying SI in *Sambucus*. Funding: DGAPA: IN213025; FQ: 5000-9198.

Poster 74. **Analysis of translational regulation in *Arabidopsis thaliana* in response to phosphate deficiency**

Ana Karen Gervacio-Montecillo[1], Sandra Isabel González-Morales[1], Luis Herrera-Estrella[1,2,*]

[1]Unidad de Genómica Avanzada (UGA-LANGEBIO) del Centro de Investigación y de Estudios Avanzados del IPN, Irapuato, Gto., 36821, México. [2]Plant and Soil Science Department, Institute of Genomics for Crop Abiotic Stress Tolerance, Texas Tech University, Lubbock, TX, 79409, United States.
*Email: lherrerae@cinvestav.mx

Phosphorus (P) is an essential macronutrient for plant development. Although soils typically contain high levels of P, plant growth and productivity are often limited due to its low availability. The P low availability is caused by different factors as plants only take and metabolize P in the form of inorganic phosphate (Pi). Under Pi deficient conditions, plants activate a wide range of adaptative responses to cope with the lack of this nutrient. These responses have been extensively studied at the genetic level, primarily focusing on transcriptional regulation. However, it is well established that the mRNA abundance does not always correlate with protein levels, highlighting the need for further investigation into post-transcriptional regulatory mechanisms, such as regulation at the translational level. Recent studies on translational efficiency, using polysome profiling, in *Arabidopsis thaliana* plants exposed to Pi deficiency suggest that this layer of regulation plays a significant role in the adaptive response. In particular, the enhanced translation of mRNAs that encode proteins involved in expanding the root system—such as those promoting lateral root growth and increasing root hair length and density—appears to be a key component of the plant's strategy to improve Pi acquisition. This work aims to investigate mRNAs subject to translational regulation and to identify transcript regions responsible for this process. In addition, considering that the identified genes are mainly linked to root architectural changes under phosphate deficiency, we will evaluate the translational efficiency of mRNAs across distinct root cell types in *Arabidopsis thaliana* under low phosphate conditions.

Poster 75. Identification of microRNAs of *Carnegiea gigantea*, a Cactaceae with determinate growth of the primary root.

Kenia A. Galván-Alcaraz[1], Gustavo Rodríguez-Alonso[2], Svetlana Shishkova[1]*

[1]Institute of Biotechnology, UNAM. Cuernavaca, Morelos, México. Av. Universidad 2001, Col. Chamilpa, Cuernavaca, Morelos, México.
[2]Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México. Av. Universidad 1001, Col. Chamilpa Cuernavaca, Morelos, México.
*Email: svetlana.shishkova@ibt.unam.mx

Cactaceae is a family of plants distributed throughout the American continent and is distinguished by numerous developmental adaptations. One of these adaptations is the determinate growth of the primary root, caused by the exhaustion of the root apical meristem (RAM) and the consequent differentiation of all root-apex cells. The determinate growth of primary and lateral roots in Cactaceae has been suggested to be an adaptation to arid and semi-arid environments, since it promotes the rapid development of a shallow and branched root system. To further explore genetic regulation of the determinate growth, smallRNAseq of primary root apex on initial (active RAM) and terminal (exhausted RAM) stages, the whole root and the shoot of *Carnegiea gigantea*, a Sonoran Desert columnar cactus a.k.a. 'Saguaro' was performed. A total of 176 sequences of microRNA precursors and 138 sequences of mature microRNA were identified using computational methods, among them 114 precursors belong to conserved families and 62 were precursors of possible novel microRNAs. A total of 141 mature microRNA and microRNA* sequences, that is, including 5´and 3´ sequences from the same precursor, were expressed in the root apex of *C. gigantea,* of these 39 sequences had a differential expression on the initial and terminal stages, which suggests their involvement in the determinate root growth. We will also present results of the analysis of conservation degree of the *C. gigantea* microRNAs identified in this work and of *Seleniceres undatus* and *Selenicerus monacanthus* publicly available microRNAs (http://www.pitayagenomic.com/). This work was partially funded by the DGAPA-PAPIIT-UNAM project IN208824.

Poster 76. Functional analysis of the cytokinin biosynthesis *IPT* genes during tomato flower and fruit development

Gustavo Alonso Luna Zuñiga, Eduardo Martínez Estrada, Stefan de Folter*

[1]Unidad de Genómica Avanzada (UGA-Langebio), Cinvestav, Km. 9.6 Libramiento Norte, Carretera Irapuato-León, CP 36824 Irapuato, Gto, Mexico.
*Email: stefan.defolter@cinvestav.mx

Isopentenyl transferases (IPTs) catalyze the initial step in the biosynthesis of cytokinins, phytohormones that regulate essential processes like cell division, delayed senescence, root development, and abiotic stress responses. Despite their importance, there is limited research on the specific function of the IPTs in tomato flowering and fruit development. This study aims to analyze the function of all 6 *IPT* genes during tomato flower and fruit development using CRISPR-Cas gene editing technology. To achieve this, the homologous *IPT* genes in tomato were identified through a literature review and bioinformatics tools. After identification, specific CRISPR-Cas9 guides were designed for the *IPT1-IPT6* genes and used to construct CRISPR vectors for each gene. Tomato cotyledons were then transformed with the vectors to generate mutant lines. These mutant lines will be genotyped by sequencing to determine the mutation events in each line. Subsequently, the phenotypes of flowers and fruits from at least two alleles per gene will be analyzed. We anticipate identifying mutants with altered inflorescence development (either an increase or reduction), variations in fruit size, and differences in floral branching. The results will provide insight into the function of each *IPT* gene in tomato.

Poster 77. 6mA methylation and gene regulation during chloroplast biogenesis in *Agave angustifolia* Haw.

Luis F. Núñez-Becerril and Clelia De-la-Peña*

Unidad de Biotecnología, Centro de Investigación Científica de Yucatán, Calle 43 No. 130 x 32 y 34 Col. Chuburná de Hidalgo, 97205 Mérida, Yucatán, México.
*Email: clelia@cicy.mx

N^6-methyladenine (6mA) is emerging as an epigenetic mark in eukaryotes, with increasing evidence supporting its role in genome regulation. In plants, 6mA has been identified in plastid genomes, particularly within genes associated with photosynthesis, suggesting a potential role in chloroplast biogenesis. This study comparing *in vitro* plantlets green (photosynthetic) and albino (non-photosynthetic somaclonal variation) phenotypes of *Agave angustifolia* Haw. to investigate the relationship of 6mA with chloroplast genes. To explore 6mA's involvement, we combine computational predictions with experimental validation. Using plant-optimized models (CNN6mA, EpiSemble), we identified 6mA-enriched regions in key plastid-encoded photosynthetic genes (*psbB*, *psbC*, *atpA*). These *in silico* findings will be validated through Single Molecule Real Time sequencing (SMRT-seq) and 6mA-specific immunoprecipitation assays. Additionally, chloroplasts will be isolated across developmental stages via sucrose density gradients, followed by NAME-seq profiling to assess correlations between 6mA dynamics and transcriptional activity. By researching the relationship between 6mA patterns and gene expression in chloroplasts, this study aims to uncover an epigenetic layer involved in regulating plastid development and photosynthesis. These results may contribute to a broader understanding of gene regulation in non-model monocots.

Poster 78. **Silencing pAMT transcripts by applicaction of dsRNA in habanero peppers fruits**

Arellano Ordoñez Estefania, Guevara González Ramón Gerardo, Cedillo Jiménez Christopher Alexis*

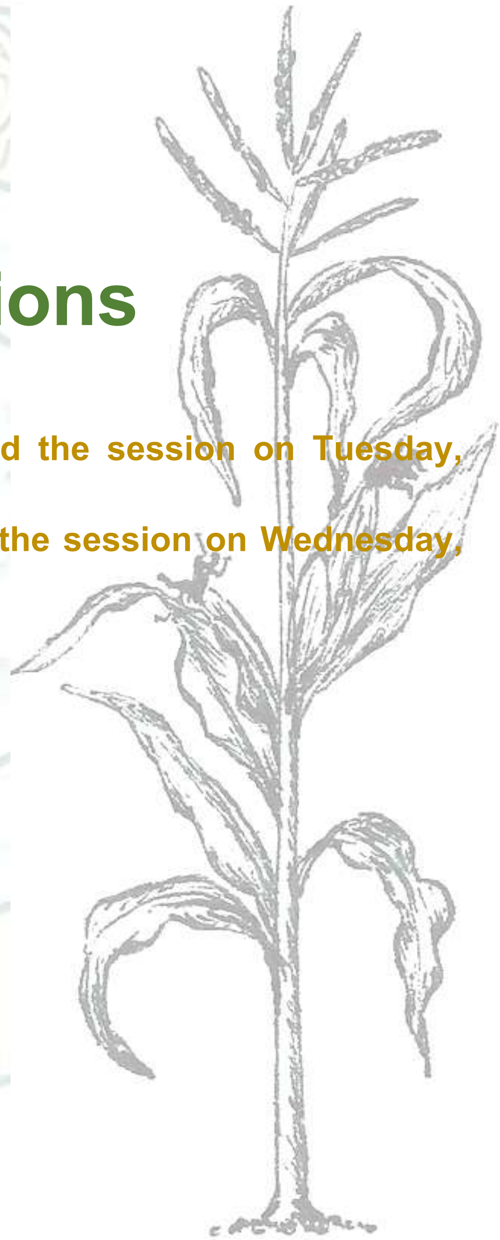
Centro de Investigación Aplicada a Biosistemas Universidad Autónoma de Querétaro, México.
*Email: c.cedilloc@gmail.com

Double-stranded RNA (dsRNA) has emerged as a biotechnological tool that modulates gene expression through the RNA interference (RNAi) mechanism, without generating permanent modifications in the genome. Its application has been explored in different species.This study evaluated the inhibition of putative aminotransferase (*pAMT*) gene transcripts in *C. chinense* fruits through the exogenous application of dsRNA, seeking to redirect the synthesis of capsaicinoids towards the formation of capsiate, a non-pungent analogue with nutraceutical potential. Fruits were selected 10 days post-anthesis (DPA) and treated by direct injection into the peduncle with three concentrations of dsRNA: T1 (1000 pmol), T2 (100 pmol), T3 (10 pmol), in addition to a control group treated with sterile nuclease-free water. The fruits were harvested at 20 DPA for molecular evaluation. The results showed a decrease in the transcript in the three treatments compared to the control, with 88.99% (T1), 37.93% (T2), and 28.94% (T3), respectively. Furthermore, these findings suggest the potential of RNAi-mediated gene silencing as a viable tool to modify the production of secondary metabolites in plants, representing a first step toward the development of biotechnological strategies aimed at managing the biosynthesis of capsaicinoids and capsinoids, which have economic, pharmaceutical, and agroindustrial significance.

Plant Biotic Interactions

Posters

Participants with even numbers will attend the session on Tuesday, October 21.
Participants with odd numbers will attend the session on Wednesday, October 22.



Poster 79. **The common bean (*Phaseolus vulgaris*) non-nodulating mutant *nnod*(1895) is affected in the initial step of rhizobial infection during the N-fixing symbiosis**

Karina San Vicente-Trujillo, Rocío Reyero-Saavedra, Gladys E. Jiménez-Nopala, Mario Ramírez, Sara I. Fuentes, Alfonso Leija, Mishael Sánchez-Pérez, Lourdes Girard and Georgina Hernández*

Centro de Ciencias Genómicas, UNAM. Avenida Universidad s/n, col. Chamilpa 62210. Cuernavaca, Morelos.
*Email: gina@ccg.unam.mx

Legumes are a major source of non-animal protein and constitute one of the primary components of the human diet in regions such as Latin America and Eastern Africa. A notable advantage of legumes is their capacity to establish symbiotic interactions with nitrogen-fixing bacteria known as rhizobia. Through this symbiotic nitrogen fixation (SNF) process, legume plants can thrive in nitrogen-deficient soils. The initial step of the SNF interaction involves molecular communication between the legume plant and compatible rhizobia. This begins with the secretion of flavonoids by the plant, which in turn induce the biosynthesis of Nodulation Factors (NF) in the bacteria. Our research group conducted a screening for symbiotic mutants in common bean (*Phaseolus vulgaris*), using an EMS mutant population derived from the BAT93 wild-type(WT) genotype. From this screening, we identified and selected the non-nodulating mutant *nnod*(1895) for further characterization. The *nnod*(1895) plants displayed impaired root hair deformation critical for bacterial entrapment and for root infection. Compared to the WT, *nnod*(1895) exhibited an increase in non-effective root hair deformations, suggesting a disruption in the early stages of SNF To identify the genetic basis of the mutant phenotype, we performed a comparative whole-genome sequence analysis between WT and *nnod*(1895). Among the candidate genes, the ISOFLAVONE SYNTHASE(IFS) gene in *nnod*(1895) was found to carry a high-impact variant resulting in the loss of the start codon. Previous studies have shown that mutations in IFS in other legumes prevent nodulation, as isoflavones are essential signaling molecules for the initiation of symbiosis. We analyzed root exudates from *nnod*(1895) plants and observed that they failed to induce rhizobial nod gene expression. Ongoing work aims to further characterize this mutant through the analysis of flavonoid and isoflavonoid profiles in *nnod*(1895) root exudates, as well as through genetic complementation experiments using the IFS gene.

Poster 80. **Analysis of the spatio-temporal activity of the synthetic promoters DR5 and TCSn in *Phaseolus vulgaris* roots inoculated with *Rhizobium etli***

Libni A. Castrejón-Noguerón, José Erik Cruz-Valderrama and Rosana Sánchez-López*

Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Campus Morelos, Avenida Universidad 2001, Colonia Chamilpa, Cuernavaca 62210, México.
*Email: rosana.sanchez@ibt.unam.mx.

Legumes have the capacity to establish a symbiotic relationship with rhizobia, which leads to the *de novo* formation of N_2-fixing nodules on roots. Nodule development involves well-coordinated infection and organogenesis. Epidermal infection is based on the formation of a rhizobia microcolony in a curled responsive-root hair, from which an infection thread is formed, allowing rhizobia entry into the root. In parallel, cortical cells in the infection site become proliferative, forming a nodule primordium. In *P. vulgaris* roots first cell division occurs in the subepidermal (S-E) cells. Further, cells differentiate leading to the nodule organogenesis. Rhizobia is delivered into cells of the central tissue of the maturing nodule, in an intracellular symbiosome, which allows the bacterial differentiation in bacteroids-expressing a nitrogenase complex, responsible for the N_2-fixation activity. Plant phytohormones auxins and cytokinins are key regulators of a battery of plant cell processes, including cell division and nodule development. For a better description of the dynamics between epidermal infection, cell division and hormone-responses, in this work we explored the spatio-temporal activity of the auxin- and cytokinin-responsive synthetic promoters DR5 and TCSn, respectively, in *P. vulgaris* nodulation. DR5- or TCSn-driven activity was traced as GUS histochemical staining in *P. vulgaris* roots non- or inoculated with *Rhizobium etli*. DR5 activity was observed in the root apex, the lateral roots in formation, the rhizobia-infected site and the nodule organogenesis process. Analysis of TCSn activity is in progress. We are also performing a microscopical analysis of transgenic roots expressing the nuclear marker NLS-Turquoise driven by promoters DR5 or TCSn. Partially supported by Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica/UNAM (IN218524 to R.S.L.) and a M.Sc fellowship (66d224a9cdea600538501e6d by Secretaria de Ciencia, Humanidades Tecnología e Innovación, SECIHTI) to L.A.C.N. and a postdoctoral fellowship by Programa de Becas Posdoctorales UNAM 2023-2025 to J.E.C.V. This work is based primarily on the Proyecto de Maestría en Ciencias Bioquímicas-UNAM by Libni Anajared Castrejón Noguerón.

Poster 81. **Analysis of the interaction between *Trichoderma atroviride* and *Phytophthora capsici***

David Cruz Ríos*, Fernando Uriel Rojas Rojas, Julio C. Vega-Arreguín

Laboratorio de Ciencias Agrogenómicas, Escuela Nacional de Estudios Superiores Unidad León, Universidad Nacional Autónoma de México (ENES-León UNAM), Blvd. UNAM 2011, León, Guanajuato 3768, México.
*Email: dcsonne24@gmail.com

Trichoderma atroviride is a fungus used as a biocontrol agent because of its ability to control plant pathogenic microorganisms, either freely in the rhizosphere or as an endophyte. It has the capacity to release cell wall-degrading enzymes or act through parasitism. The objective of this study was to characterize the interaction between *T. atroviride* and *P. capsici*, as well as to evaluate whether a glucanase encoded in the genome of *T. atroviride* is expressed during the parasitic interaction with *P. capsici*. The interaction between the two microorganisms (*T. atroviride* and *P. capsici*) was studied on potato-fructose agar (PFA) and potato-sucrose agar (PSA), with discs of each microorganism placed 2.5 cm apart. It was observed that in some replicates of the interaction on PFA medium, the fungal mycelium overgrew that of *P. capsici*, suggesting a parasitic interaction. In contrast, in the medium containing sucrose as the carbon source, inhibition by metabolites was noted, as the mycelia were suppressed without physical contact. Subsequently, a sample of the mycelia was taken from the interaction where *T. atroviride* mycelium was overgrowing that of *P. capsici* (PFA medium) for microscopic analysis. It was observed that the fungal hyphae wrapped around those of *P. capsici*, further suggesting parasitic interaction by *T. atroviride* toward *P. capsici*. In the genome of *T. atroviride*, the presence of a gene encoding a glucanase enzyme was identified. Oligonucleotides were designed, and the presence of the gene was confirmed by PCR. Subsequently, RNA was extracted from the mycelium of the interaction between *T. atroviride* and *P. capsici*, cDNA was synthesized, and PCR was used to detect the expression of transcripts of the glucanase gene. However, the transcripts could not be detected, suggesting that this glucanase may not be involved in the parasitic interaction. Currently, the ability of *T. atroviride* to prevent infection by *P. capsici* in cucumber, tomato, and chili plant roots is under study.

Poster 82. **Taxonomic and functional overlap of the coralloid root and seed microbiome from cycad *Dioon edule***

Suárez-Moo Pablo[1], Barona-Gómez Francisco[2], Cibrián-Jaramillo Angelica[3*]

[1]Unidad de Química-Sisal, Facultad de Química, Universidad Nacional Autónoma de México, 97356 Sisal, Yucatán, Mexico.
[2]Evolution of Microbial Chemodiversity Laboratory,Institute of Biology, Leiden University, 2333 BE, Leiden, the Netherlands.
[3]Naturalis Biodiversity Center, Darwinweg 2, 2333 CR, Leiden, Paises Bajos.
*Email: angelica.cibrianjaramillo@naturalis.nl

Cycads have specialized apogeotropic roots of small coral-like shapes, termed coralloid roots, which contain nitrogen-fixing cyanobacteria. High bacterial diversity and functional characterization of the coralloid root microbiome have been previously reported, while the seed cycad microbiome and its overlap with that of the coralloid root microbiome broadly remained largely unexplored. In this study, we employed an enrichment method and shotgun genomic sequencing to examine the taxonomic and functional overlap of microbial communities in the coralloid roots and seeds of the cycad *Dioon edule*. We analyzed the metagenomes of coralloid roots and seeds from the same individuals (six in total) and characterized the genomes of endophytes isolated from both plant structures. Our metagenomic investigation led to the detection of 166 and 175 bacterial genera in coralloid root and seed microbiome, respectively. The genera *Bacillus*, *Staphyloccocus*, *Serratia*, *Sphingomonas*, and *Rhizobium* were the most abundant in coralloid root, while *Bacillus*, *Rhodanobacter*, *Paenobacillus*, *Sphingomonas*, and *Burkholderia* were dominant in the seed microbiome. A total of 146 genera (75%) were shared between plant structures. Genes associated with functions such as plant cell wall degradation, phytohormone production, phosphate transport and solubilization, ferrous iron uptake were found in both coralloid roots and seeds. Endophytic species from the genera *Rhizobium* and *Serratia* were isolated from both plant structures and shared genes associated with plant growth promotion. The findings of this study contribute to understanding the ubiquity and importance of endophytes in the adaptation of *Dioon edule* to its local environment. Furthermore, the comparison between the coralloid root and seed microbiomes provides insights into the vertical transmission of microbial communities across successive generations via seeds.

Poster 83. **Host factors promoting increased arbuscular mycorrhizal benefit in maize**

Ruairidh J. H. Sawers[1*], Patrick Sydow[1], Sergio Pérez Limón[1], Meng Li[1], Melanie G. Perryman[1], Daisy Osoro[1], Alexandria Tran[2], Aswathy Sebastian[3], M. Rosario Ramírez Flores[4] and Jagdeep Sidhu[5]

[1]Department of Plant Science, Pennsylvania State University, State College, Pennsylvania, USA.
[2]School of Information Sciences, University of Illinois Urbana-Champaign, Champaign, Illinois, USA.
[3]Huck Institute of Life Sciences, Pennsylvania State University, State College, Pennsylvania, USA.
[4]Bioscience Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee, USA.
[5]Department of Agronomy, Horticulture and Plant Science, South Dakota State University, Brookings, South Dakota, USA.
*Email: rjs6686@psu.edu

Arbuscular mycorrhizal fungi (AMF) are ubiquitous in cultivated soils, forming symbiotic relationships with the roots of major crops. Studies in controlled conditions have demonstrated the potential of AMF to enhance the growth and nutrient efficiency of host plants. It is difficult, however, to estimate the benefit in the field, not least because of the lack of AMF-free controls. We report the use of maize genetic mutants to generate AMF incompatible *sentinel* plants as a baseline against which to evaluate the impact of the symbiosis. To estimate the effect of AMF and characterise the impact of host genotype on symbiotic outcome, we have selectively incorporated AMF-incompatibility into genetic mapping populations. Evaluating these populations in temperate and tropical fields, we have estimated the contribution of AMF at ~30% of total yield. We have found that the AMF effect can vary dramatically even within a single field in response to heterogeneity in soil fertility. We have used QTL analysis to identify regions of the genome linked to differential performance in the presence or absence of arbuscular mycorrhizal symbiosis. Combining QTL with allele-specific transcriptome analysis, we are identifying candidate genes implicated in specific aspects of host response to AMF. The genetic architecture of host response in our populations suggests trade-offs between growth with or without AMF. As such, as we look to reduce agricultural inputs and make better use of AMF and other soil microbes to promote efficient resource use, we may need to consider not only shifts in management practice but also the nature of crop varieties themselves. Supported by USDA project *The genetic basis of maize response to arbuscular mycorrhizal fungi* (2022-67013-38264) and CONABIO project *Impact of native arbuscular mycorrhizal fungi on maize performance* (N˚62, 2016-2018). RJHS is funded by USDA Hatch #PEN04734 Accession #1021929.

Poster 84. Characterization of *Botrytis cinerea* infection in *Lotus japonicus* and effect of nodulation on the defense response

Jessica Samantha Segura Muñoz [1,2], Ivette García Soto[2], Yordan Jhovani Romero Contreras[2] and Jesús Montiel González[2*]

[1]Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos. Av. Universidad 2001, 62210, Cuernavaca, Mor.
[2]Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México. Av. Universidad 2001, 62210, Cuernavaca, Mor.
*Email: jmontiel@ccg.unam.mx.

Legumes establish a symbiotic relationship with nitrogen-fixing bacteria known as rhizobia. This interaction leads to formation of root nodules, where nitrogen fixation occurs, enabling plants to grow in nitrogen-deficient soils. However, legumes are exposed to pathogens such as *Botrytis cinerea*, a necrotrophic fungus infecting over 1,400 plant species, including legumes. While *B. cinerea* infections are well documented, molecular mechanisms involved in legumes remain poorly understood. Additionally, it is unclear whether nodulation influences resistance to this pathogen. To address these questions, we developed a protocol to analyze *B. cinerea* infection in leaves of the model legume *L. japonicus*. Using epifluorescence microscopy, we observed that spore germination occurred between 1–2 days post-inoculation (dpi), followed by mycelial growth at 3–5 dpi. This response was accompanied by reactive oxygen species (ROS) production in the infected area, showing a typical defense response. Nodulated *L. japonicus* plants showed greater resistance to *B. cinerea* infection than the non-nodulated plants grown in nitrogen-supplemented medium. Additionally, *L. japonicus* mutants defective at different symbiotic stages exhibited variable susceptibility to *B. cinerea*. To identify genes involved in both processes, we are conducting RNA-seq analyses of mock and *Botrytis* infected leaves from *Lotus* plants grown under symbiotic and non-symbiotic conditions. This approach will provide insights into molecular mechanisms coordinating symbiosis and defense in *L. japonicus*. This study presents novel evidence on the molecular responses during *B. cinerea* infection in a legume and shows the benefits of nodulation against necrotrophic attacks. J.S.S.M. is a master's student in the Master's Program in Sciences at the Universidad Autónoma del Estado de Morelos and received the fellowship with CVU-2054613 from the Secretaría de Ciencia, Humanidades, Tecnología e Innovación (SECIHTI). This work was supported by Secretaría de Ciencia, Humanidades, Tecnología e Innovación (SECIHTI, grant CBF2023-2024-834 to J. M.) and DGAPA-UNAM – Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica (PAPIIT, grant IA200125 to J. M.).

Poster 85. EXPA1 and EXPA2, cell wall α-expansins, are required for symbiotic infection and nodule development in *Lotus japonicus*

Elizabeth Monroy Morales[1], Ivette García Soto[1], Beatrice Lace[3], Mads Vestergaard[2], Jessica Segura[1], Catalina Bucarey Ruiz[1], Niels Sandal[2], Thomas Ott[3], Jens Stougaard[2] and Jesús Montiel[1]*

[1]Centro de Ciencias Genómicas, UNAM, Cuernavaca, México.
[2]Department of Molecular Biology and Genetics, Aarhus University, DK-8000 Aarhus C, Denmark.
[3]Cell Biology, Faculty of Biology, University of Freiburg, 79104 Freiburg, Germany.
*Email: jmontiel@ccg.unam.mx.

The establishment of symbiotic nitrogen fixation (SNF) in legumes begins with a molecular dialogue between rhizobia and the host plant, leading to bacterial entry through either intracellular infection threads or intercellular routes. In *Lotus japonicus*, both infection strategies occur: *Mesorhizobium loti* utilizes root-hair infection threads, whereas *Agrobacterium pusense* IRBG74 colonizes intercellularly. The successful progression of these processes depends on dynamic remodeling of plant cell walls. Expansin proteins are known to loosen cell wall structures through a non-enzymatic mechanism that relies on pH-induced wall relaxation, and they have been proposed to participate in facilitating rhizobial infection and nodule development. In this study, we explored the function of two cell wall α-expansins, EXPA1 and EXPA2, in the symbiotic interaction between *L. japonicus* and rhizobia. Despite exhibiting distinct expression patterns in different root tissues, both expansins were recruited to key cellular sites involved in rhizobial entry and nodule organogenesis. Notably, EXPA1 accumulated strongly in dividing cortical cells during nodule formation. Functional analyses of single mutants disrupted in either *EXPA1* or *EXPA2* revealed significant impairments in rhizobial colonization and nodule development under both infection pathways. Furthermore, the double mutant lacking both *EXPA1* and *EXPA2* displayed severely compromised bacterial invasion and dramatically reduced nodule numbers, indicating their essential, partially overlapping roles. Our findings indicate that EXPA1 and EXPA2 play key roles in facilitating both intracellular and intercellular rhizobial infection and subsequent nodulation in *L. japonicus*, suggesting that cell wall remodeling may be a critical component of the symbiotic process. This work was supported by the Mexican Secretaría de Ciencia, Humanidades, Tecnología e Innovación (SECIHTI, grant CBF2023-2024-834 to J. M.) and Dirección General de Asuntos del Personal Académico (DGAPA)-Universidad Nacional Autónoma de México (UNAM) – Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica (PAPIIT, grant IA200723 and IA200125 to J. M.).

Poster 86. Colonizaion of *Trichoderma atroviride* in *Lotus japonicus* roots and impact on root nodule symbiosis

Yordan J. Romero-Contreras, Samantha Segura Muñoz, Catalina Bucarey Ruiz, Ivette García-Soto, Aileen Peña Tenorio, and Jesús Montiel*

[1]Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México, Av. Universidad 2001, 62209, Cuernavaca, Morelos, México.
*Email: jmontiel@ccg.unam.mx

Plant–microbe interactions are fundamental for plant development and adaptation to abiotic conditions. The legume-rhizobia symbiosis constitutes a highly specialized association that enables the formation of root nodules, where biological nitrogen fixation occurs. In parallel, fungi of the genus *Trichoderma* are well known for their beneficial effects on plant growth and the activation of defense responses. However, limited information is available regarding the interaction within the *Trichoderma*–legume complex and its symbiotic relationship with rhizobia. This lack of information prompted us to analyze the interaction of *Trichoderma* with the model plant *Lotus japonicus*, as well as its impact on rhizobial colonization. We conducted dual and tripartite interactions involving *Mesorhizobium loti*, *Agrobacterium pusense* IRBG74, and *T. atroviride*. The progression of fungal colonization was monitored by confocal microscopy and molecular biology analyses. *Lotus* roots were colonized by *T. atroviride* within the first three days post-inoculation, accompanied by an increase in primary root length. However, the presence of *Trichoderma* notably reduced nodule formation compared to plants only inoculated with rhizobia. Moreover, fungal colonization by *T. atroviride* was diminished in the presence of rhizobia, which negatively affected plant development. Altogether, these results suggest a negative interference of *T. atroviride* on the legume–rhizobium symbiosis. Additionally, a transcriptomic analysis of the legume–fungus interaction is currently in progress, aiming to elucidate the molecular mechanisms underlying this interaction. Nevertheless, it remains essential to determine whether the co-inoculation of these microorganisms could provide benefits, such as pathogen suppression or other positive effects derived from microbial interactions. This work was supported by Secretaría de Ciencia, Humanidades, Tecnología e Innovación (SECIHTI, grant CBF2023-2024-834 to J. M.) and Dirección General de Asuntos del Personal Académico (DGAPA)-Universidad Nacional Autónoma de México (UNAM) – Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica (PAPIIT, grant IA200125 to J. M.).

Poster 87. **The role of volatiles emitted by the entomopathogenic fungus *Beauveria bassiana* in interaction with plants**

Adame-Garnica SG[1], Castro-Mercado E[1], Ríos-Chávez P[2], García-Pineda E[1], Valencia-Cantero E[1], Macías-Rodríguez L[1]*

[1]Instituto de Investigaciones Químico Biológicas, UMSNH, Ciudad Universitaria, General Francisco J. Múgica S/N, Morelia, Mich., México.
[2]Facultad de Biología, UMSNH. Ciudad Universitaria, General Francisco J. Múgica S/N Morelia, Mich., México.
*Email: lourdes.macias@umich.mx

Microbial volatile organic compounds (MVOCs) are key signaling compounds that mediate plant and soil microbe interactions (Ryu et al., 2004). MVOCs promote growth and trigger systemic resistance in plants. *Beauveria bassiana* is an entomopathogenic fungus that parasitize and kill insects. In addition, *B. bassiana* plays a role in nutrient cycling, plant growth promotion, and in controlling disease (Quesada-Moraga et al., 2023). Despite the multiple ecosystem services provided by *B. bassiana*, the role of volatiles emitted by the fungus on plants remains obscure. Thus, the objective of this study was to provide information about the effect of VOCs emitted by *B. bassiana* on the physiological and biochemical responses in plants. A system with separate compartments was used to assess the effect of fungal VOCs on the growth, reactive oxygen species (ROS) accumulation, and in the reinforcement of cell wall constituents in sorghum (*Sorghum bicolor*) seedlings. Moreover, quantification of indole-3-acetic acid (IAA), jasmonic acid (JA), salycilic acid (SA), and 4-coumaric acid were performed in leaves by gas chromatography-mass spectrometry; and total phenols were estimated by Folin-Ciocalteu reagent. Together, the results showed that peroxide anion and superoxide hydrogen play an important role during plant and fungus interaction, and the pool of VOCs and 3-methylbutanol, as the most abundant compound emitted by *B. bassiana*, stimulate the plant growth and phytohormones content in leaves. Furthermore, significant rises in the concentration of 4-coumaric acid and phenols were also observed, suggesting their role in reducing oxidative stress by scavenging ROS. In conclusion, VOCs emitted by *B. bassiana* stimulate various physiological and biochemical responses in plants, contributing to overall plant health.

Poster 88. Fungal endophytes associated to *Asclepias curassavica* influence the oviposition and foraging in the monarch butterfly (*Danaus plexippus*)

Bravo-Monroy DS[1], Hernández-Ornelas JE[2], García-Juárez PT[1], Ramírez-Ordorica A[1], Mendoza-Cuenca LF[2], Macías-Rodríguez L[1]*

[1]Instituto de Investigaciones Químico Biológicas, UMSNH, Ciudad Universitaria, General Francisco J. Múgica S/N, Morelia, Mich., México.
[2]Facultad de Biología, UMSNH. Ciudad Universitaria, General Francisco J. Múgica S/N Morelia, Mich., México.
*Email: lourdes.macias@umich.mx

Many insects exhibit high sensitivity to odors that signal the presence of mates, rivals, food sources, and suitable habitats. Notably, adult oviposition and larval feeding are critical phases in the insect life cycle, where plant chemistry significantly influences ecological outcomes. The coevolutionary relationship between monarch butterflies (*Danaus plexippus*) and milkweed plants (*Asclepias* spp.) offers a compelling case study. Plant secondary metabolites serve as important cues in host plant acceptance, guiding both oviposition decisions and chemical sequestration. However, the specific role of leaf-emitted volatiles in female oviposition choices remains insufficiently understood. Moreover, plants and insects maintain complex and intimate associations with microbial communities, which can modulate plant defenses and, consequently, impact insect physiology and behavior. This study aimed to elucidate the role of volatiles released by milkweed and its associated fungal endophytes on monarch oviposition and foraging behavior. Volatile compounds emitted by plants and fungi were analyzed using solid-phase microextraction coupled with gas chromatography-mass spectrometry (SPME-GC-MS). Results revealed that milkweed emits a diverse array of alcohols, aldehydes, ketones, and organic acids. Fungal endophytes isolated from the plants were identified as *Cladosporium silenes*, *C. halotolerans*, *C. cladosporioides*, and *Penicillium brocae*. Notably, *P. brocae* produced a rich blend of terpenes that strongly stimulated both egg-laying and larval herbivory. These findings suggest that certain microbial symbionts associated with milkweed release infochemicals that influence host plant selection by monarch females. This highlights the intricate interkingdom interactions among microbes, plants, and insects that shape ecological dynamics and behavioral outcomes.

