57th Annual Meeting of the Phytochemical Society of North America

August 4-8, 2018
Universidad Autónoma de San Luis Potosí
(Autonomous University of San Luis Potosí)
San Luis Potosí, México

TABLE OF CONTENTS

PSNA 2018 Local Organizing Committee

- Manuel Fermín Villar Rubio (Universidad Autónoma de San Luis Potosí)
- Jorge Fernando Toro Vázquez (Universidad Autónoma de San Luis Potosí)
- Ma. Guadalupe Beatriz Zapata Zapata (Universidad Autónoma de San Luis Potosí, Facultad de Ciencias Químicas)
- Denisse Atenea de Loera Carrera (Universidad Autónoma de San Luis Potosí, Facultad de Ciencias Químicas)
- Rodolfo González Chávez (Universidad Autónoma de San Luis Potosí, Facultad de Ciencias Químicas)
- Lluvia Itzel López López (Universidad Autónoma de Coahuila)
- Cristina Cabrera González (Universidad Autónoma de San Luis Potosí, Facultad de Ciencias Químicas)
- Elva Ceyli Medina Velázquez (Universidad Autónoma de San Luis Potosí, Facultad de Ciencias Químicas)
- Marcela Esmeralda Cervantes Rojas (Universidad Autónoma de San Luis Potosí, Facultad de Ciencias Químicas)
- Vicente Osvaldo Ordorica Rivera (Universidad Autónoma de San Luis Potosí, Facultad del Hábitat)

PSNA 2018 Executive Advisory Board

- Bernd M. (Mark) Lange (President, Washington State Univ.)
- Deyu Xie (Vice President & President Elect, North Carolina State Univ.)
- Mark Berhow (Immediate Past President, USDA, ARS, NCAUR)
- Argelia Lorence (Secretary, Arkansas State Univ.)
- Dhirendra Kumar (Treasurer, East Tennessee State Univ.)
- Lloyd W. Sumner (Univ. of Missouri, Columbia)
- Franck Dayan (Colorado State Univ.)
- Elvira Demejia (Univ. of Illinois)
- Reinhard Jetter (Univ. of British Columbia)
- Dejan Nikolic (Univ. of Illinois at Chicago)
- Cecilia McIntosh (East Tennessee State Univ.)
- Li Tian (Univ. California, Davis)
- Fred Stevens (Oregon State Univ.)
- Michael Sullivan (USDA, ARS)
- Dorothea Tholl (Virginia Polytechnic Institute and State Univ.)
- Toni Kutchan (Danforth Center)
- Armando Alcazar Magana (Oregon State Univ.)
- Soheil Mahmoud (Univ. British Columbia)
- Philipp Zerbe (Univ. California, Davis)
- Björn Hamberger (Michigan State Univ.)

Thank you to PSNA 2018 Sponsors!

CONAGEN

Agilent

JEOL

SHIMADZU
Excellence in Science

CTR®
SCIENTIFIC

Distribuidora Rodval, s.a. de c.v.

EbioQUIM

QRO
Químicos y Reactivos
de Querétaro S.A. de C.V.

SIETE BARRIOS
cerveza artesanal

IS
ANALÍTICA

CERVECERÍA SAN JOSÉ

SLP
PROSPEREMOS JUNTOS
Gobierno del Estado 2015-2021
CONSEJO POTOSINO DE CIENCIA Y TECNOLOGÍA

SLP
PROSPEREMOS JUNTOS
Gobierno del Estado 2015-2021
SECRETARÍA DE TURISMO

Restaurants

American

Carl´s Jr. Morales
Venustiano Carranza Av. 2455
444 833 0295

Chili's
Venustiano Carranza Av.1105
418 103 6563

Domino´s
Venustiano Carranza Av. 1705
444 817 1155

Little Caesars Pizza
Venustiano Carranza Av. 2380
444 813 3165

Wing Stop
Venustiano Carranza Av. 1905
444 211 5891

Asian

Oriental Grill
Cordillera Real 255
444 825 6828

Saigon Brasserie
Juan de Oñate 620
444 811 2626

Shiroi-ie
Venustiano Carranza Av.1335
444 128 9990

Bars & Pubs

Frailes
Universidad Av. 165
444 812 7375

Kaktuz Gourmet & Cantina
Real de Lomas 350
Int 209 Plaza Covalia
444 388 025

Loreto Bar
Huasteca 305
444 127 5567

La Oruga y la Cebada
Universidad Av. 350
444 812 4508

La Traza
Huasteca 305 Int. 27
444 167 4419

Coffee & Bakery

Los Bisquets Obregón
Venustiano Carranza 2020
444 817 7612

Las Castañas
Venustiano Carranza Av. 1325
444 833 5040

El Encanto Caffé
Calle Naranjos 360
444 426 0219

The Italian Coffee Company
Arista 1130
444 811 3745

Starbucks
Venustiano Carranza Av. 1996
01 800 288 0888

Vips
Venustiano Carranza Av. 2090
vips.com.mx

Ice Cream & Smoothies

TipiÓka
Huasteca 295
444 825 4647

Dairy Queen
Montes Aconcagua 301
444 244 0673

Santa Clara
Venustiano Carranza Av. 1400
01 800 101 0800

Italian

Bella Italia
Venustiano Carranza Av.
1809-B
444 817 0146

Goodfellas
Venustiano Carranza Av. 2355
444 244 5008

Tiberius
Venustiano Carranza Av. 1047
444 817 2126

International Cuisine

Cielo Tinto
Venustiano Carranza Av. 700
444 814 0040

TrasHumante
Venustiano Carranza Av. 2030
444 811 6093

La Gran Vía
Venustiano Carranza Av. 560
444 812 3633

Mexican

Antojitos El Pozole
Alfredo M. Terrazas 600
444 813 1100

El Artista del Taco
Nereo Rodríguez Barragán
1380-A8. Plaza del Valle
444 8136255

El Atrio Restaurante
Avanzada 480
444 379 3171

El México de Frida
Valentín Gama 646
444 811 4603

El Sauce
Gustavo Adolfo Beker 101-A
444 813 6804

La Nueva Parroquia
Venustiano Carranza Av. 780
444 814 7032

Casa Epitafio
Río Kennedy 165
444 962 9932

Richards
Venustiano Carranza Av. 2325
444 841 3400

Seafood

La Viga
Cordillera Real 403
444 825 0511

Santo Mar
Guadalcazar 116
444 811 6243

Steakhouse

Bife El Almacén
Visa Hermosa 116
444 151 6010

Casablanca
Venustiano Carranza Av. 1325
444 811 7000

Gaucho Argentino
Cuauhtémoc Av. 1325
444 813 2850

Vegetarian

Go Free Healthy Bar & Shop
Montes Aconcagua 301-7
444 246 1869

Culture and leisure

Centro de las Artes de San Luis Potosí
Calzada de Guadalupe 705
444 137 4100

Museo Interactivo Laberinto de las Ciencias y las Artes
Boulevard Antonio Rocha
444 102 7800

Museo Nacional de la Máscara
Villerías No 2
444 812 3025

Centro Comercial El Dorado
Nereo Rodríguez Barragán Av. 450
444 811 2707

Citadella
Salvador Nava Av. 3125
444 811 7089

Plaza Covalia
Real de Lomas 350
444 198 5790

Map from FCQ to Hotel Real Plaza

Faculty Map


E
N
C
D
F
A
B
G
H
CCTD
Library
I
P
M
N
L
K
J
Building Location
FCQ Library
Rogelio Jiménez Auditorio
P
Teacher parking

Posgrado
Facultad de Ciencias Químicas

Para mayor información comunicarse a:

Coordinación General de Posgrado
coordposgrado@cq.uaslp.mx
Tel: +52 (444) 826 24 40 al 46 Ext. 6540

UASLP
Universidad Autónoma
de San Luis Potosí

FACULTAD DE
CIENCIAS QUÍMICAS

PSNA 2018 Conference Program August 4-8, 2018

All oral presentation will be held in the Rogelio Jiménez Auditorium at Facultad de Ciencias Químicas

Saturday, August 4, 2018

6:00 pm-8:00 pm Welcome Reception, Real Plaza Hotel
Sponsored by:

Químicos y Reactivos de Querétaro S.A. de C.V.

Distribuidora Rodval, s.a. de c.v.

Sunday, August 5, 2018

8:00 am-5:00 pm Conference Registration, Rogelio Jiménez Auditorium Lobby

8:30 am-9:00 am **Opening Ceremony**

9:00 am-9:45 am **Plenary Symposium: Luis Manuel Peña**, Unidad de Biotecnología, Centro de Investigación Científica de Yucatán
Metabolomics in natural product research

9:45 am-10:00 am Break

Symposium I: Plant-environment interactions

10:00 am-10:45am **Keynote: Martin Heil,** Departamento de Ingeniería Genética, CINVESTAV-Irapuato
Volatile organic compounds and extracellular DNA induce immunity in plants

10:45 am-11:15 am David Gang, Washington State University, *Metabolome changes in individual insects infected with Candidatus Liberibacter species from citrus and solanaceaous crops*

11:15 am-12:00 pm **Neish Awardee Presentation: Jennifer Wisecaver**, Purdue University, *Co-expression not co-location links genes to specialized metabolic pathways in plants*

12:00 pm-12:30 pm Sangeeta Dhaubhadel, London Research and Development Centre, *Soybean isoflavonoids and disease resistance*

12:30 pm-1:30 pm Lunch, Facultad de Ciencias Químicas Garden
Sponsored by:

SLP PROSPEREMOS JUNTOS Gobierno del Estado 2015-2021 | Consejo Potosino de Ciencia y Tecnología

Symposium II: Plant-environment interactions

1:30 pm-2:15pm **Keynote: Juan Francisco Jiménez Bremont,** División de Biología Molecular, Instituto Potosino de Investigación Científica y Tecnológica AC
Functional analysis of Glycine-Rich Domain Proteins (AtGRDP1 and AtGRDP2) in plant abiotic stress tolerance

2:15 pm-3:00 pm **Neish Awardee Presentation: Colleen Doherty**, North Carolina State University, *Time, temperature, and the secret life of plants at night*

3:00 pm-3:20 pm Short company talks
Sponsored by:

3:20 pm-3:45pm Break

Symposium III: Molecular and industrial phytochemistry

3:45 pm-4:30 pm **Keynote: Angel Alpuche Solis,** División de Biología Molecular, Instituto Potosino de Investigación Científica y Tecnológica AC
Plants and algae as bioreactors for biopharmaceutical production against respiratory diseases

4:30 pm-5:00 pm Yezhang Ding, University of California San Diego, *Multiple genes recruited from hormone pathways underlay maize defense*

5:00 pm-7:00 pm Dinner on your own

7:00 pm-9:00 pm Poster Session I with Refreshments, Facultad de Ciencias Químicas Library
Sponsored by:

Monday, August 6, 2018

8:00 am-5:00 pm Conference Registration, Rogelio Jiménez Auditorium Lobby

9:00 am-9:45 am **Plenary Symposium: Peter Facchini,** Department of Biological Sciences, University of Calgary
Opium poppy: An update

9:45 am-10:00 am Break

Symposium IV: Biochemistry

10:00 am-10:45 am **Keynote: Alisa Huffaker,** Section of Cell & Developmental Biology, University of California San Diego
Integrated genetic and biochemical approaches to define layers of maize immune metabolism

10:45 am-11:15 am Mark Lange, Washington State University, *Structure-function of monoterpene functionalization reactions*

11:15 am-12:00 pm **Elsevier Awardee Presentation: Philipp Zerbe**, University of California Davis, *Genome-wide discovery of terpenoid chemical defense mechanisms in maize (Zea mays)*

12:00 pm-12:30 pm Toshiaki Umezawa, Kyoto University, *Downregulation of p-coumaroyl ester 3-hydroxylase in rice leads to altered lignin structures and improves biomass usability*

12:30 pm-1:30 pm Lunch, Facultad de Ciencias Químicas Garden

Symposium V: Biochemistry

1:30 pm-2:15 pm **Keynote: Victor Manuel Loyola-Vargas,** Unidad de Bioquímica y Biología Molecular de Plantas; Centro de Investigación Científica de Yucatán
The crosstalk of auxins and cytokinins during the somatic embryogenesis of Coffea canephora

2:15 pm-2:35 pm Narayanan Srividya, Washington State University, *Identification and characterization of acyl activating enzymes required for decoration reactions in the biosynthesis of Taxol and other Taxanes*

2:35 pm-2:55 pm Sandra Irmisch, University of British Columbia, *A novel anti-diabetic compound from plants: Biosynthesis, gene discovery, and metabolic engineering of montbretin A*

2:55 pm-3:15 pm Sean Johnson, Michigan State University, *Systematic diterpene synthase discovery across Lamiaceae*

3:15 pm-3:45 pm Break
Sponsored by: CONAGEN

3:45 pm-4:45 pm PSNA Business Meeting

4:45 pm-7:00pm Dinner on your own

7:00 pm-9:00 pm Poster Session II with Refreshments, Facultad de Ciencias Químicas Library. At the end of this session all posters should be removed.
Sponsored by: IS ANALÍTICA, Cervecería San José

Tuesday, August 7, 2018

8:00 am-5:00 pm Conference Registration, Rogelio Jiménez Auditorium Lobby

9:00 am-9:45 am **Plenary Symposium: Rachel Mata,** Departamento de Farmacia, Facultad de Química, Universidad Nacional Autónoma de México
α-Glucosidases inhibitors from Mexican medicinal plants and fungi

9:45 am-10:00 am Break

Symposium VI: Bioactive natural products

10:00 am-10:45 am **Keynote: Rosalba Encarnación Dimayuga,** Fundación de Farmacognosia y Medicina Complementaria y Alternativa de Baja California Sur
Traditional medicine as a potential source of drugs

10:45 am-11:30 am **Keynote: Ashutosh Sharma,** Department of Bioengineering, Tecnológico de Monterrey, Querétaro
Current status and challenges in the research and development of phytomedicine in Mexico

11:30 am-12:00 pm Armando Alcazar, Oregon State University, *Characterization of phytochemical constituents in Centella asiatica extracts by High Resolution Mass Spectrometry for fingerprinting, quantitation and correlation with biological activity*

12:00 pm-12:30 pm Matthew Mattozzi, Conagen Inc, *From discovery to market: Fermentation for ingredients and natural products*

12:30 pm-1:30 pm Lunch, Facultad de Ciencias Químicas Garden
Career Panel/Workshop, Facultad de Ciencias Químicas classroom
Sponsored by:

SLP PROSPEREMOS JUNTOS Gobierno del Estado 2015-2021 | Consejo Potosino de Ciencia y Tecnología

Symposium VII: Phenotyping, metabolomics, proteomics

1:30 pm-2:15 pm **Keynote: Ana Paulina Barba de la Rosa,** Instituto Potosino de Investigación Científica y Tecnológica A.C.
Proteomic analysis of wild and domesticated Opuntia cladodes

2:15 pm-3:00 pm **Neish Awardee Presentation: Toshihiro Obata**, University of Nebraska Lincoln, *Multi-enzyme complexes of plant mitochondrial tricarboxylic acid cycle – identification to functional analysis*

3:00 pm-3:30 pm Argelia Lorence, Arkansas Biosciences Institute, *Molecular mechanisms underlying the enhanced biomass and abiotic stress tolerance phenotype of high ascorbate Arabidopsis lines*

3:30pm-4:00pm Transportation to Tangamanga park

4:00pm-6:00pm Visit to UASLP Botanic Garden

Wednesday, August 8, 2018

8:00 am-9:00 am Conference Registration, Rogelio Jiménez Auditorium Lobby

9:00 am-9:45 am **Plenary Symposium: Pam Weathers,** Department of Biology and Biotechnology, Worcester Polytechnic Institute
From plant to patient: oral delivery of Artemisia sp. to treat malaria and schistosomiasis

9:45 am-10:00 am Break

Symposium VIII: Biotechnology

10:00 am-10:45 am **Keynote: Sergio Rosales-Mendoza,** Laboratorio de Biofarmacéuticos Recombinantes, Facultad de Ciencias Químicas; Sección de Biotecnología, Centro de Investigación en Ciencias de la Salud y Biomedicina; Universidad Autónoma de San Luis Potosí
Green vaccines: using plant cells to produce orally deliver antigens

10:45am-11:15 am Fred Stevens, Oregon State University, *Agricultural and cosmetic applications of glucosinolate breakdown products from Meadowfoam seed meal*

11:15 am-11:45 am Reinhard Jetter, University of British Columbia, *Waxes coating fern fronds: Fatty acid derivatives and secondary metabolites*

11:45 am-12:15 pm Reinier Gesto Borroto, Universidad Autónoma del Estado de Morelos, *DNA barcoding on natural populations of the Mexican species Galphimia glauca Cav. (Malpighiaceae)*

12:15 pm-1:30 pm Lunch, Facultad de Ciencias Químicas Garden

1:30 pm-2:00 pm Presentation PSNA 2019

2:00 pm-7:00 pm Tour by Plant Biotechnology Building, Facultad de Ciencias Químicas, UASLP

Tour by your own in downtown (see touristic information)

7:00 pm-8:00 pm Traditional "Callejoneada" through historic downtown, Edificio Central UASLP

8:00 pm-11:00 pm Award Banquet, Caja Real de la UASLP
Sponsored by:

11:00pm Close of PSNA 2018

Speakers Abstracts

Sunday, August 5, Morning

Plenary Symposium

Metabolomics in natural product research

Luis M. Peña Rodríguez[1]

[1]*Unidad de Biotecnología, Centro de Investigación Científica de Yucatán Calle 43 No. 130, Colonia Chuburná, Mérida, Yucatán, México 97200*
Email address: lmanuel@cicy.mx

One of the most popular strategies for isolating bioactive natural products (BNPs) from plant crude extracts is the bioassay-guided fractionation; this process can be described as “find and follow” the pharmacological activity along the fractionation process using biological assays, with the final aim to purify and identify the BNPs responsible for the activity. Although this strategy has proven to be successful, it can at times be repetitive, time-consuming, labor-intensive and, consequently, expensive, since it can require the application of a wide variety of chromatographic techniques and numerous biological evaluations.
In order to speed up the isolation of BNPs from plant crude extracts, while reducing the amounts of chemical and biological waste, new strategies have been developed; these include correlating the variation of the chemical composition (i.e. chromatographic profile) of the crude extract and/or the purified fractions with their biological activity using chemometric analysis methods, which allow the detection of BNP’s without isolating them. Chemometric studies have been previously used to detect the BNP’s responsible for the antioxidant, cytotoxic, tyrosinase inhibition, and lymphocyte proliferative capacity activities of phytochemically-known plants.
In this presentation we’ll discuss several examples to show how the combination of chemometric analyses and biological activity data can be used to detect BNP’s in crude extracts from plants.

