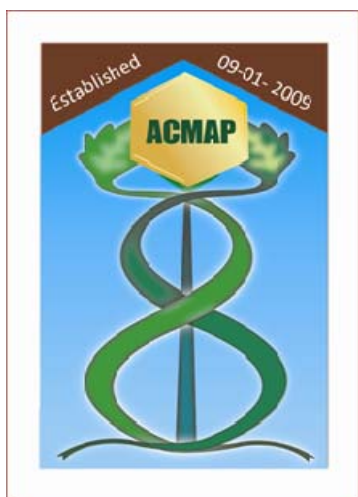


# 1<sup>st</sup> Annual Conference of the American Council for Medicinally Active Plants (ACMAP)

July 20-23, 2010. Cook Campus Center. George H. Cook Campus.

Rutgers, The State University of New Jersey. New Brunswick, NJ.

Hosted by ACPMAP and the New Use Agriculture and Natural Plant Products Program (NUANPP, Rutgers University).



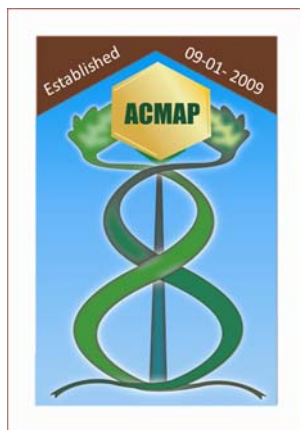
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*D*ear Colleagues:

21 July 2010

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#### FIRST ACMAP CONFERENCE

At Rutgers University  
New Brunswick, NJ  
20 – 23 July 2010

It is our genuine pleasure to welcome you all on behalf of the 2010 Annual Conference of the American Council for Medicinally Active Plants (ACMAP), being organized at Rutgers University, New Brunswick, NJ, from 20 July to 23 July 2010. To apprise you about the founding of ACMAP, the final decision on establishing this scientific society was made on 28 July 2009 in St. Louis, MO, during the Annual Conference of the American Society for Horticultural Science. Hence, ACMAP was established on September 1, 2009 and registered in the State of Florida, following several years of planning, discussing, as well as weighing and testing various opportunities and situations by the founding members and other ACMAP well-wishers. The ACMAP Board of Directors was established and its respective members were elected during January 2010.

This first annual conference of the ACMAP is designed to provide a forum for exchanging contemporary research/educational information on medicinal and other bioactive plants emphasizing their ethnobotany and bioprospecting, plant conservation, biotechnology research and improvement of medicinally active plants; phytomedicine-based industry emphasizing on the opportunities, challenges and future directions; global, regional and local regulatory issues relevant to the production of bioactive plants and products industry; current trends in botanicals; phytochemical, phytomedicinal, and biomedical research; and growing/production of medicinal and other bioactive plants. Therefore, with this much diversity of information out there, this conference has been designed to draw together professionals in medicinal and bioactive plants with researchers and scientists from universities, governments, and private sector laboratories and other entities involved in active research, education and other professional activities relevant to human/animal health and well-being.

We are genuinely excited to present a knowledge-packed scientific program featuring an impressive roster of well recognized researchers/professionals in their respective specialties. We extend our sincere thanks to all presenters and participants for their respective contributions for the success of this unique first event in ACMAP history. It is anticipated that this conference will initiate new collaborative linkages and reinforce the existing ones. Please feel comfortable to contact any of our organizers if we can help make your ACMAP participation more exciting and rewarding scientifically and professionally. Again, thank you all and welcome to the first conference of ACMAP.