Poster 89. Analysis of the *Lotus japonicus* microbiome depending on rhizobial colonization types: intracellular vs. intercellular

Fernando Morales Crisóstomo[1*], Eber Daniel Villa Rodríguez[2], Ivette García Soto[1] and Jesús Montiel González[1*]

[1]Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México, Av. Universidad 2001, 62210, Cuernavaca, Mor., México.
[2]Department of Molecular Biology and Genetics, Aarhus University, Denmark. DK-8000, Aarhus C, Denmark.
*Email: fernando_mc@ciencias.unam.mx; jmontiel@ccg.unam.mx.

The legume *Lotus japonicus* establishes symbiotic interactions with rhizobia through two distinct colonization modes: intracellular and intercellular. *Mesorhizobium loti* promotes intracellular colonization, while *Agrobacterium pusense* IRBG74 colonizes intercellularly. The plant microbiota, particularly in legumes, is influenced by factors such as root exudates and genetic signaling pathways related to nodulation. Mutations affecting these pathways can alter the microbiota, indicating a close relationship between nodulation and microbial communities. We hypothesize that the mode of rhizobial colonization affects the composition of the *L. japonicus* microbiota. We expect differences in microbial diversity and abundance within the rhizosphere, rhizoplane, and endosphere, depending on whether the colonization is intracellular or intercellular. To test this, we evaluated the microbiota of *L. japonicus* inoculated with three bacterial strains: *M. loti* R7A (intracellular), *A. pusense* IRBG74 (intercellular), and a mutant strain of *A. pusense* IRBG74-∆, which adopts an intracellular and intercellular colonization mode. Plants were grown in Magenta boxes containing perlite and desert soil from Sonora under four conditions: mock control (no inoculation), *M. loti* R7A, *A. pusense* IRBG74, and *A. pusense* IRBG74-∆. After five weeks plants were harvested for microbiano DNA extraction from the rhizosphere, rhizoplane, root endosphere and shoot endosphere. To confirm that the genomic DNA extracted from plant tissues contained amplifiable microbial DNA, several PCRs were performed using primers specific to the V5-V7 region of the 16S rRNA gene. Preliminary results showed that plants inoculated with intracellularly colonizing bacteria (*M. loti* R7A and *A. pusense* IRBG74-∆) exhibit a better nodulation efficiency and bigger shoot length compared to plants colonized intercellularly by *A. pusense* IRBG74. These findings provide insights into the complex interactions between rhizobia, host plants, and the microbial communities associated with legumes.

This project was funded by SECIHTI (CBF2023-2024-834 to J. M.) and DGAPA-UNAM (PAPIIT IA200723 y IA200125 to J. M.)

Poster 90. How can ozone affect pollination syndromes in strawberry plants?

Quintana-Rodríguez, Elizabeth[1*], Padilla-Rizo, Beatriz[1], Hernández-Cumplido Johnattan[2] and Orona-Tamayo, Domancar[1]

[1]CIATEC, León, Guanajuato, Omega #201 Industrial Delta, 37545.
[2]Facultad de Ciencias, UNAM, Coyoacán, CDMX, Coyoacán, Investigación Científica, CU, 04510.
*Email: equintana@ciatec.mx.

Ozone is one of the main pollutants that causes severe damage to health and the ecosystem. Floral characteristics, such as morphology, color, scents, are named pollination syndromes and have evolved to attract specific types of animal pollinators. These syndromes help plants maximize their reproductive success by ensuring efficient pollen transfer through specialized relationships with pollinators. Several studies have shown that ozone can alter pollination syndromes, such as the aromas emitted by flowers, resulting in pollinators not easily locating them. However, the effect on the nectar produced and other characteristics such as the polyphenols present in the flower has not been evaluated. The industrial corridor of Guanajuato is both highly polluted by industrial activities and highly agricultural, making it a good study model for analyzing the effects of ozone on pollination syndromes. Irapuato is one of the main strawberry producers in the country and the plant depends on insect pollination services. To evaluate the effects of ozone, strawberry plants were exposed to ozone concentrations recorded in the Guanajuato industrial corridor. In the strawberry plants exposed to ozone, the aromas emitted by flowers were analyzed, and the number of visits made by pollinators was assessed. In addition, the effect of ozone on the quantity of nectar produced by flowers was analyzed, and soluble solids and reducing sugars were quantified. It was observed that ozone interferes with plant-pollinator interactions, with an 82% decrease in visits found in plants treated with this pollutant. Analysis of the aromas emitted by flowers showed differential changes in plants exposed to ozone that could explain the decrease in visits. After 24 hours of ozone exposure in strawberry plants, oxidation of the plant's leaves was observed. Within the quantification of soluble solids and reducing sugars in nectar and fruit, a significant decrease in these components was found. Ozone negatively affects pollination services by altering the aromas emitted and, consequently, pollinator visits. Furthermore, it negatively affects the concentration of sugars in nectar and, ultimately, the quality of the fruits produced.

Poster 91. **Comparative analysis of the spatio-temporal activity of *MtNPL* and *PvNPL800* promoters in *Phaseolus vulgaris* transgenic roots infected with *Rhizobium etli***

Mariana Robledo-Gamboa, Libni Anajared Castrejón Noguerón, José Erik Cruz-Valderrama and Rosana Sánchez-López*

Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Campus Morelos, Avenida Universidad 2001, Colonia Chamilpa, Cuernavaca 62210, Mexico.
*Email: rosana.sanchez@ibt.unam.mx

Most members of the Fabaceae family (legumes) have developed the ability to establish a symbiotic relationship with nitrogen-fixing bacteria, known as rhizobia. This interaction involves well-coordinated processes in the root, such as rhizobial-infection, cell division and nodule organogenesis, all of them depend on cell wall remodeling events. Enzymes such as the nodule pectate lyase (NPL) are involved in those activities[1.] Epidermal infection is mediated by a de novo transcellular structure, the infection thread (IT), which expands from the apical region of a root hair-bearing a rhizobial microcolony, towards the underlying cortical region, whose cells have in turn restarted cell division.[2]. Our research group is interested in the functional role of NPL in the dynamics between the IT formation/elongation and the initial cell divisions, which in *P. vulgaris* roots occur in the subepidermal (S-E) cells underlying the infection site[2]. Herein, we report the identification of the putative orthologue of *MtNPL* gene in *P. vulgaris, PvNPL800*. We also describe a comparative analysis of the spatio-temporal activity of both *MtNPL* and *PvNPL800* promoters in uninoculated and rhizobia-inoculated roots. We also present a preliminary analysis of the cellular distribution of *Mt*NPL-GFP and *Pv*NPL800-GFP in *P. vulgaris* roots infected with *R. etli*. The emphasis of this study is to provide insights into the timing of nodule pectate lyase expression, during IT elongation along the root hair, its proximity to root hair basal plasma membrane, and the cell wall remodeling in dividing S-E cells to be invaded by the IT. [1] Su C. *et al*., (2023). Transcellular progression of infection threads in Medicago truncatula roots is associated with locally confined cell wall modifications. Curr Biol. 33(3):533-542.e5.[2] Monroy-Morales E. *et al*., (2022). Visualization of the crossroads between a nascent infection thread and the first cell division event in Phaseolus vulgaris nodulation. Int J Mol Sci. 23(9):5267 This proyect was partially supported by Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica/UNAM (PAPIIT/UNAM to R.S.L.) and a M.Sc fellowship (application no. 67f5cda85a8f4e53dbda1437 by Secretaria de Ciencia, Humanidades Tecnología e Innovación, SECIHTI) to M.R.G. and a postdoctoral fellowship by Programa de Becas Posdoctorales UNAM 2023-2025 to J.E.C.V. This work is based primarily on Proyecto de Maestría en Ciencias Biológicas-UNAM by Mariana Robledo Gamboa.

Poster 92. The common bean mutant (*Phaseolus vulgaris*) non-nodulating mutant: *nnod*(2114) is affected in the infection thread development during symbiosis with rhizobia

Rocio Reyero-Saavedra, Gladys E. Jimenez-Nopala, Karina San Vicente-Trujillo, Mario Ramirez, Yañez, Sara Isabel Fuentes Membreño, Alfonso Leija Salas, Litzy Ayra, Pablo Pelaéz Hernandez, Lourdes Girard Cuesy, José Erick Cruz Valderrama, Rosana Sánchez López, Georgina Hernández Delgado*

Centro de Ciencias Genómicas, UNAM. Avenida Universidad s/n, col. Chamilpa 62210. Cuernavaca, Morelos.
*Email: gina@ccg.unam.mx.

In the legume-rhizobia N2-fixing symbiosis, rhizobia infect the legume roots through the root hairs, forming a structure called "infection thread (IT)". The IT guides rhizobia from root hair tip to the root cortex cells, where they are released into nodules —the nitrogen-fixing organs. Over the past 20 years, advances in legume genetics and genomics, specially through mutant characterization in model legumes like Medicago and Lotus have significantly improved our understanding of the genes involved in this process. In contrast, few studies have focused on agriculturally important legumes such as common bean (*Phaseolus vulgaris*). Only one common bean N2-symbiotic mutant has been genetically and molecularly characterized. Our group has screened for common bean non-nodulating (*nnod*) mutants, from an EMS mutant population derived from the BAT93 (WT) genotype. One of *nnod* mutants selected that is being characterized is the *nnod*(2114) mutant line, which presents defects in the IT development. In this mutant, after rhizobia inoculation the ITs are formed but their development is blocked and these are aborted and do not reach the root cortex cells, except in very few occasions that result in the formation of small, non-infected, pseudo-nodules. Comparative WT vs. mutant whole genome sequence analysis, revealed a "high impact variant" in the *LIN/CERBERUS* gene from the *nnod*(2114) mutant, that presents a premature stop codon in its coding sequence. This gene codes for a Ubiquitin E3 ligase important for guiding the development of the IT during the N2-fixing symbiosis. In order to confirm that the *PvLIN* gene is responsible for the *nnod* (2114) mutant phenotype, we are performing genetic complementation experiments using the *Agrobaterium rhizogenes* genetic transformation to generate transgenic common bean roots/nodules [4]. In addition, using this system we have observed that WT plants with silenced expression of *PvLIN* in transgenic roots/nodules showed a similar altered phenotype as the *nnod*(2114) mutant. Also, translational fusions of the *PvLIN* with marker genes coding sequences are being carried out to analyze the PvLIN peptide synthesized by the *nnod*(2114) mutant as compared to that from the WT.

Poster 93. Deciphering host recognition and invasion mechanisms in the mistletoe–tree interaction

Domancar Orona-Tamayo[1*] and Elizabeth Quintana-Rodriguez[1]

[1]CIATEC. Omega # 201 Col. Industrial Delta C.P. 37545 León, Guanajuato, México. Email: *dorona@ciatec.mx.

Psittacanthus calyculatus is a hemiparasitic mistletoe present in most of the natural ecosystems in Mexico, infecting trees of ecological and commercial interest. In central Mexico, nearly 65% of the woodland is affected by this mistletoe infection, endangering endemic specimens such as mesquite trees (*Prosopis laevigata*). Successful infection of *P. calyculatus* mistletoe in a host tree depends on different mechanisms for the establishment; such as, germination conditions, growth seedlings, metabolic signatures in haustorium development, and enzymes involved in bark layer degradation and xylem connection. Information about mechanisms to establish the parasitism between mistletoe in a host tree is scarce. This work aimed to study the molecular dialogue between *P. calyculatus* mistletoe and the mesquite host tree in the initial infection stages. We analyzed the biomolecules involved in haustorium development, and enzymes related to the host bark degradation, and proteomic profiles. Additionally, we analyzed the damage perception in the host, with the study of early immune responses in the host branches adding extracellular DNA (exDNA; from the host and mistletoe) to emulate cell rupture by parasite penetration. The proteomic analyses revealed proteins with different accumulation and cellular processes in infective stages. Activities of the cell wall-degrading enzymes such as cellulase and β-1,4-glucosidase were primarily active in haustorium development. At the same time, xylanase, endo-glucanase, and peptidase were highly active in the haustorium penetration and xylem connection. Patterns of auxins and cytokinin showed spatial concentrations in infective stages and were involved in haustorium development. On the other side, we determined that mesquite trees perceive self-exDNA (mesquite) and non-self exDNA (mistletoe) with the synthesis of $O_2^{\bullet-}$, H_2O_2, flavonoids, ROS-enzymes system, MAPKs activation, spatial concentrations of JA, SA, ABA, and CKs, and auxins. Our data indicate that self and non-self exDNA application differs in oxidative burst, JA signaling, MAPK gene expression, and scavenger systems[3]. These results are the first evidence of proteins, cell wall-degrading enzymes, phytohormones, and the host perception mediated by exDNA are involved in early infection for the *Psittacanthus* genus.

Poster 94. Functional roles of pectin methyl esterase isoforms in intra- and intercellular symbiotic infection of *Lotus japonicus*

Catalina Bucarey Ruiz, Carmen Julia Zapien Oliva, Elizabeth Monroy Morales and Jesús Montiel González*

Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México. Av. Universidad 2001, 62210, Cuernavaca, Morelos.
*Email: jmontiel@ccg.unam.mx

Lotus japonicus is a legume capable of forming nitrogen-fixing symbiotic relationship with rhizobia. Depending on the microbial partner, root infection can occur via two mechanisms: intracellularly by *Mesorhizobium loti* or intercellularly by *Agrobacterium pusense* IRBG74. Both pathways require cell wall remodeling. In this study, we investigated the role of pectin methylesterases (PMEs), a family of cell wall-modifying enzymes during the symbiotic interaction between *L. japonicus* and rhizobia. A total of 69 PME genes were identified in the *L. japonicus* genome. Based on phylogenetic analysis and expression profiling, four candidate genes, *Lj*PME1–*Lj*PME4, were selected for characterization. Transcriptomic data revealed *Lj*PME2 and *Lj*PME4 are highly expressed in root hairs during early infection by rhizobia, while *Lj*PME1 and *Lj*PME3 show increased expression under both intracellular (*M. loti*) and intercellular (*A. pusense* IRBG74) colonization. To assess their roles in the symbiotic process, we identified and genotyped insertional mutants from the *LORE1* collection. Homozygous lines were obtained for three candidate genes. Phenotypic analysis revealed reduced nodule number in *Ljpme2-1* and *Ljpme3-1* mutants compared to the Gifu wild-type, suggesting a key role for these *PMEs* in nodulation. To investigate gene expression dynamics and protein localization, transcriptional and translational fusions to molecular markers were introduced into hairy roots. Confocal microscopy showed *Lj*PME2 promoter activity in root hairs and epidermal cells, consistent with a role in symbiotic signaling and infection. These results indicate that specific PME isoforms contribute to cell wall modification essential for rhizobial infection and are critical for nodule development in *Lotus japonicus*. These findings enhance the understanding of molecular mechanisms underpinning legume–rhizobia symbiosis. This work was supported by Secretaría de Ciencia, Humanidades, Tecnología e Innovación (SECIHTI, grant CBF2023-2024-834 to J. M.) and DGAPA-UNAM – Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica (PAPIIT, grant IA200125 to J. M.).

Poster 95. **Plant growth promoting bacteria effect on germination and development of Lisianthus seedlings (*Eustoma grandiflorum,* Gentianaceae)**

Jeanette Guadalupe Cárdenas-Valdovinos[1], Silvia Luna-Suárez[1], Hortencia Gabriela Mena-Violante[2*]

[1]Centro de Investigación en Biotecnología Aplicada, Instituto Politécnico Nacional (CIBA-IPN), Ex-Hacienda San Juan Molino, Carretera Estatal Tecuexcomac–Tepetitla Km 1.5, C.P. 90700, Tepetitla de Lardizábal, Tlaxcala, México.
[2]Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional, Instituto Politécnico Nacional, (CIIDIR-IPN Unidad Michoacán) Justo Sierra Ote. No. 28, Centro, C.P. 59510, Jiquilpan de Juárez, Michoacán, México.
*Email: hmena@ipn.mx

Eustoma grandiflorum (Raf.) Shinn. (Gentianaceae), better known as Lisianthus, is an ornamental plant native to northern Mexico, which stands out for the diversity of colors in its flowers and its long duration in vase, which makes it ideal as a cutting variety. The cultivation of this species is strongly influenced by environmental factors such as radiation and temperature and is vulnerable to attack by various pests and diseases, which can delay its growth and flowering. For this reason, the need to implement biotechnological approaches aimed at optimizing the process of Lisianthus propagation has been seen. Plant Growth Promoting Bacteria (BPCV) have been used as bioinoculants to improve plant growth; in this sense, the plant growth-promoting efficiency of rhizospheric and endophytic bacteria isolated from *Swertia ciliate* (Gentianaceae) has been reported whose potential as indol-producers, nitrogen fixers and phosphate solubilizers allow their viable application for sustainable cultivation systems. The objective of this study was to evaluate the effect of rhizospheric native isolates of *Gentiana biscupidata* (GBE1 and GBS9) on the germination and development of Lisianthus seedlings. The experiment was carried out in a growth room with a totally randomized design with 5 treatments: control (water), *Bacillus subtilis (*BSS), GBE1, GBS9, and GBE1+GBS9 consortium. The experiment was maintained for 3 months and biometric variables such as height, number of leaves, leaf area, root length and total fresh and dry biomass were measured. The results showed that GBE1 isolate and GBE1+GBS9 consortium promoted early germination and seedling survival. This consortium promoted most of the biometric variables, evidencing a synergistic effect and confirming the positive effect of inoculation with native BPCV on Lisianthus development.

Poster 96. MeJA triggers *Capsicum annuum* response against *Phytophthora capsici* through defense-related genes activation in early stages of infection

Aarón Barraza[1], Antonio Pérez-Aranda[2], M. Goretty Caamal-Chan[1*]

[1]Secihti-Centro de Investigaciones Biológicas del Noroeste, SC. Instituto Politécnico Nacional 195, Playa Palo de Santa Rita Sur, La Paz, B.C.S., C.P. 23096, Mexico.
[2]Centro de Investigaciones Biológicas del Noroeste, SC. Instituto Politécnico Nacional 195, Playa Palo de Santa Rita Sur, La Paz, B.C.S., C.P. 23096, Mexico.
*Email: mcaamal@cibnor.mx

Phytophthora capsici causes root and stem rot disease in *Capsicum*. The use of elicitors to trigger defense responses to biotic stresses to characterize molecular plant defense mechanisms is a regular approach. This work assessed the elicitation role in the defense-response activation in *Capsicum*. We followed a foliar SA, MeJA, or BABA challenge of *P. capsici*. The infection response throughout time during disease development in seedlings treated with elicitors was assessed by phenotypic analyses. Moreover, the activation of jasmonate/ethylene (JA/ET) and salicylic acid (SA) defense-response signaling pathways were analyzed by qPCR. Furthermore, a transcriptomic analysis was performed with the MeJA treatment, which also conferred an increase in tolerance in our experimental design matrix. The RNA-seq, coupled with transcriptomic analyses, strongly supports the notion that MeJA triggers a rapid response to biotic stress, resulting in reduced disease symptoms. Moreover, MeJA treatments challenged with *P. capsici* increase the expression level of *LOX2*, *PDF1*, and *ETR1* genes. The transcriptomic analysis showed that MeJA upregulates genes associated with cell wall modifications and ethylene biosynthesis in local infection. All together strongly suggests that in the early infection stage, the JA-dependent signaling triggers defense mechanisms against *P. capsici*, supporting the fact that early induction and high-intensity defense modulated by the right signaling pathway can induce a specific and direct defense response.

Poster 97. Identification and classification of pathogens effectors in agronomical relevant crops using meta-transcriptomics, proteome structural data, and artificial intelligence

J. Noé García-Chávez[1], José Saúl Villa Pérez[1], Yemeen Ayub[2], Jimena Jazmin Hurtado Olvera[1], Guillermo Rodríguez-Guerrero[1], Paola I. Carrera-Gutierrez[1], Jérôme Jean Verleyen[1,3], Nelly Selem-Mojica[4], Antonio Hernandez-López[1,5], Carlos Yebra-Montes[1], Elizabeth Munch[2], Dan Chitwood[2], and Alejandra Rougon-Cardoso*[1,5].

[1]Laboratory of Agrigenomic Sciences, Universidad Nacional Autónoma de México, ENES- León, 37689-León, Mexico, [2]Michigan State University, East Lansing MI, USA [3] Biotechnology Research Institute, UNAM, 62210-Cuernavaca, Mexico. [4]Centro de Ciencias Matemáticas, UNAM. [5]PlanTECC National Laboratory, ENES-León, 37689-León,
*Email: arougon@enes.unam.mx

Phytopathogenic nematodes cause annual losses of hundreds of billions of USD in agronomic crops. The plant-pathogen interaction involves a two-way communication in which the plant recognizes the pathogen and mounts a defense response, while the pathogen manipulates plant's biology to create an environment for its own growth and reproduction. Pathogens can secrete molecules known as effectors, which alter host cell functions. Although some families of effectors are well-known, a group of "pioneer" effectors has not been successfully classified using traditional methods *e.g.* protein similarity or functional domain identification, highlighting the need for new methods. Furthermore, many more effectors remain to be discovered. Therefore, this project aims to improve the methodology of searching and classifying proteins related to parasitism by integrating transcriptomic and protein structural data with artificial intelligence, allowing for more precise and comprehensive identification. To achieve this, a meta-analysis of public RNA-seq data of infected plants and pathogen's predicted protein structures is used to train AI models to identify and classify novel effectors. The results will be strengthened using comparative genomics analyses. The objectives of this study are to: i) identify pathogen-specific transcriptional signatures in plants undergoing infection; ii) identify specific transcriptional signatures in plants in response to different families of effectors and pioneer effectors; iii) correlate gene expression profiles of phytopathogenic nematodes with plant's transcriptomic signatures to identify and classify new effectors; iv) prioritize effector candidates for experimental validation using transcriptomic, structural, and comparative genomic data. This work was supported by DGAPA-UNAM postdoctoral fellowship granted to JNGC, UNAM-PAPIIT IN223024, and *Research Projects in the Cloud* UNAM-AWS.

Poster 98. Microtubule reorganization in subepidermal cells correlates with early stages of infection thread formation in *Phaseolus vulgaris* root hairs

José Erik Cruz-Valderrama., Wendy M. Escalante-Ruiz, F. Catalina A. Bucarey-Ruiz and Rosana Sánchez-López*

Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Campus Morelos, Avenida Universidad 2001, Colonia Chamilpa, Cuernavaca 62210, México.
*Email: rosana.sanchez@ibt.unam.mx.

The legume plants can establish a finely tuned symbiotic relationship with soil bacteria, called rhizobia. This association leads to the *de novo* formation of a root organ named nitrogen-fixing nodule. Once a rhizobial microcolony is formed in the infection chamber of a responsive-root hair, a nascent infection thread (IT) can be observed. Simultaneously, the cortical cells beneath the infection site become actively dividing to form a nodule primordium which further derives in a mature nitrogen-fixing nodule. *Phaseolus vulgaris* displays initial cell divisions in the subepidermal (S-E) cell layer immediately underneath the rhizobial infection site. Previously, we described that at early stages of the IT formation, the nuclei of those S-E cells tend to have a central position, indicating a cell cycle activation. Thus, the dynamics between the early steps of infection and the pre-mitotic phases in S-E cells is a relevant feature. In this research, we used a YFP-tagged tubulin A (AtTUA6), as a molecular tool to describe the cortical microtubule (MT) organization in the S-E cells underlying an infection site in *P. vulgaris* transgenic roots, and assessed the angle of arrangement of MT fibers and the parallelnes between them, two indicators that can be used to evaluate changes in the MT arrays in S-E cells at different stages of the root hair infection. Our results show that in S-E cells underneath a root hair forming a microcolony, YFP-AtTUA6-labelled MT show a typical cortical arrangement of fibers transverse to the long axis of the root, with an orientation oblique angle similar to that observed in equivalent S-E cells from uninoculated roots. Whereas in S-E cells underlying root hairs carrying an emerging or elongated IT, the cortical MT organization is no longer observed, instead it tends to have fewer transverse MTs, YFP-AtTUA6-labelled fibrils are apparently fragmented and appeared to be disorganized, suggesting MT rearrangements, as it has been described at early stages of cell cycle progression. This project was partially supported by Programa de Apoyo a Proyectos de Investigación e Innovación Tecnológica/UNAM (PAPIIT/UNAM) IN204721 and IN218524 to R.S.L. and a postdoctoral fellowship by Programa de Becas Posdoctorales UNAM 2023-2025 to J.E.C.V.

Poster 99. **How ozone can affect pollination syndromes in strawberry plants?**

Quintana-Rodríguez, Elizabeth[1]*, Padilla-Rizo, Beatriz[1], Hernández-Cumplido Johnattan[2] and Orona-Tamayo, Domancar[1]

[1]CIATEC, León, Guanajuato, Omega #201 Industrial Delta, 37545.
[2]Facultad de Ciencias, UNAM, Coyoacán, CDMX, Coyoacán, Investigación científica, CU, 04510.
*Email: equintana@ciatec.mx

Ozone is one of the main pollutants that causes severe damage to health and the ecosystem. Floral characteristics, such as morphology, color, scents, are named pollination syndromes and have evolved to attract specific types of animal pollinators. These syndromes help plants maximize their reproductive success by ensuring efficient pollen transfer through specialized relationships with pollinators. Several studies have shown that ozone can alter pollination syndromes, such as the aromas emitted by flowers, resulting in pollinators not easily locating them. However, the effect on the nectar produced and other characteristics such as the polyphenols present in the flower has not been evaluated. The industrial corridor of Guanajuato is both highly polluted by industrial activities and highly agricultural, making it a good study model for analyzing the effects of ozone on pollination syndromes. Irapuato is one of the main strawberry producers in the country and the plant depends on insect pollination services. To evaluate the effects of ozone, strawberry plants were exposed to ozone concentrations recorded in the Guanajuato industrial corridor. In the strawberry plants exposed to ozone, the aromas emitted by flowers were analyzed, and the number of visits made by pollinators was assessed. In addition, the effect of ozone on the quantity of nectar produced by flowers was analyzed, and soluble solids and reducing sugars were quantified. It was observed that ozone interferes with plant-pollinator interactions, with an 82% decrease in visits found in plants treated with this pollutant. Analysis of the aromas emitted by flowers showed differential changes in plants exposed to ozone that could explain the decrease in visits. After 24 hours of ozone exposure in strawberry plants, oxidation of the plant's leaves was observed. Within the quantification of soluble solids and reducing sugars in nectar and fruit, a significant decrease in these components was found. Ozone negatively affects pollination services by altering the aromas emitted and, consequently, pollinator visits. Furthermore, it negatively affects the concentration of sugars in nectar and, ultimately, the quality of the fruits produced.

Poster 100. Role of flotillin from *Phaseolus vulgaris* during a mutualistic interaction with *Rhizobium tropici*

Pamela Jiménez-Chávez[1], Ana I. Chávez-Martínez[1], Janet Palacios-Martínez[1], Fernando Lara-Rojas[2], Ana Yasmín Martínez-Ocampo[3] and Luis Cárdenas[1*]

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, México, [2]Departamento de Interacciones Planta-Insecto, Centro de Desarrollo de Productos Bióticos Instituto Politécnico Nacional, Yautepec, Morelos, México.
[3]Facultad de Ciencias, Universidad Autonóma de Morelos, Cuernavaca, Morelos, México.
*Email: luis.cardenas@ibt.unam.mx

The symbiotic relationship between bacteria of genus Rhizobium and the plant of common bean (*Phaseolus vulgaris*), is one of the most common successful plant-microbe interactions. The bacteria are internalized in the root through specialized cells such as the root hair; to do so, they induce the invagination of the plasma membrane and cell wall of root hairs resulting in the formation of a structure called infection thread and subsequently penetrate the root cortex to develop a new organ known as nodule, where the bacteria will be release and internalized, so they can differentiate into bacteroides and be able to carry out the nitrogen fixation. It was shown that membrane microdomains are important for membrane shaping, trafficking and signal transduction. Flotillins are lipid microdomains components in both vegetal and animal cells and in plants were shown to play important role in plant-microbe interaction. In this study, thanks of bioinformatic analysis, we identified one flotillin gene in *P. vulgaris* genome, also we identified flotillin genes in different legumes. Later, we conducted transcriptional profiling in common bean roots to determine the specific expression patterns of flotillin during nodulation. Also, we determinate the subcellular localization of flotillin during the interaction with rhizobia. In addition to, we analysed the promoter activity of flotillin during different nodulation stages. Our results reveal that flotillin is expressed in the early stages of nodulation and throughout the development of the nodule. Subcellular localization indicates that flotillin have a role in vesicular trafficking and root hair polar growth.

Poster 101. Plant-microorganism interactions under simulated microgravity conditions in *Arabidopsis thaliana*

Luz Alejandra López Rodríguez, Jean Philippe Vielle Calzada*

The Advanced Genomics Unit (UGA-Langebio).
*Email: vielle@cinvestav.mx

Arabidopsis thaliana is a relevant model to investigate how microorganism-plant interactions are influenced by simulated microgravity. Plants rely on their microbiota in distinct zones (rhizosphere, phyllosphere, and endosphere), where these interactions are critical for growth. Under normal gravity conditions, these relationships are classified as mutualistic, antagonistic, or neutral, modulated by signaling pathways activated by biotic and abiotic elicitors that regulate plant development. On Earth, microgravity can be simulated using clinostats. Continuous rotation disrupts gravitational perception in plants by preventing statolith sedimentation, thereby interfering with auxin-dependent signaling pathways. This study aims to evaluate how simulated microgravity alters the rhizosphere microbial community of *Arabidopsis* and how these changes affect its growth and development. We will expose seedlings to simulated microgravity and inoculate them with a synthetic community of bacteria and filamentous fungi. After 15 days, metagenomic sequencing will compare the microbial composition to that of seedlings under normal gravity. Our results will provide key insights into plant-microbiome dynamics in space-like conditions. Understanding these interactions is essential for future applications, such as efficient plant cultivation during long-duration space missions, enabling sustainable food production and astronaut well-being.

Poster 102. **Comparative study of volatile organic compounds from two *Candida* species and their interaction with *Arabidopsis thaliana***

Francisco Jerónimo García[1], Patricia Romero Arellano[1], Edgar Guevara Avendaño[1], David Camarena Pozos[2], Randy Ortiz Castro[1]*

[1]Instituto de Ecología, A.C., Red de Estudios Moleculares Avanzados, Xalapa, Veracruz, México.
[2]Dirección de Investigación y Soluciones Tecnológicas, CIATEC A.C., León, Guanajuato, México.
*Email: randy.ortiz@inecol.mx

Plant–microorganism interactions represent a sustainable alternative for agriculture through the use of plant growth-promoting microorganisms (PGPMs). Although volatile organic compounds (VOCs) have been extensively studied in bacteria and filamentous fungi, yeasts particularly those of the *Candida* genus have received little attention as sources of bioactive VOCs, and their role in plant interactions remains largely unexplored. In this study, we evaluated the biological activity of volatiles emitted by *Candida tropicalis* and *Candida berthetti*. Both strains were morphologically characterized by scanning electron microscopy and growth curves at different optical densities; their ability to produce indole-3-acetic acid (IAA) was also confirmed. The plant–microorganism interaction was assessed using *Arabidopsis thaliana* under conditions of direct contact and VOC exposure in compartmentalized systems. *C. berthetti* promoted plant growth under both conditions, whereas *C. tropicalis* showed phytopathogenic activity in direct contact but acted as a growth promoter when plants were exposed to its volatiles. Analysis using the *A. thaliana DR5:GUS* reporter line indicated that *C. berthetti* increased auxin levels in both conditions, while *C. tropicalis* reduced auxin levels in direct contact but stimulated them via VOC exposure. VOC profiles were analyzed by gas chromatography–mass spectrometry (GC–MS), revealing differential profiles and compounds potentially associated with the growth-promoting activity. These findings highlight the potential of certain yeasts as sources of bioactive volatile metabolites capable of modulating plant growth through hormonal signaling pathways, offering promising biotechnological tools for more sustainable agricultural practices.

Poster 103. Alkaloids as part of a specific response to biotic stress in *Argemone mexicana*

Blanca Catalina Solís Arias, José Armando Muñoz-Sánchez, Felipe Vázquez Flota*

Centro de Investigación Científica de Yucatán, A.C. Calle 43 No. 130 x 32 y 34. Col. Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México.
*Email: felipe@cicy.mx

Plants, like other living organisms respond to environmental stimuli, whether abiotic, such as water stress, or biotic, including damage by pathogens or insects. Such stimuli could trigger a series of signaling events leading to a molecular response, either in a generalized or specific way, aimed to adapt to the new condition. Alkaloids are metabolites synthesized from amino acids that could provide a chemical defense to plants, given their toxic effects on insects and microorganisms. Among the alkaloids produced from tyrosine, the benzylisoquinoline alkaloids (BIAs) are particularly notable. The genus *Argemone* (family Papaveraceae) has the capacity to synthesize (BIA). *Argemone mexicana*, known as cardo santo or chicalote, produces several of these alkaloids, including berberine and sanguinarine, being the most representative[2]. Both alkaloids are distributed throughout the plant in different tissues. Berberine accumulates in aerial and underground tissues, although it can also be detected in immature seeds, whereas sanguinarine is detected only in roots and mature seeds, under regular conditions. However, this accumulation is altered in interaction with fungal microorganisms. Noteworthy, fungal infection induces the presence of sanguinarine in leaves. Using this model, we would explore the role of alkaloids in plant-microbe interactions. The possible involvement of alkaloids has been previously observed as the above mentioned accumulation of sanguinarine in leaf tissues with fungal lesions. However, the molecular part regarding the transcriptional activity of the alkaloid biosynthetic genes in this condition is unknown. An approach to analyze these changes is the use of RNA NGS. In this study, we analyze transcriptomic changes in *Argemone mexicana* in response to fungal stress, with a focus on the expression of genes involved in alkaloid biosynthesis. Supported by SECIHTI, CBF-2023-2024-1879

Poster 104. ***PvPLDα2A* acts as a negative regulator of rhizobial symbiosis in common bean (*Phaseolus vulgaris* L.)**

Ronal Pacheco, Noreide Nava, Ana Chávez-Martínez, Janet Palacios-Martínez, Luis Cárdenas and Carmen Quinto*

Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Avenida Universidad 2001, Colonia Chamilpa 62210, Cuernavaca, Morelos, México.
*Email: carmen.quinto@ibt.unam.mx

The legume-rhizobia symbiosis induces the formation of new root organs called nodules, where biological nitrogen fixation occurs. Physiological studies have demonstrated the participation of phospholipases D (PLD) in the establishment of this symbiotic interaction. PLDs are phospholipid-degrading enzymes involved in various plant processes, such as defense responses to bacterial and fungal pathogens. However, their functional characterization in the legume-rhizobium symbiosis has been little explored. In this study, the functional characterization of the common bean PLD, *PvPLDα2A* in the symbiotic interaction between *Phaseolus vulgaris* and the rhizobacteria *Rhizobium tropici* was carried out. An *in silico* analysis showed that the common bean genome encodes 22 PLDs and that *PvPLDα2A* transcription is induced by rhizobia in roots and nodules, which was confirmed by qPCR analysis. Downregulation of *PvPLDα2A* by RNAi enhanced rhizobial infection and increased the number of nodules on bean roots. On the other hand, overexpression of this gene reduced the number of nodules formed. These data together suggest that PvPLDα2A acts as a negative regulator of legume-rhizobial symbiosis, which is consistent with a recent study in soybean (2). Our findings confirm the importance of PLDs and lipid metabolism in the regulation of the legume-rhizobium symbiosis and contribute to the understanding of the molecular mechanisms underlying this process. Acknowledgments: we are grateful for the support of UNAM-PAPIIT (IN204024 to CQ) and by Secretaria de Ciencia, Humanidades, Tecnología e Innovación SECIHTI CF-2023-I-297, (to CQ) and the 749422 scholarship (to RPS).