Symposium I: Plant-environment interactions

Keynote speaker

Volatile organic compounds and extracellular DNA induce immunity in plants

Martin Heil[1]

[1]*Departamento de Ingeniería Genética, CINVESTAV-Irapuato, km 9.6 Libramiento Norte, 36824 Irapuato, Guanajuato, México*
Email address: mheil@ira.cinvestav.mx

Research into plant resistance to biological enemies has traditionally focused on non-self recognition, that is, the perception of foreign molecules termed microbe (or pathogen-) associated molecular patterns (MAMPs/PAMPs) and herbivore-associated molecular patterns (HAMPs). More recent studies focused on damaged-self recognition and identified damage-associated molecular patterns (DAMPs) as important triggers of innate immunity. DAMPs are delocalized or fragmented endogenous molecules whose appearance in the extracellular space indicates tissue rupture in organisms from plants to mammals. Plants respond to damage caused by herbivores or infection with the release of a highly diverse set of volatile organic compounds (VOCs). So-called green-leaf volatiles (mainly C6 alcohols, aldehydes and esters) are released within seconds because in the disrupted tissue, precursors become exposed to already existing enzymes. Green-leaf volatiles and other VOCs act as plant DAMPs which exert direct anti-microbial activities and serve as signals that trigger plant immunity in distal parts of the same plants and – under certain circumstances – in neighboring plants (then causing the phenomenon of 'plant-plant communication'. Most recently, we identified fragments of extracellular DNA (eDNA) as another DAMP in plants that – as in mammals – triggers early immunity-related signals such as ROS and MAPKs. In bean (*Phaseolus vulgaris*), eDNA triggered phenotypic immunity to insect herbivores and a bacterial pathogen in a species-specific manner, that is, eDNA obtained from the same species triggered stronger immunity than eDNA from the congeneric *Phaseolus lunatus*. VOCs and eDNA represent two classes of DAMPs in plants that bear an as yet underestimated potential to be used as 'plant vaccines' in future biocontrol strategies.

Metabolome changes in individual insects infected with Candidatus Liberibacter species from citrus and solanaceaous crops

David Gang[1]

[1]*Institute of Biological Chemistry, Washington State University, WA 99164, USA*
Email address: gangd@wsu.edu

Citrus Greening Disease (also called Huanglongbing or HLB) of citrus (spread by Asian citrus psyllid) and Zebra Chip disease of potato (spread by potato psyllid) are devastating diseases caused by fastidious (so far unculturable) bacterial species belonging to the genus, Liberibacter. These bacteria are spread by psyllids, small, phloem sucking. Development of tools to fight these diseases includes establishment of the ability to culture them in vitro, which requires a better understanding of the bacterium:insect:plant interactions, including at the metabolic level. We compared the metabolite profiles (primary metabolites and lipids) between healthy and Liberibacter-infected psyllids using GC-TOF-MS and UPLC-TOF-MS and also examined individual insects using MALDI-MS imaging mass spectrometry (tissue imaging). The goal of these experiments was to identify changes in metabolite levels that occur upon infection and localize those changes within the insect tissues. Infection with Liberibacter and feeding on infected trees cause dramatic changes in the psyllid metabolome. Tissue imaging results demonstrated differences in specific metabolite levels across the psyllid body upon Liberibacter infection. These results point to major metabolic changes occurring within the insects in response to infection by Liberibacter species.

Co-expression not co-location links genes to specialized metabolic pathways in plants

Jennifer Wisecaver[1]

[1]*College of Agriculture, Purdue University BCHM A343C*
Email address: jwisecav@purdue.edu

Specialized metabolites serve myriad biological functions that allow organisms to interact with and manage their environment (e.g., resist abiotic stress, combat negative ecological interactions and promote beneficial ones). As these specialized metabolites are typically fast-evolving and lineage-specific, the genes and pathways involved in their biosynthesis are often unknown, hampering our ability to understand their function and evolutionary history. Critically, this also limits the potential utility of specialized metabolites in agricultural, pharmaceutical, and biotechnological applications. We hypothesized that genes within a specialized metabolic pathway would form tight associations (modules) with each other in co-expression networks, facilitating their identification. To evaluate this hypothesis, we used 10 global co-expression data sets, each a meta-analysis of hundreds to thousands of experiments, across eight plant species to identify hundreds of co-expressed gene modules per data set. In support of our hypothesis, 15.3 to 52.6% of modules contained two or more known SM biosynthetic genes, and module genes were enriched in SM functions. Moreover, modules recovered many experimentally validated SM pathways, including all six known to form co-localized, biosynthetic gene clusters (BGCs). In contrast, bioinformatically predicted BGCs (i.e., those lacking an associated metabolite) were no more co-expressed than the null distribution for neighboring genes. These results suggest that most predicted plant BGCs are not genuine SM

pathways and argue that BGCs are not a hallmark of plant specialized metabolism. We submit that global gene co-expression is a rich, largely untapped resource for discovering the genetic basis and architecture of plant natural products.

Soybean Isoflavonoids and Disease Resistance

Sangeeta Dhaubhadel[1,2]*, Arjun Sukumaran[1]

[1]*London Research and Development Centre, Agriculture and Agri-Food Canada, London ON, Canada N5V 4T3*
[2]*Department of Biology, University of Western Ontario, London ON, Canada N6A 5B7*
**Corresponding author, email address: sangeeta.dhaubhadel@canada.ca*

Soybeans are one of the most predominantly grown grain legumes worldwide. However, one deterrent to maximizing its yield is the pathogen, *Phytophthora sojae*, which causes stem and root rot disease. Many strategies have been implemented throughout the years to combat the pathogen such as use of pesticides and certain agricultural practises. However, these have been largely ineffective in completely preventing *P. sojae* infection. An alternative strategy would be to improve the innate resistance of soybeans by promoting increased isoflavonoid glyceollin production. Glyceollins are soybean-specific antimicrobial agents which are derived from the isoflavonoid branch of the general phenylpropanoid pathway. Soybeans produce 6 forms of glyceollins. These forms are the result of the differential prenylation on either the C2 or C4 carbon of glycinol. Here, we have identified 11 putative GmPTs in this study, and their expression under pathogen stress and tissue-specific, subcellular localization, and enzymatic activity determined. These analyses identified a new a glycinol 2-dimethylallyl transferase GmPT01 that is induced rapidly in response to stress, and is associated with a QTL linked with resistance to *P. sojae*. Increased knowledge of components of the isoflavonoid pathway will allow for more precise manipulation of glyceollin production, thus in effect, increasing soybean resistance to *P. sojae* and other diseases and pests.

Symposium II: Plant-environment interactions

Keynote speaker

Functional analysis of Glycine-Rich Domain Proteins (AtGRDP1 and AtGRDP2) in plant abiotic stress tolerance

Juan Francisco Jiménez-Bremont[1]

[1]*División de Biología Molecular, Instituto Potosino de Investigación Científica y Tecnológica AC, Camino a la Presa de San José 2055, C.P. 78216, San Luis Potosí, San Luis Potosí, México*

Proteins containing glycine-repeats sequences, arranged in (Gly)n-X signatures, have been reported in a wide variety of organisms including plants, mammalians, fungi, and bacteria. Plant glycine-rich proteins have been associated to stress response. Previously, we identified the Arabidopsis thaliana AtGRDP1 and AtGRDP2 genes, which encode protein with a glycine-rich domain. We carried out the characterization of the AtGRDP1 and AtGRDP2 genes using null mutants and over-expression lines. Under salt stress conditions, 35S::AtGRDP1 and 35S::AtGRDP2 over-expression lines displayed higher tolerance, whereas their mutants showed the opposite phenotype. Regarding AtGRDP1, 35S::AtGRDP1 over-expressing lines showed tolerance to abscisic acid (ABA), resembling a well-known ABI phenotype, whereas the disruption of *AtGRDP1* gene resulted in ABA hypersensitivity, mimicking the ABI3-overexpression phenotype. Concerning AtGRDP2, the 35S::AtGRDP2 over-expression lines had increased growth rate and early flowering time, and accumulated higher levels of indole-3-acetic acid. Our data reveal an important role for AtGRDP1 and AtGRDP2 in Arabidopsis stress response, and suggest a connection between these genes and plant hormones.

Time, Temperature, and the Secret Life of Plants at Night

Doherty, Colleen J.[1]

[1]*Department of Molecular and Structural Biochemistry, North Carolina State University Raleigh, NC 27603, USA*
Email address: colleen_doherty@ncsu.edu

Plants are attuned to the recurring daily and seasonal changes in their environment. Almost all aspects of a plant's physiology and metabolism are orchestrated to precise times in the daily cycle. This daily fluctuation in metabolism impacts plant interactions with the environment including biotic and abiotic stress responses. An identical stress, given to the same plant at different times of day can produce different outcomes at both the molecular and physiological levels. In part, these differences are due to the changing molecular landscape that occurs throughout the diel cycle. In addition, temporal control of stress responses may provide a selective advantage, ensuring efficient use of resources only when necessary. These factors result in time of day and seasonal variations in stress sensitivity. There is an urgent need to understand the fundamental role of time in heat stress responses because warmer nighttime temperatures are a significant risk to crop productivity. Climate changes are driving increases in nighttime temperatures, and warming nights are a major factor in yield loss in rice and other crops. Understanding the specific molecular mechanisms that are sensitive to mild increases in nighttime temperature is a first step towards improving resilience to changing temperature patterns. By analyzing the transcriptional and metabolic response to warmer nights in field-grown rice plants and in model systems, we have developed transcriptional regulatory networks to identify how the perception of time modulates this response. We are working to develop network maps that will allow us to identify candidate markers that can be used in the field to aid in the selection of plants tolerant to increasing nighttime temperatures. Our ultimate goal is to understand the molecular intersection between time and temperature stress and

the underlying network of temperature responses to improve plant tolerance to increasing temperatures at all times of day throughout the year.

Symposium III: Molecular and industrial phytochemistry

Keynote speaker

Plants and algae as bioreactors for biopharmaceutical production against respiratory diseases

Ángel Gabriel Alpuche Solis[1]

[1]*Laboratorio de Biología Molecular de Plantas, División de Biología Molecular, Instituto Potosino de Investigación Científica y Tecnológica, A.C. Camino a la Presa de San José 2055, San Luis Potosí, 78216, México*
Email address: alpuche@ipicyt.edu.mx

The use of plants and algae as new platforms for the production of prophylactic or therapeutic recombinant proteins is an approach that several biotechnological companies are adopting based on the reduction of time, production costs and side effects. The vaccines have saved thousands of lives and we have moved on from the live attenuated whole cell organisms to the acellular vaccines based on genetic engineering technology.
Acute respiratory infections (ARI´s) represent a major health problem since they can rank second worldwide in terms of morbidity and mortality in children under 5 years old. The main causative agents are viruses responsible for 50-60% of ARIs; among the most frequent are human Orthopneumovirus (hOPV), Metapneumovirus (hMPV), Respirovirus (hREV), Rubulavirus (hRUV) and Influenza Virus (IV). In this talk we will share some of our results on the development of antiviral or antigenic proteins produced on plants and algae against respiratory diseases as a proof of concept and the collaboration with national and international companies with the aim of transferring the technology for human uses.

Multiple genes recruited from hormone pathways underlay maize defense regulation

Y. Ding[1]*, S. Mafu[2], K.M. Murphy[2], B. Yang[3], Q. Wang[4], R.J. Schmitz[5], S.A. Christensen[6], Z. Shen[1], K.A. Kremling[7], E.S. Buckler[7,8], S.P. Briggs[1], J. Bohlmann[9], A. Huffaker[1] , P. Zerbe[2], E.A. Schmelz[1*]

[1]Section of Cell and Developmental Biology, University of California at San Diego, La Jolla, CA 92093, USA
[2]Department of Plant Biology, University of California Davis, One Shields Avenue, Davis, CA 95616, USA
[3]Department of Genetics, Development and Cell Biology, Iowa State University, Ames, IA, 50011, USA
[4]College of Agronomy, Sichuan Agricultural University, Chengdu 611130; Institute of Ecological Agriculture, Sichuan Agricultural University, Chengdu 611130, China
[5]Department of Genetics, University of Georgia, Athens, GA 30602
[6]Chemistry Research Unit, Center for Medical, Agricultural, and Veterinary Entomology, Department of Agriculture–Agricultural Research Service (USDA–ARS), Gainesville, FL 32608; [7]Department of Plant Breeding and Genetics, Cornell University, Ithaca, NY14853;
[8]United States Department of Agriculture-Agricultural Research Service, Robert W. Holley Center for Agriculture and Health, Ithaca, New York 14853, USA
[9]Michael Smith Laboratories, University of British Columbia, Vancouver, BC V6T 1Z4, Canada
**Corresponding author, email address: yeding@ucsd.edu*

Gene duplication and divergence from conserved pathways enables the production of diverse plant specialized metabolites that mediate biotic and abiotic stress protection. In maize, hormone precursors essential for gibberellic acid (GA) production are also required for acidic *ent*-isokaurene-derived antibiotics, termed kauralexins. To better understand the partitioning of phytohormone and defense pathways, we employed a series of combined approaches including forward genetics, *in vitro* biochemistry and mutant analyses to define the kauralexin biosynthetic pathway. Kauralexin genes encoding *ent*-copalyl diphosphate synthase 2, *ent*-kaurene synthase-like 2 and *ent*-kaurene oxidase 2, each contain signatures consistent with recent GA pathway derivation via gene duplication and functionalization. Distinct from GA biosynthetic genes, two closely related CYP71 family P450s (ZmCYP71Z16/18) oxidize both *ent*-isokaurene and *ent*-kaurene to produce committed kauralexin monoacids. To avoid unregulated phytohormone production, significant pools of A-series kauralexins do not rely on the GA precursor *ent*-kaurene but instead are produced from corresponding B-series kauralexins by a steroid 5α reductase-related enzyme. In total, five maize enzymes define a genus-specific inducible production of kauralexins and demonstrate how modifications in phytohormone biosynthetic genes enable genesis of defense pathways that mediate maize disease resistance.

Monday, August 6, 2018

Plenary Symposium

Opium poppy: An update

Peter Facchini[1]

[1]*Department of Biological Sciences, University of Calgary, Calgary, Alberta, Canada and Epimeron Inc., Calgary Alberta, Canada*

Agricultural production of opium poppy (*Papaver somniferum*) remains the only commercial source for the narcotic analgesics morphine and codeine, in addition to the feedstock used to produce semi-synthetic opiates including the widely prescribed painkillers oxycodone and hydrocodone, and the opioid antagonists naloxone and naltrexone. Morphine derived from the illicit cultivation of opium
poppy can also be converted to heroin, which has long had negative impacts on humankind and is contributing significantly to the current opioid epidemic in North America. Opium poppy also produces other important benzylisoquinoline alkaloids including the cough suppressant and potential anticancer drug noscapine, the vasodilator papaverine, and the antimicrobioal agent sanguinarine. Ongoing research since the 1960s has led to a remarkable appreciation for the biochemistry of benzylisoquinoline alkaloid metabolism. Elucidation of alkaloid biosynthetic pathways and associated physiological processes in opium poppy is essential for the development of new production technologies not only in plants, but also in microorganisms and beyond. Recent research progress involving alkaloid biosynthesis in opium poppy will be presented with a focus on unexpected discoveries and their biotechnological deployment.