Let us make it a great beginning

Anand K. Yadav

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American Council for Medicinally Active Plants, Inc. (ACMAP), P.O. Box 1761, Titusville, FL 32781, USA. Telephone: 321-861-3493, Fax: 321-861-2925, e-mail: [info@acmap.org](mailto:info@acmap.org), Website: [www.acmap.org](http://www.acmap.org)

Welcome to Rutgers, The State University of New Jersey,

We are pleased and honored to host the first annual conference of the American Council for Medicinally Active Plants (ACMAP). Our aim was to provide a national forum for those scientists, businesses, industries, and others interested in natural products to share ideas, discoveries, experiences and needs. As interest continues to grow in the use of foods and natural products to improve health, nutrition and an array of other applications, the requisite science becomes increasingly complex. As a result, a merging of disciplines is occurring, linking agriculture to health, nutrition and medicine. Transdisciplinary teams from partnering universities and businesses are beginning to tackle current questions. From botanical authentication and traceability from the forest or field to final product; from understanding the complex chemistry in the plant to the actual chemistry of the bioactives after consumption; from the challenges of our concepts of what may work, to the how and why, a richness of new discoveries awaits us all. This program was designed and ACMAP's intent is to bring together scientists working in botany, agronomy/horticulture, plant biology, chemistry, food science, health and nutrition, pharmacognosy and medicine together.

Therefore, on behalf of our Rutgers programs in New Use Agriculture and Plant Products Program and the Global Institute for BioExploration, the New Jersey Agricultural Experiment Station and Schools of Environmental and Biological Sciences and Pharmacy, and the many Rutgers based programs and faculty, staff and students involved in the study of natural products, welcome to Rutgers.

On behalf of the scientific program committee we hope that you'll find this conference and this venue most productive and rewarding.

Jim Simon, Program Chair, and Host

Director, New Use Agriculture & Natural Plant Products and Associate Director, Sustainable Development, GIBEX

## **ACMAP 2010 Program Meeting Committees**

### ***Scientific Program Committee***

Chair: Jim Simon  
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 Member: Rao Mentreddy  
 Member: Jeff Adelberg  
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 Member: Gary Stutte  
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### ***Field Trip***

Peggy Brennan  
 Lou Cooperhouse  
 Ed Dager  
 Jennifer Johnson-Cicalese  
 Nick Vorsa  
 Andy Wyenandt



	<i>Multi-purpose Room C</i>	<i>Multi-purpose Room B</i>
Concurrent Sessions	<b>Human Health, Phytochemistry and Bioactives</b> Convener: Agnes Rimando, USDA-ARS	<b>Production and Post-Harvest Processing</b> Convener: Lyle E. Craker, UMass-Amherst
PM 1:00-1:30	Gemma Casadesus, Case Western Reserve University, Cleveland, OH Modulation of cognition and stress/survival pathways by resveratrol and pterostilbene in the age-accelerated SAMP8 mouse	Jean Giblette, High Falls Foundation, Inc., Philmont NY Strategies in domestic production of Chinese medicinal plants
1:30-1:50	Lap Ho, Mt. Sinai School of Medicine, NYC, NY Development of brain-targeting grape-derived polyphenolics for Alzheimer's disease prevention and therapy	Zoe Gardener, UMass-Amherst Production of Chinese medicinal plants in the northeastern United States
1:50-2:10	Young-Cheul Kim, UMass-Amherst Effects of bioactive Isoflavone daidzein on adipocyte differentiation and insulin sensitivity	Robert Wick, UMass-Amherst Downy mildew of basil
2:10-2:40	Coffee, African Herbal Tea Break	
	<i>Multi-purpose Room C</i>	<i>Multi-purpose Room B</i>
Concurrent Sessions	<b>Human Health, Phytochemistry and Bioactives</b> Convener: Agnes Rimando, USDA-ARS	<b>Production and Post-Harvest Processing</b> Convener: Jorge Ferreira, USDA-ARS
2:40-3:00	Agnes M. Rimando, USDA-ARS Colon cancer preventive activity and bioavailability of pterostilbene analogs in severe combined immunodeficiency mice	Jorge Ferreira, USDA-ARS Pre- and post-harvesting procedures effects on <i>Artemisia annua</i> concentration of artemisinin and leaf antioxidant capacity
3:00-3:20	Prahlad Parajuli, Wayne State University, Detroit, MI <i>Scutellaria</i> : Anti-tumor efficacy and molecular mechanisms	Karina Knudsmark Jessing, University of Copenhagen Biomedicine production: Degradation and ecotoxicity of artemisinin
3:20-3:40	Jay Xie, North Carolina Central University, Durham, NC Identification and characterization of Se-responsive genes in the selenium-hyperaccumulator <i>Astragalus racemosus</i>	Cedric Sims, Alabama A&M University, Normal, AL Optimum date of planting and phytochemical content of <i>Ocimum tenuiflorum</i> (basil) in North Alabama
3:40-4:00	Devanand Luthria, USDA-ARS Significance of sample preparation for the assay of bioactive phytochemicals	Idrissa Moussa, Abdou Moumouni University in Niger Republic, Niger Antimalarial plants used in Niger Republic's Traditional Medicine
4:00-5:00	<b>Networking and Poster Session</b>	