Poster 105. Identification of epigenetic markers associated with the activation of pathogen defense pathways in soybeans

Selene Yazbeth Soto Acosta, Berenice Calderón Pérez, Roberto Ruiz Medrano, Beatriz Xoconostle Cázares*

Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Av. Instituto Politécnico Nacional 2508, San Pedro Zacatenco, 07360 Ciudad de México, CDMX.
*Email: bxoconos@cinvestav.mx

Soybean (Glycine max) is a strategic crop worldwide due to its high protein and lipid content, as well as its broad agro-industrial versatility. Under biotic stress conditions, plants activate defense mechanisms mediated by the induction of the salicylic acid (SA) and methyl jasmonate (MeJA) signaling pathways. Studies in model plants have demonstrated that epigenetic mechanisms—including DNA methylation, histone modifications, and the action of microRNAs—can function as transgenerational memory of stress responses. The aim of this study is to identify epigenetic markers associated with the activation of defense pathways in soybean plants subjected to stress conditions induced by SA and MeJA. For this purpose, an experimental model was established based on foliar application of these elicitors, followed by the agronomic evaluation of morphological and phenotypic changes. For epigenomic analyses, the ATAC-seq technique (Assay for Transposase-Accessible Chromatin with high-throughput sequencing) will be employed on nuclear DNA isolated from soybean leaves. This methodology will allow mapping of chromatin-accessible regions, identification of active regulatory sites, and, ultimately, the genes potentially involved in the biotic stress response. Preliminary results show morphological alterations in treated plants, including reduced stem length, variations in the number of trifoliate leaves, and modifications in floral structure development. This study aims to contribute to the understanding of the epigenetic mechanisms regulating the adaptive response of plants to biotic stress, with potential applications in breeding programs aimed at enhancing crop resilience.

Poster 106. **Root-root interactions and their Impact on root system architecture**

William LaVoy[1], Suxing Liu[1], Kabir Hossain[1], Joshua Carpenter[2], Alexander Bucksch[1*]

[1]School of Plant Sciences, University of Arizona, Tucson, AZ 85721
[2]Department of Civil Engineering, University of Akron, Akron, OH 44325
*Email: bucksch@arizona.edu

Root system architecture (RSA) directly impacts the belowground nutrient profile that plants have access to. RSA trait development is governed by both genetic and environmental factors, influencing foraging strategies and plant performance. Emerging evidence suggests that root-root interactions between neighboring plants drive significant architectural changes yet are understudied. Investigative efforts in this field are hindered by inherent difficulties of root studies, leading to a reliance on experimentation with young plants in unrealistic growth conditions. Extending our knowledge requires research focused on mature plants in realistic conditions. To bridge the gap in our understanding, we are developing an innovative experimental platform, with a specialized growth system and analytical software to quantify the effects of root-root interactions between neighbors on the RSA in common bean (*Phaseolus vulgaris*). Our growth system utilizes a mesh-layered frame that is inserted into a large mesocosm, allowing for growth and maturation of two plants side by side. Harvesting occurs at early reproductive stages, when roots are mature, by disassembling the mesocosm and gently washing away soil, resulting in root structures supported by mesh in their natural orientation. Roots are then imaged and fed into DIRT/3D, a root image reconstruction software, generating a high-fidelity 3D point cloud of the root systems. To extract a deeper understanding, we are developing novel RSA traits that leverage spatial data to quantify localized and systemic root responses to neighboring plants. Initial results contradict long-held assumptions of competition driven root-root interactions. Instead, we observe majority of soil exploration and root growth occurs away from neighbors, avoiding competition. Once finalized, this platform will be the first experimental system capable of accurately quantifying root-root interactions in mature plants. Overcoming previous research limitations, it will provide groundbreaking opportunities for scientists to explore fundamental mechanisms shaping RSA, transforming our understanding of plant competition, cooperation, and resource allocation.

Poster 107. *In vitro* nematicidal activity of BSA-Cystatin microparticles against eggs and juveniles (J2) of *Meloidogyne incognita*

Manzo-Valencia, María Karina[1]; Vaca Salazar, Xitlali Sol[2]; Olalde Portugal, Víctor[1]; Valdés Rodríguez, Silvia E.[1]*

[1]Department of Biotechnology and Biochemistry, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional. Km 9.6 Libramiento Norte Carretera Irapuato-León, 36824, Irapuato, Guanajuato, México.
[2]Life Sciences Division, Universidad de Guanajuato. Ex Hacienda El Copal km 9, carretera Irapuato-Silao, A.P. 311, C.P. 36500, Irapuato, Guanajuato, México.
*Email: silvia.valdes@cinvestav.mx

Meloidogyne incognita is a phytoparasitic nematode responsible for severe yield losses in different crops. The use of proteinase inhibitors, such as Cystatin, has emerged as a promising biotechnological approach for controlling phytopathogens. Nevertheless, their limited environmental stability reduces their overall effectiveness. In this study, we evaluated the impact of albumin-cystatin (BSA-Cystatin) microparticles on the development of *M. incognita* under *in vitro* conditions. Approximately 3,000 *M. incognita* eggs suspended in 0.5 mL sterile distilled water were incubated in the presence of BSA-Cystatin microparticles at final concentrations of 50 and 100 µg/mL. Control treatments included BSA microparticles without cystatin and solutions devoid of cystatin. Additionally, purified cystatin was employed at a concentration of 1.4 µg/µL as a positive control. All treatments were incubated at 28 °C for 7 days, after which egg hatching and eggs integrity were assessed. Furthermore, approximately 1,000 second-stage juveniles (J2) were exposed to the same treatments, and mortality was evaluated after 3 and 7 days of incubation. Juveniles were considered dead if they remained immobile when stimulated with a fine fiber brush and exhibited a typical rigid posture. Statistical analysis of hatching and mortality data was performed using one-way analysis of variance (ANOVA), with subsequent mean comparisons conducted through the Least Significant Difference (LSD) test, considering $p < 0.05$ as statistically significant. Treatment with purified cystatin significantly impacted egg integrity and inhibited hatching. Furthermore, most of the J2s exposed to cystatin exhibited a rigid, needle-like posture and failed to respond to stimulation, suggesting high mortality rates. In contrast, BSA microparticles without cystatin showed no nematicidal activity. The effects of BSA-cystatin microparticles on egg hatching and J2 viability are currently being analyzed. These preliminary results suggest that cystatin exhibits strong nematicidal properties, and that encapsulating it in protein microparticles could provide a controlled-release system, enhancing stability and efficacy for the sustainable management of nematodes in agriculture.

Poster 108. Effect of albumin-cystatin microparticles in the control of *Fusarium oxysporum* in tomato plants (*Solanum lycopersicum*) under greenhouse conditions.

Vaca Salazar, Xitlali Sol[2]; Manzo-Valencia, María Karina[1]; Olalde Portugal, Víctor[1]; Valdés Rodríguez, Silvia E.[1]*

[1]Department of Biotechnology and Biochemistry, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional. Km 9.6 Libramiento Norte Carretera Irapuato-León, 36824, Irapuato, Guanajuato, México.
[2]Life Sciences Division, Universidad de Guanajuato. Ex Hacienda El Copal km 9, carretera Irapuato-Silao, A.P. 311, C.P. 36500, Irapuato, Guanajuato, México.
*Email: silvia.valdes@cinvestav.mx

Fusarium oxysporum causes vascular wilt in tomato plants, reducing fruit production by 60% to 70%, resulting in high economic losses. The use of chemical fungicides in agriculture has led to fungal resistance, environmental pollution, harm to human health, and the elimination of beneficial organisms. The development of new sustainable alternatives for controlling phytopathogenic fungi is crucial to addressing the primary challenges in agriculture. The amaranth cystatin application has demonstrated its effectiveness for controlling early blight produced by *Alternaria alternata* in tomato plants under greenhouse conditions. The objective of this study is to evaluate the curative and preventive effects of albumin-cystatin microparticles (BSA-cystatin) against *F. oxysporum* in tomato plants var. Rio Grande under greenhouse conditions. Each trial consisted of 180 plants with a Randomized Block (RBD) design, with 9 plants per treatment. In the curative trial, tomato plants were inoculated in the soil with (1×10^8 spores) of the pathogenic fungi *F. oxysporium; w*hen symptoms appeared (8 days after inoculation), the cystatin treatments were applied. A total of ten treatments were used: BSA-cystatin microparticles (100 µg per plant), microparticles without cystatin, purified cystatin (1.4 mg/mL per plant), unpurified cystatin (335 µg per plant), and a control with just water. Each one of these treatments was inoculated with or without the pathogenic fungus. In the preventive trial, tomato plants were treated with the same above-described cystatin treatments, and one month later, they were inoculated with *F. oxysporum* (1×10^8 spores) or only water in the control plants. Morphological, physiological, and agronomic parameters were evaluated at 60 and 90 days after inoculation and treatments application. The severity of damage caused by *F. oxysporum* was evaluated in all tomato plants on a scale of 0 to 5, where 0 represented 0% infection, 1: 1-10%, 2: 11-25%, 3: 26-50%, 4: 51-75%, and 5: 76-100% infection. Our results indicate that cystatin and BSA-cystatin microparticles prevent and control the disease caused by *F. oxysporum*. In addition, cystatin treatments stimulated root growth.

Poster 109. **Polymorphism analysis of miRNA precursors between two gene pools of *Phaseolus vulgaris***

Nancy Trujillo[1], Sara Lugo[1], Daniel Cruz[1], Turgut Yigit[2], Jesus Montiel[1], Damien Formey[1*]

[1]Centro de Ciencias Genómicas, UNAM, Laboratorio de Genómica Funcional de Eucariontes, Cuernavaca Morelos.
[2]Aarhus University, Department of Molecular Biology and Genetics - Plant Molecular Biology[3].
*Email: formey@ccg.unam.mx

Phaseolus vulgaris, also known as the common bean, is one of the most important legumes worldwide. Its importance resides not only on its high nutritional value but also in its ability to form symbiotic relationships with nitrogen-fixing bacteria, also known as rhizobia. This interaction is crucial for plant growth in nitrogen poor environments, resulting in the formation of specialized organs called nodules; these structures are responsible for biological nitrogen fixation, whose efficiency varies among genotypes, divided in two main genetic pools: The Andean and Mesoamerican. Among the multiple regulatory mechanisms involved in nodulation, microRNAs (miRNAs) play a key role. These small non-coding RNAs (21 - 24 nt) regulate gene expression post-transcriptionally and are involved in numerous developmental and environmental response processes, including legume-rhizobium symbiosis. It has also been observed that polymorphisms in miRNA precursor sequences can alter their expression or function, potentially impacting the regulation of genes involved in nitrogen fixation. However, little is known about genotypic variation in miRNA precursors in *P. vulgaris*. That's why we aim to identify and characterize polymorphisms in pre-miRNA sequences between the G19833 (Andean) and Negro jamapa (Mesoamerican) genotypes, using a combination of bioinformatic analyses and experimental validation. Small RNA sequencing data from different *Rhizobium etli* inoculation conditions will be analyzed using plant-specific tools such as miRDeep-P and ShortStack, allowing the prediction of mature and precursor miRNAs. Comparative analysis will identify candidate polymorphisms, which will be experimentally validated through PCR and Sanger sequencing. Predicted miRNAs will be annotated using databases and published literature. This study seeks to improve our understanding of the molecular mechanisms regulating nitrogen fixation in common bean and generate knowledge with potential applications in genotype-oriented programs aimed at optimizing symbiosis under diverse environmental conditions.

Poster 110. Effect of arbuscular mycorrhizal fungi and plant growth promoting Rhizobacteria on the aromatic compounds of two basil varieties

Claudia Mariana Gómez-Mata, Rosalinda Serrato, Armando Guerrero, Víctor Olalde*

Center for Research and Advanced Studies of the National Polytechnic Institute (CINVESTAV – IPN) Unit Irapuato, Department of Biotechnology and Biochemistry, Guanajuato, México, Km. 9.6 Libramiento Norte Carr. Irapuato-León 36824 Irapuato Gto. México.
*Email: victor.olalde@cinvestav.mx

Basil (*Ocimum basilicum*) is a widely used aromatic plant in the culinary, pharmaceutical, and cosmetic industries due to the presence of aromatic compounds present in its essential oils. However, the yield of these essential oils is limited by the naturally low concentrations of these aromatic compounds. In this study, we evaluated the effects of the interaction between basil and arbuscular mycorrhizal fungi (AMF), and plant growth-promoting rhizobacteria (PGPR) on the biosynthesis of the main aromatic compounds in green and purple basil varieties. The experiment consisted of two inoculation treatments, one where basil plants were inoculated with three AMF species (*Claroideoglomus claroideum, Septoglomus deserticola*, and a mixed consortium of six AMF species), and another one where they were inoculated with four PGPR strains (one *Bacillus subtilis* strain, two *Azospirillum spp*. Strains, and one *Azotobacter sp* strain). Two months post-inoculation, plants were harvested and separated into roots and foliage. Roots were stained and microscopically analyzed to confirm AMF colonization. Aromatic compounds were extracted from the foliage by methanolic extraction. The extracts were filtered and analyzed via HPLC to quantify the concentration of target aromatic compounds. Results demonstrated significant increases in aromatic compound concentration in plants inoculated with *S. deserticola* and the AMF "Selva" consortium, as well as with *Azospirillum sp*. And *Azotobacter* sp. Strains. These findings suggest that the use of beneficial microorganisms can effectively enhance aromatic compounds biosynthesis and therefore, essential oil yield in basil, presenting promising applications for improving industrial production in sectors dependent on aromatic plant extracts.

Poster 111. Activity and oligomerization of the recombinant peptide BoDef1: a class I defensin from *achiote*

Hernández-Díaz Raquel[1], Sánchez-Cach Lucila[1], Cárdenas-Conejo Yair[2], Rivera Madrid Renata[1], Estrada Georgina[1]*

[1]Centro de Investigación Científica de Yucatán. Unidad de Biología Integrativa. Calle 43 No. 130. Col. Chuburná de Hidalgo, CP 97200, Mérida, Yucatán, México.
[2]Universidad de Colima. Laboratorio de Agrobiotecnología.
*Email: georgina.estrada@cicy.mx

Bixa rellana L. is a native plant from tropical America, mainly known for its seed paste "achiote," which contains the pigment bixin. Achiote remains important in the Mayan culture for both dietary and ceremonial uses. As sessile organisms exposed to abiotic and biotic stresses, plants have evolved immune responses to fight infections. Defensins are conserved peptides found across plants, invertebrates, and vertebrates. Mature defensins are composed of 45–55 amino acids and eight cysteines that, when oxidized, form four disulfide bridges. Their *in vitro* antimicrobial activity depends on peptide binding to phospholipids, where the γ-core loop plays a key role. Lipid interaction also promotes dimerization and oligomerization in class II defensins. Plant defensins generally display antifungal and/or antibacterial activity. BoDef1 was identified in the transcriptome of *Bixa rellana*. Its mature cDNA sequence was cloned into the pQE30 plasmid and expressed in *E. coli* Rosetta-gami. Western blot confirmed the recombinant peptide's presence. HisrBoDef1 was recovered in both the soluble fraction and inclusion bodies. Importantly, its solubility allowed for a simplified experimental workflow. A heat treatment step was introduced, and HisrBoDef1 remained soluble post-heating, indicating notable thermal stability likely due to its disulfide-rich structure. The peptide was further purified by RP-HPLC, yielding several fractions. Preliminary antifungal assays using the heat-treated fractions revealed that HisrBoDef1 affected melanin production in *Alternaria solani* at 250 µg/mL. These findings support BoDef1's biological activity and open the door for further characterization.

Poster 112. CaDEF1 and the CaDEF1_35 AAAA 38 mutant and the His tag impact in peptides oligomerization and antimicrobial activity

Aguiar Lena, Sánchez-Cach Lucila Aurelia, Rodríguez-Fuentes Nayeli, Estrada Georgina*

CICY. Centro de Investigación Científica de Yucatán, A.C. Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México.
*Email: ginaestapia@yahoo.com.mx

Defensins are small defense peptides expressed in plants in response against pathogen infection such as bacteria, fungi and others, including viruses. Some plant defensins oligomerize and different types of dimers are considered their active form. In nowadays the functional characterization of plant defensins is limited by difficulties to obtain natural peptides from plant tissues or to recombinantly expressed active peptides. In our group the Capsicum mature peptide defensin CaDEF1 and the mutant peptide CaDEF1_35 AAAA 38 have been recombinantly expressed in *E. coli* strain Rosetta-gami as heterologous system. To facilitate purification, a 6-histidine (His-tag) at N-terminus was added and the sequence IEGR was located before the defensins. Our efforts are now directed to evaluate the effect of the His-tag elimination in the oligomerization and antimicrobial activity comparatively with CaDEF1 and its γ-core mutant. The His-tag was cleaved using Factor Xa protease, and the reaction products purified by rpHPLC. Cleavage efficiency was verified using SDS-PAGE and Western blot techniques. Finally, both peptides were tested against *Alternaria solani* and *Candida albicans* to determine whether the His-tag affects their antimicrobial activity.

Poster 113. **Begomovirus infection triggers calcium signaling pathways in plants: insights into the role of cyclic nucleotide-gated ion channels**

García-Montelongo A.M.[1,2], Bonales-Alatorre E.[1]*, Cárdenas-Conejo Y.[2]*

[1]Centro Universitario de Investigaciones Biomédicas (CUIB), Universidad de Colima, Colima, México. [2]Laboratorio de Biología Biosintética y Estructural (LaBioSEM), Universidad de Colima, Coquimatlán, México.
Email: ebonales0@ucol.mx, yair.cardenas@gmail.com*

Calcium signaling is an early component of plant responses to biotic stress; however, its role in antiviral defense remains poorly understood. Begomoviruses, single-stranded DNA viruses that cause severe yield losses in crops, are associated with calcium influxes in infected cells, while some viral proteins interfere with host calcium signaling pathways, including Ca^{2+} sensors such as calmodulin-like proteins (CaMs). Among calcium-permeable channels, Cyclic Nucleotide-Gated ion Channels (CNGCs) mediate calcium influx during plant-pathogen interactions, although most studies have focused on bacterial and fungal infections, leaving their contribution to viral responses unexplored. We examined the role of the CNGC family in begomovirus infection by analyzing the expression of selected *Nicotiana benthamiana* CNGCs during Tomato yellow leaf curl virus (TYLCV) infection. Plants were subjected to three treatments (mock, *Agrobacterium*, and TYLCV), with five biological replicates per condition, and sampled at 14 and 21 days post-inoculation (dpi). Quantitative real-time PCR (qPCR) was used to measure gene expression with validated reference genes. Several *NbCNGC* genes showed differential expression in response to TYLCV. These transcriptional changes suggest that begomoviruses could influence calcium signaling via CNGCs, as described for bacterial and fungal pathogens but not previously in viruses. This study provides new insights into signaling events during plant-virus interactions and underscores the importance of calcium regulation in antiviral responses. Understanding CNGC responses to begomovirus infection could support strategies for enhancing resistance in susceptible crops.

Poster 114. **Probing the functional divergence of *TETRASPANIN8-1* and *TETRASPANIN8-2* in *Phaseolus vulgaris* during the symbiotic interaction**

Parra-Aguilar TJ, Palacios-Martínez J, Chávez-Martínez AI, Santana-Estrada O, Olivares-Grajales JE, Quinto-Hernández MA, Cárdenas-Torres L*.

Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, México.
*Email: luis.cardenas@ibt.unam.mx

The root-nodule symbiosis is the most important mutualistic interaction in legume plants for nitrogen uptake under conditions of limited nutrient availability. This symbiosis with rhizobia is established and developed through many key responses, including the recognition by plasma membrane receptors, calcium oscillations, the production of reactive oxygen species (ROS), cytoskeleton reorganization, and vesicular trafficking for symbiont accommodation. Tetraspanins (TETs) are transmembrane proteins with two extracellular loops containing several highly conserved cysteine residues. These proteins can self-organize into tetraspanin-enriched microdomains (TEMs), where they recruit specific proteins into platforms that are required for signal transduction, cell communication, membrane trafficking, and ROS generation. Additionally, tetraspanins such as AtTETRASPANIN 8 contribute to the formation of exosomes, which are extracellular vesicles derived from multivesicular bodies that carry DNA, mRNA, microRNA, and proteins that are important for intercellular and interkingdom communication during the plant immune response. In this study, we investigated the function of the *PvTET8-1* and *PvTET8-2* genes, which belong to the *Phaseolus vulgaris* TETRASPANIN 8 cluster, in the nodulation establishment and development. We analyzed the expression profiles of *PvTET8-1* and *PvTET8-2* in different plant tissues and nodulation stages using a meta data from public transcriptome reports. We also determined the subcellular localization of the *PvTET8.1-GFP* and *PvTET8.2-GFP* fusion proteins in transgenic roots during interaction with *Rhizobium tropici*, and evaluated the phenotypes associated with *PvTET8.1* and *PvTET8.2* silencing and overexpression during nodulation. Our results suggest that *PvTET8.1* promotes rhizobia colonization, while *PvTET8.2* impairs it by decreasing nodule number and nitrogenase activity. Therefore, we propose that *PvTET8.1* and *PvTET8.2* have diverged in function. *PvTET8.1* being necessary for rhizobia root internalization, and *PvTET8.2* more related to immune response, affecting the nodulation establishment. This work was funded by CONACYT No. CBF2023-2024-1032, PAPIIT IN221224 and scholarship No. CVU: 957208 to TJPA.

Poster 115. **Effect of fungicides applied in avocado orchards on the rhizospheric microbiota of *Persea americana* Mill.**

Mariela Martínez Mejía[1], Gustavo Santoyo Pizano[1*], María del Carmen Orozco Mosqueda[2], Sergio de los Santos Villalobos[3], Pedro Damián Loeza Lara[4], Edgardo Sepúlveda[5]

[1]Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo (UMSNH) Morelia 58040, México email: 1607956d@umich.mx;
[2]Instituto Tecnológico de Celaya (TecNM) Celaya 38010, México;
[3]Instituto Tecnológico de Sonora ITSON, Obregón 85000, México;
[4]Universidad de la Ciénega del Estado de Michoacán de Ocampo (UCEMICH) Sahuayo de Morelos 59103, México.
[5]Centro de Investigación Científica y de Educación Superior de Ensenada (CICESE), Ensenada, Baja California.
*Email: gustavo.santoyo@umich.mx

In modern agricultural practices, agrochemicals play an important role in protecting crops from attacks by phytopathogenic organisms. However, the indiscriminate application of these substances can pose environmental risks and indirectly affect beneficial soil microorganisms, soil fertility, and crop productivity. Considering this, the effect of the fungicides fosetyl-aluminum and thiabendazole at different concentrations was evaluated on the growth behavior of 138 bacterial isolates associated with the rhizosphere of *Persea americana* Mill. in three orchards in the state of Michoacán, as well as their ability to produce indoles. A total of 91.5% of the isolates tested were inhibited by fosetyl-aluminum (4.58 g/L); moreover, 82.4% of the inhibited strains showed the ability to produce indoles (>10 µg/ml). Scanning electron microscopy (SEM) micrographs also revealed structural damage, cell wall rupture, and irregular edges in sensitive isolates treated with fosetyl-aluminum. In contrast, bacterial populations were highly tolerant to thiabendazole, even at high concentrations (7.81 g/L), inhibiting only 3.62% of isolates. Finally, six bacteria were identified belonging to the genera *Pseudomonas, Stenotrophomonas, Serratia, Alcaligenes*, and *Achromobacter*. These isolates exhibited antagonistic activity against common fungal and oomycete pathogens of avocado, such as *Colletotrichum gloeosporioides* and *Phytophthora cinnamomi*, respectively. These results highlight the unintended toxic effect of fosetyl-aluminum against beneficial rhizobacteria and, more broadly, against the rhizomicrobiome associated with Persea americana Mill., in contrast to thiabendazole, which did not cause significant damage to the growth of the evaluated isolates.

Poster 116. **Molecular analysis of the effect of amphibian skin bacteria on *Solanum lycopersicum*, as phytostimulants and biocontrol against *Botrytis cinerea***

Adriana Jazmín Gutiérrez Pavón[1], Mario Alberto Serrano Ortega[2*], Yordan Jhovani Romero Contreras[2]

[1]Centro de Ciencias Genómicas y Facultad de Ciencias, Universidad Nacional Autónoma de México, Ciudad de México, México.
[2]Centro de Ciencias Genómicas, Universidad Nacional Autónoma de México, Cuernavaca, México.
*Email: serrano@ccg.unam.mx

Solanum lycopersicum (a globally important crop, best known as tomato) is affected by *Botrytis cinerea*, resulting in losses in field and storage. This fungus has also demonstrated resistance to agrochemicals. Three new species of bacteria isolated from the dermis of *Craugastor fitzingeri* were recently discovered. These species were tested on *Arabidopsis thaliana* and proved effective in inhibiting *B. cinerea* in the plant and phytostimulating plant growth. This project analyzes the effect of these bacteria from amphibian skin on *Solanum lycopersicum* as phytostimulants and biocontrol agents against *B. cinerea.* Therefore, tomato plants were inoculated with the three bacteria, and tests were conducted to compare the incidence of *B. cinerea* infection and physiological parameters between inoculated and control plants. The results showed that plants inoculated with these bacteria had a higher number of root hairs, increased height, and a lower incidence of *B. cinerea* infection, indicating that these bacteria could act as phytostimulants and biocontrol agents against *B. cinerea*. Transcriptional results are also being evaluated to understand this effect molecularly.

Poster 117. **Functional characterization of LBD37/38/39 and LBD16 in *Phaseolus vulgaris* during symbiosis with *Rhizobium etli***

Javier Rodríguez, Elizabeth Monroy, Martha Torres, Damien Formey*

Centro de Ciencias Genómicas, UNAM, Laboratorio de Genómica funcional de Eucariontes, Cuernavaca Morelos.
*Email: formey@ccg.unam.mx

The common bean, *Phaseolus vulgaris*, is one of the legumes capable of fixing nitrogen through symbiosis with rhizobia, which possess the nitrogenase enzyme that reduces atmospheric nitrogen. Each step of this association is tightly regulated by various genes, including transcription factors (TFs) such as LBD proteins (LOB domain proteins). LBD proteins are divided into two main phylogenetic classes. Class I members play key roles in the development of lateral organs such as leaves, inflorescences, roots, and nodules. A representative of this group, LBD16, has been implicated in the early stages of nodule formation in *Medicago truncatula* and *Lotus japonicus*. However, the initiation of nodule organogenesis occurs in different root cell layers depending on the plant species, making it essential to study the role of LBD16 directly in *P. vulgaris*. In contrast, class II members are mainly associated with plant secondary metabolism; notably, AtLBD37/38/39 regulate nitrogen nutrition by repressing key nitrogen transporter genes in *Arabidopsis thaliana*. In *P. vulgaris*, a possible homolog of these factors (PvLBD37/38/39) shows strong accumulation in efficient nodules, suggesting a potential role in the regulation of nitrogen derived from symbiotic fixation.This project focuses on the functional characterization of LBD37/38/39 and LBD16 in *P. vulgaris* during its symbiotic interaction with *Rhizobium etli*. Specifically, we aim to determine their spatial and temporal expression patterns during nodulation, evaluate the symbiotic phenotype of plants with altered transcript levels, and identify potential downstream targets regulated by these factors. By integrating these approaches, the study seeks to advance our understanding of the genetic regulation of nodulation, emphasizing the unique features of LBD16 function in *P. vulgaris* nodules and the unexplored contribution of class II LBDs to biological nitrogen fixation.

Poster 118. CRISPR/dCas9-mediated H3K4me3 deposition at the *SlPR-1* promoter activates transcription and confers resistance to bacterial canker disease in tomato

Leonardo García-Murillo, Eliana Valencia-Lozano, Nicolás Alberto Priego-Ranero, José Luis Cabrera-Ponce, Fátima Patricia Duarte-Aké, Juan Carlos Vizuet-de-Rueda, Diana Marcela Rivera-Toro, Humberto Herrera-Ubaldo, Stefan de Folter, Raúl Alvarez-Venegas*

Center for Research and Advanced Studies of the National Polytechnic Institute, CINVESTAV-IPN, Irapuato, Guanajuato, México.
*Email: raul.alvarez@cinvestav.mx

Histone posttranslational modifications (HPTMs) are tightly linked to enhanced plant defense responses that restrict pathogen infections. Among them, H3K4me3 enrichment at the promoter regions of defense-related genes has long been associated with a transcriptionally permissive chromatin state and with long-lasting immunological memory. Yet, whether H3K4me3 influences host gene expression and resistance to pathogens remains largely correlative and circumstantial. Understanding the causative role of this modification in transcription and harnessing it for crop improvement holds great promise. Here we use the CRISPR/dCas9 platform to recruit the catalytic SET domain of the tomato SlATX1 (an H3K4me3 methyltransferase ortholog to the *Arabidopsis* ATX1) to the promoter region of the tomato *SlPR-1* gene. Targeted recruitment of the SET domain resulted in *SlPR-1* transcriptional activation, PR-1 protein accumulation and increased H3K4me3 enrichment. Further, upon challenge with *Clavibacter michiganensis* subsp. *michiganensis* (*Cmm*), epigenetically edited tomato plants displayed a much stronger induction of *SlPR-1* expression. This transcriptional activation correlated with enhanced resistance to *Cmm* infection, as evidenced by reduced lesion size, lower bacterial colonization, and absence of mortality, while maintaining normal growth, flowering, and fruit production. Overall, our study provides evidence directly linking targeted H3K4me3 deposition at the *SlPR-1* promoter to transcriptional activation and disease resistance, highlighting targeted epigenome editing of plant defense genes as a promising strategy for crop improvement.

Poster 119. **Seasonal dynamics of avocado fruit (*Persea americana*) microbiota during postharvest storage**

Sánchez-Rodríguez D.[1,2], Mora-Godínez S.[1], Clorio-Carrillo J.A.[1], Obispo-Fortunato D.J.[1], López-Cano I.S.[1,2], Mayorga-Martínez A.A.[2], Hernández-Brenes C.[1,2*]

[1]Tecnológico de Monterrey, Institute for Obesity Research, NL, México.
[2] Tecnológico de Monterrey, Escuela de Ingeniería y Ciencias, NL, México.
*****Email: chbrenes@tec.mx

Access to nutritious and safe fruits and vegetables remains a global challenge, highlighting the need for eco-friendly solutions that preserve nutrients and control microbial pathogens. Avocado is an economically and nutritionally important fruit. Studies of its microbiota are scarce, despite its potential impact on fruit health, quality, and postharvest performance. Pre- and postharvest practices influence microorganisms, and their dynamics during storage remain poorly understood. This study aimed to characterize the microbiota of avocado pulp and surface from two seasons, and changes during storage for up to 35 days under export conditions (including storage at 4°C and ripening at 25°C). Avocados from two harvest seasons (May and October) were analyzed at days 0, 21, and 35 of storage. Microbial communities were profiled using 16S rRNA and ITS metagenomics, alongside quality assessments including color and texture. Metagenomic analysis revealed higher microbial diversity on the fruit surface compared to the pulp. On the surface, Firmicutes increased 11.2% by day 35 compared to day 0, while Proteobacteria increased by 14.0% on day 21 compared to day 0 and declined 18.1% by day 35 compared to day 21. Whereas *Methylobacterium*, associated with desirable flavor, decreased 16.0% by day 35 compared to day 0. For fungi, *Colletotrichum* increased on the surface (252.5%) and pulp (283.5%) by day 35 compared to day 0. Quality analysis showed a 15.56% - 51.8% reduction in firmness on day 35 compared to day 21 and a marked increase in gray zones (37–52%) and brown coloration over time. These results indicate that phytopathogenic fungi proliferate during storage, correlating with reduced fruit quality in terms of texture and color. Understanding the avocado microbiota provides new insights into postharvest biology and opens opportunities for microbiome-based strategies to improve fruit preservation and quality during storage.