Symposium IV: Biochemistry

Keynote speaker

Integrated genetic and biochemical approaches to define layers of maize immune metabolism

Alisa Huffaker[1]

[1]*Section of Cell and Developmental Biology, University of California at San Diego, La Jolla, CA, 92093, USA*

All plants synthesize complex blends of specialized metabolites which shape community interactions with other organisms. Maize (*Zea mays*) is an outstanding genetic model for studying chemical diversity within the Poaceae and commensurate effects on biotic interactions. While metabolomic approaches can identify insect and microbe-elicited metabolites, underlying biosynthetic and regulatory mechanisms can be elusive. To rapidly uncover relevant biosynthetic and regulatory genes, we apply classical forward genetic association mapping studies in combination with co-expression analyses. Association mapping readily yields metabolite-quantitative trait loci (mQTL) and statistically significant single nucleotide polymorphisms (SNPs). Paired co-expression analyses using mQTL candidates as bait can quickly implicate related genes, facilitating identification of entire biosynthetic pathways along with regulatory genes. *In vitro* biochemical approaches coupled with gene synthesis can then be leveraged to further speed the confirmation of top candidate genes. Use of maize forward genetics enables the identification of many candidate genes involved in the biosynthesis of diverse chemical classes, including the dominant fungal-elicited maize sesquiterpene acids defenses, zealexins. We have found many diverse zealexin structures along with terpene synthases and P450s involved in their production. While *in vitro* assays have confirmed enzyme function, to examine pathway deletions *in vivo*, we used CRISPR/Cas9 to create frame shift mutations in four redundant terpene synthase genes, thereby removing zealexin biosynthesis. Quadruple mutants display significant susceptibility to attack by both fungi and bacteria. Continued use of association mapping and co-expression analyses with development of maize biosynthetic mutant libraries will enable the systematic analyses of specialized metabolite pathways and complex functional roles under field conditions.

Structure-function of monoterpene functionalization reactions

B. Markus Lange[1]*, Narayanan Srividya[1]

[1]*Institute of Biological Chemistry and M.J. Murdock Metabolomics Laboratory, Washington State University, Pullman, WA, 99164, USA*
**Corresponding author, email address: lange-m@wsu.edu*

Terpenoids form the largest class of secondary (or specialized) metabolites in plants. This enormous diversity is due to (1) the modular assembly of building blocks, (2) rearrangement and elimination reactions modifying these backbones, and (3) functionalization and conjugation reactions that generate additional structural complexity. We have a long-standing interest in the structure-function relationships that determine the functionalization of volatile and semi-volatile terpenoids synthesized in glandular trichomes (specialized anatomical storage structures), which contribute substantially to the chemical diversity in the angiosperm lineage. We will discuss the biochemical properties of relevant biocatalysts, arranged by enzyme family (cytochromes P450, short- and medium-chain alcohol dehydrogenases, aldo/keto reductases and others). We will also discuss the implications of newer findings on our understanding of the evolution of pathways toward complex terpenoids. While the examples for this work are drawn from enzymes involved in the decoration of terpenoids, the conclusions are equally relevant to enzymes involved in the biosynthesis of other secondary/specialized metabolites.

Genome-wide discovery of terpenoid chemical defense mechanisms in maize (Zea mays)

Philipp Zerbe[1]

[1]*Department of Plant Biology, University of California Davis, One Shields Avenue, Davis, CA, USA. Email address: pzerbe@ucdavis.edu*

Major staple crops, including rice (Oryza sativa) and maize (Zea mays), deploy complex networks of terpenoid metabolites as key components of their chemical defense against herbivore and pathogen attack. To gain a deeper knowledge of the biosynthesis, distribution and physiological activity of crop terpenoid defenses we integrate genomics-enabled pathway discovery with metabolite profiling and multi-gene co-expression analyses for efficient cross validation of enzyme functions. Using this approach, we identified numerous novel terpene synthases and cytochrome P450 monooxygenases that govern the biosynthesis of terpenoid chemical defenses in maize. Among these pathways, we identified a new class of maize protective diterpenoids, named dolabralexins. Dolabralexin biosynthesis involves the sequential activity of two diterpene synthases (ZmAN2 and ZmKSL4) that together with the P450 ZmCYP71Z16 form the diterpenoid epoxide epoxy-dolabranol not previously identified in plants. Much of the epoxy-dolabranol is further converted into trihydroxydolabradiene (THD), which occurs as the predominant metabolite in the roots of several field-grown maize cultivars. Oxidative stress and elicitation with major maize Fusarium pathogens induce dolabralexin accumulation in root tissues, and dolabralexins significantly inhibit pathogen growth in vitro. These findings highlight the potential of gene-to-metabolite approaches for the discovery of previously unrecognized defense metabolites. Discovery of dolabralexins expand the known chemical space of diterpenoid defenses as genetic targets for understanding and ultimately improving maize resilience.

Downregulation of p-*COUMAROYL ESTER 3-HYDROXYLASE* in rice leads to altered lignin structures and improves biomass usability

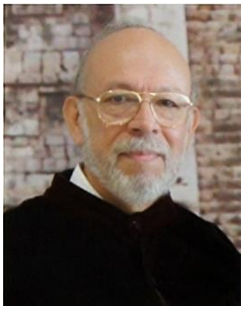
Toshiaki Umezawa[1,6]*, Yuri Takeda[1], Yuki Tobimatsu[1], Steven D. Karlen[2,3], Taichi Koshiba[1], Shiro Suzuki[1], Masaomi Yamamura[1], Shinya Murakami[1], Mai Mukai[1], Takefumi Hattori[1,a], Keishi Osakabe[4], John Ralph[2,3], Masahiro Sakamoto[5]

[1]*Research Institute for Sustainable Humanosphere, Kyoto University, Gokasho, Uji, Kyoto 611-0011, Japan*
[2]*U.S. Department of Energy Great Lakes Bioenergy Research Center, University of Wisconsin-Madison, Madison, WI 53726;*
[3]*Department of Biochemistry, University of Wisconsin-Madison, Madison, WI 53706, USA*
[4]*Faculty of Bioscience and Bioindustry, Tokushima University, Kuramoto-cho, Tokushima 770-8503, Japan*
[5]*Graduate School of Agriculture, Kyoto University, Sakyo-ku, Kyoto 606-8502, Japan*
[6]*Research Unit for Development of Global Sustainability, Kyoto University, Gokasho, Uji, Kyoto 611-0011, Japan*
[a]*Present address: Faculty of Bioscience and Bioindustry, Tokushima University, 2-1 Minamijosanjima-cho, Tokushima 770-8513, Japan*
**Corresponding author, email address: tumezawa@rish.kyoto-u.ac.jp*

p-Coumaroyl ester 3-hydroxylase (C3´H) is a key enzyme involved in the biosynthesis of lignin. Although the crucial role of C3´H in lignification and its manipulation to upgrade lignocellulose have been investigated in eudicots, limited information is available in monocotyledonous grass species despite their potential as biomass feedstocks. Here we address the impacts of *C3´H*-deficiency on the structure and properties of grass cell walls. *C3´H*-knockout rice mutants generated via CRISPR/Cas9-mediated mutagenesis were severely dwarfed. In contrast, *C3´H*-knockdown lines generated via RNAi-mediated gene silencing, even with ~0.5% of residual expression levels, reached maturity without displaying a severely impaired growth phenotype. Cell wall structural analysis revealed that lignins in *C3´H*-knockdown rice cell walls were largely enriched in *p*-hydroxyphenyl (H) lignin units, and substantially reduced in the normally dominant guaiacyl (G) and syringyl (S) lignin units. Interestingly, however, the enrichment of H units was limited to being within the non-acylated lignin units, with grass-specific γ-*p*-coumaroylated lignin units remaining apparently unchanged on the residual G and S units. Suppression of *C3´H* also resulted in relative augmentation in tricin residues in lignin as well as substantial reduction in wall cross-linking ferulates. Collectively, our data demonstrate that *C3´H* expression is an important determinant, not only of lignin content and composition, but also of the degree of cell wall cross-linking. Lastly, we demonstrated that *C3´H*-suppressed rice displays remarkably enhanced biomass saccharification.

Symposium V: Biochemistry

Keynote speaker



The crosstalk of auxins and cytokinins during the somatic embryogenesis of Coffea canephora

Víctor M. Loyola-Vargas[1], Johny R. Avilez-Montalvo[1], Geovanny Nic-Can[2], Rosa M. Galaz-Ávalos[1], Fátima Duarte-Aké[1], Ruth Márquez-López[1], Ariana Pérez-Hernández[1], Ligia Brito-Argaez[1], Víctor M. Aguilar-Hernández[1].

[1]*Unidad de Bioquímica y Biología Molecular de Plantas; Centro de Investigación Científica de Yucatán; Mérida, Yucatán, México*
[2]*Universidad Autónoma de Yucatán, Facultad de Ingeniería Química.*

Auxins (Aux) and cytokinins (CKs) are plant growth regulators (PGR) involved in multiple physiological processes in plants. These PGR regulate and crosstalk each other their homeostasis. The homeostasis of Aux and CKs is very relevant to understand their physiological roles. We are using somatic embryogenesis (SE) in *Coffea canephora* to understand the role of Aux and CKs in the control of cell differentiation in plants. The SE induction process involves a pre-treatment (PT) of the source plants for 14 days with 0.52 µM naphthalene acetic acid and 2.32 µM kinetin. After this PT, the leave explants are incubated in Yasuda's medium supplemented with 5 µM of benzyladenine. The first embryogenic structures appear after 21days of incubation. The amount of both Aux and CKs increases during the PT of the plants. The Aux and CKs were identified by HPLC-MS and the increment of indole-3-acetic acid (IAA) is due to *de novo* biosynthesis and accumulates in specific sites in the somatic embryo. The blocking of both, its biosynthesis or transport, inhibits the induction of SE. The relationship of CKs and Aux increased at the beginning of the PT, and decreases at the end of PT. This relationship significantly increased during the firths 24 hours of the induction of SE. The increment in the amount of Aux and CKs correlated with changes in the expression of genes related to their biosynthesis, transport, conjugation, and perception. Our actual model suggests that the relationship CKs and Aux is central to the differentiation process in plants.

[3]This project is supported by CONACyT (Grants, 257436 and 1515).

Identification and Characterization of Acyl Activating Enzymes Required for Decoration Reactions in the Biosynthesis of Taxol and other Taxanes

Narayanan Srividya[1]

[1]*Institute of Biological Chemistry and M.J. Murdock Metabolomics Laboratory, Washington State University, Pullman, Washington*
Email address: narayanan.srividya@wsu.edu

Members of the genus Taxus accumulate diterpenoid natural products termed taxanes (500 different structures characterized to date), of which taxol® (paclitaxel) has risen to particular prominence as one of the most successful anti-cancer treatments of all time. Taxane structures are generally characterized by a high degree of decoration with multiple acyl functional groups. The decoration of the taxadiene core toward different taxane end products requires the activation, as coenzyme A ester, of straight-chain alkyl, branched-chain alkyl, or aromatic carboxylic acids. To identify the determinants of chemical complexity in taxane biosynthesis, a better understanding of the catalytic specificity of acyl activating enzymes (AAEs) for different carboxylic acids is urgently needed.
In this study, we quantified the major taxanes (characterized by differences in their acyl decoration) that accumulate in the medium of Taxus x media cell suspension cultures. Publicly available transcriptome data sets acquired under various experimental conditions then served as a resource for identifying AAE genes with possible functions in the activation of carboxylic acids with relevance for the biosynthesis of different taxane structures. cDNAs of 19 AAE candidates were amplified and subcloned into appropriate expression vectors. The functional evaluation of the corresponding recombinant proteins, which were assayed with 29 different carboxylic acids, demonstrated preference of certain candidates for the activation of benzoate, hexanoate, butyrate, 2-methyl butyrate or 2-methyl-2-butenoate. Our efforts have led to the characterization of a set of AAEs sufficient to activate carboxylic acid precursors for all major taxanes. We can now build on these discoveries to develop metabolic engineering approaches that favor the accumulation of pharmaceutically relevant taxol (contains two benzoyl moieties) over other taxanes (which contain both benzoyl and alkyl side chains).

A novel anti-diabetic compound from plants: Biosynthesis, gene discovery, and metabolic engineering of montbretin A

Sandra Irmisch[1]*, Seohyun Jo[1,2], Christopher R. Roach[1], Sharon Jancsik[1], Macaire Man Saint Yuen[1], Lufiani L. Madilao[1], Mark O'Neil-Johnson[3], Russel Williams[3], Stephen G. Withers[1,4], Joerg Bohlmann[1,2*]

[1] *Michael Smith Laboratories,* [2] *Department of Botany,* [4] *Department of Chemistry, University of British Columbia, Vancouver, BC, Canada*
[3] *Sequoia Sciences, St. Louis MO, USA*
**Corresponding author, email address: sirmisch@msl.ubc.ca*

Plant specialized metabolism serves as a rich resource of biologically active molecules for drug discovery. The acylated flavonol glycoside montbretin A (MbA) and its precursor mini-MbA are potent inhibitors of human pancreatic α-amylase (HPA) and are being developed as drug candidates for the treatment of type-2 diabetes. MbA occurs in corms, which are the below-ground storage organs, of the ornamental plant montbretia (Crocosmia x crocosmiiflora), but a system or process to obtain large quantities of MbA is not currently available. Improved MbA production thus requires knowledge of its biosynthesis from the flavonol core myricetin. Metabolite profiling and enzyme assays showed that MbA formation and accumulation occurs during early stages of corm development. We established myricetin 3-O-rhamnoside (MR), myricetin 3-O-glucosyl rhamnoside (MRG) and myricetin 3-O-(6'-O-caffeoyl)-glucosyl rhamnoside (mini-MbA) (MRG-Caff) as the first three intermediates formed during MbA biosynthesis. Contrasting transcriptomes of young and old corms revealed a set of differentially expressed UDP-sugar-dependent glycosyltransferases (UGTs) and BAHD-acyltransferases (BAHD-ATs). cDNA cloning and enzyme characterization identified CcUGT1 and CcUGT2 as the enzymes that catalyze the consecutive glycosylation of myricetin to produce MR and of MR to give MRG, respectively. In addition, we identified two BAHD-ATs, CcAT1 and CcAT2 that catalyze the acylation of MRG to complete the formation of mini-MbA. Transcript profiles of CcUGT1, CcUGT2, CcAT1 and CcAT2 during corm development matched the metabolite profile of MbA accumulation. Expression of these enzymes in Nicotiana benthamiana resulted in the formation of a surrogate mini-MbA, which supports the potential of MbA and mini-MbA metabolic engineering and production in a heterologous plant system.

Systematic diterpene synthase discovery across Lamiaceae

Sean R. Johnson[1]

[1]*Institute of Biological Chemistry and M.J. Murdock Metabolomics Laboratory, Washington State University, Pullman, Washington 99164-6340, USA*
Email address: John6121@msu.edu

Members of the mint family (Lamiaceae) accumulate a wide variety of industrially and medicinally relevant diterpenes. While there are more than 7000 plant species in Lamiaceae, diterpene synthase (diTPS) genes have been characterized from just 11. The Mint Genome Project has recently sequenced leaf transcriptomes from 48 phylogenetically diverse Lamiaceae species, more than doubling the number of mint species for which transcriptomes are available. We have used a gene discovery workflow informed by transcriptome data, chemotaxonomic data, and enzyme sequence data to mine Lamiaceae for diterpene synthases with novel activities. Genes were selected for functional characterization with an intent to roughly evenly sample the sequence homology space, and to focus on species where diTPS genes were found in the transcriptome, but from which no diterpene structures had been previously reported. Novel activities were found for four class I enzymes, and two class II enzymes. Enzymatic work also guided the discovery of diterpenes in a species where none had previously been reported. The results give insights into evolution and diversification of diterpene biosynthesis in mints and establish a comprehensive foundation for continued investigation of diterpene biosynthesis in Lamiaceae.

Tuesday, August 7, 2018

Plenary Symposium

α-Glucosidases Inhibitors from Mexican medicinal plants and fungi

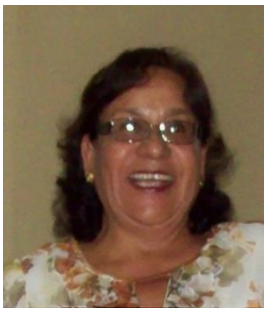
Rachel Mata[1]

[1]*Departamento de Farmacia, Facultad de Química, Universidad Nacional Autónoma de México, 04510, Mexico City, México*

Type 2 diabetes mellitus is a major public health concern worldwide. The foundations of diabetes management include lifestyle intervention along with pharmacological therapy. The major classes of oral antidiabetic medications include biguanides, sulfonylureas, meglitinide, thiazolidinedione, dipeptidyl peptidase 4 inhibitors, sodium-glucose cotransporter inhibitors, and α-glucosidase inhibitors (AGIs). AGIs are not recognized as the first line anti-diabetic therapy, however, they have significant importance for treating the prediabetes condition, and in combination with other oral drugs or insulin are very useful for severe diabetes. In elderly patients the therapy of choice comprises sulfonylureas and AGIs. Furthermore, they reduce cardiovascular events by 49%. In a program devoted to discover new antidiabetic therapies we have isolated a few plants and fungi metabolites with *α* -glucosidase inhibitory properties. The most relevant aspects concerning their origin, chemistry and pharmacology will be discussed.