Day 1 ends at 5:00pm. Dinner on Your Own

<b>Day 2: Thursday, July 22, 2010</b>		
<b>Thurs, July 22</b>	<b>Location: Cook College Campus Center, School of Environmental &amp; Biological Sciences</b>	
AM 7:00-8:30	<b>ACMAP Board Meeting - Breakfast</b>	
8:15-9:00	<b>Registration, Coffee and African Herbal Teas and Posters all on display</b>	
9:00-9:40	<p style="text-align: center;"><b>Plenary session II: <i>Multi-purpose Room C</i></b></p> <p style="text-align: center;"><b>Elucidating and manipulating alkaloid biosynthesis in the opium poppy</b></p> <p style="text-align: center;">Toni M. Kutchan, Donald Danforth Plant Science Center, St. Louis, MO</p>	
9:40-10:15	Networking	
	<i>Multi-purpose Room C</i>	<i>Multi-purpose Room B</i>
Concurrent sessions	<b>Commercialization</b> Convener: Peggy Breenan, Rutgers University, NJ	<b>Ethnobotany and Botanical Sourcing</b> Convener: Jim Simon, Rutgers University, NJ
10:15-10:45	Peter Gillies, Director, NJ Institute for Food, Nutrition and Health, Rutgers University  Alternative sourcing of omega-3 fatty acids: The role of biotechnology	Lena Struwe, Rutgers University, NJ  On the importance of being vouchered, identified, classified, and properly documented in botanical bioprospecting
10:45-11:05	Ilya Raskin, Rutgers University, NJ  Botanicals for metabolic syndrome and inflammation	Karen Reeds, University of Pennsylvania, Philadelphia, and Princeton Research Forum, Princeton NJ  <i>Hypericum perforata</i> L. (St. John's wort) in Renaissance herbal medicine: assessing the historical evidence for use of hypericum as an anti-depressant in early modern Europe
11:05-11:25	Rodolfo Juliani, Rutgers University, NJ  Linking education, research and development: The chemistry and quality of spices and medicinal plants from Ghana and Liberia	Glenda Smith, Univ. of Alabama, Birmingham, AL  African American use of herbs in health care
11:25-11:45	Lou Cooperhouse, Rutgers University, NJ  Business incubation resources: Supporting technology commercialization: Case study of the Rutgers Food Innovation Center	Frankie Hutton, Bowie State University  Rose lore: Essays in semiotics and cultural history
11:45 – 1:00	<b>Networking lunch</b> (Cold Buffet lunch open only to paid registrants) and <b>Posters to View</b>	
	<i>Multi-purpose Room C</i>	<i>Multi-purpose Room B</i>
Concurrent Sessions	<b>Sustainability and Economics</b> Convener: Rao Mentreddy, Alabama A&M University	<b>Biotechnology of Medicinally Active Plants</b> Convener: Fabricio Medina-Bolivar, Arkansas State University
PM 1:00-1:30	Jim Simon, Rutgers University, NJ  Enterprise development in rural areas: The case for natural products as sustainable income generating activities using a market-first science-driven model	Pam Weathers, Worcester Polytechnic Institute  Artemisinin: <i>in planta</i> production and therapeutic options
1:30-1:50	Edward Fletcher, Strategic Sourcing Inc., Banner Elk, NC  Increasing marker constituent levels of medicinal plant species <i>ex situ</i> to enhance market value: Two case studies: <i>Echinacea purpurea</i> L. and <i>Hydrastis canadensis</i> L.	Gnana Viji, Fort Valley State University, GA  Molecular characterization of peach [ <i>Prunus persica</i> (L.) Batsch] germplasm in the United States using microsatellite markers