Poster 120. The ATP-binding cassette transporter A3 regulates root nodule development in *Phaseolus vulgaris*

Carolina Cervera-Torres[1], Kalpana Nanjareddy[2*], ManojKumar Arthikala[2]

[1]Posgrado en Ciencias Biológicas, Escuela Nacional de Estudios Superiores Unidad Morelia, Universidad Nacional autónoma de México (UNAM), Morelia 58190, Michoacán, México.
[2]Ciencias Agrogenómicas, Escuela Nacional de Estudios Superiores Unidad León, Universidad Nacional Autónoma de México (UNAM), León 37689, Guanajuato, México.
*Email: kalpana@enes.unam.mx

Rhizobial bacteria and leguminous plants form a symbiotic relationship crucial for biological nitrogen fixation. Recently, this symbiosis has gained significant interest as a tool for enhancing sustainable crop production. Therefore, is essential a detailed analysis of the molecular mechanisms regulating this symbiosis. Gene families, such as the ABC transporter family, may be involved in regulating the various stages of this symbiosis. In this study, we identified 144 ABC transporters in *Phaseolus vulgaris* and categorized them into eight subfamilies. We also analyzed their expression during symbiosis with both rhizobia and mycorrhizae. Our findings indicated that the PvABCA3 transporter exhibited the most significant change in expression in the presence of symbionts. Consequently, we proceeded with a functional analysis of PvABCA3. This analysis involved examining the spatiotemporal expression of the gene's promoter and performing silencing via RNA interference (RNAi) in *P. vulgaris* roots during symbiosis with *Rhizobium*. Spatio-temporal expression analysis of the *PvABCA3* gene promoter revealed its expression within infected nodule cells. Furthermore, in PvABCA3-RNAi roots, while root hair length did not significantly differ from that of control roots, root hair density and overall length were notably affected. The symbiotic phenotype in PvABCA3-RNAi roots showed that infection thread progression was interrupted in the epidermal cells of the root hairs. Most infection events were not associated with cortical cell divisions, and the number of nodules was reduced by 80%. These results strongly suggest that the ABCA3 transporter plays a critical role in rhizobial infection and root nodule development in *P. vulgaris*. The authors wish to acknowledge the CONACyT: CF-MI-20191017134234199-316538 to M.K.A for funding this work and partially supported by DGAPA/PAPIME-UNAM grant no. IN216321 to K.N and IN213221 to M.K.A.

Poster 121. Rational design of spray-induced gene-silencing agents to suppress *Fusarium* spp.

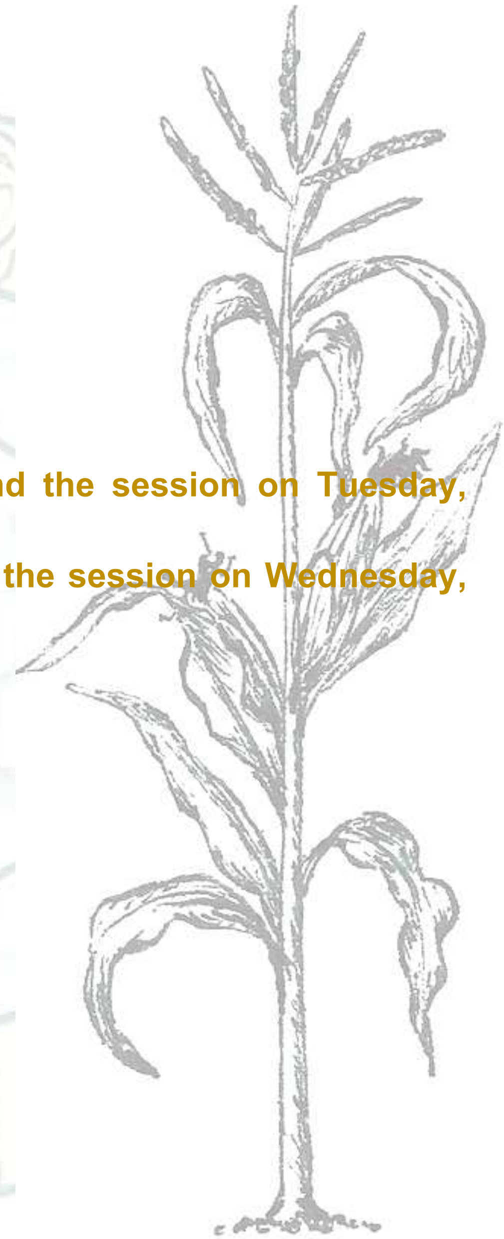
Lizbeth Jazmín Moreno Alegría[1], Jorge Adrián Cirilo Reyes[1], Carbajal Valenzuela Ireri Alejandra[2], Cedillo Jiménez Christopher Alexis[2]*

[1]Facultad de Química, Universidad Autónoma de Querétaro, México.
[2]Centro de Investigación Aplicada a Biosistemas Universidad Autónoma de Querétaro, México.
*Email: c.cedilloc@gmail.com

A transgene-free route to control fungal phytopathogens via spray-induced gene silencing is provided by RNA interference (RNAi). A workflow to design a small interfering RNA (siRNA)–based active ingredient against *Fusarium* spp., focused on targets whose knockout is lethal or required for virulence, is presented. First, the presence of Argonaute homologs in representative *Fusarium* genomes is confirmed to ensure an operational RNAi pathway. Next, candidate genes are ranked by essentiality/virulence evidence, and transcript presence in local isolates is ascertained by RT-PCR followed by qPCR. *In-silico* design of siRNA/dsRNA is then performed following canonical rules (5′ U/A bias, 30–52% GC, low internal stability at the 5′ antisense, absence of long GC stretches), thermodynamic fitness is evaluated, and off-/non-targets are screened across host crops, beneficial organisms, and *Fusarium* genomes. a shortlist of guide sequences, prioritized by predicted efficacy and biosafety, is generated for formulation and delivery optimization. As a case study, candidate guides against a conserved *Fusarium* gene are outlined to illustrate decision points and acceptance thresholds at each step. Subsequent work to be undertaken includes *in-vitro* mortality and virulence assays, dose–response and exposure timing, and iterative refinement to eliminate residual intra-genus off-targets. A reproducible, safety-aware path toward RNA-based crop protection products targeting *Fusarium* spp. is provided by this workflow.

Plant Metabolism
Posters

Participants with even numbers will attend the session on Tuesday, October 21.
Participants with odd numbers will attend the session on Wednesday, October 22.



Poster 122. **Structural insights into the functional regulation of DPE2 during bean fruit development**

Gerardo Contreras-Ruiz, Lilia Angélica Bernal Gracida and J. Eleazar Martínez Barajas*

Departamento de Bioquímica, Conjunto E, Facultad de Química, UNAM. Ciudad de México 04510, México.
*Email: emtz@unam.mx

The pericarp of bean (*Phaseolus vulgaris* L.) fruit is a transient storage tissue; it accumulates starch that is later degraded to provide carbon and energy to the growing seeds. Disproportionating enzyme 2 (DPE2; EC 2.4.1.25) catalyzes the transfer of glucose units from maltose to soluble glucans and is considered a key component in this process. Sequence analysis of DPE2 identified several putative post-translational modifications, including lysine acetylation and serine/threonine phosphorylation, which may impact enzymatic activity. Using *in silico* modeling, we also identified two carbohydrate-binding modules of the CBM20 type in the N-terminal region, which are known to mediate substrate recognition. To investigate how DPE2 is regulated, both the enzyme activity and abundance were evaluated during the mobilization of starch stored in the fruit pericarp. Our results show that both parameters increase during this period, and these increases are enhanced when sucrose flow is reduced. No major changes in the kinetic properties of DPE2 were observed. Using a starch-binding assay, we found that the affinity of DPE2 for its substrate may increase during the starch mobilization phase, particularly under limited sucrose availability. To determine which component of starch DPE2 interacts with, we repeated this assay using amylopectin, amylose, and dextrin. We observed a higher affinity toward amylose. These findings indicate that the DPE2–substrate interaction is regulated by developmental cues and is also responsive to environmental conditions that influence sucrose availability during seed development. We believe that uncovering the regulatory mechanisms of DPE2 activity will yield valuable insights into how plants coordinate carbon remobilization during seed filling and could guide future strategies to optimize source–sink dynamics in legume crops. Acknowlegements. This research was supported by DGAPA (IN205323) and FQ (PAIP 5000-9127). GCR is a recipient of SECIHTI scholarship.

Poster 123. Exploring the role of chloroplast redox networks in nutrient allocation and reserve mobilization in bean fruit pods

Bernal Gracida LA[1], Pérez-Ruiz, JM[2], Cejudo, FJ[2], Martínez-Barajas E[1]*

[1]Departamento de Bioquímica, Conjunto E, Facultad de Química, UNAM. Ciudad de México 04510, México.
[2]Instituto de Bioquímica Vegetal y Fotosíntesis, Universidad de Sevilla and CSIC, España.
*Email: emtz@unam.mx

Chloroplasts possess a complex redox network involving two major systems: the ferredoxin-dependent thioredoxin reductase (Fd-FTR) and associated thioredoxins, which relies on photoreduced ferredoxin, and the NADPH-dependent thioredoxin reductase C (NTRC). These two systems are interconnected via the redox regulation of 2-Cys peroxiredoxins (Prxs), which play a crucial role in maintaining chloroplast function. NTRC is key to sustaining the redox state of 2-Cys Prxs, thereby preserving the reducing capacity of the chloroplast thioredoxin pool, hence ensuring the proper light-dependent regulation of photosynthetic enzymes involved in carbon assimilation. The fruit pod is a photosynthetic structure that plays a vital role for the efficient allocation of nutrients to developing seeds. It also serves as a temporary storage organ, mobilizing reserves during rapid seed growth. These processes require tight biochemical coordination in response to nutrient availability. It is well-known the key role of redox regulation in reflecting the cellular energy state and controlling numerous metabolic and signaling pathways, however, its role in reserve mobilization within the bean fruit pod remains unexplored. To investigate redox dynamics in bean (*Phaseolus vulgaris*) fruit pods, we analyzed two genotypes that differ in the abundance of key proteins involved in the chloroplast redox system. The impact of these differences was evaluated under normal fruit development and under conditions of limited photosynthate availability. Redox status was assessed through the redox state of representative chloroplast proteins, including Prxs, γ-ATPase, rubisco activase (RCA), fructose-1,6-bisphosphatase (FBPase), and ADP-glucose pyrophosphorylase (AGPase), as well as by measuring the NADPH/NADP$^+$ ratio.
Acknowledgements. This research was supported by Grant DGAPA-PASPA, DGAPA (PAPIIT IN205323), CONAHCYT (CY000767), Facultad de Química, UNAM (PAIP 5000-9127), and Grant PID2023-146573NB-I00, funded by MICIU/AEI/ 10.13039/501100011033, Spain, and by ERDF/EU.

Poster 124. The activity of AGPase control starch synthesis in the pericarp of bean fruits

Lilia Bernal, Eleazar Martínez*

Departamento de Bioquímica, Facultad de Química, UNAM.
*Email: emtz@unam.mx.

Seed development demands a substantial supply of metabolic resources over a short time frame. Photosynthetic activity is often insufficient to meet this demand, leading to incomplete seed development in some cases. Moreover, adverse environmental conditions during this critical phase frequently reduce photosynthesis, significantly impacting crop productivity. In bean plants (*Phaseolus vulgaris*), starch is transiently accumulated in the fruit pericarp and later mobilized to support seed growth. Given the considerable genetic variability in starch accumulation in the pericarp, this study aimed to investigate the biochemical basis underlying this variation using two contrasting genotypes differing in pericarp starch content. Despite the marked differences in starch accumulation, both genotypes received comparable amounts of sucrose, and the concentrations of sucrose, glucose, and fructose in the pericarp were also similar. However, the genotype with higher starch levels exhibited reduced concentrations of glucose-1-phosphate (G1P), glucose-6-phosphate (G6P), and fructose-6-phosphate (F6P), suggesting more efficient utilization of these intermediates in starch biosynthesis. ADP-glucose pyrophosphorylase (AGPase), which catalyzes the conversion of G1P to ADP-glucose, is a key regulatory enzyme in this pathway. Its activity is modulated by 3-phosphoglycerate (activator) and inorganic phosphate (inhibitor), granting AGPase a high control coefficient over starch synthesis. In line with this, our results indicate that the differences in starch content between genotypes correlate with significant differences in both AGPase protein levels and enzymatic activity. Ongoing experiments are focused on determining whether these differences also involve distinct regulatory properties of AGPase. Acknowledgements. This research was supported by CONAHCYT (grant CBF2023-2024-767); DGAPA (grant IN205323) and Facultad de Química, UNAM (PAIP 5000-9127).

Poster 125. **Phytochemical evaluation of *E. beecheyanum* a medicinal plant from the Purépecha plateau**

Gema Hernández-Maldonado[1], Flor de Fátima Rosas-Cárdenas[1], Hortencia Gabriela Mena-Violante[2]*, Jeanette Guadalupe Cárdenas-Valdovinos[1]

[1]Centro de Investigación en Biotecnología Aplicada, Instituto Politécnico Nacional (CIBA-IPN), Ex-Hacienda San Juan Molino, Carretera Estatal Tecuexcomac–Tepetitla Km 1.5, C.P. 90700, Tepetitla de Lardizábal, Tlaxcala, México.
[2]Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional, Instituto Politécnico Nacional, (CIIDIR-IPN Unidad Michoacán) Justo Sierra Ote. No. 28, Centro, C.P. 59510, Jiquilpan de Juárez, Michoacán, México.
*Email: hmena@ipn.mx.

Wild plants are important for medicinal uses in the Purépecha Plateau. This applies to hierba del sapo (*Eryngium beecheyanum*); these plants are traditionally used for treatment of diabetes, dermatological and renal conditions and are not cultivated for their use. Land use changes have caused the decline of its wild populations; it is important to develop strategies to maintain these species and obtain phytochemical information that allow it´s sustainable use and study. This study aimed to characterize the phytochemical profiles of the wild medicinal plant species: *Eryngium beecheyanum*, collected from wild populations in Cherán, Michoacán, Mexico. Two complementary analytical techniques were utilized: (1) SPME-GC-MS for the identification of volatile compounds directly from plant material, and (2) HPTLC for the qualitative analysis of metabolites in ethanolic extracts. In its volatile constitution *Eryngium beecheyanum* showed a predominance of germacrene D, terpinolene, and caryophyllene. HPTLC profiles were obtained using ethanolic extracts detecting phenolic compounds in this species, such rosmarinic acid. Also, different contents were found in different tissues such as pedicels, flowers and bracts. The results highlight the distinct chemical richness of the species and its potential for further studies on bioactivity and conservation. This information permits us to standardize HPTLC and SPME-GC-MS techniques for further phytochemical analysis and to conduct genetic analysis related to phytochemical biosynthetic routes in the following experiments. The characterization provided scientific support for traditional uses and represents a step forward toward the standardization of natural extracts with therapeutic and ethnobotanical value.

Poster 126. Effect of nitrogen on PEP and NEP RNA Polymerases in plantlets of *Agave angustifolia* Haw.

Ligia M. Che-Aguilar and Clelia De-la-Peña*

Unidad de Biotecnología, Centro de Investigación Científica de Yucatán, Calle 43 No. 130 x 32 y 34 Col. Chuburná de Hidalgo, 97205 Mérida, Yucatán, México.
*Email: clelia@cicy.mx

Nitrogen is one of the most important macronutrients for plant growth and development, serving as an essential structural element of amino acids, nucleic acids, and chlorophyll. Plants primarily absorb nitrogen in the form of ammonium (NH_4^+) and nitrate (NO_3^-), with approximately 75% of total nitrogen allocated to the chloroplasts – organelles where it supports protein synthesis and the assembly of photosystems I and II. In addition to its structural role, nitrogen regulates fundamental physiological and metabolic processes essential for photosynthetic efficiency and the development of the photosynthetic apparatus, directly affecting biomass production and plant growth. This project investigates how nitrogen availability and chemical form influence the functional stability of the chloroplast, with a focus on plastidial RNA polymerases: PEP (plastid-encoded polymerase) and NEP (nuclear-encoded polymerase). These enzymes are responsible for transcribing genes that control chloroplast development, photosynthetic machinery, and overall organelle maintenance. We hypothesize that both the form and concentration of nitrogen modulate the activity of these polymerases, thereby impacting chloroplast gene expression, protein synthesis, and pigment accumulation. To investigate this, green and albino plantlets of *Agave angustifolia* were grown under different nitrogen treatments, differing in NO_3^- and NH_4^+ sources and concentrations. Additionally, specific inhibitors such as rifampicin, flavopiridol, and 5,6-dichlorobenzimidazole 1-β-D-ribofuranoside (DRB) were used to change RNA polymerase activity and reinforce albino phenotypes. These treatments facilitate the comparative analysis of PEP and NEP abundance and activity across developmental context and nitrogen conditions, using immunodetection methods including Western blotting. Furthermore, the expression of key plastidial genes (*psbA, psaB*, and *rpoA*) is being assessed to elucidate the combined effects of nitrogen supply and transcriptional inhibition on chloroplast gene regulation. Together, these findings will provide insight into how nutrient signals are integrated into the genetic programming of chloroplast biogenesis.

Poster 127. Transport modulation of the alkaloid transporter from *Argemone mexicana*, AmMATE1, by serine rational mutagenesis

Javier Adrián Morales-Morales, Lloyd Jorge A. Loza-Muller, Felipe Vázquez-Flota*

Centro de Investigación Científica de Yucatán. Mérida, Yucatán. México. C. 43 No. 130-x 32 y 34 Col, Chuburna de Hidalgo, Pinzón II.
*Email: felipe@cicy.mx

Argemone mexicana is a medicinal plant traditionally used for therapeutic purposes due to its ability to produce bioactive alkaloids, such as sanguinarine and berberine, which are widely utilized in the pharmaceutical industry. As part of the metabolic regulation of these alkaloids, plants rely on a variety of control mechanisms, including transporters, which play key roles in the storage, relocalization, and excretion. For this reason, it is essential to biochemically characterize these transporters. One effective approach is rational site-directed mutagenesis due that it allows us to assess the functional importance of specific residues, either by suppressing or enhancing protein activity. In the present study, an in-silico search was conducted on the XT1 transcriptome from A. mexicana to identify MATE-like transporters. One candidate, AmMATE1, was selected and cloned into the pUC19 vector for site-directed mutagenesis. The mutated constructs were subsequently introduced into the yeast expression vector pYES-DEST52. Transport assays have already been performed using the wild-type AmMATE1 protein, thus, 12-point mutations will allow us to assess the effect of phosphorylation in its activity. Based on NETphos 3.1 predictions, six serine residues (S75, S158, S262, S322, S336, and S494) were selected and each was substituted by alanine (phospho-deletion) and by glutamate (phospho-mimic). Transport assays with these point mutations will allow us to know the effect on the transport activity of AmMATE1.
Supported by SECIHTI CBF2023-2024-1879

Poster 128. Opuntia gums and mucilage's: metabolomic and structural characterization, properties, and applications in artwork

Emanuel Bojórquez-Quintal[1*], Stefania D´Ottavio[2], Matteo Nava[3], Gianfranco Diretto[3], Carlos Cruz-Cárdenas[4], Rodrigo Chan-Navarro[5], Alexis E. Chávez-García[1], Olimpia Alonso-Pérez[1], Luis Rojas-Abarca[1], Alejandro Meza-Orozco[1], Marisela Ayala-Flores[1], Esteban Sanchez-Rodriguez[1]

[1]Laboratorio de Análisis y Diagnóstico del Patrimonio, El Colegio de Michoacán, Cerro de Nahuatzen 85, Jardines del Cerro Grande, 59370 La Piedad, Michoacán, México.
[2]Sapienza Università di Roma, Piazzale Aldo Moro 5, 00185 Roma, Italy.
[3]GREEN Biotechnology Laboratory, BIOAG Division, SSPT Department, ENEA Casaccia, Via Anguillarese 301, 00123 Rome, Italy.
[4]Centro Nacional de Recursos Genéticos-Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias, Tepatitlán de Morelos, Jalisco, México. [5]Universidad Autónoma de Nuevo León, Facultad de Ciencias Químicas, Cd. Universitaria, Av. Universidad s/n. C. P. 66451, Nuevo León, México.
*Email: emanuel@colmich.edu.mx; jedabojorquezqu@conacyt.mx

Plant gums and mucilage's are bioorganic substances synthesized and/or exuded from various plants, generally from their stems, bark, roots, leaves and seeds, and produced in response to adverse conditions, such as extreme biotic and abiotic stress. Humans have used these substances since ancient times for different applications, such as adhesives, medicinal uses, food preservatives and artworks [2,3]. However, the metabolic profile, chemical structure and properties of Opuntia's gum and mucilage remain poorly explored; in addition, their application as pigment and dye binders for artwork has not studied despite its pre-Hispanic use. This work presents the characterization of gums, mucilage, and tissues of *Opuntia sp.* by HPLC-ESI(+/−)-HRMS, DART-MS (+/-), NMR, FTIR-ATR, SEM-EDS and UV-VIS. Gums from cladodes of *O. ficus indica* (wild), *O. cochinera*, and *O. streptacantha*; and precipitated mucilage from cladodes of *O. ficus indica* (domestic), cuticle, and freeze-dried cladode tissue (without cuticle) were analyzed. The mineral and organic chemical composition was different between species and tissues. Opuntia's gum and mucilage are composed of complex heteropolysaccharides, with minor components such as lipids, 4 principal minerals, and 104 bioactive compounds; also, they exhibit biological and antioxidant activity. Mockups were created to examine the binding properties of Opuntia's gum and mucilage compared to Arabic gum, used in watercolors. Sixteen organic and inorganic pigments were dispersed in the three binders (1:1 ratio) and applied to lime-based and protein-based substrates. Samples were analyzed using FORS colorimetry, FTIR-ATR spectroscopy, optical microscopy and SEM-EDS analysis to assess chromatic behavior and binder interactions for pictorial retouching on mural and canvas paintings.

Poster 129. Modulation of AmABCB2, an auxin transporter, from *Argemone mexicana* through interaction with the immunophilin AmTWD1

Lloyd Jorge A. Loza-Muller, Omar Diaz-Herrera, José Armando Muñoz-Sánchez, Felipe Vázquez-Flota*

Centro de Investigación Científica de Yucatán. Mérida, Yucatán. México. C. 43 No. 130-x 32 y 34 Col, Chuburna de Hidalgo, CP 97205.
*Email: felipe@cicy.mx

Auxins are small molecules playing fundamental roles in plant growth, development and flowering. To do so, auxin functions through the generation of local concentration maxima and minima through the Polar Auxin Transport (PAT) which is partially regulated by ATP Binding Cassette B transporters (ABCB). *Argemone mexicana* is a non-model plant native to northern Mexico that produces pharmacologically important alkaloids such as berberine and sanguinarine. To better understand the development of this species and exploit its medicinal potential, studying auxin transport modulation is fundamental. In previous work, we reported the functional characterization of AmABCB2, an auxin transporter, from *A. mexicana*. ABCB auxin transporters have been reported to be regulated in part by immunophilins-a group of proteins that act as cis-trans-peptidylprolyl isomerases (PPI). In this study, we performed a sequence search of the *A. mexicana* XT1 transcriptome for putative immunophilins using the Pfam domain PF09312. We identified two candidates, which we named AmTWD1 and AmTWD2. AmTWD1 showed the highest sequence identity (73%) to AtTWD1, an immunophilin from *Arabidopsis thaliana* known to modulate the activity of the auxin transporter AtABCB1; based on this homology, we selected AmTWD1 for further study. AmTWD1 was cloned into the entry vector pCR8/GW/TOPO and subsequently subcloned into the destination vector pEG103-GFP for functional characterization. Auxin transport assays were conducted in conjunction with AmABCB2, and subcellular localization of AmTWD1 was assessed in mesophyll protoplasts from *Nicotiana benthamiana*. Here, we present the effect of AmTWD1 on the modulation of AmABCB2-mediated auxin transport, along with its subcellular localization.
Supported by SECIHTI CBF2023-2024-1879

Poster 130. Effect of growth regulators on phytic acid accumulation in soybean seeds

Cruz Pauseno G[1], Calderón Pérez B[1], Ruiz Medrano R[1], Lira Carmona M. R[2], Aguilar Granados A[3], Xoconostle Cázares B[1*]

[1]Centro de Investigación y de Estudios Avanzados, CDMX, México.
[2]Unidad de Investigación Biomédica Oncológica Genómica, IMSS, CDMX, México.
[3]Centro de Investigación y de Estudios Avanzados, UGA, México.
*Email: bxoconos@cinvestav.mx

The role of plant growth regulators, particularly gibberellins (GA) and abscisic acid (ABA), has been extensively described in the regulation of plant growth and development, as well as in responses to abiotic stress conditions. Interestingly, these regulators have also been associated with the accumulation of defense-related metabolites, among them phytic acid (PA), also known as inositol hexakisphosphate. PA serves important functions as the primary phosphorus reserve in seeds; however, it also acts as a cation-chelating agent, forming insoluble complexes that reduce mineral bioavailability and impair digestion in monogastric animals. In this context, PA synthesis represents a plant defense mechanism against herbivory. For example, soybean seeds can accumulate up to 5% PA of their dry weight, negatively affecting their digestibility. In fact, in the livestock feed industry, specific pretreatments are recommended to reduce PA content and thereby improve nutritional utilization. Since PA metabolism can be modulated by growth regulators, in this study we evaluated PA accumulation in soybean plants treated with GA or ABA, with the aim of exploring strategies to reduce its content. PA levels were quantified in leaves and seeds of the Huasteca 400 variety, comparing GA- or ABA-treated plants with a control group. The results of this evaluation demonstrate the adaptability of this genotype to hormonal treatments and suggest that this approach could represent a promising alternative to decrease PA content in soybean, thereby contributing to the development of seeds with enhanced nutritional value for both animal and human consumption.

Poster 131. **Functional analysis of three transcription factor genes possibly involved in the regulation of carotenoid biosynthesis in chili pepper fruits (*Capsicum annuum* L.)**

Zarate-Villagomez, A.J[1], Martínez, O.[2], Ochoa-Alejo, N.[1]*

[1]Departamento de Ingeniería Genética, Unidad Irapuato, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (Cinvestav). Irapuato, Guanajuato, 36824, México.
[2]Unidad de Genómica Avanzada (Langebio), Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (Cinvestav). Irapuato, Guanajuato, 36824, México.
*Email: netfali.ochoa@cinvestav.mx

During chili peppers (*Capsicum* spp.) fruit ripening, substantial amounts of carotenoids are biosynthesized and accumulated in pericarp. These pigments, responsible for the characteristic yellow, orange, and red coloration of mature fruits, function as accessory pigments in photosynthesis and protect against oxidative stress by scavenging reactive oxygen species (ROS). The growing interest in *Capsicum* fruits stems from the nutraceutical properties of carotenoids, which exhibit potent antioxidant and anti-inflammatory activities, stimulate the immune system and help prevent cardiovascular diseases and certain cancers. While the carotenoid biosynthesis pathway in *Capsicum* species has been widely investigated, the transcriptional regulation of genes encoding carotenoid biosynthetic enzymes remains poorly understood compared to climacteric species like tomato (*Solanum lycopersicum*). In this study, we investigated the potential regulatory function of three transcription factors (TF) genes (*ERF113*, *WRKY31* and *WRKY20*) that may be involved in the transcriptional control of carotenoid biosynthesis during chili pepper fruit ripening. These genes were selected through RNA-Seq co-expression analysis correlating their expression profiles with those of the carotenoid structural genes *β-carotenoid hydroxylase* (*β-CHX*) and *capsanthin/capsorubin synthase* (*CCS*). Promoter analysis revealed putative binding sites for these TFs in *β-CHX* and *CCS*. Functional characterization was performed using virus-induced gene silencing (VIGS) in *Capsicum annuum* 'Tampiqueño 74' (Serrano-type) employing TRV-based constructs carrying partial sequences of each candidate gene. Molecular analyses and phenotypic evaluation at 1-, 3-, and 12-weeks post-inoculation confirmed effective and stable gene silencing. Silencing of *ERF113*, *WRKY31*, and *WRKY20* led to a significant reduction in total carotenoid content, suggesting their positive regulatory role in carotenoid biosynthesis. Future work includes HPLC chemical profiling to quantify specific carotenoids at different fruit developmental stages, and the exploration of interactions between the TF proteins and the regulatory sequences of *β-CHX* and *CCS*, aiming to elucidate the transcriptional network controlling carotenoid accumulation in chili pepper fruits. Financial support: Conacyt Project CB280755.

Poster 132. Quaternary structure of pepper defensin CaDEF 1 and its mutant CaDEF1 35-RGFR-38/35-AAAA-38 and their role in haemolysis in vitro

Perla Karina Salazar, Lucila A. Sánchez Cach, Georgina Estrada*

Centro de Investigación Científica de Yucatán A.C. CICY. Calle 43 #130 32 y 34, Chuburná de Hidalgo. CP 97205, Mérida, Yucatán, México.
*Email: ginaestapia@yahoo.com.mx

Plants have established multi-level defense mechanisms, that can be constitutive or inducible tightly coordinated to achieve resistance against pathogen infections. Plant defensins were initially classified as a new group of thionins and subsequent studies showed that these peptides were structurally much closer to mammalian and insect defensins. Defensins exhibit highly variable amino acid sequences, except for the 8 conserved cysteines, plant defensins are small around 45-54 amino acid residues, almost all are cationic and are members of the αβCS 3D structural family stabilized by four disulfide bonds. Several defensins have been characterized as dimers and the dimer is considered the active form of theses peptides. In our research group the Capsicum defensin CaDEF1 and its mutant 35-RGFR-38/35-AAAA-38 have been heterologously expressed in *E. coli.* Thanks to the analysis of the quaternary structures of defensins, the determined protein-lipid interactions and the dependence on the described structural motifs it seems possible to desing recombinant peptides with modified lipid or membrane interactions. Beyond antimicrobial activity, our aim in this work was to compare CaDEF1 and its mutant haemolytic activity. Their protein-lipid interactions are probably necessary for antimicrobial activity, but even those interactions might be involved in the erythrocyte membrane disruption in CADEF1 haemolysis caused by the less cationic CADEF1 35-RGFR-38/35-AAAA-38 γ-core mutant was not expected.

Poster 133. **Establishment of *in vitro* cultures for micropropagation and biochemical study of fructans in *Agave amica* cv. Pearl Double**

Hernández-Zerpa Héctor M[1], Ochoa-Alejo Neftalí[2], & López Mercedes G[1]*

[1]Departamento de Biotecnología y Bioquímica, Unidad Irapuato, Centro de Investigación y de Estudios Avanzados del IPN (Cinvestav-IPN), Irapuato, Mexico.
[2]Departamento de Ingeniería Genética, Unidad Irapuato, Centro de Investigación y de Estudios Avanzados del IPN (Cinvestav-IPN), Irapuato, Mexico.
*Email: mercedes.lopez@cinvestav.mx

Agave amica, an endemic monocot from Mexico, presents a promising biotechnological model due to its rapid development. Floral tissues cultured on MS medium with 2,4-D, NAA and BAP induced dedifferentiation and produced friable callus. Intracellular spherocrystals of fructooligosaccharides (FOS) were confirmed by TLC and FT-MIR, identifying products of fructosyltransferases like 1-kestose, neokestose, and possible isoform of nystose or DP5. Sorbitol (1-3%) enhanced fresh biomass without affecting FOS accumulation, while mannitol caused necrosis. In somatic embryogenesis, 3% sucrose promoted callus induction, 3-4% enhanced friability and 6% in suspension cultures maximized monocot-like somatic embryos production. Finally, a dual bipolar embryogenic pathway is proposed. These findings support the use of *Agave amica* as an extraordinary model to study fructan metabolism and somatic embryogenesis optimization.

Poster 134. Regulatory role of the transcription factor gene *Solyc07g053230* (*SlMYB15*) in the biosynthesis of flavonoid compounds in tomato (*Solanum lycopersicum* L.) variety Micro-Tom

Cortés-Solís Cristóbal[1], López Mercedes G.[2], & Ochoa-Alejo Neftalí[1]*

[1]Departamento de Ingeniería Genética, Unidad Irapuato, Centro de Investigación y de Estudios Avanzados del IPN (Cinvestav-IPN), Irapuato, Mexico.
[2]Departamento de Biotecnología y Bioquímica, Unidad Irapuato, Centro de Investigación y de Estudios Avanzados del IPN (Cinvestav-IPN), Irapuato, Mexico.
*Email: netfali.ochoa@cinvestav.mx

Tomato (*Solanum lycopersicum* L.) is an important source of antioxidants due to dietary relevance and industrial applications. Flavonoids are antioxidant compounds of great interest synthesized from phenylalanine. These compounds play specific roles of protection and development in plants, while also offering various health benefits to humans (nutraceuticals), such as preventing some types of cancer, reducing adipose tissue, and decreasing oxidative stress in cells. The Micro-Tom variety presents high concentrations of flavonoids in the fruit's exocarp, their synthesis is tightly regulated by transcription factors from the MYB family. In tomato, the transcription factor gene *SlMYB15* has been already identified; it is a homolog of *CaMYB31* in chili pepper (*Capsicum annuum* L.), which regulates the biosynthesis of capsaicinoids (pungent compounds) in the fruit. However, the role of *SlMYB15* in the flavonoid biosynthesis in tomato has not yet been characterized. This study analyzes the expression levels of the *SlMYB15* gene and of target genes within the flavonoid biosynthesis pathway (*Ca4H*, *4Cl*, *ChI*, *FS*, *F3H*, *F3GR*), along with the quantification of relevant flavonoids in different fruit tissues (exocarp, mesocarp and endocarp) of the Micro-Tom tomato throughout its development and ripening stages, as well as their spatial and temporal identification and visualization.