Symposium VI: Bioactive natural products

Keynote speaker



Traditional medicine as a potential source of drugs

Dra. Rosalba Encarnación Dimayuga[1]

[1]*Fundación de Farmacognosia y Medicina Complementaria y Alternativa de Baja California Sur, México*
Email address: farmecalbcs779mail.com

Traditional medicine has been a very important source of drugs since the man has used medicinal plants to solve their health problems. Traditionally, Mexico is a very biodiverse country. Its richness in the terrestrial and marine flora and fauna is extraordinary. Approximately 5 % of the flora has been investigated as source of active compounds and much more less is related with the marine organism. In the Traditional Medicine most of the sources are related to medicinal plants and very few to marine organisms. However, the pharmacognositc research performed in the natural products of BCS, showed the potential there is in the plants of arid zones and in the marine organisms of Gulf of California.

Keynote speaker

Current status and challenges in the research and development of phytomedicine in Mexico

Ashutosh Sharma[1]

[1]*Department of Bioengineering Tecnologico de Monterrey, Queretaro, México*

Phytomedicine with therapeutic potential has played a significant role throughout the human history. Although its usage greatly diminished during the dawn of the scientific era, there is a revival of interest in its potential by late 20th century, especially in the development of new drugs. Medicinal plants are an important part of traditional medical systems in Mexico. Medicinal plants are usually regarded as part of a culture's traditional knowledge. Mexico has been considered the 4th megadiverse country in the world. More than 6000 medicinal plant species have been in used for the traditional healing system in Mexico. More than 60% of the Mexican population still uses medicinal plants as alternative medicine. However, high-quality phytomedicines use is still dependent on the international market and import. Mexico produces a large number of medicinal plant species for the commercial use but most of them are being exported to other countries, and the major part of the medicinal plant market is still based on the regional market and local plant collectors. The use of phytomedicine is increasing significantly all over the world. However, Mexico is still facing important challenges in the research and development of local phytomedicines due to

lacked governmental strategy, planning and multidisciplinary research focus. The integration between the academic institution, government, and private sector is still under development. In this study, we discuss the the current situation, challenges and future of phytomedicine in Mexico.

Characterization of phytochemical constituents in Centella asiatica extracts by High Resolution Mass Spectrometry for fingerprinting, quantitation and correlation with biological activity.

Armando Alcazar Magana[1,4]*, Kirsten Wright[2], Maya Caruso[2], Charles Murchison[2], Amala Soumyanath[2], Joseph Quinn[2,3] Jan F. Stevens[4,5], Claudia S. Maier[1]

[1]*Department of Chemistry, Oregon State University, 153 Gilbert Hall, Corvallis, Oregon 97331, USA*
[2]*Department of Neurology, Oregon Health & Science University, 3181 SW Sam Jackson Park Road, Portland, Oregon 97239, USA*
[3]*Department of Neurology and Parkinson's Disease Research Education and Clinical Care Center (PADRECC), Portland Veterans Affairs Medical Center, Portland, Oregon 97239, USA*
[4]*Department of Pharmaceutical Sciences, Oregon State University, 1601 SW Jefferson way, Corvallis, Oregon 97331, USA*
[5]*Linus Pauling Institute, Oregon State University, 2900 SW Campus way, Corvallis, Oregon 97331*
**Corresponding author, email address: alcazara@oregonstate.edu*

Plants produce a diverse set of secondary metabolites, which play a pivotal role in defense, communication, and regulation of primary metabolic pathways. In addition, these compounds may possess biological activities beneficial to human health. *Centella asiatica* (CA) is an Ayurvedic botanical reputed to improve brain function and cognition. CA-derived products are marketed as dietary supplements ('Gotu Kola') to 'support the nervous system'. Extraction methodology, geographic origin, genetics, developmental stage, and post-harvest processing all affect chemical composition of extracts, severely impacting the reproducibility of preclinical and clinical trials. The overall goal of this project is to develop a standardized CA product containing appropriate levels of active compounds against age-related cognitive decline.

For quantification analysis, the combination of liquid chromatography and high-resolution orthogonal acceleration hybrid quadrupole time-of-flight (oa Q-TOF) mass spectrometry, in single ion monitoring and parallel reaction monitoring (PRM) mode, increases the confidence in the detection and quantification of phytochemicals in plant extracts. For qualitative purposes, high-resolution tandem mass spectrometry in data-dependent acquisition (DDA) mode supported by high throughput data mining tools is particularly suitable for screening extracts and documenting known and unknown phytochemical constituents.

By using parent ion monitoring, PRM and DDA, we quantified 25 known phytochemicals found in aqueous extracts of CA and assigned 117 compounds to phenolic acids, caffeoyl quinic acids, triterpenoids among others by online database searching and matching.

In order to identify active compounds in CA, we are exploring a bioassay-guided fractionation approach in combination with flow-injection mass spectrometry of fractions to correlate biological effect to relative concentration of individual phytochemicals or closely related phytochemicals. We expose human neuroblastoma MC65 cells to CA subfractions and measure the protective effects of the phytochemical mixtures in the fractions against amyloid β-cytotoxicity.

This project is supported by National Institutes of Health grant # R01AT008099.

From Discovery to Market: Fermentation for Ingredients and Natural Products

Matthew Mattozzi[1]

[1]*Senior Scientist, Business Development at Conagen Inc. 15 DeAngelo Drive, Bedford, Massachusetts, United StatesConagen Inc.*
Email address: matt.mattozzi@conagen-inc.com

Plants and their secondary metabolites have served as fragrances, flavors, and pharmaceuticals for millennia. While the discovery of new secondary metabolites is extremely important, cost barriers such as limited availability and high molecule complexity can prevent their adoption by consumers. The emerging field of synthetic biology is beginning to democratize this process. By determining the metabolic pathways by which plants make small molecules and re-constructing them an organism amenable to fermentation, we can greatly increase the availability of natural products. Here we present case studies in the natural flavorings (peach lactones) and sweeteners (steviol glycosides) space and how Conagen has produced natural product molecules heterologously via fermentation. Current and future applications of bioproduction will be discussed, and several case studies will be presented that illustrate the impact already being felt in the food and beverage space. We are interested in working with academics to sponsor projects for natural product and metabolic pathway discovery.

Symposium VII: Phenotyping, metabolomics, proteomics

Keynote speaker

Proteomic Analysis of wild and domesticated *Opuntia* cladodes

Ana Paulina Barba de la Rosa[1]

[1]*Instituto Potosino de Investigación Científica y Tecnológica A.C. Camino a la Presa San José 2055, Lomas 4a sección, San Luis Potosí, SLP, 78216 México*

The *Opuntia* genus is widely distributed in America, but the highest richness of wild species are found in Mexico, as well as the most domesticated *Opuntia ficus-indica*, which is the most important crop in agricultural economies of arid and semiarid areas worldwide. During domestication process, the Opuntia morphological characteristics were favoured, however, there is a gap regarding Opuntia molecular mechanisms that enable them to grow in extreme environmental conditions and how the

domestication processes has changed them. To obtain more insights about the *Opuntia* molecular changes through domestication, a shotgun label-free analysis was carried out to characterize the proteomes of five Opuntia species selected by its domestication degree. Most of the changes were observed in glucose, secondary, and 1C-metabolism pathways, which correlate with the observed protein, fibre and phenolic compounds accumulation in Opuntia cladodes. Regulatory proteins, such as 14-3-3 proteins were highly accumulated. 14-3-3 proteins are important proteins in regulation of protein function through protein-protein interaction. The Op14-3-3 was cloned, characterized and possible protein clients were detected. Our results provide new valuable data that will help to the understanding of the molecular changes amongst wild and domesticated Opuntia species.

Multi-enzyme complexes of plant mitochondrial tricarboxylic acid cycle – identification to functional analysis

T. Obata[1]

[1]*Department of Biochemistry and Centre for Plant Science Innovation, University of Nebraska-Lincoln, 1901 Vine Street, Lincoln, NE 68588, USA*
Email address: tobata2@unl.edu

Plant mitochondrial metabolism alters dramatically in the day and the night. Metabolic flux through the tricarboxylic acid (TCA) cycle changes accordingly, between the cyclic and non-cyclic flux modes. This daily alteration is likely regulated by the modification of existing enzymes as the abundance of the enzymes shows little diurnal oscillation. We hypothesize that multi-enzyme complex is one of the mechanisms involved in the daily metabolic changes. TCA cycle enzymes in animal and bacteria form multi-enzyme complexes that catalyze sequential reactions and channel intermediates between the enzymes (metabolon). It is theorized to enhance (even thermodynamically unfavorable) reactions and to direct metabolic flux by protecting intermediates from competing reactions. Additionally, interactions of the non-sequential enzymes also affect enzyme activities of them in bacteria. However, the interactions among the TCA cycle enzymes and other proteins were unexplored.
All possible binary interactions between 38 proteins composing mitochondrial TCA cycle were tested using a compromised score generated from (semi-)quantitative scores obtained by three independent methods, namely affinity purification-mass spectrometry (AP-MS), split luciferase and yeast two hybrid (Y2H) assays. The analysis revealed very dense interaction network with 158 interactions. These include the interactions of catalytic subunits mediating sequential reactions of the pathway. Isotope dilution experiments demonstrated that these complexes mediate citrate and fumarate channeling in isolated mitochondria. Additionally, 125 extra-pathway interactions were suggested from the analysis of AP-MS results. Further AP-MS analysis using plants expressing StrepII-tagged citrate synthase (CSY4) revealed the interaction between CSY4 and respiratory complex I subunit B13 in the illuminated leaves. The B13 knock-down Arabidopsis lines showed reduction in malate-dependent respiration. We are currently trying to elucidate the functions of these detected interactions in the regulation of mitochondrial metabolism.

Molecular Mechanisms Underlying the Enhanced Biomass and Abiotic Stress Tolerance Phenotype of High Ascorbate Arabidopsis Lines

Argelia Lorence[1,3]*, Nirman Nepal[1], Jessica P. Yactayo-Chang[1], Lucia M. Acosta-Gamboa[1], Karina Medina-Jiménez[2], Mario A. Arteaga-Vazquez[2]

[1]Arkansas Biosciences Institute, Arkansas State University, 4301 W Markham St #821, Little Rock, AR 72205, USA
[2]Universidad Veracruzana, INBIOTECA No. 101, Colonia Emiliano Zapata, ZIP 91090, Xalapa, Veracruz, México; [3]Department of Chemistry and Physics, Arkansas State University, P.O. Box 419, State University, AR 72467, USA
**Corresponding author, email address: alorence@astate.edu*

Myo-Inositol oxygenase (MIOX) is first enzyme in the inositol route to vitamin-C (L-ascorbate, AsA). MIOX Arabidopsis over-expressers have elevated AsA and display enhanced biomass and increased tolerance to abiotic stresses. The molecular mechanisms underlying this phenotype are not well understood. In this work RNA-Seq analysis, RT-qPCR, LC/MS, microscopy, and physiological measurements were used to study gene expression profiles and to find biological significance of the differentially expressed transcripts in the high AsA line compared to controls. I*n silico* and RT-qPCR analysis indicate increased expression of transcripts involved in auxin biosynthesis, hydrolysis, transport, and metabolism, which are supported by elevated auxin levels and their effect on epidermal cell elongation in the MIOX over-expressers. Additionally we detected upregulation of transcripts involved in photosynthesis. In support of this finding we measured increased efficiency of the photosystem II and lower non-photochemical quenching in the transgenics. These changes in auxin metabolism and efficient photosynthesis are likely explanations for the enhanced biomass accumulation and growth rate of the MIOX line. Multiple gene families conferring plants tolerance to cold, drought, and heat stresses were found to be elevated in the MIOX over-expressers. Increased expression of amylase and increased glucose levels in the high AsA line possibly confer tolerance to cold stress and act as signal molecules to initiate biotic defense responses. Interestingly, we detected upregulation of transcripts involved in defense hormones biosynthesis (e.g. jasmonates), defense proteins (e.g. defensin), secondary metabolites (e.g. glucosinolates), and transcription factors that are known to be important for biotic stress tolerance in the high AsA line. Negative effects of downregulation of transcripts in pathogen defense response seems to be compensated by elevated salicylic levels in the MIOX line. Further *in vivo* auxin quantification and sensitivity analysis, and bioassays challenging the MIOX line with nematodes will be carried out in follow up studies.

Wednesday, August 8, 2018

Plenary Symposium

From plant to patient: oral delivery of Artemisia sp. to treat malaria and schistosomiasis

Pamela Weathers[1]

[1]*Dept. Biology and Biotechnology, Worcester Polytechnic Institute, Worcester, MA 01609, USA*

Malaria and schistosomiasis (bilharzia) are serious neglected tropical diseases afflicting millions. Two *Artemisia* species, *A. annua* and *A. afra*, were compared in DRC with current therapeutics, artemisinin combination treatment (ACT, ASAQ), and praziquantel (PZQ), against malaria and schistosomiasis, in two clinical trials of 1,000 and 800 patients, respectively. *Artemisia*-treated patients received 1L/day of dry leaf/twig infusions for 7 days with 28-day follow-up. To double-blind the trials, placebos for each standard drug and the tea infusion were included. Artemisinin content of *A. afra* was negligible, but therapeutic responses of patients in both trials were statistically equivalent to *A. annua*-treated patients. Malaria: parasitemia and gametocytes were measured microscopically; recrudescence vs. reinfection was measured by nested-PCR. Asexual parasites cleared after 24h, but took up to 14 days in *Artemisia* vs. ASAQ-treated patients. D14-28 no *Artemisia*-treated patients had detectable gametocytes; 10 ASAQ-treated patients had ametocytes at D28. Cure rates were lowest for children aged 5-15 at 95% and 86% for *Artemisia* and ASAQ-treated patients, respectively. Recrudescence for *Artemisia* and ASAQ-treated patients was 0.4% vs. 1.6%, respectively. Bilharzia: Within 14 days of treatment, all *Artemisia*-treated patients had no bilharzia detectable eggs in fecal smears, but PZQ-treated patients required 21 days
treatment; all 800 patients were egg free by 28 days. Both adult and pediatric patients treated with *A. annua* responded better compared to PZQ treatment. There were no major side effects, from either *Artemisia*. No major gender differences were observed. Both trials showed efficacy that was not artemisinin dependent. *A. annua* and *A. afra* provided faster, more effective treatment of both malaria and schistosomiasis than the standard ACT and PZQ drugs, and should be considered for global use.

Symposium VIII: Biotechnology

Keynote speaker

Green vaccines: using plant cells to produce and orally deliver antigens

Sergio Rosales-Mendoza[1,2]

[1] Laboratorio de Biofarmacéuticos Recombinantes, Facultad de Ciencias Químicas, Universidad Autónoma de San Luis Potosí,. Av. Dr. Manuel Nava 6, San Luis Potosí, 78210, México
[2] Sección de Biotecnología, Centro de Investigación en Ciencias de la Salud y Biomedicina, Universidad Autónoma de San Luis Potosí, Av. Sierra Leona 550, Lomas 2ª. Sección, San Luis Potosí, 78210, México
Email address: email: rosales.s@uaslp.mx

The use of innovative platforms to produce vaccines cheaply and deliver them through non-invasive routes could expand their social benefits. For more than two decades of research, oral vaccines that rely on genetically engineered plants expressing antigens have been explored to treat or prevent high-impact diseases. The development of several vaccine candidates designed to treat infectious and non-communicable diseases will be described. Plant-made antigens were capable to induce humoral responses in test mice and ongoing efforts are focused on testing the immunoprotective potential of the vaccine candidates in the final target species and transferring the technology to biotech companies.

Agricultural and Cosmetic Applications of Glucosinolate Breakdown Products from Meadowfoam Seed Meal

J.F. Stevens[1]

[1]Department of Pharmaceutical Sciences and the Linus Pauling Institute, Oregon State University, 435 Linus Pauling Science Center, 2900 SW Campus Way, Corvallis, Oregon 97331, USA
Email address: fred.stevens@oregonstate.edu

Meadowfoam (*Limnanthes alba*) is an oilseed crop in the Willamette Valley of Oregon. Meadowfoam seed oil has commercial value as an ingredient of cosmetic products. A waste product of the oilseed extraction, the seed meal is a rich source of the glucosinolate, glucolimnanthin (up to

4% by weight), and has been used by Oregon farmers to control weeds as an alternative to field burning, with variable success. We investigated the problem and found that the enzymatic breakdown products of glucolimnanthin exert the actual herbicidal activity, not glucolimnanthin itself. Because the technology used in the commercial oil extraction destroys enzyme activity, we developed methods for enzymatic conversion of glucolimnanthin into its corresponding isothiocyanate or nitrile degradation products [1-3]. We are also exploring pharmaceutical and cosmetic applications of meadowfoam-derived phytochemicals. We studied the breakdown products of glucolimnanthin, *i.e.*, 3-methoxyphenyl-acetonitrile (MPAN) and 3-methoxybenzylisothiocyanate (MBITC), in a skin model of UV-induced DNA damage. The 3-dimensional model consisted of co-cultured human epidermal keratinocytes and human dermal fibroblasts grown to form a multilayered dermis and epidermis (termed 'skin equivalents'). When exposed to UVB-light, the DNA in the skin equivalents develops damage through dimerization of pyrimidine nucleobases. We measured the resulting cyclobutane pyrimidine dimers (CPDs) as an index of UVB-induced DNA damage. We found that both MPACN and MBITC, applied topically in solution, protected against DNA damage [4]. These data demonstrate that MBITC and MPACN exhibit promising anti-photocarcinogenic and anti-photoaging properties in the skin microenvironment and could be used for application in cosmetic products.

Waxes coating fern fronds: Fatty acid derivatives and secondary metabolites

Reinhard Jetter[1]

[1]*Department of Botany, University of British Columbia, 6270 University Blvd, Vancouver V6T 1Z4, Canada*
Email address: jetter@botany.ubc.ca

Fern fronds are coated by true cuticles resembling those of higher vascular plants. However, it is not clear how far wax compositions vary between fern species, and between ferns and gymno-/angiosperms. Here, the cuticular wax components of five fern species occurring in British Columbia were analyzed by GC–MS and GC–FID. Esters were found to be the most abundant compound class by weight in all species, with high chain length and isomer diversities. The fern waxes further comprised very–long–chain primary alcohols, fatty acids, aldehydes, alkanes, secondary alcohols, ketones, and some secondary metabolites such as fernene and β–sitosterol. The secondary alcohols and ketones identified in the fern waxes had typcial polyketide structures, different from those found in other vascular plants like Arabidopsis thaliana. This result, together with other characteristics of the fern wax mixtures, has significant implications on the evolutionary history of wax composition and biosynthesis.

DNA barcoding on natural populations of the Mexican species *Galphimia glauca* Cav. (Malpighiaceae)

Reinier Gesto-Borroto[1, 2], Jessica P. Yactayo Chang[3], Alexandre Cardoso Taketa[1], Argelia Lorence[3*], María Luisa Villarreal[1**]

[1]*Laboratorio de Investigación en Plantas Medicinales, Centro de Investigación en Biotecnología;*
[2]*Centro de Investigación en Dinámica Celular, Instituto de Investigación en Ciencias Básicas y Aplicadas, Universidad Autónoma del Estado de Morelos, Avenida Universidad 1001, Colonia Chamilpa, C.P. 62209, Cuernavaca, Morelos, México*
[3]*Arkansas Biosciences Institute, Arkansas State University, P.O. Box 639, State University, Arkansas, 72467, USA*
**Corresponding author, email address: alorence@astate.edu*
***Corresponding author, email address: luisav@uaem.mx*

Galphimia glauca Cav. (Malpighiaceae) is a plant used popularly in Mexico since prehispanic times to treat different illnesses, including central nervous system disorders. *G. glauca* is distributed widely in the country; however the scientific studies about the pharmacological properties of the plant, have been limited to populations located in certain localities of Mexico. The first studies made on a natural population were carried out in Doctor Mora, Guanajuato and demonstrated that the plant has anxiolytic and sedative properties in both, mice and humans. The compounds responsible for these properties were isolated and identified as *nor*-secofriedelanes triterpenes and were named as galphimines. In a previous work, our group demonstrated that seven populations of *G. glauca* collected in different ecosystem presented diverse metabolic profiles and biological activities, related to the presence of galphimines. In that work, DNA barcode approaches involving *matK, rbcL* and *rpoC1* genes were used with the aim of finding out if these populations belong to the same species. That study showed that the seven analyzed populations belong to at least three potential species of the genus *Galphimia*. In this new work the molecular analysis using DNA barcodes was performed in eight populations botanically classified as *G. glauca*. In this study new markers, *its1* and *its2*, were used, in addition to *matK, rbcL* and *rpoC1*. A phylogenetic analysis was performed using Neighbor-Joining (NJ), Maximum Likelihood (ML) and Maximum Parsimony (MP) methods for all five genes. The resulting phylogenetic trees obtained by NJ, ML and MP methods showed strongly supported clades with high bootstrap values for all gene sequences individually and combined. Overall, these results suggest that the studied populations of *G. glauca*, potentially belong to five different species of the genus *Galphimia*.

Poster Abstracts

All posters should be put up before Poster Session I at Facultad de Ciencias Químicas Library at 7:00 pm on Sunday, August 5th and should remain up until after Session II, August 6th

P1

Phytochemical diversity of *Piper auritum* (Kunth)

Camilo Guerrero-Perilla[1], Massuo J. Kato[1]

[1]*Instituto de Química, Universidade de São Paulo, Av. Prof. Lineu Prestes, 748, Bloco 11 Térreo, 05508-000, São Paulo, SP, Brasil*

Piper auritum (Piperaceae family) is a shrub native to Central America and it is widely used in folk medicine and also in culinary. Its essential oil has exhibited important insecticide and anti-leishmanial activities while leaf infusions are used for treatment of bronchitis and throat irritations. Despite the several studies on its volatile compounds, there is no further information on the phytochemical composition of this species. Thus, the aim of this work is to describe the metabolite diversity in different organs of this species and also on different developmental stages of the plant. The methanolic extracts of roots, leaves, and fruits were fractionated by flash column-chromatography, and the major compounds were further purified and analyzed by means of HPLC-DAD-ESI-HRMS as well as by ^{1}H and ^{13}C NMR (1D and 2D). Apart of the asarone and safrol, which were described from essential oils by several authors, the extracts were characterized by HPLC-ESI-MS and NMR data to contain the amides pellitorine and 1-cinnamoylpyrrolidine; phenylpropanoids dillapiole, elemicin, and asarone; the aristolactames cepharanone B, aristolactam AII; the piperolides 4,6-dimethoxy-5-*E*-phenylbutenolide, 4,6-dimethoxy-5-*Z*-phenylbutenolide and a cyclobutane-photodimer of the piperolides. Additionally, the minor compounds longicaudatin, arboreumine, piperlongumine, dihydropiperlongumine, faragamide, pipermethysine, 1-hexadecanoylpyrrolidine, hydroxykavain, cepharadione A, norcepharadione B, debromocymopolone, and dehydropipernonaline were identified by intepretation of LC-MS/MS data. The seedling chemistry has important differences as compared to the adult individuals with the lack of piperolides and presence of further compounds. While most of these compounds were reported for the first time for the species, the overal data indicated that it has a complex phytochemical composition based on the production of secondary compounds from different classes of natural products.

P2

Identification of phenolic compounds by HPLC with Diode Array Detector (DAD) and Time of Flight Mass Spectrometry (MS-TOF) in in vitro cultures of the cacti Mamillaria candida and Turbincarpus laui

A. Reyes-Martínez[1], M. Antunes-Ricardo[1], J.A. Gutierrez-Uribe[2], M.S Santos-Diaz[1]

[1]Centro de Investigación y Estudios de Posgrado, Facultad de Ciencias Químicas., Universidad Autónoma de San Luís Potosí, Av. Manuel Nava No. 6, Zona Universitaria C.P 78210 San Luis Potosí, S.L.P., México
[2]Centro de Biotecnología-FEMSA., Tecnológico de Monterrey, Av. Eugenio Garza Sada 2501 Sur, C.P. 64849 Monterrey, N.L., México

P3

Glycosidic acids from the Brazilian Jalap Root (Operculina hamiltonii) with Purgative Activity

A. Moreno-Velasco,[1] R. Pereda-Miranda[1], J. Castañeda-Gómez[2], N. R. Jiménez-Bárcenas[1], R. Costa Simas[3], S. Guimarães Leitão[3]

[1]Departamento de Farmacia, Facultad de Química, Universidad Nacional Autónoma de México, Ciudad Universitaria, Mexico City 04510, México.
[2]Grupo Químico de Investigación y Desarrollo Ambiental. Programa de Licenciatura en Ciencias, Facultad de Educación. Universidad Surcolombiana, Nieva, Colombia.
[3]Faculdade de Farmacia, Universidade Federal do Rio de Janeiro, CCS, Bloco A, Ilha do Fundão, 21941-590, Rio de Janeiro, Brazil.

Brazilian Jalap root, a traditional medicinal plant complex still considered to be a useful treatment for enteric disorders due to its purgative activity, can be found as a crude root drug and powders sold by herbalists in traditional markets as well as an ingredient in some over-the-counter phytopharmaceuticals as pills, syrups, and hydro-alcoholic extracts retailed by drug stores in Brazil. Analysis of methanol-soluble resin glycosides from “batata-de purga” or “batatão”, O. hamiltonii (G. Don) D.F. Austin & Staples, the jalap root with yellow flowers and disseminated in the Northern Brazil, was assessed by generating HPLC and 13C-NMR spectroscopic profiles of the glycosidic acids obtained through saponification. Two glycosidic acids, operculinic acids M and N, and the known operculinic acid A, all having a common pentasaccharide moiety and 11- or 12-hydroxy fatty acid aglycones of different lengths were isolated by semipreparative recycling HPLC. The known pentasaccharide operculinic acid B and turpethic acid C were also isolated in addition to minor operculinic acid O, a new tetrasaccharide of 12-hydroxyheptadecanoic acid. Their structures were elucidated by high-field NMR spectroscopy and electrospray ionization mass spectrometry.

P4

Antidiabetic Effects of Naringanin-7-O-glucoside from Edible Chrysanthemum "Kotobuki" and Naringenin by Activation of the PI3K/Akt Pathway and PPARγ

Atsuyoshi Nishina[1]

[1]*College of Science and Technology, Nihon University, 1-5-1 Kandasurugadai, Chiyoda, Tokyo 101-0062, Japan*
Email address: nishina.atsuyoshi@nihon-u.ac.jp

Obesity is directly associated with type 2 diabetes, hypertension, cardiovascular injury, and cancer. On the other hand, adipocytes secrete adipocytokines and are known to affect metabolism and the function of many tissues. To date, Yamamoto identified that hot water extracts of edible chrysanthemum (EC) showed anti-diabetic effects such as improvement in insulin resistance and the down-regulation of the blood glucose level and liver lipid content in a type II diabetes model mouse. In the present study, we attempted to identify the antidiabetic components in EC. The methanol fraction was prepared from the dried hot water extract of edible chrysanthemum “Kotobuki”. The biological activities of extracts and isolated compounds were evaluated by measurement of the lipid accumulation levels and size of cells during adipogenesis by insulin, with thiazolidine (Rosiglitazone), an antidiabetic agent, as the positive control. The expression and phosphorylation levels of the related proteins in 3T3-L1 cells were measured by electroblotting. The methanol fraction from EC that showed relatively strong biological activity was purified by chromatography to obtain acacetin-7-O-glucoside, apigenin-7-O-glucoside, kaempferol-7-O-glucoside, and naringenin-7-O-glucoside. Among the isolated compounds and their aglycones, naringenin (NA) and naringenin-7-O-glucoside (NAG) up-regulated the intracellular lipid accumulation and secretion of adiponectin and down-regulated the diameter of 3T3-L1 cells during adipogenesis. Following measurements of the expression and/or phosphorylation levels of adipogenesis- and intracellular signaling-related proteins of NA and NAG, their effects were different from that of Rosiglitazone used as a positive control. Because the PPARγ antagonist BADGE and PI3K/Akt inhibitors wortmannin and LY29004 inhibited the intracellular lipid accumulation by NA and NAG associated with adipogenesis, it was considered that NA and NAG showed the above-mentioned activities via the activation of PPARγ as well as phosphorylation of the PI3K/Akt pathway. From the results of this study, it was deduced that NA and NAG were major antidiabetic components of EC.

P5

Antidiabetic in vitro and in vivo evaluation of cyclodipeptides isolated from bacteria

B. Ovalle-Magallanes[1], M. Lozano-González[1], R. Mata[1]

Facultad de Química, Universidad Nacional Autónoma de México, Ciudad de México 04510, México

Cyclodipeptides (CDP) of the diketopiperazine family of secondary metabolites produced by many bacteria and fungi. This class of compounds has recently gained interest due to its wide array of

biological activities, but, to our knowledge, there are no reports on the association between CDP and their effect in diabetes. In the present study we present the isolation from bacteria of four CDP [Cyclo (Pro-Leu), 1; Cyclo (Pro-Phe), 2; Cyclo (Pro-Leu), 3; Cyclo (Pro-Tyr), 4], and their respective synthesis. Furthermore, we analyzed their α-glucosidase inhibition potential using an in vitro enzymatic assay, and evaluated the most active compound in the sucrose, glucose and insulin tolerance tests in ICR mice. When given intragastrically, CDP 1 decrease sucrose and glucose levels in the postprandial state at the doses of 10 mg/kg and 30 mg/kg. In addition, preliminary results show that this compound favor insulin release from β-pancreatic cells.

P6

Preliminary qualitative and quantitative determination of flavonoids and antioxidant capacity from *Struthanthus quercicola* extracts

C. S. Arjona-Ruiz[1], D. A. de Loera-Carrera[1], R.M. Gamboa-León[2]

[1] *Facultad de Ciencias Químicas, Universidad Autónoma de San Luis Potosí, Av. Dr. Manuel Nava Núm. 6 Zona Universitaria San Luis Potosí, S.L.P. 78210 México*
[2] *Coordinación Académica Región Huasteca Sur, Universidad Autónoma de San Luis Potosí, Km. 5 Carretera Tamazunchale-San Martín, Tamazunchale, S.L.P. 79960 México*

Although medical plants have been used since ancient times to treat human diseases, nowadays there is growing interest in the health benefits of herbs and botanicals. In 2013, importance of alternative treatments as fundamental primary health service worldwide was accentuated by The Traditional Medicine Strategy proposed by World Health Organization (WHO). Antioxidants have an important role in the treatment and prevention of several diseases such as diabetes mellitus type 2 and cancer. Many secondary metabolites have shown towering antioxidant capacities, among them flavonoids are the most known due their diverse biological activities. *Struthanthus quercicola* has been used by *Tenek* Mexican ethnic group as an antidiabetic plant. Previous studies had reported and described the antidiabetic effect of Rutine, a flavonoid isolated from *Struthanthus subtilis* extracts, nevertheless, limited data is available concerning the effectiveness of this genus extracts. In the present work, preliminary qualitative and quantitative determination of flavonoids and antioxidant capacity from *S. quercicola* extracts is reported. The analysis of the results obtained indicates that there is a relation between the host and the flavonoid content, and consequently its possible antidiabetic activity. Furthermore, chemical and pharmacological screening looking for the hypoglycemic principles in the plant species should be intensified.

P7

Characterization of Betalain Extracts from Cultivated Vegetable Amaranth (*Amaranthus* spp.) for Use as a Natural Food Colorant

Chance W. Riggins[1], J.E. Howard[1]

[1]*Department of Crop Sciences, University of Illinois, 1201 West Gregory Drive, Urbana, Illinois 61801, USA*

Consumer demands for healthier foods with natural ingredients have evoked trends in the food industry to replace synthetically-produced colorants such as FD&C Red 40 with naturally-derived alternatives. Amaranth (*Amaranthus* spp.) is a particularly appealing color source due to the ability of certain species and/or cultivars to produce significant concentrations of betalains—more specifically magenta-red pigments called betacyanins. In addition, amaranth species grown for grain, leafy vegetables, and ornamental purposes are emerging specialty crops in the U. S. due to their generally favorable growth characteristics (e.g., drought and heat tolerance, low maintenance), high nutritional values, and wide variation in pigmentation patterns. Amaranth is generally considered an underutilized crop with high genetic diversity, thus there is great potential for further exploration and characterization of desirable traits and potential new uses. A study was conducted to explore the betalain diversity of different vegetable amaranth species and cultivars in comparison to the exclusive source of betalains in the food industry, red beets (*Beta vulgaris*). Above-ground biomass from 40 different amaranth genotypes was analyzed via HPLC and UV-VIS spectrophotometry to identify lines with high pigment concentrations and unique betalain compositions. Amaranthine and isoamaranthine were identified as the major color constituents of all extracts, with total betacyanin concentrations ranging from 0 to 73 mg betacyanins/100g fresh weight. In contrast to beets, none of the genotypes analyzed contained significant amounts of betaxanthins which impart a yellow hue to extracts. Colorimetric analyses were performed on extracts of the most pigmented genotypes to characterize their unique bright magenta color in reference to commercially available beet extracts. Data from this preliminary investigation are now being used to guide further chemical and agronomic research on select vegetable amaranth types to ascertain their economic potential in the United States and suitability for commercial betalain extraction for expanding colorant markets.

P8

Additional a-glucosidase inhibitors from Malbranchea flavorosea (Leotiomycetes, Ascomycota)

D. Rebollar-Ramos[1], M. L. Macías-Ruvalcaba[2], M. Figueroa[1], H. A. Raja[3], M. González-Andrade[4], R. Mata[1]

[1]*Facultad de Química, Universidad Nacional Autónoma de México, México, Ciudad de México 04510, México*
[2]*Instituto de Química, Universidad Nacional Autónoma de México, México, Ciudad de México 04510, México*

University of North Carolina at Greensboro, Greensboro, NC 27402, USA
[4]Facultad de Medicina, Universidad Nacional Autónoma de México, México, Ciudad de México 04510, México

From the rice-based culture of Malbranchea flavorosea, three new compounds namely flavoroseoside B (5-desoxy-5-chloroflavoroseoside) (2), 4-hydroxy-2-O-a-ribofuranosyl-5-methylacetophenone (3), and (S)-3,4-dihydro-3-(1H-indol-3-ylmethy)-4-methyl-1H-1,4-benzodiazepine-2,5-dione (4), along with three known compounds, rosigenin (5), massarilactone B (6), and riboxylarinol B (7) were obtained. The structures were determined by spectroscopic methods. Compound 4 and its synthetic analog 3,4-dihydro-3-(1H-indol-3-ylmethy)-1-methyl-1H-1,4-benzodiazepine-2,5-dione (9) inhibited the activity of Ruminococus obeum a-glucosidase enzyme. Molecular docking and dynamic studies revealed that compounds 4 and 9 might bind to this a-glucosidase at the catalytic center. Phylogenetic analysis using internal transcribed spacer region revealed that Malbranchea flavorosea ATCC 34529 is related to Myxotrichum spp.