Poster 135. Analysis of the possible function of class II *Trehalose Phosphate Synthase* (*TPS*) *5* and *TPS6* genes by their overexpression in *Arabidopsis thaliana*

Rodrigo Alexis Martínez-Gallardo[1], Gabriela Cabrales-Orona[2], Norma Martínez-Gallardo[1], Ma. Barbara Lino Alfaro[1], Luis E. González de la Vara[1] and John P. Délano Frier[1*]

[1]Centro de Investigación y de Estudios Avanzados del IPN, Unidad Irapuato, Irapuato, Guanajuato, México. Libramiento Norte Carretera Irapuato León Kilómetro 9.6, Carr. Panamericana Irapuato León, 36821.
[2]Department of Plant and Soil Science, Institute of Genomics for Crop Abiotic Stress Tolerance (IGCAST), Texas Tech University, Lubbock, TX 79409, USA.
*Email: john.delano@cinvestav.mx

Light-activated photosynthetic processes are essential for carbon (C) assimilation in plants. The latter requires a signaling network to regulate C distribution, storage and use during growth and development. A central component of this network is trehalose-6-phosphate (T6P). This key signaling molecule is synthesized by Class I TREHALOSE PHOSPHATE SYNTHASE (TPS) enzymes (TPS1–4) in *Arabidopsis thaliana*. In contrast, the roles of Class II TPS genes (TPS5-11) remain largely unknown. In this study, the analysis of transgenic *A. thaliana* lines overexpressing (OE) *AtTPS5* and *AtTPS6*, Class II genes were found to produce morphological alterations compared to wild type (WT) plants, as well as distinct phenotypes across multiple developmental-related traits, including seed germination, root development, rosette vegetative growth, flowering time, inflorescence size, seed yield and sensitivity to exogenous abscisic acid, trehalose and sucrose. OE plants also displayed significant alterations in ATPase activity in roots and leaves, detected by densitometric quantification of phosphothreonine-immunodetected bands (~100 kDa) at 7 and 15 days after germination, a finding that proposed a link between these genes and the regulation of ATP synthesis. Additionally, an ongoing transcriptomic profiling in roots and shoots that was performed to gain a better understanding of the physiological roles that Class II TPS proteins may play in plant development and stress responses, revealed an unexpected induction of genes involved in jasmonic acid, wounding and biotic stress-activated responses as well as in membrane transport. In contrast a general down-regulation of auxin and ethylene signaling and photosynthesis-related processes, the latter mostly in roots, as well as of peroxide and T6P phosphatase-related genes. Differences in transcript abundance between both OE lines, i.e., induced in AtTPS5, repressed in AtTPS6, was detected in categories representing flavonoid/phenylpropanoid metabolic and MAPK signaling pathway genes in addition to those involved in the response to oxygen containing compounds (oxylipins?).

Poster 136. **Functional expression of the reluctant S-RNase from *Nicotiana alata*: overcoming expression challenges in *E. coli***

López-Rodríguez ID[1], Portillo-Morales R[1], Rios-Carrasco S[2], Bustos-Jaimes I[3], Cruz-García F[2], Juárez-Díaz JA[1*]

[1]Departamento de Biología Celular, Facultad de Ciencias, Universidad Nacional Autónoma de México (UNAM), Ciudad Universitaria, Cd.Mx. 04510, México.
[2]Departamento de Bioquímica, Facultad de Química, UNAM, Ciudad Universitaria, Cd.Mx. 04510, México.
[3]Departamento de Bioquímica, Facultad de Medicina, UNAM, Ciudad Universitaria, Cd.Mx. 04510, México.
[*]Email: j.a.juarezdiaz@ciencias.unam.mx

Flowering plants are the most diverse group of land plants, occurring in nearly every habitat on Earth. This diversity is partly due to their varied sexual strategies for generating and dispersing offspring. Some of these strategies promote genetic variability by preventing inbreeding, which is critical since ~90% of angiosperms are hermaphrodites, bearing flowers with both female and male functional organs. Among these strategies, self-incompatibility (SI) is one of the most efficient systems for avoiding self-fertilization, preventing self-pollen from achieving self-fertilization through precise biochemical recognition. SI is generally controlled by a single polymorphic *locus* (*S locus*) encoding both female (pistil) and male (pollen) recognition determinants. The most widespread SI system –found in Solanaceae– is the gametophytic S-RNase-based type, where the pistil determinant is a stylar-specific T2-ribonuclease: the S-RNase. In *Nicotiana alata*, S-RNases act as cytotoxins in self-pollen, but their activity depends on the reduction of one of their four disulfide bonds by a pistil-expressed thioredoxin, NaTrxh. To understand how NaTrxh-mediated reduction enhances S-RNase activity, recombinant proteins are essential for *in vitro* structural and biochemical assays. This has been challenging due to the enzyme's potential toxicity and its complex tertiary structure, in which disulfide bonds are critical. Previous attempts have yielded limited success. We now report a promising expression of *N. alata* S-RNase in *E. coli* by co-expressing two enzymes that promote correct disulfide bond formation. The recombinant enzyme exhibits *in vitro* ribonuclease activity; its *in vivo* function is under evaluation. This will enable structural analysis of the S-RNase in its "active" form –reduced by NaTrxh– and elucidation of the highly specific S-RNase-NaTrxh complex. Such insights will not only deepen our understanding of the molecular basis of SI in *N. alata* but also, as a collateral outcome, provide valuable information on the functional specificity of plant thioredoxins –particularly NaTrxh. Funding: PAPIIT IN207823.

Poster 137. Unconventional protein secretion in plants: The Nβ motif of NaTrxh as a versatile targeting signal

Zaragoza-Gomez A[1,2], Sánchez-Jiménez MP[3], Fiordelisio T[4], Juárez-Díaz JA[1*]

[1]Departamento de Biología Celular, Facultad de Ciencias, Universidad Nacional Autónoma de México (UNAM), Ciudad Universitaria, Cd.Mx. 04510, México.
[2]Posgrado en Ciencias Biológicas, UNAM. Ciudad Universitaria, Cd.Mx. 04510, México.
[3]Departamento de Ecología Funcional, Instituto de Ecología, UNAM, Ciudad Universitaria, Cd.Mx. 04510, México.
[4]Departamento de Ecología y Recursos Naturales, Facultad de Ciencias, Universidad Nacional Autónoma de México (UNAM), Ciudad Universitaria, Cd.Mx. 04510, México.
[*]Email: j.a.juarezdiaz@ciencias.unam.mx

The proper functioning of cellular physiological processes relies on accurate protein trafficking, whether to specific cellular compartments or to the extracellular space (ECS). For the traffic to the ECS, the best characterized route is the conventional protein secretion (CPS) pathway, which begins with the co-translational translocation of secretory proteins into the lumen of the endoplasmic reticulum (ER). These proteins are then transported to the Golgi apparatus (GA) and finally released into the ECS via secretory vesicles upon fusion with the plasma membrane. Entry into the CPS requires the presence of an N-terminal signal peptide (SP), recognized by the signal recognition particle (SRP), which mediates their co-translational translocation into the ER. However, numerous proteins have been identified in the ECS despite lacking an SP, revealing the existence of unconventional protein secretion (UPS) pathways. While UPS has been mainly studied in animals and fungi, nearly half of the plant secretome consists of SP-lacking proteins. NaTrxh is an example of such a protein. NaTrxh is a stylar extracellular thioredoxin from *Nicotiana alata* that lacks an SP but contains an 11-amino acid N-sequence –Nβ motif. This motif directs its post-translational translocation into the ER before continuing through the GA and secretory vesicles to the ECS. Unlike SPs, the Nβ motif is non-hydrophobic and therefore might not be recognized by SRP. The chaperone HsP70-1 is the best promising candidate to replace SRP role in this process, which is currently investigated in this study.Interestingly, other plant thioredoxins possess Nβ-like motifs, some of which may be involved in directing them to different cellular destinations. This suggests that the Nβ motif functions as a versatile transit peptide, with specific variations determining different cellular localization. Funding: PAPIIT IN207823.

Poster 138. Scientific education with natural pigments: A botanical and cultural proposal

Estrada-Alburez, N.

University San Carlos de Guatemala, Guatemala City, University City, Zone 12.
Email: nicolealburez@gmail.com

Plant pigments such as chlorophylls, anthocyanins, and carotenoids are bioactive compounds essential for photosynthesis, antioxidant defense, and ecological signaling. They also hold cultural value, having been traditionally used as medicines, dyes, and food. This project promotes the educational use of natural pigments as a pedagogical and andragogical tool to teach botany and phytochemistry. Through workshops for middle school students and the general public, participants extract pigments from leaves, flowers, and fruits using accessible methods (maceration, boiling, natural solvents) to create watercolors and textile dyes. The activities explore pigment types, cellular localization, physiological functions, and the role of mordants in color fixation. They also encourage reflection on the ancestral knowledge of Mayan communities, who pass down plant wisdom through generations. The workshops have proven effective in sparking curiosity, encouraging active learning, and bridging science, art, and culture. Quiroz et al. (2018) highlight how such activities help contextualize scientific knowledge and support meaningful learning in school and community environments. The long-term goal is to expand this methodology to diverse schools and collaborate with phytochemistry researchers to bring their work into classrooms, strengthening the dialogue between scientific and local knowledge.

Poster 139. Characterizing phosphoenolpyruvate carboxylase (PEPC) genes in *Arabidopsis thaliana*

Calista Mrozinski, Edward V. McAssey, Karolina Heyduk*

University of Connecticut, Storrs CT, USA
*Email: karolina.heyduk@uconn.edu

Phosphoenolpyruvate carboxylase (PEPC) is found in all plants and provides intermediate metabolites to the citric acid cycle. In some plants—those that have evolved carbon concentrating mechanisms like C_4 and CAM photosynthesis—PEPC is also responsible for fixing atmospheric CO_2 into a four-carbon acid. There are two main gene families of PEPCs in plant genomes: plant-type, which is involved in citric acid provisioning and photosynthetic function, and bacterial-type, which has a largely unknown role but has been shown to be involved in seed development and, in rare cases, is used for CAM photosynthesis. However, bacterial-type PEPCs are found in all plant genomes and are relatively conserved, suggesting some retained function across all plants. Here we screen t-DNA mutants of all four PEPC genes in *Arabidopsis thaliana*, including the bacterial-type gene, and characterize their effect on photosynthesis, biomass, and malate abundance under well watered and drought stress conditions. We also assess the effects in mutants of PEPC kinase, a gene which is dedicated to phosphorylating PEPC. Phosphorylation of PEPC is crucial for function as a photosynthetic enzyme in C_4 and CAM plants but its importance and role in C_3 is relatively unknown. Together our results demonstrate variable effects across mutant lines on photosynthesis and help to better characterize the roles of PEPC in a C_3 context. Our results can help us better understand the evolution of PEPC as it was recruited from its ancestral C_3 function into more specific photosynthetic roles in C_4 and CAM species.

Poster 140. Engineering a genetically optimized platform for limonoid production in model strains

Keren Priscila López Hernández[1,2], Humberto Javier Constantino Méndez[1,2] and Laura A. Espinosa Barrera[1*]

[1]Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán A. C., Mérida, Yucatán, C.P. 97203.
[2]Universidad Politécnica de Chiapas, Carretera Tuxtla-Gutiérrez, Portillo Zaragoza Km 21-500, Col. Las Brisas, C.P. 29150.
*Email: laura.espinosa@cicy.mx

Limonoids are secondary metabolites found in the Meliaceae and Rutaceae families, widely studied for their bioactive properties, ranging from pesticide applications to potential anticancer therapies. Their biosynthesis originates from the terpene pathway, with squalene serving as a structural precursor that leads to a diverse array of compounds capable of interacting with biological targets. However, the enzymes responsible for this pathway remain poorly characterized, limiting their biotechnological exploitation. This work focuses on the identification of key proteins involved in limonoid biosynthesis using genomic and bioinformatic tools, as a foundational step toward designing a synthetic biology-based production platform. We propose the construction of a modified operon in Escherichia coli, aimed at the functional expression of these proteins to evaluate their capacity to generate limonoids and their potential as a sustainable alternative to conventional pesticides in local agricultural systems.

Poster 141. Physicochemical and metabolomic characterization of nanostructured materials derived from *Prosopis juliflora*

Omar Patiño-Rodríguez[1], Aida A. Rodríguez-Hernández[1], Glenda Pacheco-Vargas[2], Rosalia A. González-Soto[2]

[1]SECIHTI -CEPROBI, Instituto Politécnico Nacional, Yautepec, MOR, México.
[2]CEPROBI, Instituto Politécnico Nacional, Yautepec, MOR, México.

Prosopis juliflora (mesquite) is a species widely distributed in arid and semi-arid regions and is recognized as one of the most invasive plants worldwide. Its successful propagation is linked to its remarkable adaptability, efficient seed dispersal mechanisms, and the absence of effective natural control. While its invasiveness poses ecological challenges, the sustainable utilization of this species as a source of biomass and metabolites for agro-industrial applications has been proposed as a viable strategy to mitigate its environmental impact and simultaneously create added value. Traditionally, *P. juliflora* has been valued for its broad ethnobotanical applications, which are supported by its high content of bioactive compounds, including polyphenols, alkaloids, and dietary fiber, many of which exhibit functional and therapeutic potential. These attributes make the species an attractive candidate for the development of novel plant-base materials. In recent years, nanostructuring of plant materials has emerged as an innovative approach to obtain matrices with enhanced physicochemical and functional properties. Such nanostructured materials are of growing interest due to their potential disruptive applications, particularly in the food and pharmaceutical industries, although their structural and compositional features remain largely unexplored. In this study, we produced nanostructured materials from *P. juliflora* flour and characterized them through a multidisciplinary approach. Metabolomic profiling was performed using (LC-MS), physicochemical characterization included proximate composition analyses, and microstructural properties were examined by confocal microscopy. This integrative strategy allowed us to evaluate both the compositional diversity and the physicochemical behavior of the nanostructured matrix. The results reveal, for the first time, the structural and metabolomic landscape of nanostructured materials derived from *P. juliflora*. These findings provide a foundation for exploring its application as a sustainable, plant-based resource with potential uses in food formulation, nutraceutical development, and pharmaceutical systems. This study therefore highlights the global relevance of *P. juliflora* as a promising raw material for innovative biomaterial research and valorization.

Poster 142. Connecting molecular regulation to the therapeutic potential of spices: The correlation between secondary metabolites with pharmacological properties of *C. cyminum*, *N. sativa*, and *T. foenum-graecum* and their miRNA expression profiles

Reyes-Calderón Almendra, Fernández-Cortés José Miguel, Angulo-Bejarano Paola Isabel*, and Sharma Ashutosh*

Tecnologico de Monterrey, School of Engineering and Sciences, Ave. Eugenio Garza Sada 2501 Sur, Col: Tecnológico, Monterrey, N.L., México, 64700.
*Email: pangulobe@tec.mx; ashutosh.bt@gmail.com

Plants are one of the noblest living beings on Earth since they form symbioses with all other organisms in their habitat. They ensure resilience and adaptability as they respond to biotic and abiotic stresses. Plants have contributed to human being survival not only as an inexhaustible source of food and shelter but eventually transforming modern medicine and the industries that underlie them. Spices such as cumin (*Cuminum cyminum*), black cumin (*Nigella sativa*) and fenugreek (*Trigonella foenum-graecum*) are products of great commercial relevance due to their applications in gastronomy, the food industry, cosmetics and pigments, in addition to their uses in traditional herbal medicine for their anti-inflammatory, antioxidant, antimicrobial, neuroprotective, antidiabetic, and analgesic properties, to name a few. It is well known that the pharmacological and nutraceutical properties of plants depend on their expression of secondary metabolites, which are classified into different categories based on their biosynthesis, structure, and function. The phytochemical composition of plants depends on factors such as spatio-temporal status and maturity. MicroRNAs (miRNAs) are small non-coding RNA molecules that play crucial post-transcriptional regulatory roles in eukaryotic organisms, including the modulation of secondary metabolite synthesis pathways in plants. The present study provides an exploratory mapping of the metabolic fingerprint of commercial essential oils of cumin, black cumin, and fenugreek derived from their qualitative analysis of antioxidant activity (Folin-Ciocalteu, DPPH, and ABTS), quantitative phytochemical profile by GC-MS, and their relationship with the expression of miRNAs linked to key enzymes in the synthesis of secondary metabolites. Due to their complex structure, the synthetic production of economically important secondary metabolites represents a current challenge, so understanding the mechanism of action of the biosynthetic pathways is imperative.

Poster 143. Functional transcriptomic analysis and impact of drought stress on phytochemical content and expression of flavonoid-related genes in *Artemisia ludoviciana* Nutt.

José Miguel Fernández-Cortés[1], Dianella Iglesias[1], Andrea Amy Balderas-Robles[1], Marcos de Donato[1,2], Paola Isabel Angulo-Bejarano[1,*], and Ashutosh Sharma[1,*]

[1]Plant Biotechnology Lab, Tecnologico de Monterrey, School of Engineering and Sciences, Ave. Eugenio Garza Sada 2501 Sur, Col: Tecnológico, Monterrey, N.L., México, 64700.
[2]The Center for Aquaculture Technologies, San Diego, CA 92121, USA
*Email:pang ulobe@tec.mx; ashutosh.bt@gmail.com

Artemisia ludoviciana Nutt. is an aromatic perennial and medicinal herb, native of North America. The aerial parts of the plant are widely used in traditional Mexican medicine for the treatment of different diseases, including diabetes. Numerous secondary metabolites related to its medicinal properties have been identified, of which monoterpenoids, sesquiterpenes, and flavonoids are the most significant. Despite the research conducted on *A. ludoviciana*, there is no transcriptomic data available on the plant to support the synthesis of these compounds in the plant. RNA sequencing of *A. ludoviciana* leaves was conducted to produce 8.4 GB of the plant's transcriptomic data. After *de novo* filtering and assembly of the raw data, 52,561 transcripts were generated, of which a total of 29,854 transcripts (56.79%) were annotated against public databases using Blast2GO. Further KEGG pathway analysis led to the mapping of several transcripts associated with the biosynthesis pathways of major plant secondary metabolites. The expression of key flavonoid-related genes (*CHI*, *F3'MO*, *FSI*, *F3'OMT*) was also evaluated in *A. ludoviciana* after severe water stress conditions. Drought significantly decreased the relative water content (32.78%), total chlorophyll, and carotenoids, while increasing hydrogen peroxide accumulation, showing stress in the plant. The phenol and flavonoid content of the plant also decreased under these conditions. However, the expression of all flavonoid-related genes was upregulated, suggesting an active response of the plant's flavonoid pathway to water stress and potentially contributing to the plant's drought tolerance. This work provides valuable insights into the functional gene annotation, the biosynthetic pathways of secondary metabolites, and the molecular response of *A. ludoviciana* to drought stress. These insights provide the basis for further research into the molecular mechanisms behind the synthesis and accumulation of bioactive compounds in *A. ludoviciana*, as well as strategies to maintain or enhance the plant's medicinal properties.

Poster 144. *In silico* approach for the development of a folate-binding platform for quantifying folates

Miranda Montes-Garza[1], Ernesto Sánchez-Gómez[1], Julieta Ortega-López[1], Itzel Aviña-Ávalos[1,2], Daniel Cantú-Ruiz[1,2], Diaz de la Garza, R[1,2], Perla A. Ramos-Parra[1], Sara M. Garza-Aguilar[1*]

[1]Escuela de Ingeniería y Ciencias, Tecnológico de Monterrey, Monterrey, Nuevo León, México.
[2]Institute for Obesity Research, Tecnológico de Monterrey, Monterrey, Nuevo León, México.
*Email: samaga@tec.mx

Tetrahydrofolate and its derivatives (Vitamin B9) are related compounds structurally that act as essential coenzymes in one-carbon transfer reactions involved in the synthesis of amino acids, purines and other metabolites in all cells. Currently, folate detection relies on the use of folate-binding protein (FBP) purified from bovine milk; however, the presence of plant FBP suggests alternative sources with yet unexplored potential. Sequences of FBP from animals and plants were retrieved and compared, analyzed their subcellular localization, phylogenetic relationship and primary sequence (using Geneious V4.8, MEME Suite, Pfam and DeepLoc 2.0). Key regions of post-translational modifications were identified for animal FBP and compared with plant sequences in the alignment. Eleven animals and 46-plant sequences were retrieved and verified as FBP homologs. The multi-alignment revealed 32% of identity between animal and plant FBPs. The *Hs*FBP is extracellular, and its structure is characterized by 8 disulfide bonds. On the other hand, the all-plant analyzed sequences were predicted with endoplasmic reticulum location and their analysis showed at least 4 possible disulfides bonds conserved with animals. Glycosylation is the principal post-translational modification in *Hs*FBP. Although its role in folate binding remains unclear, studies suggest that it does not affect binding capacity but rather enhances the solubility of the unbound protein. Three glycosylation sites are known in *Hs*FBP, Asn69, Asn161 and Asn201. The site Asn201 was found in 80% of animal FBPs, while in plant FBPs, only 24% showed it. The other sites were not found in the plant sequences. The results suggest that plant-derived FBPs could serve as a viable alternative for folate detection. Their recombinant production in *Escherichia coli* is proposed as an efficient and sustainable strategy. This approach will enable the development of a folate quantification platform based on recombinant proteins for efficient quantification.

Poster 145. **Metabolic changes in avocado (*Persea americana*) during postharvest storage: implications for quality and nutritional properties**

López-Cano I.S.[1,2], Mora-Godínez S.[1], Sánchez-Rodríguez D.[1,2], Clorio-Carrillo J.A.[1], Obispo-Fortunato D.J.[1], Mayorga-Martínez A.A.[2], Hernández-Brenes C.[1,2*]

[1]Tecnológico de Monterrey, Institute for Obesity Research, NL, México.
[2]Tecnológico de Monterrey, Escuela de Ingeniería y Ciencias, NL, México.
*****Email: chbrenes@tec.mx

Avocado (*Persea americana*) is a fruit with high nutritional value for human well-being due to its profile and content of healthy lipids. It is rich in monounsaturated fatty acids (MUFAs) and other bioactive compounds such as phytosterols (PSs) and tocopherols (TCs). During postharvest storage, the fruit´s biochemical composition is affected by metabolic processes, such as respiration rate, ethylene production, maturation, and oxidative degradation, as well as environmental factors. This study aimed to characterize the effect of storage on the nutritional quality (fatty acids and phytosterols) and physicochemical properties of avocados. The pulp of avocados from 0, 21, and 35 days stored under an export simulation design, including storage at 4°C, followed by an incubation at 25°C for 7 days, was analysed. Fatty acids (FA) and PSs were quantified using gas chromatography coupled to a flame ionization detector (GC-FID). The results showed that oleic acid (50.4-55.1%) was the most abundant FA. Also, almost all the FAs rapidly increased by day 21, followed by a decline on day 35 (P<0.05, ANOVA test). β-sitosterol was the most abundant phytosterol (83.2 - 85.6 %), and the avocados showed the highest phytosterol content on day 21 (5607.6 ± 647.3 ppm), which then decreased by 16.7%. The respiration rate increased during storage, and firmness decreased, and were correlated with significant changes in metabolites. Avocado metabolites of nutritional interest and quality (firmness and color) associated with avocado ripening and deterioration during storage are reported. These findings evidence the need for novel postharvest strategies to control avocado ripening and preserve the nutritional quality of avocados, maximizing the use of the fruit and increasing access to healthy foods.

Poster 146. **Wound healing activity of *Bursera simaruba* leaves ethanolic extract in human periodontal ligament cells**

Luis E. Puc-Kantún[1,2], Diana Y. Escalante-Réndiz[1], Angelica A. Serralta-Interian[1,2], Leydi M. Carrillo-Cocom[1], Rafael Rojas-Herrera[1], Beatriz A. Rodas-Junco[2,3*]

[1] Facultad de Ingeniería Química, Universidad Autónoma de Yucatán. Periférico Nte. Km. 33.5, T.C. 13615, Col. Chuburná de Hidalgo Inn, Mérida 97203, Yucatán, México.
[2]Laboratorio de Células Troncales. Facultad de Odontología, Universidad Autónoma de Yucatán. Calle 61-A x Av. Itzaes Costado Sur "Parque de la Paz", Col. Centro, Mérida 97000, Yucatán, México.
[3]SECIHTI-Facultad de Ingeniería Química, Universidad Autónoma de Yucatán, Mérida, Yucatán, México.
*Email: beatriz.rodas@correo.uady.mx

Plants synthesize secondary metabolites as an adaptive mechanism against environmental stress. Beyond their physiological roles, some compounds exhibit pharmacological activities such as antioxidant and wound healing. Such properties may benefit dentistry, particularly in healing after invasive oral soft tissue procedures involving the periodontal ligament. Medicinal plants, therefore, represent a promising source of bioactive compounds. In Mexico, the genus *Bursera* comprises around 89 species, some of which are used in traditional medicine. Within this genus, *Bursera simaruba* is the most widely distributed species and is used to treat skin injuries. However, its wound healing activity has not yet been evaluated.This study evaluates the wound healing activity of *B. simaruba* leaves ethanolic extract (LEE) by assessing its capacity to promote *in vitro* migration of human periodontal ligament cells (hPLC). Cell migration was evaluated through the scratch assay. Cells were cultured until they formed a monolayer, which was scratched and then treated with the LEE at concentrations between 1 and 300 μg/mL. Wound closure was monitored through microphotographs taken at 0, 24, 48, and 72 h and quantified using ImageJ software. The LEE yield was 7.5% (w/w), with a total phenolic content of 128 μg/mg. At 1 μg/mL, the extract enhanced cell migration by 36% after 72 h compared to the control group, which could be explained due to the phenolic compounds content. These results suggest that the LEE stimulate the migration of hPLC at low concentrations, supporting the wound healing activity atributed to *B. simaruba* by the traditional medice. However, concentrations between 10 and 300 μg/mL induced morphological alterations; therefore, it may have a cytotoxic effect.

Poster 147. *Baccharis conferta* Kunth as a source of specialized metabolites with pharmacological applications

Chuzeville-Munguía C[1], Cano-Europa E[2], Gutiérrez-Román AS[1], Medina-Pérez V[1], Trejo Tapia G[1*]

[1]Instituto Politécnico Nacional, Centro de Desarrollo de Productos Bióticos, Morelos, 62739 México.
[2]Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Ciudad de México, 11340, México.
Email: gttapia@ipn.mx

Specialized metabolites (SMe) are essential for plant ecology and represent valuable resources for drug development, supporting disease prevention, treatment, and recovery. *Baccharis conferta* Kunth (Bc) (Asteraceae), commonly known as “hierba del carbonero”, is traditionally used in Mexico for its anti-inflammatory and analgesic properties, which are mainly attributed to flavonoid and terpenoid content, particularly cirsimaritin and kingidiol, respectively. The low abundance of SMe hinders their isolation and characterization, underscoring the need for standardization protocols to ensure the quality of active compounds. The objective of this work was to identify and evaluate a partially purified fraction of Bc with therapeutic application. The fraction was obtained from the aerial parts of Bc. Open column chromatography with TLC verification yielded a partially purified flavonoid fraction (PFF), while cirsimaritin (CIR) was identified and quantified by HPLC. Therapeutic evaluation was conducted in NIH Swiss mice, where $HgCl_2$-induced acute kidney injury was assessed by renal morphological analysis. The PFF consisted mainly of CIR (85%), with a concentration of 55 mg/g, as well as hispidulin and pectolinaringenin as flavonoids, and kingidiol as a terpenoid. Morphological analysis of the renal cortex was performed with a PFF concentration of 20 mg/kg, which significantly reduced the extent of renal damage by decreasing edema and cellular atrophy in the proximal and distal tubules, as well as in the glomeruli. The activity of CIR and other constituents of PFF may have a positive impact on the functional integrity of renal cells, supporting proper organelle function. The use, optimization, and synthesis of SMEs through synthetic biology represent pharmacologically relevant strategies, as applied knowledge and understanding of the pathways of the target metabolites could lead to increased yield and greater potency, safety, and control.

Poster 148. Functional validation of the achiote *BoCCD1-1* gene in bixin biosynthesis through heterologous expression in tomato

Cinthia Berenice Romano Martin, Diana Laura Sierra Ulín, Luisa López Ochoa, Margarita Aguilar-Espinosa, Renata Rivera Madrid*

Unidad de Biologia Integrativa, Centro de Investigación Cientifica de Yucatán, A.C. Calle 43 No. 130, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México.
*Email: renata@cicy.mx

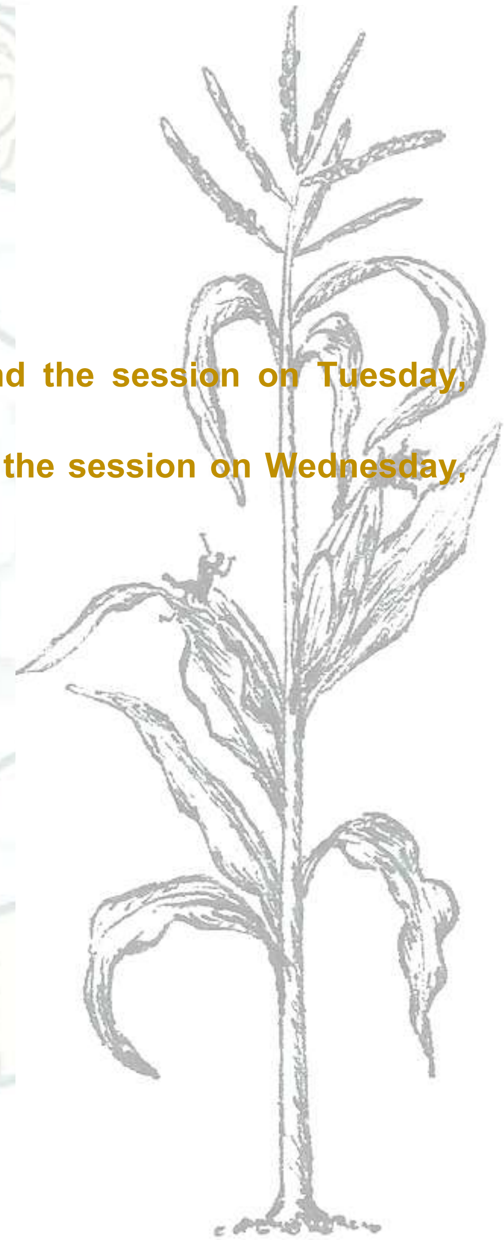
Achiote (*Bixa orellana* L.) is the main natural source of bixin, an apocarotenoid widely used in the food, cosmetic, textile, and pharmaceutical industries. However, the biosynthetic pathway of bixin has not yet been fully elucidated. Studies have proposed that this pathway involves the sequential action of three types of enzymes: carotenoid cleavage dioxygenases (CCDs), aldehyde dehydrogenases (ALDHs), and SABATH methyltransferases. The BoCCD1-1 enzyme has been identified as a key candidate in the first stage of synthesis, participating in the cleavage of lycopene, the direct precursor of bixin. The present study aimed to generate transgenic lines of *Solanum lycopersicum* cv. Micro-Tom plants expressing the *BoCCD1-1* gene under the control of the E8 promoter, which is specific to ripening fruits, where tomato exhibits its highest lycopene accumulation. This strategy enables us to evaluate the potential involvement of BoCCD1-1 in bixin synthesis using tomato as a model system, as its fruit is a significant source of lycopene, the precursor of bixin. To this end, the gene was cloned into the binary vector pMONlink and subsequently introduced into Micro-Tom plants via genetic transformation mediated by *Rhizobium radiobacter* GV2260. Seeds of *Solanum lycopersicum* cv. Micro-Tom seeds were germinated to obtain cotyledon explants, which were infected with the construct-bearing strain and subjected to a regeneration process on selective media containing kanamycin. As a result, fully developed shoots were obtained, of which six plants were successfully acclimated in their substrate. Transformed tomato plants are currently being analyzed. This study represents an important step toward the functional validation of BoCCD1-1 in bixin biosynthesis. In the future, this approach could contribute to the design of biotechnological strategies for the sustainable production of bixin, a natural pigment in high demand.

Plant Development

Posters

Participants with even numbers will attend the session on Tuesday, October 21.

Participants with odd numbers will attend the session on Wednesday, October 22.



Poster 149. The role of mir156 and *SQUAMOSA PROMOTER BINDING PROTEIN-LIKE* transcription factors in *Arabidopsis* embryo development

Victoria Vela- Aguilera[1*,] Andrea Tovar- Aguilar[1], Alma Armenta- Medina[2], Stewart-Gillmor[1]

[1]Unidad de Genómica Avanzada, Centro de Investigación y Estudios Avanzados del Instituto Politécnico Nacional (Cinvestav), Irapuato, Guanajuato, México.
[2]Centro Nacional de Recursos Genéticos, Instituto Nacional de Investigadores Forestales, Agrícolas y Pecuarias (INIFAP), Tepatitlán de Morelos, Jalisco, México.
*Email: stewart.gillmor@cinvestav.mx

MicroRNAs (miRNAs) are key regulators of plant development. miR156 is among the most conserved miRNAs and is known to target *SQUAMOSA PROMOTER BINDING PROTEIN-LIKE (SPL)* transcription factors. Transcriptomic analyses of *Arabidopsis dicer like 1* and *serrate* mutants revealed the upregulation of *SPL2*, *SPL3*, *SPL10*, and *SPL11* at the globular stage, pointing to the possibility that the loss of miR156 in *dcl1* mutants caused the *dcl1* phenotype. However, recent results from our laboratory have shown that these *SPL* genes do not contribute to the *dcl1* phenotype. We are currently investigating an alternative hypothesis: that upregulation of miR156 (rather than downregulation as in *dcl1*) affects cell identity in early embryogenesis. To investigate how miR156 regulates early embryonic events via specific *SPL* targets, we are combining reporter gene assays and analysis of gain- and loss-of-function mutations in miR156 and *SPL* genes. We will examine *mir156* loss-of-function mutants, miR156-overexpressing lines, and higher order *spl* mutants, along with miR156-senstive *SPL-GUS* reporter lines. Our findings will elucidate how miR156-mediated SPL repression orchestrates embryonic patterning and will provide crucial insights into the mechanisms controlling early embryogenesis.

Poster 150. Nitrogen and auxin-cytokinin crosstalk in somatic embryogenesis of *Daucus carota*

Laura C. Fuentes-Vázquez, Monserrat Hernández-Solís, Rosa M. Galaz-Ávalos, Víctor M. Loyola-Vargas*

Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, Mérida, México.
*Email: vmloyola@cicy.mx

Somatic embryogenesis (SE) is a complex biological process that occurs naturally in some species, which generates somatic embryos that develop from somatic cells without the intervention of meiosis or fertilization. SE can be induced *in vitro*, during which the explant responds to stimuli that trigger a signaling response, causing a change in the cell genetic program. Among the factors that modulate this signaling are the nitrogen (N) source and plant growth regulators (PGR), such as auxins (Aux) and cytokinins (CK), which interact closely and are key coordinators during cell morphogenesis and transdifferentiation. However, the biochemical and molecular mechanisms that trigger the signal for somatic to embryogenic cell transition are not yet elucidated. This study investigated the role of N source by the effect of different NO_3^-/NH_4^+ ratios (1:0, 0:1, 2:1, 1:1, 1:2, and 1:1) during SE in *D. carota*, evaluating their influence on morphological response, expression of genes involved in Aux and CK homeostasis, as well as on the N assimilation pathway and endogenous content of these PGR. Preliminary results indicate that a balanced NO_3^-/NH_4^+ (2:1) ratio favors the induction of somatic embryos, whereas the presence of NH_4^+ alone drastically decreases the efficiency of the process. These findings provide a basis for understanding the signaling and interaction networks between N and PGR that enable the formation of somatic embryos.

Poster 151. Study of the effect of NO_3^-/NH_4^+ ratio on somatic embryogenesis

Monserrat Hernández-Solis, Laura C. Fuentes-Vazquez, Marina G. Hernández-Chacón, Rosa M. Galaz-Avalos, Víctor M. Loyola-Vargas*

Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, Mérida, México.
*Email: vmloyola@cicy.mx.

Plant totipotency in vegetal cells is a unique characteristic that allows events such as somatic embryogenesis (SE), in which one or more somatic cells undergo a series of molecular, biochemical, and physiological changes that make them capable of regenerating whole embryos like those obtained from zygotic embryogenesis. Although this process has been studied and exploited since the 1950s, establishing efficient models such as *Daucus carota* is unique for each genotype. It is affected by many factors that make it challenging to develop efficient protocols. One of the factors that points to have a considerable effect on SE is the source of nitrogen, since besides being an essential nutrient it is involved in critical signalling networks of plant growth regulators such as auxins and cytokinins, the main regulators linked to the establishment of SE in several models, including *Coffea canephora*, a species of great commercial importance worldwide. This work aims to elucidate the effect of five different NO_3^-/NH_4^+ ratios during the induction of Se in *C. canephora* and *D. carota*, as well as analyse the endogenous auxin and cytokinin content. The results show a marked effect of the NO_3^-/NH_4^+ ratio on the embryogenic response of both species, where the 2:1 NO_3^-/NH_4^+ ratio is optimal for obtaining somatic embryos; while using only one of the two nitrogen sources, the response was low in the case of NO_3^- and null in the case of NH_4^+. The endogenous content of both auxins and cytokinins was affected throughout each treatment, which points to the possible effect of nitrogen on the signals that trigger the establishment of embryogenic potential during SE induction.

Poster 152. Arabidopsis gynoecium development: confocal imaging of the cell cycle and cytokinin signaling

Juan Ramos-Pulido, Stefan de Folter*

Unidad de Genómica Avanzada, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (Cinvestav), Irapuato, Guanajuato, CP 36824 México.
*Email: stefan.defolter@cinvestav.mx.

In angiosperms, the gynoecium is the female reproductive structure, whose function is to contain the ovules and promote their fertilization. In Arabidopsis, the gynoecium develops from a small primordium with meristematic properties to a complex organ with internal and external specialized tissues. Due to its anatomical and developmental complexity, it is challenging to study in vivo the developmental processes of the gynoecium. Although efforts have been made, there are still unanswered questions, especially those related to cell proliferation of internal tissues. Here, we study the early gynoecium and the establishment of medial tissues through confocal microscopy and specific dissection techniques. This research focuses on the analysis of cell proliferation using the *P*lant *C*ell *C*ycle *I*ndicator (PlaCCI), which is based on a combination of phase-specific fluorescent reporters. With this line, we observed that early gynoecium proliferation is complex and has long cell cycle dynamics. Moreover, we demonstrated the role of cytokinins (CK) in the proliferation of the early gynoecium and the medial tissues with the new version of the CK signaling reporter *TCSn::GFP* and mutant lines. Overall, we observed a coincidence between the region of greatest proliferation and maximum CK signaling, an increase in proliferation attributed to BAP treatment, formation of ectopic tissue with CK signaling, and alterations in the gynoecium primordium in mutant lines. Together, our results provide imaging insights into cell cycle progression, meristematic maintenance, and clear evidence of the role of CK in cell proliferation of the medial domain of the Arabidopsis gynoecium.

Poster 153. The future of habanero chili: innovations in *in vitro* regeneration and genetic improvement

Palma Solís, Renata Valentina; Canto Flick, Adriana; Guzmán Antonio, Adolfo; Pastrana Pérez, Jacobo; Torres Ruiz, José Rubén; Santana, Nancy*

Unidad de Biología Integrativa. Centro de Investigación Científica de Yucatán, México.
*Email: buzzy@cicy.mx

Over the last century, agriculture has evolved toward a more detailed approach, taking into account aspects such as pest management, disease control, and postharvest practices, which has generated uncertainties in production. In this context, habanero chili plays a key role in the culture and economy of Yucatán, being fundamental both in traditional gastronomy and in international export markets. Habanero chili production not only generates local income but also faces technical challenges, particularly in plant regeneration through in vitro culture, given the recalcitrance of the *Capsicum* genus. This difficulty has motivated research on in vitro morphogenesis, a process that involves the development of organized structures under controlled conditions and depends on multiple factors, such as genotype and culture medium. This has posed challenges for science, especially in the field of genetic improvement and plant biotechnology. A key technique in conventional breeding is androgenesis, which enables the production of haploid plants without the need for sexual reproduction, thereby saving time and space. In vitro androgenesis reprograms embryogenesis to create plants from pollen, using simple technologies and environmental control, allowing the generation of specific genetic combinations. Furthermore, in vitro conservation and the production of virus-free plants have been facilitated through the manipulation of plant tissues. Despite its complexity, recent innovations such as genetic editing and the optimization of protocols have improved efficiency in the regeneration of different Capsicum varieties. The present study aims to explore these advances applied to reproductive organs of habanero chili (*Capsicum chinense* Jacq.), with an emphasis on in vitro androgenesis. The analysis of these factors will contribute to genetic improvement and the optimization of techniques for producing plants with desirable traits.

Poster 154. **Direct somatic embryogenesis from Habanero pepper leaves (*Capsicum chinense* Jacq.)**

José Rubén Torres-Ruiz[1], Carlos Alberto Lecona Guzmán[2], Lorena Rodríguez López[1], Renata Valentina Palma Solís[1], Adriana Canto-Flick[1], and Nancy Santana Buzzy[1]*

[1]Centro de Investigación Científica de Yucatán (CICY), Unidad de Biología Integrativa (UBI) Mérida Yucatán; México.
[2]Tecnológico Nacional de México-Campus Tuxtla Gutiérrez. Laboratorio de Cultivo de Tejidos Vegetales. Carretera Panamericana Km 1080 S/N, Tuxtla Gutierrez, Chiapas, México.
*Email: buzzy@cicy.mx

Habanero chili peppers (Capsicum chinense Jacq.) are a crop of great agricultural importance in Mexico, due to their high consumption both fresh and processed, serving as raw material for various industries (Shafiq et al., 2022). This species is recognized as severely recalcitrant to in vitro regeneration, which limits the application of biotechnological tools in the propagation and genetic improvement of the crop (Pijeira and Santana 2024). Somatic embryogenesis (SE) is a morphogenetic process through which perfectly bipolar structures are induced that have no vascular connection with the parent tissue, and can occur directly or indirectly (Fambrini et al., 2022). This study consisted of evaluating the behavior of three habanero chili genotypes during somatic embryogenesis from leaf tissue. Leaf segments from the Mayan Kauil, Mayan Ba'alche, and hybrid (H52) varieties were used in MS medium with 9.05 µM 2,4-dichlorophenoxyacetic acid (2,4-D) for 30 days to induce embryos. Subsequently (after 30 days), the explants were transferred to liquid MS medium containing 4.52 µM of 2,4-D, supplemented with Zeatin (6.84 µM) or dimethylaminopurine-2-iP (3.16 µM), respectively. Histological analyses revealed that when treated with Zeatin and 2-iP, the embryogenic structures showed a better-defined morphology at different stages of development (globular, heart, and torpedo). Better organization of parenchyma cells was observed, improving the formation of meristematic centers and identifying different cellular patterns, which are fundamental for optimal development and cotyledon formation. However, treatment with 4.52 µM of 2,4-D and 4.75 µM of 2-iP was the most favorable, as embryos with better morphology were observed. No significant effect of genotype was observed in the process. The three varieties responded similarly.

Poster 155.

~~**Poster cancelled**~~

Poster 156. The effect of nucleolar stress on primary root growth in wild-type and the *plt1plt2* double mutant of *Arabidopsis thaliana*

Elsy Naomi Miranda Vargas[1,2], Selene Napsucialy-Mendivil[l] , Joseph G. Dubrovsky[1], Svetlana Shishkova[1], Gustavo Rodríguez-Alonso[3*]

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México (UNAM), Cuernavaca, Morelos, Mexico. C.P. 62210.
[2]Universidad Politécnica del Estado de Morelos (UPEMOR), Jiutepec, Morelos, Mexico. C.P. 62574.
[3]Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México. C.P. 62210.
*Email: gustavo.rodriguezal@docentes.uaem.mx

Root development is essential for plant nutrient assimilation and water uptake, among other functions. The root apical meristem (RAM) is a dynamic and highly organized root zone in which stem cells divide, generating cells that enter the transit-amplifying domain and then elongate and differentiate. The establishment of root zonation depends on genetic regulatory networks, including one mediate by the PLETHORA (PLT) transcription factors. In *Arabidopsis thaliana,* the PLT transcription factors underlie root development by controlling root zonation, cell proliferation, and cell differentiation. The double *plt1 plt2* loss of function mutant plants develop short primary and lateral roots with determinate growth due to RAM exhaustion, highlighting their essential role in root development. Although significant progress has been made in elucidating the molecular mechanisms underlying PLT-mediated root growth, the gene regulatory networks governed by these transcription factors remain poorly defined. Our previous computational analyses suggested that PLTs may play a role in regulating ribosome biogenesis. This process, which occurs in the nucleolus, is essential for sustaining the high rates of cell proliferation in the RAM. In this study, we induced nucleolar stress in both wild-type and *plt1 plt2* mutants to examine its impact on the growth of the primary root. The results obtained indirectly suggest that the PLT transcription factors are involved in the regulation of nucleolus-dependent cellular processes. We thank DGAPA-PAPIIT-UNAM, grants IN203024 and IN208824.

Poster 157. Transcriptome analysis of the functional megaspore of *Arabidopsis thaliana*

Guerrero-Díaz H. [1] , González-Jaime, M.I.[1], Tovar-Aguilar A.[1], Badillo-Corona J.A.[1], Choi, J.[2], Gehring M.[2], & Durán-Figueroa, N.[1]*

[1]UPIBI-IPN, Mexico City, Mexico, Av. Acueducto, La Laguna Ticomán, Gustavo A. Madero, 07340.
[2]Whitehead Institute for Biomedical Research, Cambridge, MA, USA, 455 Main St, 02142.
*Email: nduranf@ipn.mx

The development of the female gametophyte in *Arabidopsis thaliana* begins with the specification of the Megaspore Mother Cell (MMC), which undergoes meiosis to produce the Functional Megaspore (FM). This haploid cell subsequently performs three mitotic divisions, giving rise to a mature gametophyte composed of seven cells of four types: three antipodals, two synergids, one Egg Cell (EG), and one binucleate Central Cell (CC). Although transcriptomes of several gametophytic cell types have been described, the molecular landscape of the FM, the initial haploid cell that seeds female gametophyte development, has remained unexplored. Here, we present the first characterization of the FM transcriptome by isolating cell-specific protoplasts through fluorescence-activated cell sorting (FACS). From ~250 protoplasts, we extracted RNA and generated two independent biological replicates. From one library, we obtained 19,038,137 reads, and from the other, 20,306,158 reads, both generated as paired-end sequences using Illumina technology. These datasets reveal that the female gametophyte (FM) expresses approximately 60% of the *Arabidopsis thaliana* genome. Comparative transcriptome analysis between the megaspore mother cell (MMC) and FM identified at least 977 differentially expressed genes, including 397 up-regulated and 580 down-regulated. Furthermore, specific comparisons of FM with the Central Cell (CC) and Egg Cell (EG) uncovered an unconventional transcriptional landscape, highlighting the involvement of diverse regulatory pathways ranging from epigenetic mechanisms to hormonal signaling. This dataset provides a unique molecular snapshot of the FM and represents a critical step toward understanding the transcriptional program that initiates female gametophyte formation in flowering plants.

Poster 158. Post-embryonic cell proliferation in the primary root apical meristem of *Mammillaria* species

Juan M. Palacios-Corona[1], Selene Napsucialy-Mendivil[2], Perla Calderón-Delgado[2], Joseph G. Dubrovsky[2], Svetlana Shishkova[2], Gustavo Rodríguez-Alonso[2]*

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México. Cuernavaca, Morelos, México, C. P. 62210,
[2]Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México, C. P. 62210.
*Email: rodriguezal.gustavo@gmail.com

Most cacti exhibit determinate growth in the primary root due to root apical meristem (RAM) exhaustion soon after germination. In some genera, such as *Pachycereus* and *Stenocereus*, primary-root growth is temporarily sustained by the mitotic activity of the RAM cells before its exhaustion. Here, we analyzed root growth and cell proliferation in the primary root of several *Mammillaria* species (*M. haageana* ssp. *san-angelensis*, *M. coahuilensis*, *M. melaleuca* and *M. viridiflora,* among others), a diverse genus with a high number of species and remarkable morphological diversity within the Cactaceae family. Briefly, seeds were germinated *in-vitro* under controlled conditions, and seedlings or their primary roots were subsequently cleared and mounted for microscopic analysis. Mitotic figures in the primary root apex were evidenced with Orcein or Sytox-Green staining, which allowed us to reliably detect cell division. We observed activation of cell proliferation in the RAM of species with a primary root longer than 5 mm (such as *M. viridiflora*), which continued to grow for several days after germination. This finding suggests that primary root growth in this species is the result of both cell proliferation and subsequent elongation, leading to sustained development for approximately 5-6 days after germination. In contrast, we did not find mitotic figures in species with very short primary root (<1mm; *M. haageana* ssp. *san-angelensis*, *M. coahuilensis* and *M. melaleuca*), which showed growth for only a few hours after germination before cell differentiation at the root apex. This study shows that, even within a single genus of Cactaceae, there is high variability in post-embryonic root development. Such variability may have important implications for ecological performance and might reflect adaptations to specific microenvironments in their natural habitats. We thank DGAPA-PAPIIT-UNAM (grants IN208824 to SS and IN203024 to JGD).

Poster 159. Transcriptomic and functional characterization of transcription factors across different stages of trap development in *Utricularia gibba*

García-Luna Viridiana[1], Oropeza-Aburto Araceli[1], Herrera-Estrella Luis R.[1,2*]

[1]Center for Research and Advanced Studies of the National Polytechnic Institute (UGA-LANGEBIO), Irapuato, Mexico, Libramiento Norte Carretera Irapuato León, Irapuato, Gto.
[2]Texas Tech University, Lubbock, Texas, 2500 Broadway, Lubbock, TX 79409, United State.
*Email: lherrerae@cinvestav.mx

The carnivorous trait has independently evolved at least nine times in angiosperms, enabling plants to thrive in nutrient-deficient environments. It is characterized by the ability to acquire nutrients from animals through specialized leaf-derived structures known as traps, which capture and digest prey. Understanding the molecular modifications underlying trap development has attracted considerable interest. Transcriptomic analyses have revealed differentially expressed genes between carnivorous and non-carnivorous plants, as well as genes preferentially expressed in traps compared to other plant organs. Transcription factors (TFs), as key regulators of gene expression, play essential roles in shaping plant physiology, morphology, and evolutionary trajectories. They have facilitated plant adaptation to diverse environments and the emergence of novel structures. *Utricularia gibba*, an aquatic carnivorous plant belonging to the Lentibulariaceae family, possesses bladder-shaped traps capable of suctioning prey through the rapid release of elastic energy stored in the trap wall. This suction mechanism is unique within the genus *Utricularia* and represents one of the fastest movements in nature. Although its genome and transcriptome have been studied, the regulatory genes controlling trap development remain largely unknown. To address this, we aim to identify transcription factors involved in trap development and to characterize the transcriptional dynamics across nine defined developmental stages of *U. gibba* traps. Our objective is to determine the key TFs associated with trap development and to perform functional studies on selected candidates.

Poster 160. **Expression pattern defined by Cactaceae *PLETHORA1* promoter in transgenic *Arabidopsis thaliana* is opposite to that of *AtPLT1***

Julieta Olvera Berruecos, Elizabeth Cordoba Martínez, Pablo Villa Núñez, Svetlana Shishkova*

Departamento de Biología Molecular de Plantas; Instituto de Biotecnología, Universidad Nacional Autónoma de México. Cuernavaca, Morelos, México.
*Email: svetlana.shishkova@ibt.unam.mx

Most angiosperms, including model species *Arabidopsis thaliana*, show indeterminate growth of the primary root due to the persistent root apical meristem (RAM) activity. In contrast, many Cactaceae species possess determinate root growth; that is, the RAM cells divide only for a short time and then the RAM becomes exhausted and all cells of the root apex differentiate. PLETHORA (PLT) transcription factors are master regulators of plant embryonic and postembryonic development. *A. thaliana* has six partially redundant *PLT* genes, four of them are important for the establishment and maintenance of the RAM. *plt1 plt2* double mutants show determinate primary root growth as a consequence of the RAM exhaustion. Here, we present the results of analysis of the promoter activity of a *PLT1a* putative ortholog from *Carnegiea gigantea* (Cactaceae) in transgenic *A. thaliana.* For this, 2.5 kb promoter region of the *pCgPLT1a* was fused with the coding sequence of a ß-glucuronidase (GUS) or green fluorescent protein (GFP). The *CgPLT1a* promoter, similarly to the orthologous *pAtPLT1*, was active in the RAM of *A. thaliana* seedlings 1 day after germination (dag). Nevertheless, an expression pattern opposite to that of *AtPLT1* was detected in *pCgPLT1a::GUS* primary root of 3 and 5 dag plants: *pCgPLT1a::GUS* expression showed a gradient with a strong signal in the differentiation zone of the *A. thaliana* primary root, while the gradient of *AtPLT1* expression and activity with a maximum in the meristematic zone was reported for the *A. thaliana* primary root. Likewise, *CgPLT1a* promoter was active in the RAM of the recently emerged, but not older lateral roots. Moreover, we also observed *pCgPLT1a::GUS* expression in the shoot organs of 3 and 5 dag plants. Our results suggest the PLT involvement in the regulation of determinate root growth in Cactaceae. This work was partially funded by the DGAPA-PAPIIT-UNAM project IN208824.

Poster 161. Molecular phenology of the flower-to-fruit transition in *Vanilla planifolia* Andrews (Orchidaceae): a bioinformatic approach to identifying transcriptional regulation cores

Hernández-Miranda O. A.[1,4], Campos-Contreras, J. E.[1], Rosas-López U. Y.[2], Sandoval-Zapotitla E.[2], Acosta-Carmona M. A.[3], Salazar-Rojas, V. M.[1]*

[1]Facutad de Estudios Superiores Iztacala, UNAM, Estado de México, México.
[2]Jardín Botánico, Instituto de Biología, UNAM, Ciudad de México, México.
[3]Nodo de Ciencia Ciudadana Xanath, Puntilla Aldama, San Rafael, Veracruz. México.
[4]Posgrado en Ciencias Biológicas, UNAM, Ciudad de México, México.
*Email: adnbic@gmail.com

Developmental processes in angiosperms are often studied using model species such as *Arabidopsis thaliana*. However, non-model plants exhibit distinctive phenomena, including the flower-to-fruit transition (FFT). In *Vanilla planifolia*, this transition is characterized by a a unique post-pollination syndrome (PPS). Unlike model species, in which ovule fertilization occurs shortly after pollination, vanilla ovules require about 25 days post-pollination (DPP) to develop. As a result, vanilla is highly sensitive to environmental fluctuations, and incomplete pollination frequently leads to ovary abscission and reduced fruit set. The limited genomic resources available for vanilla, together with the particularities of its PPS, pose challenges for identifying the genetic networks and regulatory genes involved in this process. To address this gap, this study proposes the integration of molecular phenology with coexpression network analysis to identify transcriptional regulatory hubs associated with FFT in *V. planifolia*. The workflow comprises four phases: (1) molecular phenological profiling of gene expression and anatomical changes; (2) data mining and orthology-based approaches to detect ancestral genes; (3) construction and characterization of FFT coexpression networks; and (4) integrative analyses to define conserved transcriptional regulatory cores associated with FFT. This approach will provide bioinformatic tools valuable not only for vanilla but also for other non-model species. By identifying conserved transcriptional regulatory cores and candidate genes for functional validation, the study offers new insights into the molecular mechanisms governing FFT in orchids. More broadly, these findings contribute to understanding developmental transitions in non-model plants, highlighting the evolutionary conservation of regulatory networks while providing a foundation for applied research in plant biology.

Poster 162. Proteomic approach during the induction of somatic embryogenesis in *Coffea canephora*

Ana Odetth Quintana-Escobar and Víctor Manuel Loyola-Vargas*

Centro de Investigación Científica de Yucatán, Unidad de Biología Integrativa. Calle 43, No. 130 x 32 y 34, CP, Mérida 97205, México.
*Email: vmloyola@cicy.mx.

Plant growth regulators (PGR) are essential for somatic embryogenesis (SE) in different species, and *Coffea canephora* is no exception. In our study model, we have previously elucidated the participation of various genes involved in SE using different strategies; however, we have not yet employed a proteomic approach. This research aims to contribute to the understanding of the primary cellular pathways involved in the development of SE in *C. canephora*. The process of our model consists of two stages: (1) preconditioning in MS medium with auxin (NAA) and cytokinin (KIN), and (2) induction in Yasuda liquid medium added with cytokinin (BA). Therefore, in this study, we analyzed different days of the SE induction process using shotgun label-free proteomics. A total of 1,630 proteins were identified across different sampling days of the process, with the majority accumulating during the induction stage. We found that some of the most enriched pathways during this process were the biosynthesis of amino acids and secondary metabolites. Eighteen proteins were found to be related to auxin homeostasis, and two to cytokinin metabolism, including ABC, BIG, ILR, LOG, and ARR. Ten proteins and transcription factors related to SE were also identified, including SERK1, SKP1, nuclear transcription factor Y, MADS-box, and calreticulin. Additionally, 19 proteins were identified as being related to other processes of plant development, among which the 14-3-3 and PP2A proteins stand out. This is the first report on the proteomic approach to elucidate the mechanisms that operate during the induction of SE in *C. canephora*. So, our findings provide the groundwork for future, more in-depth research.

Poster 163. Epiphytic *Rhipsalis* cacti: is cell proliferation activated during growth of the primary root?

Perla E. Calderón-Delgado[1,2], Selene Napsucialy-Mendivil[1*], Joseph G. Dubrovsky and Svetlana Shishkova[1*]

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México (UNAM), Cuernavaca, 62210, Morelos, Mexico.
[2]Universidad Politécnica del Estado de Morelos (UPEMOR), Jiutepec, 2550, Morelos, Mexico.
*Email: selene.napsucialy@ibt.unam.mx; svetlana.shishkova@ibt.unam.mx

Determinate growth of the primary root was reported for many Cactaceae species. Remarkably, the primary root of epiphytic cacti from the Rhipsalideae tribe only grows for 24 to 48 hours after germination (hag) and reaches 1 to 2 mm in length. The present study aimed to assess whether this minimal primary-root growth in *Rhipsalis* species occurs solely through cell elongation or if cell proliferation in the root apical meristem (RAM) is activated. Four *Rhipsalis* species, *R. elliptica*, *R. baccifera*, *R. paradoxa*, and *R. cruciformis,* were used in this study. Analysis of cleared primary roots revealed that the RAM structure was similar in the four species, and the epidermal cells exhibited a length gradient, with longer cells transitioning to shorter cells towards the apex. To visualize mitotic figures, using Sytox green dye and confocal microscopy, we adjusted a protocol to implement it for cacti roots. This analysis allowed us to visualize cell divisions in the primary roots of *R. baccifera* and *R. cruciformis,* whereas no cell divisions were detected in *R. elliptica* and *R. paradoxa* primary roots. In agreement with these data, we also found an increase in the number of epidermal cells and the primary root length from 36 to 48 hag in *R. baccifera* and *R. cruciformis*. However, these traits did not change for the primary root of *R. elliptica* and *R. paradoxa* seedlings. In summary, our results suggest that in *R. baccifera* and *R. cruciformis* the proliferation is activated in the RAM, that is, the limited primary-root growth occurs due to cell proliferation and cell elongation. In contrast, the primary root of *R. elliptica* and *R. paradoxa* seedlings grows only by cell elongation. This developmental flexibility increases plant fitness in specific habitats. This work was funded by DGAPA-PAPIIT-UNAM grants IN203024 and IN208824.

Poster 164. A *cis*-apocarotenoid signal influences plastid biogenesis and meristem maintenance

Sierra, J[1], Oltehua, O[1], Agreda-Laguna, K.A[1], Napsucialy-Mendivil, S[1], Dubrovsky, J.D[1], McQuinn, R.P[2], and León P[1]*

[1]Instituto de Biotecnología, Universidad Nacional Autónoma de México, Mexico
[2]Australian Research Council Centre of Excellence in Plant Energy Biology, Research School of Biology, Australian National University, Australia.
*Email: patricia.leon@ibt.unam.mx

Plastids function as central metabolic hubs and environmental sensors in plants. Their developmental and physiological integration relies on retrograde signals that regulate nuclear gene expression. Recent evidence implicates carotenoids and their cleavage products, apocarotenoids, as retrograde signals controlling plastid biogenesis and plant development. Characterization of the *Arabidopsis* carotenoid-deficient mutant *clb5* led to the identification of a plastid-derived cis-apocarotenoid signal (ACS1) that disrupts plastid differentiation, impairs lateral organ formation in leaves and roots, and compromises shoot and root apical meristem maintenance. The associated defects are reversed under conditions that lower ACS1 accumulation, such as low light, demonstrating its dynamic regulatory role. ACS1 accumulation alters the expression of hundreds of nuclear genes, including many plastid housekeeping genes, and correlates with an early block in plastid biogenesis—after DNA replication but before transcriptional activation. This disruption impairs chloroplast formation and starch accumulation in amyloplasts. Notably, genes encoding chlororibosomal proteins are among the most affected, leading to a global inhibition of plastid translation and, consequently, defects in leaf and root development. We further demonstrate that ACS1 accumulation interferes with SAM and RAM activity, altering meristem progression and lateral organ development, which translates into defects in leaves, flowers, and roots. The role of ACS1 in floral development reveals a previously unrecognized layer of plastid-to-nucleus regulation that is conserved in other plants, including tomato. These findings uncover a novel plastid-derived signal that shapes organelle biogenesis and plant developmental programs.

Poster 165. **Morpho-histological analysis of the shoot apical meristem and early leaf development of the jipi palm (*Carludovica palmata* Ruiz & Pavón)**

Dafne V. Bacab-Caamal[1], Ángela F. Kú-González[1], Hugo A. Méndez-Hernández[1], Gastón Contreras-Jiménez[2], María E. Álvarez-Buylla[3], María J. Abraham-Juárez[4], Víctor M. Loyola-Vargas[1], Fulgencio Alatorre-Cobos[1*]

[1]Centro de Investigación Científica de Yucatán, Unidad de Biología Integrativa, Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo, 97205, Mérida, Yucatán, México.
[2]Laboratorio de Microscopía y Microdisección Láser, Instituto de Ecología, Universidad Nacional Autónoma de México, Ciudad de México, 04510, México.
[3]Laboratorio de Genética Molecular, Epigenética, Desarrollo y Evolución de Plantas, Instituto de Ecología, Universidad Nacional Autónoma de México, Ciudad de México, 04510, México.
[4]Langebio-Cinvestav Sede Irapuato, Km 9.6 Libramiento Norte Carretera. Irapuato, Guanajuato 36821, México.
*Email: fulgencio.alatorre@cicy.mx

Early leaf development in angiosperms remains poorly understood, particularly in non-model species. Most existing studies have focused on *Arabidopsis thaliana*, limiting our knowledge of species with diverse leaf architectures. Histological evidence suggests that the initial stages of leaf morphogenesis differ between simple and compound leaves. The shoot apical meristem (SAM), a dome-shaped structure established during embryogenesis, functions as a stem cell niche that sustains continuous organogenesis and gives rise to all post-embryonic aerial organs. However, its study is hindered by its small size and internal location. *Carludovica palmata*, commonly known as jipi palm, is a monocot species of the Cyclanthaceae family characterized by compound palmate leaves. Despite its commercial relevance, it remains largely understudied. In this study, we characterized the morpho-histological changes of the SAM during both embryonic and vegetative stages, as well as during the first two plastochrons of leaf development, using optical and electronic microscopy combined with bioimage analysis techniques. Embryos at 100 days after anthesis (DAA) and seedlings between 34 and 38 days after germination (DAG) were analyzed. In both stages, the SAM exhibited isodiametric cells with spherical, centrally located nuclei and progressive vacuolization. Clarification methods combined with Feulgen staining and digital super-resolution imaging revealed distinct chromatin patterns, supporting the proposal of stage-specific histological zonation models. Moreover, the immunolocalization of *At*STM, *Zm*PIN1a, and IAA allowed the identification of early primordium stages (P0 and P1). An atypical division observed in the L1 layer may represent a key event in the onset of leaf organogenesis. These findings provide novel insights into the structural dynamics of the SAM and contribute to a broader understanding of early leaf development.

Poster 166. Establishment of an efficient system for the protoplast isolation from two pineapple (*Ananas comosus* L.) varieties for breeding purposes

Stephanie López-Erosa, Alejandrina Pereira-Patrón, Adriana Canto-Flick y Nancy Santana Buzzy*

Centro de Investigación Científica de Yucatán (CICY), Unidad de Biología Integrativa (UBI) Mérida,Yucatán, México.
*Email: buzzy@cicy.mx

Pineapple is a crop of great importance in various industries, which motivates the development of new varieties that meet the growing demand. Conventional propagation techniques present some limitations due to self-incompatibility and operational challenges, which has prompted the development of genetic improvement techniques. The use of protoplasts (cells without walls) is a crucial aspect of plant biotechnology, particularly in somatic hybridization to develop new germoplasm. While there have been notable advances in the study of protoplasts in a number of species, research in pineapple remains relatively limited. The objective of this study is to establish an efficient protocol for the isolation of viable protoplasts from two pineapple varieties using leaf explants. In this study, an efficient protocol was established for the isolation of viable protoplasts from leaf explants of the MD-2 and Cayena Lisa varieties. Enzymatic combinations of cellulase (1.0 and 2.0%), pectinase (0.1 and 0.5%), and hemicellulase (1.0%) were evaluated, determining yield and viability through optical microscopy and fluorescein diacetate staining. The optimal combination was 2.0% cellulase + 0.1% pectinase, achieving in MD-2 a yield of 4.05×10^6 protoplasts g^{-1} fresh tissue and 83% viability after 10 h of digestion, and in Cayena Lisa 1.9×10^6 protoplasts g^{-1} fresh tissue and 81% viability after 8 h of digestion.

Poster 167. Role of auxin signaling in modulating spike architecture and development in barley

Zambrano-Pereira, A., Demesa-Arevalo, E.*

Centro de investigacion y estudios avanzados del Instituto Politécnico Nacional, Irapuato, Guanajuato, Mexico, Km 9.6 Libramiento Norte Carretera Irapuato-León.
*Email: edgardemesa@cinvestav.mx.

Barley (*Hordeum vulgare* L.) is a cereal of global agronomic importance whose yield is directly linked to the development of its spike architecture, a trait that varies between two-row and six-row varieties. The development of this complex inflorescence is a process that involves the differentiation of different meristems, among which is a type of meristem unique to barley: the triple spikelet meristem (TSM). Spike development in barley is a complex morphogenic process that directly impacts grain yield and is finely regulated not only by molecular mechanisms but also by phytohormones such as auxins, with ARFs being the main transcription factors that execute auxin responses. Despite its importance, the precise spatial and temporal patterns of auxin signaling in barley spikelet meristems are not yet fully understood. Therefore, we propose to determine the role of key auxin response factors in signaling during barley spike development. To unravel this regulation, the research will combine a comprehensive characterization of auxin response patterns and the identification of ARF transcriptional regulators using available tools such as single-cell RNA sequencing. Validation of their expression and study of their dynamics and distribution in situ and functional evaluation using CRISPR/Cas9 gene editing. The results of this project will contribute significantly to the understanding of hormonal regulatory networks in barley inflorescence development and lay the foundation for future genetic improvement strategies aimed at optimizing crop yield and adaptability.

Poster 168. **Functional analysis of the *SPT-like* gene in tomato**

Eduardo Delgadillo Téllez, Eduardo Martínez-Estrada and Stefan de Folter*.

Unidad de Genómica Avanzada (UGA-Langebio), Cinvestav, Km. 9.6 Libramiento Norte, Carretera Irapuato-León, CP 36824 Irapuato, Gto, México
*Email: stefan.defolter@cinvestav.mx

Arabidopsis thaliana has been used as a model organism to study genetic networks involved in gynoecium development; however, little is known about these processes in other species. Tomato (*Solanum lycopersicum*) is a model species that serves to investigate the development and ripening of fleshy fruits. In this study, the tomato variety Micro-Tom was used to analyze the expression and function of the SPATULA-like (SPT-like) transcription factor, a member of the bHLH family that has not been studied in tomato until now. In addition, this factor seems to share certain functions with the previously reported SlSPT transcription factor. Mutant tomato lines (loss-of-function) were generated using CRISPR–Cas9 to characterize the function of *SPT-like* gene. In addition, a construct with the *SPT-like* promoter with the GUS and GFP reporter genes was made to analyze their expression patterns. Here we report our last findings about the *SPT-like* gene function that allows us to find potential redundancy and specific roles for each gene.