P9

Characterization of SIP68: A Cytosolic UDP-Glucosyltransferase Enzyme that Interacts with Tobacco SABP2

Dhirendra Kumar[1], Saroj Lohani[1], Abdulkareem Odesina[1], Olivia Simo[1]

[1]Department of Biological Sciences, East Tennessee State University, Johnson City, TN 37614, USA

SIP68, a UDP-glucose: flavonoid glucosyltransferase has a conserved PSPG Box that is characteristic of family 1 glycosyltransferases. It was identified in a yeast two-hybrid screen in which tobacco SABP2 was used as bait. SABP2 is an important component of the salicylic acid-mediated pathogen response pathway in tobacco and many other plants. SABP2 is a methyl salicylate esterase that that catalyzes the conversion of SAR signal, methyl salicylate into salicylic acid. Glucosides are a ubiquitous class of secondary metabolites involved in roles ranging from the protection of plants against pathogens and herbivory to the physical appearance of plants, transportation of metals, symbiotic agents between plants and microorganisms, and acting as sexual hormones. The recombinant SIP68 showed relatively high activity with several flavonoids including kaempferol and quercetin while it failed to show any activity when salicylic acid was used as a substrate. Expression of SIP68 transcripts is modulated upon pathogen infection. The confocal microscopy of eGFP tagged SIP68 transiently expressed in *Nicotiana benthamiana* leaves showed that it is likely expressed in the cytoplasm. Subcellular fractionation using differential centrifugation of the tobacco leaves transiently expressing SIP68-+eGFP confirmed that SIP68 is localized in the cytosol. To study the role of SIP68 in plant stress signaling, transgenic lines with altered SIP68 expression were generated using RNAi and CRISPR Cas9. Some of these lines are currently being used to assess the role of SIP68 in stress response. Studying SIP68 will help to improve our general understanding of how plants respond to biotic and abiotic stresses.

P10

Anti-inflammatory and metabolic effects of *Calea urticifolia* lyophilized aqueous extract in high-fat diet-induced obesity C57BL/6 mice.

E.O. Segura Esparragoza[1], E. García Chávez[2], L. Vega Loyo[3], G. A. Soto Peña[4], E. Estrada Muñiz[3]

[1]Posgrado en Ciencias Farmacobiológicas, Facultad de Ciencias Químicas, Universidad Autónoma de San Luis Potosí, Av. Dr. Manuel Nava No.6, San Luis Potosí, S.L.P., México
[2]Instituto de Investigación de Zonas Desérticas (IIZD-UASLP)
[3]Centro de Investigación y Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN), unidad Zacatenco
[4]Facultad de Ingeniería (UASLP)

Traditional Mexican Medicine has been used since ancient times as a provider of primary health care at the community level, in San Luis Potosí, *Calea urticifolia* known as "negrito" by the Xi'iuy ethnic group, in San Luis Potosí, México, is traditionally used to decoction its leaves as a therapeutic remedy to treat diabetes, ulcers gastric and inflammatory processes. Through of promoting the study and scientific validation of traditional medicine through public policies that allow the design and operation of strategies for the purpose of their inclusion in health services and to strengthen them; the aim was to was to evaluate the inflammatory and metabolic regulation of lyophilized aqueous extract of *Calea urticifolia* in an *in vivo* model of low grade inflammation. Chronic low-grade inflammation in male C57BL/6 mice high fat diet-induced obesity was generated in 20 weeks and treated for 12 weeks with 0.55, 2.75, 5.5 or 11 mg/kg of lyophilized aqueous extract (CuAqE). Body weight, food and water intake of mice was monitored weekly throughout the study period. Fasting blood Glucose, Insulin, HbA1c, Leptin, Resistin, C-Peptide, GLP-1, Adiponectin TNF-α, IL-6, IFN-γ, IL-1β, IL-4, IL-10, IL-12p70, VEGF, Triglycerides and cholesterol (total, HDL and LDL) were measured. HOMA-IR, HOMA-β or %β, insulin sensitivity, insulin disposition index and QUICKI were calculated. The group of mice with a high-fat diet leads to the development of obesity and low-grade inflammation, which was reflected by the induction of hyperglycemia, insulin resistance and dyslipidemia during 32 weeks. CuAqE modulates the dose-dependent energy metabolism; decreases caloric intake, body weight, insulin, glucose, proinflammatory cytokines such as TNF-α and Il-6, leptin, triglycerides and cholesterol, also increases adiponectin and anti-inflammatory cytokines such as IL-10 and IL-4 ($p<0.001$), improves insulin resistance. The aqueous extract of *Calea urticifolia* could be considered as a potential treatment to treat the metabolic disorders present in obesity and related diseases.

P11

Development of a green methodology for the extraction of saponins from *Dioscorea composita*

G. Guerrero-Luna[1], J. Reyes-Melchor[2], M. G. Hernández-Linares[3,4], M. Rodríguez-Acosta[4], A. Carrasco-Carballo[1], J. Sandoval-Ramírez[5]

[1]*Doctorado en Ciencias Químicas-BUAP, Ciudad Universitaria, 72570 Puebla, Pue,*
[2]*Licenciatura en Biotecnología;*
[3]*Instituto de Ciencias;*
[4]*Laboratorio de Investigación del Jardín Botánico;*
[5]*Facultad de Ciencias Químicas-BUAP, Puebla, México*

Saponins are steroid or triterpenoid glycosides that occur in a wide variety of plants, and used for diverse purposes for humans. In Mexico, the roots of Dioscorea composita (commonly named as barbasco), is found mainly at the humid semi-tropical zone; for example, at the Sierra Norte of Puebla. Extensive research has been carried out on its steroid derivatives, saponins and sapogenins. Nowadays, some biological activity as antifungal, antiviral, anti-inflammatory, antimicrobial, anticancer, has been reported. It has been highlighted in the literature and according to the results of our research, that the carbohydrates present in saponins can potentiate their bioavailability and activity. Therefore, it is important to continue on the research for a fast, less expensive and environmentally friendly methodologies (green chemistry) to obtain greater quantities of the valued saponins. A green process for the extraction of dioscin using aqueous phase, short extraction times and purification. The methodology is applicable to other substrates, thus it is possible to selectively obtain water-soluble compounds. The obtained dioscin has been used in the evaluation of its antiproliferative activity, and as a raw material for obtaining steroidal anticancer analogs.

P12

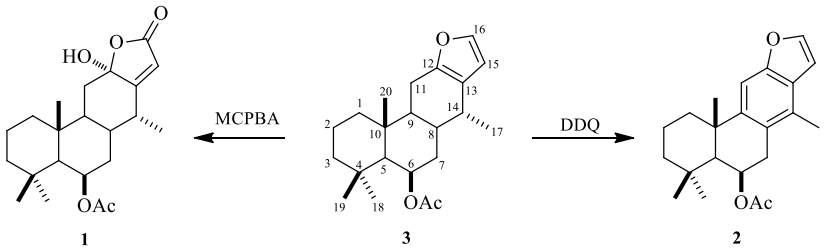
Oxidative Derivatization of 6*β*-Acetoxivouacapane from *Caesalpinia platyloba*

Gabriela Servín-García[1], Armando Talavera-Alemán[1], Mario A. Gómez-Hurtado[1], Gabriela Rodríguez-García[1], Carlos M. Cerda-García-Rojas[2], Pedro Joseph-Nathan[2], Rosa E. del Río[1*]

[1]*Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Morelia, Michoacán 58030, México.* [2]*Departamento de Química, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Apartado 14-740, Mexico City 07000, México*
**Corresponding author, email address: ndelrio@umich.mx*

Secondary metabolites are frequently isolated in low yields, limiting further studies to establish their potential applications. Fortunately chemical modifications provide solutions to this kind of situations which include strategies like structural changes or total synthesis. In some cases natural compounds as raw materials for organic synthesis have some advantages to explore complex molecular structures. Of relevance is that many natural products contain one or more stereogenic centers and their optical purity is probably the most important and valuable structural characteristic. Terpenoids usually are chiral and their structural variations provide a wide range of uses, including those directed to industry. Cassane diterpenes comprise the representative group of natural products in the *Caesalpinia* genus. When a furan ring is present in the cassane skeleton, they are known as furan cassanes or vouacapanes. Several of these compounds present biological activity, including cytotoxic and anti-inflammatory activities. As example, caesalpinolide-E (**1**), together with 6*β*-acetoxyvouacapan-8(14)-9(11)-diene (**2**) were isolated from *C. bonduc*, the

cytotoxic potential determination of **1** was possible, but in the case of **2** this was not possible due to low concentration in plant. In a previous work, the absolute configuration determination of 6*β*-acetoxyvouacapane (**3**) isolated from *C. platyloba* was reported. Isolation of **3** in good yields permitted the first photooxidative study in vouacapanes, providing diterpenic lactones. In the present work oxidative studies of **3** are described. Column chromatography of the CH_2Cl_2 extract of the leaves of *C. platyloba* provided **3**, whose oxidative treatment, using MCPBA and DDQ gave known lactone **1** and diene **2** in 70% and 40% yield, respectively. Physical and spectroscopic data were concordant with those reported. This result represents an alternative to prepare **1** and **2** for investigations directed towards chemical and biological studies. In addition, a chemical relation establishment between *C. platyloba* and *C. bonduc* is possible through this study, despite its native ecosystems are in Mexico and India, respectively.



1 **3** **2**

P13

Absolute Configuration Determination of Epoxythymol Derivatives from *Ageratina glabrata* by Vibrational Circular Dichroism

Héctor M. Arreaga-González[1], Gabriela Rodríguez-García[1], J. Martín Torres-Valencia[2], J. Jesús Manríquez-Torres[3], Rosa E. del Río[1], Carlos M. Cerda-García-Rojas[4], Pedro Joseph-Nathan[4], Mario A. Gómez-Hurtado[1]

[1]*Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo, Morelia, Michoacán 58030, México*
[2]*Área Académica de Química, Universidad Autónoma del Estado de Hidalgo, Mineral de la Reforma, Hidalgo 42184, Mexico*
[3]*Área Académica de Medicina, Instituto de Ciencias de la Salud, Universidad del Valle de México, Tezcatlipoca 2301, Saltillo, Coahuila 25204, Mexico*
[4]*Departamento de Química, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Apartado 14-740, Mexico City 07000, Mexico*
**Corresponding author, email address: magomez@umich.mx*

Vibrational circular dichroism (VCD) is an efficient chiroptical spectroscopic method for the absolute configuration (AC) determination of natural products. Measurements are performed in solution, which is perhaps a main advantage of this method, although low sensibility of vibrational absorptions is associated to the methodology. A comparison of the experimental and calculated spectra is required. Thus, a conformational distribution exploration by the Monte Carlo protocol,

using the MMFF level of theory is suggested after which energy and geometry optimizations by density functional theory (DFT) calculations are required. This is followed by calculation of VCD and IR spectra from Boltzmann-averaged abundant conformers, and finally, a statistical comparison of the calculated and the experimental spectra provides a quantitative spectra similarity and the level of confidence for the correct enantiomer of the studied molecule. The first configurational study of an epoxythymol derivative was performed using this methodology a few years ago. In continuation with our research dealing with the constituents of the *Ageratina* genus, the AC determination of (+)-(8*S*)-10-benzoyloxy-6-hydroxy-8,9-epoxythymol isobutyrate (**1**), (+)-(8*S*)-10-acetoxy-6-methoxy-8,9-epoxythymol isobutyrate (**2**), and (+)-(8*S*)-10-benzoyloxy-6-methoxy-8,9-epoxythymol isobutyrate (**3**) from *A. glabrata* is described herein. The initial statistical comparison results for the AC of **1** reflected a low spectroscopic similarity, and therefore an extensive systematic conformational search was carried out by rotating bonds in small dihedral angle amounts. The procedure provided the initially overlooked conformers and the new spectra comparison showed adequate level of confidence for the correct enantiomer. Rotational analyses in **2** and **3**, employing the PC model program, revealed rotational barrier energies for the C-3−O-3−C-1′−C-2′ and C-4−C-8−C-10−O-10 dihedral angles higher than 10 kcal/mol. Thus, complete conformational searches starting from four different models provided the complete conformational distribution determination. Identical conformers provided by the four searches were discarded. Subsequent DFT calculations, and VCD and IR statistical comparison provided good spectra similarities and confidence levels to establish the AC of **2** and **3**. This calculation strategy seems suitable for molecules with high conformational flexibility.

P14

Phytochemistry of prickly pear (Opuntia spp.) juices

G. Zenteno-Ramírez[1], B.I. Juárez-Flores[1], J.R. Aguirre-Rivera[1], J.A. Rendón-Huerta[2], C.I. Godínez Hernández[1]

[1]*Instituto de Investigación de Zonas Desérticas, Universidad Autónoma de San Luis Potosí, Altair 200, Colonia Del Llano, 78377 San Luis Potosí, San Luis Potosí, México*
[2]*Coordinación Académica Región Altiplano Oeste, Universidad Autónoma de San Luis Potosí, Avenida Insurgentes esquina Himno Nacional S/N, 78600. Salinas de Hidalgo, San Luis Potosí, México*

Prickly pears (Opuntia spp.) can be white, purple, red, orange or yellow, with physical and chemical differences. The objective of this study was to identify and quantify the sugars (glucose, fructose and sucrose), soluble fiber (mucilage and pectin) and secondary metabolites (total phenolic compounds, phenolic acids and flavan-3-ols, ascorbic acid and betalains) of the juice from mature cultivated and wild prickly pears. The cultivated variants were: Rojo Pelón (O. ficus-indica), Blanca or Cristalina (O. albicarpa), Amarilla Monteza, Pico Chulo, Torreoja and Sangre de Toro (O. megacantha); the wild variants were: Cardona (O. streptacantha), Charola (O. streptacantha ssp. aguirrana), Tapona and Tapón Rojo (O. robusta). The experimental design was completely randomized, with three repetitions. The results were analyzed through ANDEVA and multiple mean comparisons were carried out with the Tukey test ($p <= 0.05$). Differences ($p<0.0001$) were found in the concentration of total sugars in the juice (109.10 to 151.33 mg mL-1); glucose predominated (95.75 mg mL-1), followed by fructose (63.90 mg mL-1), and sucrose had minimum amounts only in

some variants. The mucilage predominated over the pectin. Tapón Rojo had the higher total sugars and soluble fibers content. Gallic acid was abundant in most variants. Catechin and epicatechin isomers, and procyanidins B1 and B2 were present in most variants. Ascorbic acid content was higher than 84 mg. Betacyanins stand out in red-colored juices; betaxanthins in the yellow ones. There were significant differences in the content of sugars and soluble fiber among variants. The sugar concentration did not show a direct relation to the degree of variant humanization.

P15

Pharmacopeial Identity Parameters for Achillea millefolium L. (Asteraceae) growth in Mexico

I.Y. Martínez-Aldino[1], B. Ovalle-Magallanes[1], R. Torres-Colín[2], R. Mata[1].

[1]Departamento de Farmacia, Facultad de Química, [2]Instituto de Biología, Universidad Nacional Autónoma de México, Mexico City, 04510, Mexico.

Achillea millefolium L. (Astereaceae) is a medicinal herb commonly known in Mexico as "milenrama", "mil hojas" or "ciento en rama". The infusion and essential oil of the aerial parts of the plant are used to treat stomachache, diarrhea and vomita, and as analgesic, antidiabetic, and antiespasmodic agentsb. Despite its broad use in Mexican traditional medicine, identity and composition parameters for quality control of the species growth in Mexico are lacking. Therefore, in order to establish identity tests from A. millefolium, chromatographic profiles of the species using HPLC-PDA and GC-MS were obtained. The profile of a dichloromethane fraction of the infusion was obtained using HPLC-PDA, a gradient elution of acetonitrile-water, and a C-18 reversed phase and the major compounds were identify as the guaianolide sesquiterpene lactones leucodine, achillin and matricarin. Analyses with GC-MS allowed the identification of the main components of the essential oil, being α-thujone, (+)-2-bornanone and chamazulene, the most abundant. The chromatographic profiles obtained constitute the basis for the development and validation of analytical methods useful for quality control of this species.