Poster 169. **Lateral root primordium morphogenesis and its regulation at epigenetic level**

Selene Napsucialy-Mendivil[l], Héctor H. Torres-Martínez[1,2], Gustavo Rodríguez-Alonso[3], Diana Marcela Rivera-Toro[4], Raúl Alvarez-Venegas[4], Marco Adán Júarez-Verdayes[1,5], Svetlana Shishkova[1], and Joseph G. Dubrovsky[1*]

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México (UNAM), Cuernavaca, 62210, Morelos, Mexico. [2]Current address: Department of Biology, Stanford University, Stanford, CA 94305, USA. [3]Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos. Av. Universidad 1001 Col. Chamilpa CP 62210, Cuernavaca, Morelos, México. [4]Center for Research and Advanced Studies of the National Polytechnic Institute, CINVESTAV-IPN, Unidad Irapuato, Guanajuato, Mexico. [5]Departamento de Ciencias Básicas, Universidad Autónoma Agraria Antonio Narro, Saltillo, Coahuila 25315, México.
*Email: joseph.dubrovsky@ibt.unam.mx

formation. We thank DGAPA-PAPIIT-UNAM (grants IN203024, IN208824) and CONACyT (grant A1-S-9236) for support.Lateral root (LR) is the predominant type of root in most root systems, where it fulfills many important functions. For many years, we have studied cellular and molecular bases of LR primordium (LRP) morphogenesis. Overall, developmental processes are regulated at multiple levels, including the epigenetic level. Here I will discuss the role of a histone H3 lysine 4 (H3K4) methyltransferase ARABIDOPSIS HOMOLOG OF TRITHORAX1 (ATX1) which contributes to active transcription of target genes, in *Arabidopsis thaliana* LRP morphogenesis. We examined LR development in a loss-of-function null mutant (*atx1-1*) and a mutant affected in the ATX1 catalytic domain (*atx1setm*) through bright-field and long-term time-lapse confocal microscopy, transcriptomics, and chromatin immunoprecipitation analyses. LRP initiation and morphogenesis in both mutants were severely abnormal, resulting from altered principal growth directions, extended cell cycle duration, and slower transitions between LRP stages. In *atx1-1* LRPs, the cell cycle was 2.1 times longer than that in the WT. These abnormalities included asymmetric, flattened, extended or fused LRPs. Transcriptomic analysis revealed that among the differentially expressed genes downregulated in *atx1setm*, the most enriched Gene-Ontology categories were cell wall organization and H_2O_2 metabolism, the latter included *PEROXIDASE35* (*PRX35*). The LRP morphogenesis abnormalities were similar in *prx35* and *atx1* mutants. In the *atx1setm* mutant, the deposition of H3K4me3 at the *PRX35* promoter and the *PRX35* expression were significantly reduced. Our data also showed that CDK inhibitor genes *SIAMESE-RELATED5* (*SMR5*) and *SMR7* were among the most upregulated differentially expressed genes in *atx1-1setm,* which partially explains the slow LRP morphogenesis in *atx1* mutants. Our results reveal a link between LR development and redox homeostasis controlled at the epigenetic level by ATX1, which maintains the active transcription of *PRX35* and thereby impacts root system

Poster 170. Role of Trithorax group proteins in *Arabidopsis* root regeneration

Edoardo Yáñez-Martínez, Adriana Garay-Arroyo, Elena Álvarez-Buylla, Ma De La Paz Sánchez*

Laboratorio de Genética Molecular, Epigenética, Desarrollo y Evolución de Plantas, Instituto de Ecología, Universidad Nacional Autónoma de México, 3er Circuito Ext. Junto a J. Botánico, Ciudad Universitaria, UNAM, Mexico City CdMex 04510, México.
*Email: mpsanchez@iecologia.unam.mx

Plants, being sessile, are able to regenerate different organs and tissues to cope with various forms of damage. In many cases, plant regeneration relies on their meristem activity. The *Arabidopsis* Root Apical Meristem (RAM) contains a group of undifferentiated, highly mitotic cells that are crucial for the root regenerative ability, since the Root Stem Cell Niche (RSCN) is completely regenerated in approximately 3 or 4 days after being removed. This regeneration depends on the presence of meristematic cells, and it has been reported the function of key transcriptional factors, as well as some hormones. However, the role of epigenetic factors remains unclear. Therefore, to understand how epigenetic factors contribute to root regeneration, we focused on the Trithorax Group (TrxG), which regulates gene expression by deposition of H3K4me3 and H3K36me3 epigenetic marks. We analyzed the role of three TrxG proteins, the accessory ULTRAPETALA1 (ULT1) protein and the methyltransferases ARABIDOPSIS TRITHORAX1 (ATX1) and SET DOMAIN GROUP4 (SDG4), which play important roles in the proper RSCN organization and maintenance. Our results indicate that RSCN regeneration depends on the presence of meristematic cells. When all RAM cells are excised, regeneration does not take place; instead, lateral root and root hair are produced close to the excision site, an effect that was enhanced in ULT1 loss-of-function mutants. Whereas when the RAM is partially excised, the RSCN is regenerated in 72 hrs in wild-type plants, but ULT1, ATX1, and SDG4 loss-of-function mutants display distinct capacities of regeneration associated with different morphological alterations and different forms of restoration of the quiescent center of the RSCN. Our findings suggest that plant regeneration involves complex morphological changes and that the TrxG proteins analyzed in this work function independently despite belonging to the same complex. Funding: IN206223, CBF2023-2024-433, CBF2023-2024-1002 and IN213524.

Poster 171. The role of phosphorylation of A-type ARRs 4 and 16 in their interaction with NTT

Yuliana Laguna-Montes[1], David Díaz-Ramírez[1], Stefan de Folter[2], Nayelli Marsch-Martínez[1,*]

[1]Department of Biotechnology and Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav), km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, Mexico.
[2]Advanced Genomics Unit (UGA-Langebio), Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav), km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, Mexico.
[*]Email: nayelli.marsch@cinvestav.mx

NO TRANSMITTING TRACT (NTT/WIP2) is a C2H2 transcription factor involved in the development of specific Arabidopsis tissues. In heterologous systems, NTT has been shown to interact with ARR4 and ARR16, which are type A Arabidopsis Response Regulators (ARRs) known for their negative regulation of cytokinin signaling. This pathway consists of a phosphorylation cascade that culminates in the phosphorylation of a conserved Asp residue of the ARRs. We wondered whether ARR4/16 phosphorylation is necessary for their interaction with NTT and how the NTT-ARR interaction affects the capability of NTT to regulate its target genes. BiFC protein-protein interaction assays and NanoLuc-mediated transactivation were performed using NTT and phosphomimetic (Asp→Glu) and non-phosphorylatable (Asp→Asn) variants of ARR4/16 in *Nicotiana benthamiana*. Our results show that only the phosphomimetic versions of ARR4 and ARR16 interact with NTT, whereas the non-phosphorylatable variants show no detectable interaction. Also, we observed that co-expression of the phosphomimetic ARR4 version with NTT reduces the activation of an NTT target gene, compared to NTT alone, suggesting a modulatory effect of ARR4 on NTT transcriptional function. These findings suggest that phosphorylation of type A ARRs not only functions to negatively regulate cytokinin signaling but also determines their ability to interact with other proteins and affects the regulatory effects of NTT in its targets.

Poster 172. Uncovering the design rules of the root epidermis in plant-environment interactions

E G Kocaoglan, M Yang, J Dinneny*

Department of Biology, Stanford University, CA, USA.
*Email: dinneny@stanford.edu)

Our insights into plant-environment interactions are becoming increasingly refined as the paradigm shifts from studying whole-plants to individual cell layers. The root epidermis is unique as it directly interacts with the soil and comprises hair (H) and non-hair (N) cells. H-cells express transporters for nutrient and water absorption, assist root anchorage through their physical association with soil particles, and facilitate symbiotic interactions as the first interface with the soil environment [1]. N-cell function is less studied but is thought to regulate H-cell development [2]. This project hypothesizes that the epidermis form contributes to environmental stress adaptation. Using salinity as a model stressor, we investigate epidermal form and function relationship of 11 Brassicaceae species, each adapted to diverse habitats. By integrating biodiversity and synthetic biology approaches, this study aims to define the gene regulatory networks underlying H/N cell responses and explore how cell-type-specific regulation can be leveraged to engineer resilient root systems. This work will provide foundational insights into the form-function relationships of epidermal cells and inform strategies for developing climate-resilient crops.

Poster 173. **Exploration of mechanisms of the root apical meristem maintenance dependent on threonine in *Arabidopsis thaliana***

Anil Kumar[1], Selene Napsucialy Mendivil[1], José Antonio Pedroza-García[2], Joseph G. Dubrovsky[1*]

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, UNAM, México
[2]Departamento de Bioquímica, Facultad de Química, UNAM, México
[*]E-mail: joseph.dubrovsky@ibt.unam.mx

The growth of lateral roots (LRs) and the proper function of the root apical meristem (RAM) are essential for root system development. In the *Arabidopsis thaliana* mutant, known as *moots koom1* (*mko1*) ("short root" in Mayan), the root growth is determinate, and the RAM is not maintained due to the mutation of the *THREONINE SYNTHASE 1* (*TSY1*) gene also known as *METHIONINE OVERACCUMULATOR2* (*MTO2*). In the roots of *mko1* mutant, free Thr level was found to be higher compared to WT, however, when exogenous Thr is added to the medium, the RAM re-establishes its activity and maintains root growth. These findings suggest that the distribution of free Thr in the root is compartmentalized and that the *mko1* RAM may still be deficient of Thr. Our previous data also suggest that Thr is critical for stem cell function. To address which compartment of the root tip is critical for re-establishing indeterminate growth, complementation of *tsy1-2*/*mko1* mutant was performed with constructs containing *TSY1* gene under tissue-specific promoters. We have analyzed several constructs to verify RAM recovery in the *mko1* mutant complemented with *PromoterX::TSY1:GFP* constructs. Our preliminary result showed that the RAM was reestablished when *TSY1* was expressed under *pGLV9* (active in RAM except QC), *pGLV6* (active in RAM including QC), and *pCO2* (active in distal RAM cortex excluding QC), suggesting a lack of Thr in the *mko1* RAM. Complementation by other *PromoterX::TSY1* constructs is in progress. The *mko1* mutant is not lethal possibly due to the compensation by the protein THREONINE SYNTHASE2 (TSY2). To address what developmental processes are most sensitive to Thr deficiency, we created a sesquimutant *mko1(-/-) tsy2-1(+/-).* Double-homozygous seeds were aborted, suggesting the lethality of the double-homozygous mutant. The data will be presented on RAM development in *mko1(-/-) tsy2-1(+/-).* We thank DGAPA-PAPIIT-UNAM for support (IN203024) and for AK-postdoctoral fellowship.

Poster 174. BOL transcription factor associates with glutaredoxins proteins ROXY1 and ROXY2 during flower development in Arabidopsis

Ramos-Tamayo Claudia Cecilia[1,2], Peralta-Castro Antolín[2], Guerrero-Largo Herenia[1], Brieba Luis G[2], Stefan de Folter[2] and Marsch-Martínez Nayelli[1*]

[1]Biotechnology and Biochemistry Department, Centre for Research and Advanced Studies (CINVESTAV-IPN) Irapuato Unit, Mexico.
[2]Unidad de Genómica Avanzada (CINVESTAV-IPN), Km. 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato 36824, Guanajuato, México.
[*]Email: nayelli.marsch@cinvestav.mx

The flower is a complex structure in plants, composed of a set of specialized organs that play a critical role in reproduction. The reproductive organs are stamens and gynoecia. The relationship between these two organs is essential for plant reproduction, where pollination and fertilization of the ovules give rise to the seeds. Development of the reproductive organs requires distinct molecular cues that are orchestrated through specialized signaling pathways. These pathways involve biomolecules that work together at multiple regulatory levels. Understanding these dynamic regulatory networks is crucial. However, further research is needed to discover novel interactions implicated in reproductive organ development. Here, we study the transcription factor BOLITA/DORNRÖSCHEN-like (BOL/DRNL) which marks organ founder cells, regulates embryo cell patterning, cotyledon organogenesis, and early organ development in Arabidopsis. We performed an *in-silico* analysis to identify potential protein interactors of BOL. Then, we evaluated some relevant interactors related to development *in planta*. We selected ROXY1/2 interactors, which are glutaredoxin proteins that play a role in flower development. We examined the relationship between these glutaredoxins and BOL in plants. Our study demonstrated that the BOL protein-protein interaction network contains a variety of protein partners, from key regulators of floral development to unknown proteins, and these interactions were confirmed *in planta*. Furthermore, the results suggest that the ROXY1/2 – BOL complex protein plays a crucial role during reproductive organ development. The absence of this complex severely affects stamen development, including stamen-gynoecium fusions are observed. In addition, fruit development is impacted with a significantly reduced length. Consequently, there is a reduced seed yield. These results suggest that BOL and ROXY1/2 play a crucial role in regulating reproductive development by coordinating the morphology of floral organs, fertility, and fruit development.

Poster 175. **Transcriptional regulation of auxin and cell wall genes by the transcription factor BOL**

Ana Cecilia Ascencio Bribiesca[1], Herenia Guerrero Largo[1], Stefan de Folter[2], Nayelli Marsch Martínez[1,*]

[1]Department of Biotechnology and Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav), km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, Mexico.
[2]Advanced Genomics Unit (UGA-Langebio), Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav), km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, Mexico.
[*]Email: nayelli.marsch@cinvestav.mx

Plants, unlike animals, have the capacity to form new organs throughout their life cycle. This capacity is provided by niches of pluripotent cells called meristems, which contain pluripotent cells that give rise to new tissues and organs. The correct development of new organs requires the coordination of three main general processes: cell division, differentiation and expansion. These processes in turn involve different cellular activities, among which cell wall remodeling stands out. Cell wall remodeling is promoted by the perception of the phytohormone auxin, which initiates a signaling cascade in which various cell wall remodeling proteins and enzymes are activated. In addition to changes in the mechanical properties of the cell wall, and the presence of the phytohormone auxin, precise and coordinated regulation of many genes involved in organogenesis is necessary. The main regulators of such gene expression are transcription factors, proteins that bind to DNA and activate or repress the expression of different genes. BOLITA (BOL) is an AP2/ERF-type transcription factor that is expressed in new organ founder cells and during shoot regeneration. Loss of function of this transcription factor causes malformation of aerial and vegetative and floral organs and loss of shoot regeneration capacity. When overexpressed, it causes the reduction or loss of the aerial organs of the plant, as well as their vitrification. In addition, it causes the appearance of green callus on the root, indicating that the cells lose their identity and undergo reprogramming. Global expression analyses performed in the laboratory suggest that this transcription factor modulates genes involved in different bioprocesses. Some of these are related to the auxin pathway such those as coding for the auxin transporter PIN-FORMED (PIN1) and enzymes involved in cell wall remodeling such as XYLOGLUCAN ENDOTRANSGLUCOSYLASE HYDROLASE 12 (XTH12). This work focuses on confirming and determining the functional relevance of this regulation.

Poster 176. Exploring the role of HvYABBY transcription factors in the specification of axillary meristems in barley inflorescences

Romo-Arenas O.[1], Abraham-Juárez J.[2,3], Demesa-Arévalo E.[1]*

[1]Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Unidad Irapuato, Irapuato, Guanajuato, México.
[2]Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Unidad de Genómica Avanzada, Irapuato, Guanajuato, México
[3]University of Cambridge, Cambridge, England.
*Email: edgar.demesa@cinvestav.mx

Plant architecture is shaped by meristems. In angiosperms, the transition from the vegetative to the reproductive phase occurs when the shoot apical meristem (SAM) becomes the inflorescence meristem (IM), which originates floral meristem (FM) and culminates in the production of an inflorescence. This process is more complex in barley (*Hordeum vulgare*); where other axillary meristems (AM) are generated, such as the triple spikelet meristem (TSM), the central spikelet meristem (CSM), and lateral spikelet meristems (LSM), which precede the FM development. With the aim of understanding the transcriptional changes that occur during ear development in barley, Demesa-Arévalo *et al.* (2025) integrated single-cell RNA sequencing (scRNA-seq) and multiplex fluorescent *in situ* hybridization (multi7plex FISH) to impute gene expression of over 40,000 genes. This led to the establishment of BARVISTA, a novel database that enables the exploration of expression profiles and virtual microdissection in the barley ear. By virtually-dissecting the putative founder cell regions within the IM using BARVISTA, two YABBY transcription factors (TF) were identified, and the localization of their transcripts might be relevant for TSM development. However, the function of YABBY TF remains unexplored in barley. This project aims to characterize YABBY TF by immunolocalization, gene editing, and defining their interaction with known regulators in meristems. The functional characterization of YABBY TFs in barley could contribute to unraveling the molecular mechanisms underlying the regulatory network that specify the TSM in barley ears and that has not been reported to date.

Poster 177. Exploring the transcriptomic pathways of somatic embryogenesis in *Agave tequilana* Weber var. azul

Sandra Y. Martínez-Martínez[1,2]; Sandra Gonzales-Morales[2]; June Simpson[1]*; Luis R. Herrera-Estrella[2,3]*

[1]Department of Genetic Engineering at Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav), Irapuato, Guanajuato, Mexico
[2]Advanced Genomics Unit (UGA-Langebio), Irapuato, Guanajuato, Mexico
[3]Department of Plant and Soil Science at Texas Tech University, Texas, U.S.A.
*Email: june.simpson@cinvestav.mx; lherrerae@cinvestav.mx

Somatic embryogenesis (SE) is a key tool in plant biotechnology for the propagation and genetic improvement of recalcitrant species such as *Agave tequilana* Weber var. azul. Understanding the molecular mechanisms that regulate this process is essential for optimizing regeneration protocols and enabling more efficient biotechnological applications, including genetic transformation and large-scale clonal propagation. Transcriptomic analysis provides a powerful strategy to explore the gene expression dynamics underlying the transition from totipotent cells to organized embryos. This study aimed to characterize the transcriptomic changes occurring throughout the SE process in *A. tequilana*. An *in vitro* system was established using zygotic embryos extracted from seeds, which were cultured in specific media to induce the formation of proembryogenic calli and, subsequently, somatic embryos. Four key developmental stages were selected for transcriptomic analysis: zygotic embryo (EZ), callus (CA), embryogenic callus (EC), and somatic embryos (SE). Total RNA was extracted from the collected tissues and assessed for integrity prior to cDNA library construction. Sequencing was performed using RNA-seq technology on the Illumina NovaSeq X Plus platform. The resulting reads were aligned to the *A. tequilana* reference genome, achieving high mapping efficiency. Differential gene expression analysis was conducted using specialized bioinformatics tools, enabling the identification of genes with significant expression changes across the embryogenic stages. This approach facilitated a detailed characterization of the molecular events involved in the transition from undifferentiated tissues to structured embryonic forms. The results revealed dynamic patterns of gene regulation at each developmental stage, with key changes observed during embryogenic induction. This study provides a comprehensive transcriptomic framework to better understand the molecular basis of somatic embryogenesis in *Agave tequilana*, with direct implications for its clonal propagation and genetic enhancement.

Poster 178. Revisiting the ABCE model in the establishment of floral organs in *Hordeum vulgare*

Méndez-García, J., Demesa-Arevalo, E.*

[1]Centro de investigación y estudios avanzados del Instituto Politécnico Nacional, Irapuato, Guanajuato, México, Km 9.6 Libramiento Norte Carretera Irapuato-León.
*Email: edgar.demesa@cinvestav.mx

MADS-box genes encode transcription factors (TF) that regulate multiple processes in plant development, such as determining inflorescence architecture, floral identity and seed development. Most of the knowledge of the structure, morphology and genetics of flowers has been obtained through the study of eudicot plants such as *Arabidopsis thaliana*. Monocot plants are very important to humanity due to their adaptation, diversity and their uses; they are relevant in the economy, agriculture, food, among other applications. In this work, we propose as a model a monocot plant, *Hordeum vulgare* (barley), to determine if the functions of the different ABCE classes of the canonical model of the MADS-box TF, proposed in *Arabidopsis*, are conserved. According to recent studies, the expression of class A genes in barley extends to other organs than those proposed in the canonical ABCE model. With this project we want to characterize the role of some HvMADS TF belonging to class A involved in determining floral identity. It is expected that expression patterns will be identified by immunolocalization to establish correlations with developing floral morphology. Protein-protein interactions between HvMADS of different classes will be evaluated as well. This project will contribute to the understanding of the ABCE model in monocots and our results could provide knowledge for possible applications in crop improvement.

Poster 179. **Quantification and identification of cAMP in different plant tissues**

Galaz Ávalos R. M.[1], Monribot Villanueva J.L.[2], Guerrero Analco J. A.[2] Loyola Vargas V.M.[1]*

[1]Centro de Investigación Científica de Yucatán A, C. (CICY), calle 43 No. 130 x 32 y 34, Col. Chuburná de Hidaldo, C.P. 97205, Mérida, Yucatán, México.
[2]Instituto de Ecología, A. C. (INECOL), Carretera antigua a Coatepec 351, colonia El Haya, Xalapa, Veracrúz, México. C.P. 91073.
*Email: vmloyola@cicy.mx

Adenosine 3,5-cyclic monophosphate (cAMP) was discovered in 1957 by Sutherland and is generated from adenosine triphosphate (ATP) by the action of adenylate cyclase. The cAMP functions as a second messenger in a wide range of physiological responses in animals, bacteria, fungi, and algae. For a long time, it was not accepted that cAMP was present in plant tissues, primarily because its levels are significantly lower than those found in animal cells. The improvement of analytical techniques has demonstrated that cAMP is present in plant tissues, and its levels increase in response to various stimuli. It is suggested that cAMP may be involved in the auxin response. Our interest is to determine whether cAMP mediates the response to auxins during the induction of somatic embryogenesis. To achieve this, the first step was to design a reliable and reproducible method for quantifying cAMP levels independently of the tissue and its physiological stage. We used the developed technique to determine the amount of cAMP in different *in vitro* plant culture models. Different cAMP extraction methods were evaluated. Once cAMP was extracted from the samples, the samples were lyophilized and then reconstructed with 50 µL of HPLC-grade water. The HPLC run conditions were based on those used by Zhao et al. Using this methodology, cAMP content was determined in *in vitro* cultures of *Daucus carota*, *Carludovica palmata*, *Coffea arabica*, *Solanum lycopersicum*, *Catharanthus roseus*, *Nicotiana tabacum*, *Stenocereus queretaroensis*, *Agave angustifolia*, and *Mangifera indica*. In *C. canephora* leaves, the cAMP concentration is 0.52 pmol g^{-1} FW. The identification of cAMP was performed using mass spectrometry and comparison with a standard. The data will be discussed in the context of the probable physiological role of cAMP in plants. Acknowledgment: To SECITIH (CBF-2025-I-758) for the research funding for this project.

Poster 180. Functional analysis of an uncharacterized member of the AP2/ERF transcription factor family in *Arabidopsis thaliana*

Henri Jimenez-Jimenez[1], Magali Morales-Melo[1], Cristina Betsabeth Miravel Gabriel[1], Tania Hernández-Hernández[2], Yolanda Durán-Medina, Nayelli Marsch-Martínez[*,1]

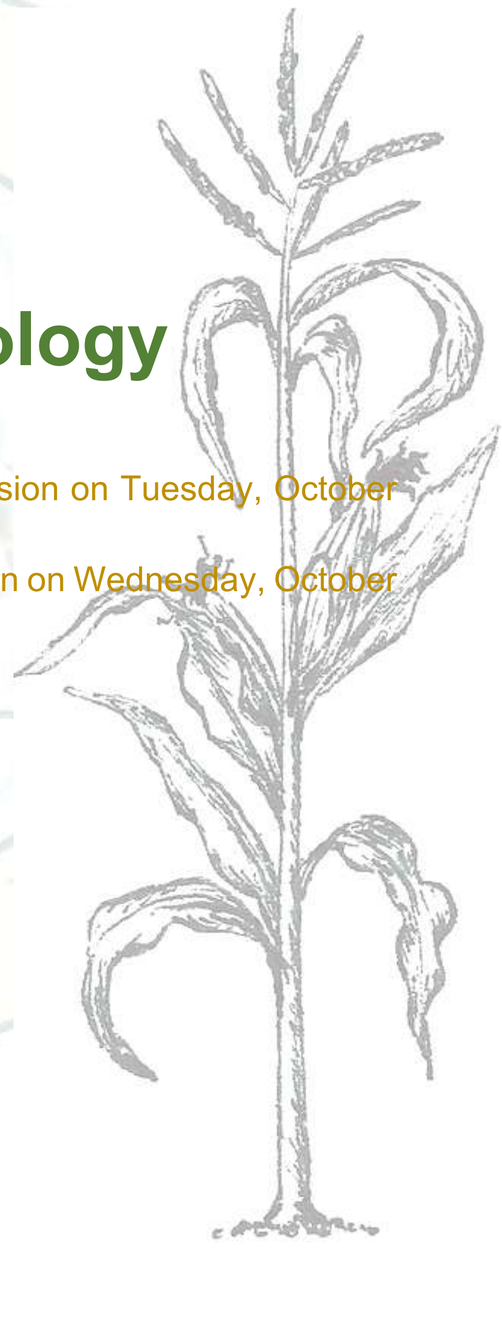
[1]Department of Biotechnology and Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute (Cinvestav), Km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, México.
[2]Desert Botanical Garden, 1201 N. Galvin Parkway, Phoenix, Arizona, Estados Unidos.
*Email: nayelli.marsch@cinvestav.mx

At least 1533 transcriptional regulators have been reported in the *Arabidopsis thaliana* genome, representing ~5.9% of its estimated total number of genes. Transcription factors belonging to the APETALA2/Ethylene Response Factors (AP2/ERF) superfamily regulate many biological processes such as growth, development of new organs, and hormonal and stress response. *DORNRÖSCHEN (DRN/ESR1), PUCHI, LEAFY PETIOLE (LEP)* and *BOLITA (BOL/DNRL/ESR2)* belong to the VIII-B subfamily of AP2/ERF transcription factors and have been found to be developmental regulators. However, the VIII-B subfamily includes more members that have been barely characterized to date. Therefore, the aim of this study is to use reverse genetics to elucidate the function of one of these genes, with yet unknown functions, in the development of *Arabidopsis thaliana* plants. For this, overexpression (using the 35S promoter), inducible overexpression and loss of function (knock-out using CRISPR-Cas9) lines were generated. Moreover, a reporter line (fusing its promoter to GUS-GFP) line was also obtained. This work is still in progress; however, the phenotypes observed at initial screens of overexpression and loss of function lines suggest that the gene may regulate developmental processes in flowers, seeds, leaves and roots. Accordingly, analyses of the reporter line showed that the gene is expressed in these organs. The latest results will be presented.

Evolutionary Plant Biology

Posters

Participants with even numbers will attend the session on Tuesday, October 21.
Participants with odd numbers will attend the session on Wednesday, October 22.



Poster 181. Toward understanding evolution of development in cacti

Ulises Rosas[1*], Joel Colchado-López[1,2], Delil A. Chincoya[1], Brenda A. López-Ruiz[1], Eduardo Torres-Flores[1,2], Luis Fernando Cisneros-Lemus[1,2]

[1]Jardín Botánico, Instituto de Biología, Universidad Nacional Autónoma de México, Tercer Circuito Exterior, Ciudad Universitaria, Coyoacán, Ciudad de México 04510, México.
[2]Posgrado en Ciencias Biológicas, Universidad Nacional Autónoma de México, CDMX, México, Ciudad Universitaria, 04510 Coyoacán.
*Email: urosas@ib.unam.mx.

The cactus family is diverse, displaying a wide range of shapes and forms. Their evolutionary novelties are gigantism, stem flattening, determination of the apical meristems, diversity of phyllotactic patterns, the extreme modification of the inflorescence, CAM photosynthesis, and the origin of an axillary meristem called the areole, which defines the family, among other traits. Despite cacti being of imperative biocultural and agronomical importance in Mexico, little attention has been paid to developing tools and genetic / genomic resources towards the understanding of key traits, some of them shared with other succulent plants, but some others unique to the family. Here we will present the general advances that we have made on developing *Mammillaria san-angelensis* as model system towards the understanding of traits such as determinate growth in the shoot and the root apical meristems, the evolution of the inflorescence, the bacterial microbiomes associated to the root of closely related *Mammillaria* species, and the whole genome sequencing and assembly of *M. san-angelensis*. Particularly within the bacterial microbiome, we will show the core bacterial microbiome that is recurrently recruited even when closely related species inhabit drastically different environments. Moreover, we will show the first draft of the *M. san-angelensis* genome, a resource that has allowed us the identification of homologue genes that we hypothesize are implicated in several aspects of plant development and evolution. Altogether we expect to draw attention to the study of fundamental processes occurring in cacti, developing models of Mexican relevance such as *M. san-angelensis*.This work was supported by PAPIIT DGAPA UNAM IN219725 to UR, SECIHTI Postdoctoral program to DAC, and UNAM-DGAPA Postdoctoral Program (POSDOC) to BALR.

Poster 182. **Molecular basis of inflorescence evolution in cacti**

Eduardo Torres-Flores[1,2] and Ulises Rosas[1*]

[1]Jardín Botánico, IB-UNAM. Tercer Circuito Exterior S/N. Ciudad Universitaria, 04510, Coyoacán, Ciudad de México, México.
[2]Posgrado en Ciencias Biológicas, Universidad Nacional Autónoma de México, Ciudad Universitaria, 04510, Coyoacán, Ciudad de México, México.
*Email: urosas@ib.unam.mx

The Cactaceae family, which comprises approximately 1,851 species, is divided into five subfamilies: Leuenbergerioideae, Pereskioideae, Maihuenioideae, Opuntioideae and Cactoideae. Leuenbergerioideae and Pereskioideae exhibit the most plesiomorphic traits within the family, including both vegetative and reproductive features. In many species of these subfamilies, inflorescences are evident and highly branched. In contrast, nearly all species within the Maihuenioideae, Opuntioideae and Cactoideae subfamilies, which display the most derived traits of the family, typically develop what has traditionally been described as solitary flowers. Thus reflects a dramatic evolutionary trend within the family toward a reduction in inflorescence complexity.In other angiosperms, inflorescence development involves the function of the PEBP TFL1-like transcription factors, such as TERMINAL FLOWER 1 (TFL1) in *Arabidopsis thaliana* and CENTRORADIALIS (CEN) in *Antirrhinum majus*. In this study, we analyze the structural and molecular evolution of the three genes encoding TFL1-like transcription factors across multiple cactus species. We identified all three TFL1-like genes in ten cactus species, and our results suggest that the second intron of cactus *TFL1* homologue gene tends to increase dramatically in length in the more derived lineages. These results point to a potential link between *TFL1* structure and inflorescence development, which can be further explored through functional analyses in cacti. Through this approach, we aim to provide a genetic explanation for the changes in inflorescence development observed along the evolutionary gradient of the cactus family. Grants: PAPIIT DGAPA UNAM IN219725.

Poster 183. Functional characterization of temperature-induced lipocalin involve plastid-nucleus retrograde signaling in *Marchantia polymorpha*

Carolina Yáñez-Domínguez[1], Leidi Itzuri Leon-García[1], Daniel Lagunas-Gómez[1], Kenny Alejandra Agreda Laguna[1], Omar Oltehua López, Luis Brieba-Castro[2] and Patricia León-Mejía[1*]

[1]Instituto de Biotecnología, Universidad Nacional Autónoma de México. Av. Universidad 2001, Col. Chamilpa. 62210, Morelos, México.
[2]Centro de Investigación y de estudios Avanzados del Politécnico Nacional, Unidad Irapuato LANGEBIO. Irapuato, Guanajuato. 36821, México.
[*]Email: patricia.leon@ibt.unam.mx

Lipocalins are proteins with a conserved barrel-shaped structure made of eight antiparallel β-strands, capable of binding small hydrophobic molecules such as retinoids and fatty acids. These proteins take part in a wide range of biological processes like cellular signaling, immune response, and other physiological processes. In plants, apocarotenoids (small hydrophobic molecules) derived from carotenes, function as plastid retrograde signals involved in regulating plastid homeostasis, as well as plant development and stress responses. The bryophyte *Marchantia polymorpha,* with gene reduction and distinct plastids, serves as an effective model for studying plastid retrograde signaling. In this work we propose that apocarotenoid signals require lipocalins as intermediates of plastids to nucleus communication, but this role is still unknown. The aim of the project is to characterize the functionality of apocarotenoid-binding proteins with structural similarity to lipocalins in *Marchantia polymorpha*, specifically the temperature-induced lipocalin TIL. These studies will allow us to understand the evolution of TIL lipocalin and their importance in plastid communication.

Poster 184. Gradual domestication of root traits in ancient maize shows an adaptation to the Neolithic soils from Tehuacán

Ivan Lopez-Valdivia[123], Alden C. Perkins[2], Hannah M. Schneider[23], Miguel Vallebueno-Estrada[14], James D. Burridge[2], Eduardo González-Orozco[1], Aurora Montufar[5], Harini Rangarajan[2], Kelly Swarts[4], Bruce F. Benz[6], Michael Blake[8], Jagdeep Singh Sidhu[2], Sergio Perez-Limon[2], Ruairidh J. H. Sawers[2], Rafael Montiel[1], Jean-Philippe Vielle-Calzada*[1] and Jonathan P. Lynch*[2]

[1]Unidad de Genómica Avanzada, Laboratorio Nacional de Genómica para la Biodiversidad, Cinvestav, Irapuato 36821 Guanajuato, Mexico
[2]Department of Plant Science, Pennsylvania State University, State College, PA 16802
[3]Department of Physiology and Cell Biology, Leibniz Institute for Plant Genetics and Crop Plant Research (IPK), OT Gatersleben, 06466, Seeland, Germany
[4]Gregor Mendel Institute of Molecular Plant Biology GmbH: Vienna, Wien, 1030,Austria;
[5]Subdirección de Laboratorios y Apoyo Académico, Instituto Nacional de Antropología e Historia, CP6010 Ciudad de México, Mexico
[6]Biology Department, Texas Wesleyan University, Fort Worth, TX 76105, USA
[7]Department of Anthropology, The University of British Columbia, Vancouver, BC, V6T 1Z4,Canada
*Email: vielle@cinvestav.mx; jpl4@psu.edu

Despite their importance in supplying nutrients, root traits related to maize domestication are not fully understood. We used laser ablation tomography to characterize the root architecture and anatomy of 5,300-year-old maize specimens from Tehuacán, Mexico, revealing exquisite preservation of their cellular organization. Anatomically, these specimens possessed thick, lignified outer cortical cells, similar to modern maize adapted to hard soils. However, their architecture lacked seminal roots, a trait characteristic of maize's ancestor, teosinte. This absence may be linked to mutations found in two genes important for seminal root development, indicating that some traits for drought adaptation were not yet fully present during early cultivation. To understand this evolutionary context, a broader reconstruction of root phenotypes and environments in the Tehuacán Valley over the last 18,000 years was conducted using ancient DNA, paleobotany, and modeling. This study revealed that a reduction in nodal root number and cortical sclerenchyma, promoting deeper rooting, was favored between 12,000 and 8,000 years before present (yrBP) due to rising atmospheric CO_2. The later advent of irrigation by 6,000 yrBP reinforced the utility of these traits by shifting nitrogen to the subsoil. Notably, an increase in seminal root number—the trait absent in the 5,300-year-old specimen—likely appeared around 3,500 yrBP. This development coincides with increased human population, agricultural intensification, and soil degradation. Together, these results suggest that root phenotypes enhancing performance under nitrogen stress were critical for maize adaptation to changing agricultural practices in the Tehuacán Valley.

Poster 185. The chloroplast lipocalin LCNP is required for photoprotection in *Marchantia polymorpha*

Daniel Lagunas-Gomez[1], Carolina Yañez-Dominguez[1], Kenny Alejandra Agreda-Laguna[1], Omar Oltehua-Lopez[1], Antolín Peralta-Castro[2], Luis G. Brieba[2] and Patricia León[1*]

[1]Instituto de Biotecnología, Universidad Nacional Autónoma de México, Av. Universidad 2001, Col., Chamilpa 62210, Morelos, México, 2 Laboratorio Nacional de Genómica para la Biodiversidad, Centro de Investigación y de Estudios Avanzados del IPN, Apartado Postal 629, Km. 9.6 Libramiento Norte Carretera, Irapuato-León, CP 36821 Irapuato, Guanajuato, México
[*]Email: patricia.leon@ibt.unam.mx

Light is essential for photosynthesis and supports most life on earth. However, too much light can lead to increased production of damaging reactive oxygen species as byproducts of photosynthesis. In some cases, photooxidative damage can cause pigment bleaching and death. To overcome that, the plants have resulted in the evolution of a remarkable suite of processes within the photosynthetic system to prevent cellular damage, such as, the process termed nonphotochemical quenching (NPQ), which play an important role in photoprotection and dissipation of excess absorbed light energy. However, little is known about the mechanics of photoprotection in mosses, in particular in *Marchantia polymorpha* a basal terrestrial plant with strategic evolutionary position. In this work, we analyzed the importance of the single gene plastid lipocalin (LCNP) and its possible involvement in photoprotection. Firstly, the subcellular location was observed thylakoid membranes and the tissue level in gemma and rhizoids and the gene mutation showed several defects, mainly huge thallus. These and additional results will be presented and discussed.

Poster 186. Plastid-to-nucleus signaling in *Marchantia polymorpha*: impact on plastid differentiation and plant development

Hernández-Muñoz, A., Agreda-Laguna, K.A., Yáñez-Domínguez, C., Oltehua, O., and León, P.*

Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, México. Avenida Universidad 2001.
*Email: patricia.leon@ibt.unam.mx

Plastids are essential plant organelles responsible for photosynthesis and the synthesis of vital metabolites. Their functionality depends on proper differentiation into specialized types, such as chloroplasts, a process that requires strict coordination between nuclear and plastid gene expression. This coordination is mediated by bidirectional communication: anterograde signaling (nucleus to plastid) and biogenic retrograde signaling (developing plastid to nucleus). The non-vascular plant *Marchantia polymorpha* possesses multiple plastid types, including chloroplasts, chloramyloplasts, and proplastids, suggesting that retrograde signaling was already established early in land plant evolution, making *Marchantia* a valuable model to study this process.Our work demonstrates that the single *MpGLK* transcription factor in *M. polymorpha* functions as a true orthologue of GLKs in vascular plants. GLK proteins are central regulators of retrograde signaling during chloroplast development. We show that *MpGLK* overexpression induces ectopic chloroplast formation, while loss of function produces plastid defects. Both overexpressing and mutant lines also display developmental phenotypes not previously associated with *GLK* activity. Notably, *MpGLK* harbors an additional N-terminal domain absent in other homologues, suggesting that ancestral GLKs possessed broader functions, possibly shared with other GARP family members, that were lost in angiosperms. Furthermore, we characterize the developmental expression pattern of *MpGLK*. Using *glk* mutants and overexpression lines, we have begun to dissect classical retrograde signaling pathways by perturbing carotenoid biosynthesis and plastid gene expression (PGE) with inhibitors such as norflurazon, lincomycin, and spectinomycin. Our results indicate that multiple retrograde signaling mechanisms are already present in *Marchantia*. These studies represent the first characterization of retrograde signaling responses in basal land plants and open new avenues for understanding the evolution of these pathways.

Poster 187. The transcriptional repressor VAL as a key regulator of development in Marchantia polymorpha

Frida Higareda Pacheco, Kenny Alejandra Agreda Laguna, Omar Oltehua López, Carolina Yáñez Dominguez, Patricia León Mejía*

Instituto de Biotecnología, Universidad Nacional Autónoma de México. Av. Universidad 2001, Col. Chamilpa. 62210, Morelos, México.
*Email: patricia.leon@ibt.unam.mx

In vascular, VAL (VP1/ABI3-LIKE) transcription factors are central repressors of gene expression during key developmental transitions, including seed maturation, germination and reproduction (Suzuki et al.,2007). Homologs of this factor are present in early land plants such as *Marchantia polymorpha,* yet their function in basal lineages remains poorly understood. Genome-wide of *Marchantia* identified more than one thousand genes containing RY motifs –the canonical sites of VAL proteins–suggesting a broad regulatory potential. To investigate *MpVAL* expression, we generated promoter-GUS fusions (MpVAL:GUS). Reporter activity was primarily detected in meristems and along the central vein, as well as in gemma cups and gemmae, but was absent in rhizoids. We conducted a functional characterization including both loss-of-function mutants and overexpression lines. Phenotypic analyses revealed that both genetic backgrounds exhibited delayed thallus growth compared to wild-type. Strikingly, overexpression lines displayed additional alterations, including increased number of rhizoids and premature germination of the gemmae within gemma cups, a phenotype not observed in either wild-type plants or mutants. Together, these results demonstrate that *MpVAL* functions as a key developmental regulator in *M. polymorpha*. Misexpression of VAL is sufficient to induce distinct morphological outcomes, underscoring the evolutionary conservation of VAL-mediated regulatory mechanisms across land plants.

Poster 188. Homologous Recombination as a Molecular Scaffold for Land Plant Adaptation: MpRAD51 at the Crossroads of Morphogenesis and DNA Damage Response

Elias Trujillo-Esquivel[1,2], Luis Manzanares[3], Annie Espinal-Centeno[4], Domingo Méndez-Alvarez[5], Luis G. Brieba[6*], and Alfredo Cruz-Ramírez[7*]

[1] Centro de Investigación y de Estudios Avanzados, Irapuato, México. email: jose.trujillo@cinvestav.mx [2] Centro de Investigación Científica y de Educación Superior de Ensenada, Ensenada, México. email: jtrujillo@cicese.edu.mx [7] Centro de Investigación y de Estudios Avanzados, Irapuato, México. Email: alfredo.cruz@cinvestav.mx

The transition of plants from water to land required major evolutionary innovations which allowed their adaptation to more genotoxic conditions. DNA repair via homologous recombination (HR) plays a crucial role in plant adaptation to harsh environments, and its evolutionary conservation underscores its essential function in this process. In this study we confirmed the conservation of genes involved in the main activities for HR across *Viridiplantae*. Among them, the recombinase RAD51 has a central role in the HR pathway due to its DNA binding, homology recognition and DNA displacement activities. Here we describe the functional characterization of the RAD51 ortholog in *Marchantia polymorpha*. In silico, the mutations within critical RAD51 domains and phosphorylation motifs identified offered insights into the possible regulatory adaptations of RAD51's function through the plant evolution. The main functions of MpRAD51 described in this pathway were confirmed biochemically. Nevertheless, beyond DNA repair, the loss of Mp*RAD51* showcased its role in the regulation of DNA-damage-induced plant development and differentiation, highlighting its importance in overcoming DNA damage and growth under genotoxic contexts. This study provides evidence of the evolutionary importance of HR in the transition from aquatic to terrestrial environments, where DNA repair mechanisms were crucial for plant adaptation and survival on land.

Plant Signaling

Posters

Participants with even numbers will attend the session on Tuesday, October 21.

Participants with odd numbers will attend the session on Wednesday, October 22.

Poster 189. **Responses of the microalga *Neochloris oleoabundans* to the self-DNA**

Edmundo Lozoya-Gloria[1]*, Mariana A. Rojas-Raya[1], Mónica A. Zárate-López[1], César A. Ávila-Hernández[1], José Luis Hernández-Flores[1], Nancy E. Lozoya-Pérez[2], Domancar Orona-Tamayo[2], Elizabeth Quintana-Rodríguez[2]

[1]Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN), Irapuato, Guanajuato, México.
[2]Centro de Innovación Aplicada y Tecnologías Competitivas (CIATEC), León, Guanajuato, México.
*Email: edmundo.lozoya@cinvestav.mx

Microalgae produce diverse metabolites and can adapt to diverse conditions. Under stress, microalgae increase metabolites production, like triacylglycerols (TAGs) among others. Self-DNA (sDNA), added or released by damaged cells, acts as a damage-associated molecular pattern (DAMP) inducing stress responses, and is recognized with a high degree of specificity. We compared the responses of *Neochloris oleoabundans* to sDNA, with those elicited by nonself-DNA (nsDNA), methyl jasmonate (MeJA), sodium bicarbonate ($NaHCO_3$), low-nitrogen condition (LN), and a combined treatment of LN+sDNA. We analyzed the peroxidase enzyme activity and polyphenols production, lipids and TAGs amounts, phytohormones, cell diameter, chlorophyll content and the expression of GPDH, LPAAT, and DGAT key genes involved in lipid biosynthesis. Results showed that peroxidase enzyme activity, polyphenols, lipids and TAGs were increased at 24 h with all elicitors, sDNA, LN and LN+sDNA, except with nsDNA. sDNA and MeJA increased gibberellic acid, isopentenyladenine, and benzylaminopurine at 24 h. Interestingly, sDNA increased lipid accumulation more rapidly than LN stress, while at 48 h LN treatment had the highest values followed by sDNA treatment. Simultaneous application of LN+sDNA resulted in less lipids than those produced by each treatment independently, suggesting distinct and possibly antagonistic stress mechanisms. Furthermore, sDNA increased the cell diameter but did not significantly modify chlorophyll content, except under LN conditions, where a decrease was observed. We propose that nucleic acids may be used as C:N:P source under stress conditions. GPDH, LPAAT, and DGAT genes expression showed variations in their transcript levels under different treatments and times. Our results indicated that the defense response after sDNA was different from LN treatments, suggesting a putative antagonistic effect of sDNA application on the LN response.

Poster 190. Revealing new mechanisms of maize Wee1 kinase regulation

Mingyar N. López-Hernández and Jorge M. Vázquez Ramos*

Facultad de Química, Departamento de Bioquímica, Universidad Nacional Autónoma de México. Ciudad de México, México, Av. Universidad 3000, Coyoacán, C.P. 04510.
*Email: jorman@unam.mx.

Cyclin-dependent kinase/cyclin (CDK/Cyc) complexes regulate cell cycle progression in eukaryotes by phosphorylating hundreds of substrates. Monomeric CDK lacks kinase activity, but cyclin association CDK promotes its basal activation. However, full activation requires phosphorylation of a highly conserved threonine residue on CDK, while in addition to being necessary for CDK activation in both humans and yeast, cyclins discriminate between different substrates. Kinase activity of CDK/cyclin complexes is also negatively regulated by phosphorylation of a highly conserved tyrosine residue. This regulatory mechanism is particularly important in vertebrates for ensuring successful mitosis and is mediated by Wee1 kinase, a highly conserved protein also present in plants; nonetheless, it is not clear whether cell cycle regulation by Wee1 is also conserved as it is in vertebrates. In fact it has been suggested that Wee1 function in plants is species-dependent. To investigate the conservation of Wee1 function in maize, in this work we have studied its effect on maize CDKA2;1a and CDKB1;1 kinases, two of the principal CDKs that control cell cycle progression in plants. Unlike results reported for *Arabidopsis thaliana*, we found that maize Wee1 phosphorylated CDKB1;1 and inhibited the kinase activity of CDKA2;1a and CDKB1;1 independently of its kinase activity. Inhibition was substrate-dependent, so we suggest that Wee1 associates with CDKs and modifies how CDKs recognize their substrates. In addition, Wee1 phosphorylation of CDKB1;1 inhibited its self-phosphorylation on the threonine residue necessary for its activation; surprisingly, this differentially affected substrate phosphorylation by CDKB1;1. Finally, both CDKA2:1a and CDKB1;1 affected differentially Wee1 activity.

Poster 191. **Maize Wee1 (ZmWee1) protein characterization during seed germination**

Elpidio García-Ramírez, Mingyar Neftalí López-Hernández and Jorge Manuel Vázquez-Ramos*

Facultad de Química, Departamento de Bioquímica, Universidad Nacional Autónoma de México, Ave. Universidad 3000, Col. Ciudad Universitaria, Coyoacán C.P. 04369, Ciudad de México.
*Email: jorman@unam.mx.

Maize seeds, as all orthodox seeds, maintains a very low metabolic status in the dry state, however, in the presence of water and under optimal conditions of light and temperature, reactivate metabolism and prepare for growth and proliferation through the cell division cycle, in which many protein factors participate; a key role in this process is played by the Wee1 protein kinase, which is ubiquitous during plant development and, through specific phosphorylation ($pThr^{14}$ $pTyr^{15}$) of Cyclin Dependent kinases (CDKs), negatively regulates cell cycle progression. In *silico* studies of ZmWee1 protein show that it has an intrinsically disordered region (contains 9.4% prolines). Wee1 potentially forms 2 disulfide bridges, and contains a PEST, an NLS and 11 CDK phosphorylation sites that could be related to 7 short lineal motifs that putatively recognize the different cell cycle cyclins. Antibodies raised against a ZmWee1 fragment recognized a ≈70 kDa protein in cell extracts of dry maize embryo axes, that decreased at 6h of imbibition, increased at 12h and then decreased again at 18-36 h. In nuclear extracts ZmWee1 was detected in nuclei from dry seeds, increased at 6h and then decreased at 12-36h. We are now focused on the study of its phosphorylation pattern using ZmCDKA-Cyclin D2-CKS pulled down complexes as the kinase source obtained at different germination times, simultaneously it will be determined the inhibitory phosphorylation status of the CDKs and the ZmWee1 kinase activity in anti-Wee1 immunoprecipitation assays during the first 36h of imbibition in nuclei.

Poster 192. **Efecto del ácido ferúlico en el crecimiento y defensa en la planta de *Arabidopsis thaliana***

Fernanda Federica Fernández-Licona[1], Randy Ortiz-Castro[1]*, Luis Arturo Ibarra-Juárez[1], José Francisco Castillo-Esparza[2]

[1]Red de Estudios Moleculares Avanzados, Instituto de Ecología, A.C. Xalapa, México. Carretera antigua a Coatepec 351, Col. El Haya, Xalapa, Veracruz. CP 91073.
[2]Red de Biodiversidad y Sistemática, Instituto de Ecología, A.C. Xalapa, México. Carretera antigua a Coatepec 351, Col. El Haya, Xalapa, Veracruz. CP 91073.
*Email: randy.ortiz@inecol.mx

El ácido ferúlico (AF) es un compuesto fenólico presente en diversas especies de plantas y se localiza en la pared celular. Este metabolito participa en interacciones alelopáticas como un regulador del crecimiento, provocando efectos como la inhibición del crecimiento de las raíces, la germinación y la expansión foliar. Por otro lado, promueve la defensa de la planta ante patógenos y plagas. Sin embargo, su efecto directo sobre las rutas hormonales en la planta modelo *Arabidopsis thaliana* permanece poco caracterizado. En el presente trabajo se evaluó el efecto del AF exógeno sobre el crecimiento y la expresión de genes marcadores de señalización hormonal en *A. thaliana* cultivada *in vitro*. Se cultivaron plántulas en medio Murashige & Skoog (MS) suplementado con AF a diferentes concentraciones (10, 20, 40, 80, 160 y 320 μM). Se emplearon líneas reporteras relacionadas con la respuesta a las auxinas (*DR5:uidA, BA3:uidA*), citocininas (*ARR5:uidA*), ácido jasmónico (*LOX2:uidA, JAZ1:uidA*) y ácido salicílico (*PR1a:uidA*). Los resultados mostraron que el FA induce un efecto de inhibición en la longitud de la raíz primaria (LRP) e inducción de raíces laterales en una manera dosis dependiente. Además, de la promoción en la formación de RLs se observó un incremento en el contenido de clorofila y una disminución en la biomasa foliar y radicular dosis dependiente. Los efectos mostrados en la formación de RLs se relacionan con un incremento en los niveles de auxinas como puede observar en el incremento en los niveles de expresión de la línea reportera *DR5:uidA* de *Arabidopsis thaliana* sugiriendo que los efectos mostrados por el AF son dependientes de una vía de señalización de auxinas. El análisis de las demás vías hormonales relacionadas con el crecimiento y defensa brindarán una mayor compresión en el entendimiento de los efectos del AF en la planta modelo de *Arabidopsis thaliana*.

Poster 193. Identification and characterization of the NPR3 protein interactome during defensive responses in *Arabidopsis thaliana*

Everardo Jair Flores Cuevas[1,2], Israel Maruri López[3], Mario Alberto Serrano Ortega[1*]

[1]Genómica Funcional de Eucariotes; Centro de Ciencias Genómicas – Universidad Nacional Autónoma de México.
[2]Programa de maestría y doctorado en ciencias bioquímicas; Instituto de Biotecnología– Universidad Nacional Autónoma de México.
[3]Center for Desert Agriculture, Biological and Environmental Science and Engineering Division (BESE), King Abdullah University of Science and Technology (KAUST), Thuwal, Saudi Arabia.
[*]Email: serrrano@ccg.unam.mx

Plants have evolved complex defense strategies involving proteins, hormones, and regulatory elements to recognize and respond to microbial threats. Salicylic acid (SA) is a key hormone orchestrating response to biotic and abiotic stresses, playing central roles in effector-triggered immunity (ETI) and systemic acquired resistance (SAR). The NPR protein family, particularly NPR1, NPR3, and NPR4, functions as SA receptors and is critical in regulating immune responses. In *Arabidopsis thaliana*, NPR1 serves as a transcriptional coactivator and key regulator of SA-responsive genes. In contrast, NPR3 and NPR4 act as substrate adaptors for the CRL3 complex and repress immune-related gene expression. Recent studies have revealed that NPR proteins form biomolecular condensates in both the cytoplasm and nucleus, a process enhanced by SA accumulation. Cytoplasmic condensates of NPR1 (SA-induced condensates or SINCs) have been linked to promoting cell survival near infection sites, yet the function and composition of nuclear NPR3 condensates remain poorly understood. Despite advances in understanding NPR function, the identities of proteins interacting with NPR3 are still largely unknown. Defining these interactions may reveal novel mechanisms governing plant immune regulation. This project aims to identify NPR3-associated proteins using two complementary approaches: the Split Ubiquitin system, a membrane-based yeast two-hybrid technique, and TurboID, a proximity labeling method that enables detection of transient and stable protein interactions in vivo. Together, these tools will facilitate mapping the NPR3 interactome and provide insight into its role in SA-mediated signaling. By integrating genetic and proteomic methods, this study will contribute to the functional characterization of NPR3 and enhance our understanding of the protein networks that fine-tune salicylic acid signaling in plants.

Poster 194. *In planta* localization and response to cytokinin during gynoecium development

Solano-Garcia, Fernando[1,2]; Ramos-Pulido, Juan[2]; de Folter, Stefan[2*]; Marsch-Martínez, Nayelli[1*]

[1]Departamento de Biotecnología y Bioquímica, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (Cinvestav), Km. 9.6 Libramiento Norte, Carretera Irapuato-León, CP 36824 Irapuato, Gto., México
[2]Unidad de Genómica Avanzada (UGA-Langebio), Cinvestav, Km. 9.6 Libramiento Norte, Carretera Irapuato-León, CP 36824 Irapuato, Gto, Mexico.
[*]Email: nayelli.marsch@cinvestav.mx; stefan.defolter@cinvestav.mx

The gynoecium is the female reproductive organ of angiosperms, and its correct development involves finely tuned hormonal signaling pathways. Among these hormones, cytokinins (CKs, adenine-derived molecules) play a central role in cell proliferation, meristem maintenance and organ patterning. In order to understand CK dynamics during gynoecium development, we investigated their localization and signaling patterns. To assess CK localization in developing gynoecium, we performed immunolocalization of trans-Zeatin (tZ), the most abundant naturally occurring CK. In parallel, to determine CK signaling patterns, we used the synthetic reporter line TCSn::GFP, which responds to CKs via the Two Component Signaling system (TCS). Our results show that tZ accumulates in specific tissues within the gynoecium which closely matches the TCSn::GFP signal, suggesting a relationship between localization and signaling output of CKs. Additionally, *in situ* hybridization assays revealed that expression of the transcription factor NO TRANSMITTING TRACT (NTT; previously suggested by our lab to regulate CK metabolism genes) overlaps with tZ accumulation and CK signaling, pointing to a possible role of NTT in modulating local CK homeostasis. Taken together, these results provide new insights into spatial coordination of CK activity during gynoecium development. Understanding the interplay between hormone distribution, perception, and gene regulation may help elucidate the mechanisms that govern reproductive organ development in plants.

Poster 195. Dual phosphorylation as a regulatory mechanism for SnRK1α1 kinase activity

Alejandra Ávila[1], Aitana López[1], Jacquelynne Cervantes[2], Rogelio Rodríguez-Sotres[1], Eleazar Martínez-Barajas[1] and Patricia Coello[1]*

[1]Departamento de Bioquímica, Facultad de Química, UNAM, México.
[2]Depto. Microbiología e Inmunología, FMVZ, UNAM, Mexico.
*Email: pcoello@unam.mx

SnRK1 (SNF1-Related Kinase 1) is a central energy sensor in plants, coordinating responses to metabolic stress, growth, and development by balancing energy homeostasis under fluctuating conditions. While phosphorylation at Thr175 in the activation loop was historically considered essential for SnRK1 activity, this study demonstrates that dual phosphorylation at Thr175 and Ser176 is indispensable for full kinase functionality. The mutational analysis (T175A, S176A, and TS/AA mutants) showed loss of kinase activity in recombinant proteins and impaired complementation in *Δsnf1* yeast and failed to restore full activity in *Arabidopsis* protoplasts. In addition, the AlphaFold modeling of the SnRK1α1 catalytic domain complexed with the AMARA peptide substrate revealed that pSer176 forms salt bridges with Arg15 of AMARA, positioning it optimally in the active site and stabilizing the kinase structure. Antibody-based profiling in *Arabidopsis* revealed at 8 am the phosphorylation at Thr175 fluctuates throughout the day in contrast the phosphorylation at Ser176 is apparently more stable thoughout light/dark cycles. Under heat stress (37°C) non-phosphorylatable mutants (TS/AA) accumulated cytoplasmic aggregates, suggesting phosphorylation regulates protein turnover and stress adaptation. This study shows how adjacent phosphorylation sites synergistically control substrate binding, structural stability, and stress-induced degradation in kinase regulation. Funding: PAPIIT: IN201922 y IN208425.

Poster 196. **Analysis of the interactions between the bHLH transcription factor AIB/JAM1 with MYB factors and MED14 in *Arabidopsis thaliana***

Elizabeth Córdoba* and María-Isabel Cruz-López

[1]Departamento de Biología Molecular de Plantas, Instituto de Biotecnología, Universidad Nacional Autónoma de México-Campus Morelos, 62210.
*Email: elizabeth.cordoba@ibt.unam.mx

Through its function as a signaling molecule, glucose (Glc) regulates gene expression at transcriptional level. It is now recognized that plants perceive and transduce sugar-derived signals by master regulators. However, most of the molecular downstream components that constitute the signaling pathways, as well the regulatory mechanisms, remain unknown. Previously, we identified the transcription factor AIB/JAM1(ABA INDUCIBLE bHLH/JASMONATE-ASSOCIATED MYC2-LIKE1) by its binding to a promoter region of the Glc-regulated *STP1* (*SUGAR TRANSPORTER PROTEIN 1*) gene, where sugar-responsive *cis*-elements were located, and then considered AIB/JAM1 as a downstream regulatory component in the Glc signaling pathway. The characterization of AIB/JAM1 allelic mutants (*jam1-1* and *jam1-2*) revealed that they are hypersensitive to Glc; they showed arrested growth, delayed germination, and lack of greening at 4% of Glc, a condition under which wild-type plants develop and grow properly. A transcriptomic analysis of the *jam1-2* mutant grown in presence of Glc, showed that the expression of genes involved in photosynthesis, cellular respiration, and in metabolism, transport, storage, and signaling of sugars were affected. These results demonstrated that AIB/JAM1 participates in the regulation of genes involved in the transition from heterotrophic to autotrophic state, during early stages of seedling establishment. To determine the mechanism of Glc expression regulation through AIB/JAM1, we explore its interaction with MYB-type transcription factors and with the MED14 subunit of the MEDIATOR complex. Structural complexes were revealed by modeling and the interactions between these factors analyzed by BiFC (Bimolecular Fluorescence Complementation) assays. The interactions between these transcriptional factors reveal the conformation of potential regulatory complexes that could mediate responses to sugars at the transcriptional level.

Poster 197. **Identification of β-glucosidase in maize coleoptiles using dextran-β-lactose derivatives**

Isabel Guerrero Cabrera[1], Liliana Hernández García[1], Eduardo Pérez-Campos[1], José Luis Hernández Morales[2], Alexis Martínez Barras[1], Carlos Alberto Matías Cervantes[2] Margarito Martínez Cruz[1]

[1]Tecnológico Nacional de México/ Instituto Tecnológico de Oaxaca/Unidad Académica de Posgrado/Laboratorio de Bioquímica. Av. Ing. Víctor Bravo Ahuja No. 125 Esq. Calzada Tecnológico. Ex-hacienda de Aguilera S/N, Sur San Felipe del Agua, Oaxaca, Oax.
[2]Facultad de Medicina Universidad Autónoma Benito Juárez de Oaxaca

The application of glycosylated and biotinylated dextran derivatives constitutes a valuable tool for the identification of β-glucosidase. This hydrolase was isolated and purified from 5-day-old maize coleoptiles, and an assay employing glycosylated and biotinylated derivatives was implemented. Maize coleoptiless (Asgrow) were collected and homogenized. Purification was performed following the protocol described by Esen (1992). The obtained fractions were analyzed by both denaturing SDS-PAGE and native electrophoresis. The purified protein was separated using native PAGE and subsequently transferred onto a nitrocellulose membrane. The membrane was incubated with the dextran-lactose-biotin derivative and developed accordingly (3). A single band of approximately 60 kDa was detected, corresponding to the β-glucosidase-β-lactose interaction, demonstrating that the methodology employed herein is effective for the identification of this hydrolase.

Poster 198. Transcriptomic analyses reveal an increase in the expression of genes related to biotic response in *bdx-1* seedlings

José Eduardo Pineda Hernández[1], Laura Figueroa Corona[2], Esther Zúñiga Sánchez[3], Alicia Gamboa de Buen[1*]

[1]Instituto de Ecología, Universidad Nacional Autónoma de México (UNAM), CDMX, México.
[2]Laboratorio Internacional de Investigación sobre el Genoma Humano (LIIGH), Universidad Nacional Autónoma de México (UNAM), Juriquilla, Querétaro, México.
[3] Facultad de Ciencias, Universidad Nacional Autónoma de México (UNAM), CDMX, México.
*Email: agamboa ecologia.unam.mx

At4g32460/BIIDXI is a gene of the DUF642 family that encodes a protein located in the primary cell wall that interacts in vitro with a Pectin methyl esterase (AtPME3). In Arabidopsis thaliana, the loss-of-function mutants for this gene (*bdx-1*) present alterations on embryonic development, seed production, germination, and fruiting. The activity of PME; an enzyme that removes methyl groups from homogalacturonans, is lower in seedlings and other tissues of *bdx-1*. At the cellular level, the endosperm cell wall exhibits a higher degree of methyl esterification, which prevents proper embryo folding. The in vitro interaction of rice DUF642 proteins with PMEs. Based on these results, we decided to perform a morphological analysis and a transcriptomic analysis of *bdx-1* seedlings. These seedlings presented a higher percentage of damage in the cotyledons and a lower number of true leaf primordia compared to WT col seedlings. The transcriptomic results indicated that there is a change in the expression of genes involved in response to biotic stress in *bdx-1* seedlings. The expression of genes related to calcium transport, including glutamate receptors (GLR2.4 and GLR2.8) and protein kinases (CRK36), were increased, while the expression of genes related to auxin synthesis decreased significantly. An increase in the expression of *Polygalacturonase Lateral Root* (*PGLR*), involved in oligalacturonides production, was also detected. These results suggest that BIIDXI could be involved in signaling processes in response to alterations in cell wall structure caused by stress during cell morphogenesis or elongation and environmental conditions. Recently, it has been shown that the protein DUF642 ENHANCED XYLEM AND GRAFTING1 (EXG1/ At3g08030) is involved in the integration of information in response to stress.

Poster 199. Multi-omic characterization of plant-derived extracellular vesicles from Mexican medicinal plants

Francisco Pérez-Aguirre[1], Noé Valentín Durán-Figueroa[1*], Jesús Agustín Badillo-Corona[1] and Héctor Quezada-Pablo[2]

[1]UPIBI, National Polytechnic Institut. CDMX, México, Av. Acueducto, La Laguna Ticoman, 07340.
[2]Hospital Infantil de México Federico Gómez (HIMFG), CDMX, México, Calle Dr Márquez 162, 06720.
[*]Email: nduranf@ipn.mx

Plant-derived extracellular vesicles (EVs), also referred to as exosomes, are nanoscale structures involved in intercellular communication through the transfer of proteins, RNAs, lipids, and metabolites. While initially described in animals, recent evidence suggests that EVs of plant origin may also play a critical role in modulating biological responses, including immune system regulation. In this project, we explore the immunomodulatory potential of EVs obtained from *Arabidopsis thaliana* and selected Mexican medicinal plants with recognized anti-inflammatory properties, including *Echinacea purpurea*, *Aloe vera*, *Curcuma longa*, and *Zingiber officinale*. EVs were isolated by differential ultracentrifugation and analyzed through proteomic, metabolomic, and transcriptomic approaches, employing mass spectrometry, nuclear magnetic resonance, and next-generation sequencing. Preliminary results show that vesicle preparations reached concentrations above 10 particles/mL, with protein yields greater than 4 mg/mL and average sizes ranging from 160–170 nm. Protein abundance was proportional to vesicle numbers, and initial proteomic profiling suggests that specific proteins could underlie the immunomodulatory effects of these vesicles. Ongoing metabolomic and transcriptomic analyses, together with in vitro assays using immune cell models, will further define their functional potential. Overall, this study provides the first multi-omic insight into plant-derived EVs from medicinal species, highlighting their promise as novel, safe, and effective platforms for therapeutic development and vaccine adjuvants.

Poster 200. Sanguinarine accumulation is restricted to necrotic lesions caused by fungus infection in leaves of *Argemone mexicana* L (Papaveraceae)

José Ignacio Laines-Hidalgo, Edgar Adrián González-Martínez, José Armando Muñoz-Sánchez, Felipe Vázquez Flota*

Unidad de Biología Integrativa, Centro de Investigación Científica de Yucatán, *felipe@cicy.mx

Argemone mexicana, known as Mexican prickly poppy, accumulates more than twenty benzyisoquinoline alkaloids, mainly berberine and sanguinarine. Whereas berberine is equally distributed in aerial and underground parts, sanguinarine is restricted to roots[1]. However, after fungal infection, the presence of sanguinarine is also detected in leaves, suggesting a role for this alkaloid in the plant-fungus interaction[2].

In this study, we monitored the distribution of sanguinarine through the leaf blade in fungus infected leaves of *A. mexicana*. Leaves displaying necrotic lesions, caused by infection, were collected and alkaloids analyzed by chromatography in specific regions: the necrotic lesion, that adjacent to the lesion, and from distal areas. Sanguinarine was only detected in the necrosed area whereas berberine accumulation remained unaffected by the fungal lesion.

Interestingly, sanguinarine was also accumulated in developmentally desiccated leaves. However, in the absence of fungus induced injuries, the alkaloid was uniformly distributed across the leaf blade. In this way, although the presence of sanguinarine in leaves seems related to tissue decay, its accumulation during fungal infection remains restricted to the colonized area. These results will be discussed in relation with process of senescence and cell death by pathogen infection.

Supported by SECIHTI, CBF-2023-2024-1879

1) Vázquez Flota F et al (2018) Rev Fitotec Mex, 41: 13-21
2) Hernández-Eleria GC et al (2025) J Plant Pathol/10.1007/

Poster 201.Morphogenetic and Phytochemical Study of *Bixa orellana* L. Cultivars from Southern Mexico

Aguilar-Espinosa M[1*], Pech-Hoil R [1*],Vallejo-Cardona A[2], Rodríguez-Campos J[2] & Rivera-Madrid R[1*].

[1]Centro de Investigación Científica de Yucatán, A. C. Calle 43 No. 130 x 32 y 34, Chuburná de Hidalgo; CP 97205, Mérida, Yucatán, México.
[2]Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A.C. Av. Normalistas 800. Colinas de la Normal. 44270 Guadalajara, Jalisco. México.
*Contact: mgf@cicy.mx

Bixa orellana L., known as achiote in Mexico, is a shrub native to Mesoamerica that has been cultivated since pre-Columbian times for its bixina pigment and medicinal properties. This study evaluates the morphological and phytochemical variability of *B. orellana* in southeastern Mexico, focusing on the quantification of bixina and geranylgeraniol (GGOH), a terpenoid with therapeutic potential. Morphological descriptors of flowers, fruits, and seeds were used to characterize the samples. The seeds were analyzed using high-performance liquid chromatography (HPLC) and gas chromatography coupled with mass spectrometry (GC-MS). GGOH profiles were validated through derivatization and spectral comparison with the NIST database. The cluster analysis identified two morphological groups, one of which exhibited favorable agronomic characteristics, such as larger fruits, higher bixina content (up to 4.03 $mg \cdot g^{-1}$ FW), and GGOH (up to 0.252 $mg \cdot g^{-1}$ FW). The principal component analysis (PCA) showed that fruit size and bioactive compounds (bixina and geranylgeraniol) were associated with the first component, while seed weight, fruit width, and the fruit length/width ratio were related to the second component. This study demonstrates the vast, untapped genetic and phytochemical diversity of this species, advancing the identification and selection of highly productive genotypes, thereby contributing to the development of crops with superior agronomic traits and therapeutic properties.