P16

Chemical and Biological Studies of Ascomycetes from Cuatro Cienegas Basin, Coahuila, Mexico

I.R. Yeverino[1], M. Figueroa[1]

[1]Departamento de Farmacia, Facultad de Química, Universidad Nacional Autónoma de México, Ciudad de México 04510, México

As part of our continuing search for novel antimicrobial compounds from unexplored habitats of Mexico, a series of 20 saprotrophic fungi were isolated from soil and sediment samples collected at the Cuatro Cienegas Basin, Coahuila, Mexico. Taxonomic diversity of fungal isolates was assessed by

nuclear ribosomal internal transcribed spacer barcoding. The organic (CHCl3-MeOH) extracts from the axenic solid (moisture rice) cultures were tested against Escherichia coli, Salmonella typhi, Pseudomonas aeruginosa, Staphylococcus aureus, Bacillus subtilis and Candida albicans. Additionally, they were dereplicated against a database containing more than 300 fungal secondary metabolites via recording UPLC retention times, UV data, and full-scan (high-resolution) mass spectra and MS/MS spectra in both positive and negative electrospray ionization modes. Bioactive-guided fractionation (growth inhibition 80% at 20 ug/mL and negative dereplication) of the scaled-up (10x) cultures led to the isolation of a series of butyrolactones derivatives from the Aspergillus sp. (CC1-1); the bis-indolyl-dyhydroxybenzoquinone cochliodinol from a fungus of the order Sordariales (CC9-6); and the ergochrome neosartorin from the Aspergillus sp. (CC7-12). Their structures were elucidated using 1D and 2D NMR and HRMS data analysis. To the best of our knowledge, this is the first report of chemical and biological studies of Ascomycetes isolated from the Cuatro Cienegas Basin.

P17

Ska Pastora and the Enzymes Along the Way to Salvinorin A

J.A. Velasco[1], X. Chen[1], D.R. Gang[1]

[1]*Institute of Biological Chemistry, Washington State University, WA 99164, USA*

Salvia divinorum (Lamiaceae), known as "Ska Pastora" to the Mazatec people of northern Oaxaca is exclusive to a small region of the Sierra Mazateca in Mexico. Salvinorin A, the main bioactive metabolite synthesized by *S. divinorum* is a neo-clerodane diterpenoid of interest to the pharmacological community because of its biological activity. Salvinorin A is the only known naturally synthesized non-nitrogenous *kappa*-opioid receptor agonist and its analogs have potential for use in the treatment of multiple neuropsychiatric conditons and various drug addictions. This compound is primarily synthesized and accumulated in the plant's glandular trichomes, though its biosynthetic pathway is not yet entirely known. Here we plan to expand on previous work which reported (-)-kolavenyl diphosphate synthase as the enzyme which catalyzes the first step in the synthesis of salvinorin A. An *S. divinorum* trichome-specific transcriptome will be will be probed with the purpose of identifying candidate enzymes for further investigation and characterization via recombinant expression and *in vitro* enzyme activity assays. Enzymes of interest include: cytochrome P450 monooxygenases, NAD-dependent dehydrogenases, NAD(P)H-dependent reductases, and acetyltransferases, among others.

P18

Assessing Flux Distribution Associated with Metabolic Specialization of Glandular Trichomes

Jordan J. Zager[1], B. Markus Lange[1]*

1 Institute of Biological Chemistry and M.J. Murdock Metabolomics Laboratory, Washington State University, Pullman, WA 99164, USA
**Corresponding author, email address: lange-m@wsu.edu*

Many aromatic plants accumulate mixtures of secondary (or specialized) metabolites in anatomical structures called glandular trichomes (GTs). Different GT types may also synthesize different mixtures of secreted metabolites, which contributes to the enormous chemical diversity reported to occur across species. Over the last two decades, significant progress has been made with characterizing genes and enzymes responsible for the unique metabolic capabilities of GTs in different lineages of flowering plants. Less is known about the processes that regulate flux distribution through precursor pathways toward metabolic end products. Here, we discuss the results from a meta-analysis of genome-scale models developed to capture the unique metabolic capabilities of different GT types.

P20

Recycling HPLC as an Analytical Methodology for the Purification of Hederifolic Acids from Ipomoea hederifolia

L.K. Gualteros-Montaño[1] , J. Castañeda-Gómez, *,[2] R. Pereda-Miranda,*,[1] S. Guimarães Leitão[3]

[1]*Departamento de Farmacia, Facultad de Química, Universidad Nacional Autónoma de México, Ciudad Universitaria, Mexico City 04510, Mexico*

[2]*Grupo Químico de Investigación y Desarrollo Ambiental. Programa de Licenciatura en Ciencias Naturales y Educación Ambiental, Facultad de Educación. Universidad Surcolombiana, Neiva, Colombia*

[3]*Faculdade de Farmacia, Universidade Federal do Rio de Janeiro, CCS, Bloco A, Ilha do Fundão, 21941-590, Rio de Janeiro, Brazil*

Scarlet morning glory or scarlet creeper (Ipomoea hederifolia Linn., Syn. Ipomoea coccinea and Quamoclit coccinea) is an annual vine belonging to the Convolvulacea family. This species occurs mostly in disturbed sites throughout North and South America. It is considered an invasive plant in coffee plantations. Ipomoea hederifolia has been described to possess anticancer, anti-inflammatory, anti-microbial, anti-oxidant and oxytoxic properties, according to indigenous systems of medicine in India. The scraped roots are eaten to treat stomach aches. Decoction of the root is used for intestinal parasites. This morning glory has been used as a mild purgative for the treatment of nervous disorders and against tumors. The methanol extract was prepared from the aerial parts and fractionated by column chromatography. Then, the fractions rich in resin glycosides were saponified to yield the mixture of glycosidic acids, which were peracetylated. Reclying HPLC of the resulting mixture was used for purification of four new glycosidic acids, hederifolic acids A and B, as heptaglycosides of 3,12-dihydroxy fatty aglycones, along with two new heptaglycosidic acids C and D, identified by electrospray ionization mass spectrometry and NMR spectroscopy as their congeners with 3-deoxy-12-hydroxy fatty acid aglycone.

P21

Understanding the contribution of the four ascorbate pathways to abiotic stress tolerance in arabidopsis using phenomics approaches

Lucia Acosta Gamboa[1]

[1]*Arkansas Biosciences Institute, Arkansas State University, 4301 W Markham St #821, Little Rock, AR 72205, USA*

L-Ascorbic acid (AsA, vitamin C) is a key antioxidant that counteracts the excess reactive oxygen species that are accumulated in response to stresses. Ascorbate is also an enzyme cofactor and a modulator of cell division, cell expansion, and plant senescence. Biosynthesis of AsA in plants is carried out by a complex metabolic network involving D-mannose/L-galactose, D-galacturonate, L-gulose, and myo-inositol as main precursors. We have previously shown by manual phenotyping that Arabidopsis lines over-expressing enzymes in the myo-inositol pathway have elevated AsA, accumulate more biomass of both aerial and root tissues, and are tolerant to abiotic stresses including salt, cold, heat, and environmental pollutants. In this work we crossed a high AsA line over-expressing a myo-inositol oxygenase gene (MIOX4) with two low-vitamin C mutants (vtc1-1 and vtc2-1) encoding enzymes in the D-mannose/L-galactose route. The purpose of making these crosses was two-fold: first test MIOX4's ability to restore the low AsA phenotype of the mutants, and second to assess the contribution of individual biosynthetic pathways to abiotic stress tolerance. We are using a powerful high throughput phenotyping platform to characterize in detail the phenotype of the Arabidopsis lines with visible, fluorescence, and near infrared sensors. Our results show that MIOX4 is able to restore the AsA content of the mutants and that high-AsA lines grow faster, accumulate more biomass, and display healthier chlorophyll fluorescence and water content profiles than controls. By studying abiotic stress in a high throughput fashion using optimized protocols, we have also shown that these high-AsA lines are more tolerant to salt and water limitation stresses than controls. Our next step is a more detailed characterization of the crosses to better understand the contribution of the various AsA pathways to abiotic stress tolerance using a combination of genetic, transcriptomic, and phenomic approaches.

P22

Phytochemical study of Peach (*Prunus persica* (L) Bafsch)

M. Soto-Hernández[2], M. Palma-Tenango[1], R. Sánchez-Fernández[2], I. Ortiz-Olvera[3], Rubén San Miguel Chávez[2]

[1]*SEPI-ESIME, Instituto Politécnico Nacional. Professional Unidad 'Adolfo López Mateos', Col. Lindavista, México D.F., C.P. 07738, México*
[2]*Programa en Botánica, Colegio de Postgraduados-Campus Montecillo. km 36.5. Carr. México-Texcoco. 56230, Montecillo, Texcoco, Estado de México*
[3]*Posgrado en Socio economía. Colegio de Postgraduados-Campus Montecillo. km 36.5. Carr. México-Texcoco. 56230, Montecillo, Texcoco, Estado de México*

In Mexico, the peach variety "criollo" (*Prunus persica* (L) Bafsch) is economically important. The fruits are traded and the producers of this species are looking for alternative uses in their orchards. Due to this, analyses of leaves, flowers and fruits of peaches from an orchard located in the town of San Martin Tequesquipan, municipality of Temascaltepec, State of Mexico, were carried out. Total phenolic content expressed as gallic acid equivalents was determined through *UV-Vis* spectroscopy. Phenolic and flavonoid content was determined through HPLC and the identification of the volatile components was done through Solid Phase Micro-Extraction in the vapor phase (HS-SPME) followed by Gas Chromatography coupled with Mass Spectrometry (GC-MS). Total phenolic content in the fruit was 6.8 mg/g D.W. There was presence of gallic acid, chlorogenic acid, vanillic acid, naringin, naringenin, hesperidin, phloretin and galangin. The volatile compounds detected in flowers, fruits and leaves of peach are mainly terpenes and aldehydes. In addition, some alkanes, esters, benzene derivatives, alcohols, lactones and a carboxylic acid were detected. There was variation in the type and content of phenolics, flavonoids and volatile compounds depending on the organ involved.

P23

Same glycosidation sequence in the resin glycosides from the Mexican and Brazilian jalaps

M. E. Montiel-Ayala[1], J. J. Lira-Ricárdez,[1] R. Pereda-Miranda[1], Suzana Guimarães Leitão[2]

[1]*Departamento de Farmacia, Facultad de Química, Universidad Nacional Autónoma de México, Ciudad Universitaria, Mexico City 04510, Mexico*
[2]*Faculdade de Farmacia, Universidade Federal do Rio de Janeiro, CCS, Bloco A, Ilha do Fundão, 21941-590, Rio de Janeiro, Brazil*

Mexican and Brazilian Jalap roots (*Ipomoea purga* and *Operculina macrocarpa*, respectively) are traditional medicinal plants with purgative activity. Ethnobotanical publications quoted their uses as antihelmintic, blood purifier, skin cleaner, anti-stroke, and as a treatment for uterine infection in the form of dried roots slices. Phytochemical studies of both crude drugs have described that resin glycosides are the principal chemical entities found in the root extracts, which represents a mixture of some different glycolipids formed by monohydroxy and dihydroxy long-chain fatty acids bounded to a lineal or branched heteropolysaccharides. *Ipomoea purga*, the authentic "jalap root", yielded two major hexasaccharides of 11S-hydroxytetradecanoic acid 11S-hydroxyhexadecanoic acid, the known purgic acids A and B. Operculinic acid J was isolated from *O. macrocarpa* and characterized as presenting 12-hydroxytetradecanoic acid as its aglycon but having the same glycosidation sequence as purgic acids. This novel glycolipid was identified as (12*S*)-hydroxytetradecanoic acid 11-*O*-β-D-quinovopyranosyl-(1→2)-*O*-β-D-glucopyranosyl-(1→3)-O-[β-D-fucopyranosyl-(1→4)]-*O*-α-L-rhamnopyranosyl-(1→2)- *O* -β-D-glucopyranosyl-(1→2)-*O*-β-D-quinovopyranoside.

P24

Partial synthesis of steroidal oximes from saponins of *Dioscorea mexicana* and their antiproliferative activity

M. G. Hernández-Linares[1], G. Guerrero-Luna[1,2], A. Carrasco-Carballo[1,2], M.A. Fernández-Herrera[4], J. Sandoval-Ramírez[3]

[1]*Instituto de Ciencias, Laboratorio de Investigación del Jardín Botánico;*
[2]*Posgrado en Ciencias Químicas;*
[3]*Facultad de Ciencias Químicas, Benemérita Universidad Autónoma de Puebla, C.U. Col San Manuel. C.U., Colonia San Manuel, Puebla, Pue., México. C.P. 72570*
[4]*CINVESTAV Unidad Mérida.*

Saponins are high-molecular-weight glycosides with a sugar moiety linked to a triterpene or steroid aglycone. They are constituent of many plants drugs and folk medicines, such as the roots of *Dioscorea composita* (Barbasco). In our research group, we have developed efficient methodologies for obtaining dioscin from Barbasco. In addition to evaluating its antiproliferative activity, the dioscin has been used as a raw material in the partial synthesis of new steroidal oximes, as analogs of steroidal anticancer alkaloids. Cancers figure among the leading causes of morbidity and mortality worldwide, an extensive research of cancer chemotherapy is generated; there is, however, a continuing need for new treatments inspired by medicinal chemistry and drug design. Synthetic chemistry has been find a solution to the problem of the limited supply of natural products by developing synthetic broadly employed to modify drug targets, especially those of natural origin. Steroidal oximes have shown high activity against cancer cell lines; however, the extraction of natural sources and the total synthesis of compounds has not sufficient, wherein the partial synthesis becomes relevant. Using as a starting material diosgenin, our group has synthesized and evaluated the spirostanic oximes with oxime group in the skeleton and the side chain with worthy results. In chemotherapy, the mechanism of action of the drugs used is based on their cytotoxic activity, inducing the tumor and non-tumor cells to a necrotic death, with undesirable side effects. Therefore, it is essential to generate drugs with great efficiency like classical, but with antiproliferative activity in tumor cell lines with low or no cytotoxic activity. The results obtained from synthetized oximes show important antiproliferative activity and scarce cytotoxic activity in cervical cancer cell lines, without cytotoxic activity in human peripheral blood lymphocytes, which profile them as promising therapeutic agents.

P25

Optimization of Fungal Treatment and Processing Parameters to Increase Glyceollin Production in Soybeans

M. Berhow[2], S Wootton[1], B. Karki[1], W.R. Gibbons[1]

[1]*Biology and Microbiology Dept. South Dakota State University, Brookings, SD, 57007*
[2]*United States Dept. of Agriculture, National Center for Agricultural Utilization Research, Peoria, IL, USA.*

As size and number of world populations continue to increase, the ability to feed those populations becomes progressively challenging. Recurrent use of antibiotics disseminates an inevitable increase in antibiotic-resistant microorganisms. This drives a need to explore sources of natural antimicrobial compounds and then to consider how to apply such compounds. One way to possibly apply natural antimicrobials is supplementing them into feeds. Studies have shown fungal infection promotes a response by soybean seeds to accrue glyceollin; however, the glyceollin titers are at low levels. In this study our goal is to evaluate and identify the optimal processing parameters to producing high glyceollin content in soybeans using the fungal metabolic process. In a previous study, our research team at South Dakota State University identified *Trichoderma reesei* NRRL 3653 as the best performing strain, stimulating the highest total glyceollin yield among several other tested strains. Hence, in this study, various processing parameters such as seed soaking time, inoculation method, incubation time, seed germination effect, seed varieties, etc. have been evaluated. Although the research is at the early stage, the preliminary findings have exhibited there is no major parallel between the seed germination and glyceollins level in soybeans. Likewise, increase in fungal incubation time increased the glyceollins production in soybeans (0 mg/g at 0 hrs Vs. 0.812 mg/g at 120 hrs of incubation). We fully expect that process optimization will allow us to enhance the glyceollin titer in soybeans considerably; thereby, boosting the commercial value of soybean and its market.

P26

Characterization of *Lippia graveolens* HBK essential oil extracted by supercritical fluids

N.A. Luna[1], O.J. Calva[1], A. De León-Rodríguez[1], A. Barrera-Pacheco[1], A.P. Barba de la Rosa[1]

[1]*Instituto Potosino de Investigación Científica y Tecnológica, Camino a la Presa San José 2055, Lomas 4 sección, CP. 78216, San Luis Potosí, México*

Mexico is the second exporter worldwide of Oregano. 90% of this production corresponds to the species *Lippia graveolens* HBK, catalogued as non-timber forest product and whose use is well regulated due to high economic importance. Commercial uses include the automotive, cosmetic, beverage, and food industries. Antibacterial, antifungal, antiparasitic, anti-inflammatory and antiviral activity has also been reported for the essential oil. The composition of the essential oil is similar amongst species showing representative differences in two monoterpenes: thymol and carvacrol. These compounds determine the quality and price of essential oil in the market. Traditionally, oregano essential oil is extracted by steam distillation or extraction with organic solvents. However, both methods can become inefficient and polluting. Extraction with supercritical fluids is considered the most efficient green technology of recent years for obtaining essential oils, allow obtaining extracts without final residues, without subsequent stages of concentration and high yields. In this work the method of obtaining of the essential oil of *L. graveolens* HBK was standardized by supercritical fluids. The essential oil was obtained from three samples of *L. graveolens* HBK from different locations and its characterization was carried out by gas chromatography- mass spectrometry (GC-MS). We show the difference in yields between the traditional extraction methods and the supercritical fluids method as well as the difference in the composition of the essential oil of *L. graveolens* HBK cultivated in different localities in the country.

P27

Identity tests of Cunila lythrifolia and Clinopodium macrostemum (Lamiaceae)

O.D. Jiménez-Martínez[1], B. Ovalle-Magallanes[1], R. Torres-Colín[2], R. Mata[1]

[1] Departamento de Farmacia, Facultad de Química, [2]Instituto de Biología, Universidad Nacional Autónoma de México, Ciudad Universitaria, Mexico City, 04510, Mexico

Cunila lythrifolia Bentham and Clinopodium macrostemum var. laevigatum are two aromatic species known as "poleo de monte" y "poleo" respectively. These species are widely used for the treatment of respiratory diseases (flu, cough and hoarseness), and to relieve digestive disorders such as diarrhea, vomiting, stomachache and dyspepsia a,b. As part of our research on Mexican Medicinal plants, the main objective of the work was to carry out a comparative study of the essential oils of these species, in order to obtain chromatographic profiles useful as identity parameters. The essential oils prepared by hydrodistillation were analyzed using gas chromatography coupled to mass spectrometry (GC-EM). The major compounds of C. lythrifolia were α-caryophyllene and β-caryophyllene; while those of C. macrostemum var. laevidatum turned out to be menthone and pulegone. The chromatographic profiles developed are the basis for chemical identity tests and quality control of these species.

P28

Extraction and study of the electrical properties of bixina

O. Cárdenas González[1], M. Rivera Mateus[1], S. J. Ojeda[1], A. Cárdenas-Chaparro[1]

[1]Laboratory of Spectroscopy and Instrumental Analysis, Research Group of Molecular-Chemistry Physics and Computational Modeling (QUIMOL), Pedagogical and Technological University of Colombia (UPTC), Central North Avenue, Tunja, Boyacá, CP 150001, Colombia

The plant Bixa orellana L., commonly known as achiote, is a native to tropical America species. From the seeds the bixin is obtained, a natural pigment whose code corresponds to E-160b according to the European Union. It has a red pigment, used in the food, cosmetic and pharmaceutical industries. Studies have shown that bixina has an antioxidant, antimicrobial, antifungal and antineoplastic potential, and recently experimental tests have been carried out, making known their possible applications in the production of solar cells sensitized with dyes. This allows to deepen the study of its electric transport properties. In this project the experimental and theoretical study of the physicochemical and electrical properties of bixin is being developed. This pigment was extracted from the species Bixa orellana L., by soxhlet, ultrasound and microwaves, comparing them with the traditional extraction method in alkaline medium. The characterization and determination of purity was carried out by different spectroscopic techniques (FT-IR, UV-Vis, 1H and 13C NMR) and quantitative analytical methods such as GC-MS and HPLC. Better results were obtained using ultrasound with 98% purity and 5% yield, while in an alkaline medium the yield was 10% with the presence of up to 50% of impurities. The physicochemical characterization of electronic transport was carried out by linear voltammetry through gate voltages, the corresponding charge migration and its effects on the molecular system. The theoretical electrical properties will be calculated by

the method of Green functions through the Hamiltonian Strong Link and Landauer formalism, which is appropriate to describe the electronic dynamics of this system when placed between metal contacts to analyze conductive or semiconductor properties of the bixina.

P29

Antiadipogenic effects of Mexican grown *Physalis ixocarpa* extracts, in human pre-adipocytes

P. Leal-Puerta[1], D.E. Cruz-Vega[1], J.E. Moreno-Cuevas[1], V. Robledo-Torres[2], M.T. González-Garza[1].

[1]Tecnologico de Monterrey, Escuela de Medicina y Ciencias de la Salud, Ave. Morones Prieto 3000 Pte., 64710, Monterrey, N.L., Mexico
[2]Departamento de Horticultura, Universidad Autonoma Agraria Antonio Narro, Calz. Antonio Narro 1923, Buenavista, 25315 Saltillo, Coah., Mexico

Obesity has become a major public health problem, this complex and multifactorial disease along with overweight, affects over a third of the world's population, and the financial cost to manage its complications is already a burden on public healthcare. Herbal extracts contain many bioactive components and therefore exert a diverse array of biological activities. According to previous data, in a korean study, aqueous extracts of fruits and stem and leaves of Korean *Physalis angulata*, inhibited lipid synthesis. The aim of this research was to evaluate the possible anti-adipogenic effects of extracts of *Physalis ixocarpa* B., var. Gran Esmeralda and var. Morado Tamazula, on the lipids synthesis, using pre-adipocytes derived from human mesenchymal stem cells (ATCC® PCS-210-010™) as a cell model. Aqueous, ethanolic and methanolic extracts from stem and leaves, and aqueous extracts from fruits were prepared. Cell viability assay was performed in order to determine non-toxic doses of the extracts. To evaluate the effects on lipid synthesis, cell cultures were incubated during 15 days with adipogenic medium plus 1, 10 or 100 μg/ml of extracts. To verify lipid production, Oil Red O staining was performed. Results showed aqueous extract from *P. ixocarpa* B. var. Morado Tamazula fruits (100 μg/ml,) and stem and leaves (10 μg/ml) inhibited 65.8% and 45% respectively. None of the methanolic or ethanolic extracts showed significant activity. Regarding *P. ixocarpa* B. var. Gran Esmeralda, only the aqueous extract from fruits at 10 μg/ml inhibited 47% of lipid accumulation. These results demonstrate that *P. ixocarpa* has anti-adipogenic effects, these are the first antiadopogenic reported from this plant, and justifying further investigations.

P30

Characterization of Juice, Skins, Seeds, and Whole Blackcurrant (Ribes nigrum) Berries After Ultrasound-Assisted Water-Based Extraction, Biochemical Inhibition of α-Amylase, α-Glucosidase and Dipeptidyl Peptidase-IV

Regina Cortez[1]

[1]*Department of Food Science and Human Nutrition, University of Illinois at Urbana-Champaign, IL 61801, USA*

The aim was to characterize phenolic compounds in four cultivars of blackcurrant (BC) (Ribes nigrum) grown in the Midwestern USA after water-based ultrasound-assisted extraction (UAE). Cultivars (Titania, D16-6-54, Consort, and D16-8-14) were dissected into parts (juice, skins and seeds). Whole berries and the separated parts were evaluated after 2, 4, 6 h UAE for total anthocyanins (TA), total tannins (TT), total polyphenols (TP), colorimetric measurements followed by HPLC quantification, LC-ESI-MS identification, and determination of the % inhibition of α-amylase, α-glucosidase and dipeptidyl peptidase (DPPIV). There was a 215% difference in hue angles between the whole berries after 2 h and 4 h UAE and 640% difference between 2 h and 6 h. Skins of Titania, Consort and D16-8-14 had the highest TA overall concentration (19.0 ± 2.0, 19.7 ± 2.7 and 20.3 ± 3.5 μg eq. cyanidin 3-O-glucoside (C3G)/mg dry weight (DW), respectively) (p 0.05). Consort and D16-8-14 had the highest concentration of TT in the whole berries with no statistical difference between them. Consort cultivar had the highest concentration of TP in whole, juice and seeds (22.0 ± 0.53, 23.1 ± 1.03, 24.0 ± 0.56 μg eq. GA/mg DW, respectively) (p 0.05). In general, seeds were rich in TP in all varieties. The highest concentrations of delphinidin 3-O-glucoside were found in skins followed by juice and whole fruit. D16-8-14 had greatest concentration of C3G in skins followed by juice. Delphinidin and cyanidin were identified in both rutinoside and glucoside forms across all extracts. BC extracts had higher levels of inhibition of α-amylase and α-glucosidase compared with DPPIV. BC was more effective at inhibiting enzymes related to breakdown of starch when compared to proteolytic enzymes. This information can be useful for extraction of BC phenolics, processing of foods and beverages and plant breeding for higher concentrations of phenolics.

P31

A group of multi-substrate terpene synthases from the roots of *Prunella vulgaris* (Lamiaceae)

Sean R. Johnson[1]*, Radin Sadre[1], Wajid W. Bhat[1], Alekzander Garcia[2], Bjoern Hamberger[1]

[1]*Department of Biochemistry and Molecular Biology, Michigan State University, 603 Wilson Rd Rm 212, East Lansing, MI 48823, USA*

[2]*Tennessee State University*

**Corresponding author, email address: John6121@msu.edu*

The mint family (Lamiaceae) is well documented as a rich source of labdane-related diterpenoids, and much effort has gone into characterizing the associated biosynthetic enzymes. Recently, some non-labdane-related diterpenoids with the unusual tetracyclic “vulgarisane” skeleton were

reported in common selfheal (*Prunella vulgaris*) from the mint family. We have cloned and functionally characterized a terpene synthase, PvHVS, from the roots of P. vulgaris, which catalyzes the formation of 11-hydroxy vulgarisane. PvHVS is unusual for a mint diterpene synthase in that it falls into a clade (TPSa) that mostly contains sesquiterpene synthases, and that it can act on multiple substrates. We further characterized three related terpene synthases, which also showed activity on multiple substrates. Promiscuity in this group of enzymes may have contributed to the evolution of diterpene synthase activity from sesquiterpene synthase activity.

P32

Antinociceptive principle from Zinnia grandiflora Nuttall (Astereaceae)

V. I. Reyes-Pérez [1], A. L. Martínez [1], A. Pérez-Vásquez [1], M. Déciga-Campos [2], R. Bye [1], R. Mata [1]

[1] Facultad de Química, [3] Instituto de Biología, Universidad Nacional Autónoma de México, Mexico City 04510, México
[2] Escuela de Superior de Enfermería y Obstetricia, Instituto Politécnico Nacional, Mexico City 11340, México

Zinnia grandiflora Nuttall (Astereaceae) is used by indigenous groups in North America, including the Navajo, Ramah Navajo, Zuni, and Acoma and Laguna to treat strokes, stomachache, nose and throat pain. This study approach toxicological, pharmacological and chemical aspects of this plant in order to contribute to its rational consumption. Toxicological study in mice was carried out through the Lorke acute toxicity test, which allowed to establish that its median lethal dose (LD50) is greater than 5 g/kg of animal weight. The antinociceptive potential of the aqueous extract was demonstrated by means of formalin test in mice; diclofenac (31.6 mg/kg) and gabapentin (30 µg/paw) as drug control. The results of this trial (10.0 - 562.3 mg/kg) revealed that the preparation has dose-dependent antinociceptive effect; the ED50 was 315.7 ± 85.5 mg/kg. The effect was higher in the inflammatory phase of the assay which was corroborated with the carrageenan-induced paw edema test in mice; the most effective dose in this trial was 100 mg/kg. The primary fractions obtained by partitioning the active aqueous extract with ethyl acetate and dichloromethane were also active; the second one was the most active with a remarkable effect at the highest dose tested (562.3 mg/kg) in the inflammatory phase of the bioassay. Hereby, the dichloromethane fraction was chemically investigated using different chromatographic procedures. These processes yielded three novel δ-elemanólide sesquiterpene lactones designed with the trivial names of zinagrandinolides D-F (2-4). Chromatographic profiles of the organic fractions of dichlorometane and ethyl acetate obtained from the infusion by partition were established by UHPLC-PDA-ESI-MS.

P33

Diterpenoid derivatization in medicinal plants

Seitaro Matsumoto[1]

[1]Researcher at Michigan State University

Diterpenoids are a diverse and widespread class of plant products with a broad range of functions in development and adaptation. They are also the known active compounds in certain medicinal plants. Medicinal diterpenoids are often unavailable for industry and research due to difficulty in chemical synthesis or limited access to the natural source. Our goal is to produce high value diterpenoids in a large amount with engineered organisms. We are focusing on diterpene synthase (diTPS) and cytochrome P450 in diterpenoid biosynthetic pathways. In particular, we are trying to identify the P450s that can oxidize and convert the diTPS product to medicinal compounds. The candidate P450 genes are mined from transcriptomes of specific tissues in the families of Lamiaceae, Thymelaeaceae and Euphorbiaceae. Two P450s are identified in Leonotis leonurus and presumed as the key genes for formation of bioactive diterpenes characteristic for this plant. A CYP71 family gene from Euphorbia poissonii oxidizes a unique macrocyclic diterpene. We also found two active P450s in the Chinese medicinal plant Daphne genkwa. The identified P450s may be key enzymes in diterpenoid biosynthesis in the medicinal plants, and also useful tools for molecular engineering to create valuable or structurally interesting diterpenoids.

P34

Isolation and characterization of three novel δ-elemanolides from an antinociceptive infusion from the aerial parts of *Zinnia grandiflora* Nuttall (Asteraceae)

V. I. Reyes-Pérez [1], A. Pérez-Vásquez [1], M. Déciga-Campos [2], R. Bye [1], R. Mata [1]

[1] *Facultad de Química, Universidad Nacional Autónoma de México, Mexico City 04510, Mexico*
[2] *Escuela de Superior de Enfermería y Obstetricia, Instituto Politécnico Nacional, Mexico City 11340, México*
[3] *Instituto de Biología, Universidad Nacional Autónoma de México, Mexico City 04510, México*

Zinnia grandiflora Nuttall (Astereaceae) is used by indigenous groups in North America, including the Navajo, Ramah Navajo, Zuni, Acoma and Laguna to treat painful complaints. From an antinociceptive infusion prepared from the aerial parts of *Z. grandiflora* three novel δ-elemanolide sesquiterpene lactones were isolated and characterized by spectroscopic (^{1}H and ^{13}C NMR, IR, UV) and spectrometric (ESI-MS) techniques. The new compounds were designated with the trivial names of zinagrandinolides D-F. The structure of zinagrandinolide D was corroborated by an X-ray diffraction analysis.

P35

Metabolic Engineering of Anthocyanins: from laboratory to field

De-Yu Xie[1], Xianzhi He[1]

[1]*Department of Plant and Microbial Biology, North Carolina State University, Raleigh, NC 27695, USA*

Production of Anthocyanin Pigment 1 (PAP1) from Arabidopsis thaliana is a master regulator of anthocyanin biosynthesis. It has been widely tested in different crops in laboratory, such as in

tobacco and tomato, for value-added agronomic traits. Whether its regulatory activity is stable in plants in the field is interesting but remains open for studies. After many years of selection, we obtained homozygous PAP1 transgenic lines (isogenic lines) of Nicotiana tabacum Xanthi, which were characterized by a high production of anthocyanins in growth chamber due to the constitutive expression of the PAP1 transgene driven by 35S promoter. In addition, we engineered red N. tabacum narrow leaf (NL) and obtained near isogenic lines. We continuously grew isogenic transgenic plants in the field two years and near isogenic lines one year. Field growth phenotype was recorded in details. The resulting data showed that red transgenic tobacco plants could continuously produce anthocyanins in all tissues in the field. Plants generally exhibited red/purple color most of time during the entire growth season. It was interesting that during the middle summers, leaves exhibited different greenish-reddish patterns, indicating that anthocyanin accumulation or biosynthesis was downregulated by environmental factors. Anthocyanin profiles were analyzed using LC-MS and cyanidin 3-O-rutinoside was identified to be a main anthocyanin molecule synthesized in the field. These data demonstrate that although PAP1 is overexpressed in plants, its regulatory activity in the field can be regulated by growth environment conditions. In addition, although anthocyanin biosynthesis activity is regulated, the principal anthocyanin composition is mainly controlled by PAP1 transgene. This study is informative to field growth of transgenic plants for targeted agronomic traits. This research was funded by R.J. Reynolds.

P36

Wound healing *in vitro* assays by *Iostephane heterophylla* extracts and HS-SPME/GC-MS-TOF analysis of the volatile compounds from its roots

M.I. Aguilar[1], K.L. Zarco[2], H. Arzate[3], S. López Letayf[4], G. Duarte Lisci[5]

Universidad Nacional Autónoma de México, [1,2]Departamento de Farmacia, Facultad de Química, [3,4]Laboratorio de Biología Periodontal División de Estudios de Posgrado, Facultad de Odontología, [5]Unidad de Servicios de Apoyo a la Investigación y a la Industria (USAII). Circuito de la Investigación, Ciudad Universitaria, CDMX, 04510, México

Roots of *Iostephane heterophylla* (Cav.) Benth ex Hemsl. (Asteraceae) popularly known as “escorcionera” have been used in traditional medicine to heal wounds, for rheumatic condition and for arthritis in topical preparations. Previous studies have shown sesqui, monoterpenes and their glycosides as main secondary metabolites of the drug. In this work, tests *in vitro* of the proliferative capacity in fibroblasts confluence assays by aqueous (decoction) and methanol extracts of the roots of “escorcionera” are shown, so as its contents of volatile compounds by HS-SPME/GC-MS-TOF analysis. The effects of the aqueous extract on proliferation of Human Periodontal Ligament-Derived Cells (hPDLs) were evaluated by the colorimetric MTT (tetrazolium) assay. Cells were treated for 96 h with increasing concentrations of the decoct (0.05, 0.5, 5 and 7.5 µg/mL). Results showed that fibroblasts proliferation by decoct was in a concentration dependent manner employing 2 and 10% fetal bovine serum. In the assay of volatile components of the roots, mono and sesquiterpenoids were the major constituents.

58th Annual Meeting of the Phytochemical Society of North America

July 20-24, 2019
East Tennessee State University
Johnson City, TN, USA