1:50-2:10	Mario Morales, Mountain State University, Beckley, WV Amendment of low-fertility acidic soils for fairywand culture in Appalachia	Rohan Patil, UMass-Amherst Engineering taxol biosynthesis in <i>Taxus</i> suspension cell culture
2:10-2:30	Lorraine Cordeiro, UMass-Amherst The role of medicinal plants in food security and nutrition	Fabricio Medina-Bolivar, Arkansas State University Natural resveratrol analogs from root cultures of peanut and muscadine grape: bioproduction, biotransformation and bioactivity
2:30-2:45	Break	
Concurrent Sessions	<b>Quality Control</b> Convener: Rodolfo Juliani, Rutgers University, NJ	<b>Ethnobotany and Plant Improvement for MAPs</b> Convener: Art Tucker, Delaware State
2:30-3:00	Rodolfo Juliani, Rutgers University, NJ Quality assurance and quality control: Making it relevant to growers and processors	Art Tucker, Delaware State, Dover, DE Genetics and biotechnology of the genus <i>Mentha</i> : a model for other polyploid species with essential oils and other plant constituents?
3:00-3:20	Qingli Wu, Rutgers University, NJ Using natural products chemistry for QC and botanical standardization	Jennifer Johnson-Cicalese, Rutgers University, NJ Breeding blueberries and cranberries for health and nutrition
3:20-3:40	Dennis Awang, White Rock, British Columbia, Canada Regulating herbal products: A North American perspective	Jim Simon, Rutgers University, NJ Breeding for active principles in aromatic plants and herbs
3:40-5:00	<b>Poster Session and Networking. Poster Session closes at 5PM</b>	
6:00 PM	<b>Social Hour</b>	
7:00- 9:00 PM	<b>Banquet (Multi-purpose Room C, open only to paid registrants)</b>	

## DAY 3: Friday, July 23: Field Day: Tentative Agenda

### Departing at 7:00AM and estimated return time, 6:00 PM

*NOTE: An additional registration fee required to cover transportation costs and packed lunch. **Schedule is tentative.***

Tour lead by Jim Simon, Rutgers.

**Stop 1:** Travelling south from New Brunswick to Cumberland County in southern Jersey, our first stop is the **Rutgers Agricultural Research and Extension Center (RAREC)**, 121 Northville Road, Bridgeton, New Jersey 08302. Local Host: Dr. Andy Wyendant, Extension Specialist in Vegetable Pathology, Department of Plant Biology and Pathology. This research facility, with 305 acres, is located near the largest vegetable, herb and horticultural production in NJ, conducts research applicable to the production of high-quality vegetable crops, ornamentals, field crops, and tree and small fruits, with special emphasis on crop protection and integrated pest management. In this visit, you'll see some of the vegetable research that is ongoing as well as studies seeking to identify control strategies against basil downy mildew, a new and devastating disease on sweet basil (Prof. Wick, Umass will be presenting a paper on this topic);

For more detailed information, directions and more: <http://njaes.rutgers.edu/centers/quickinfo.asp?RAREC>

**Stop 2:** We next head into the city of Bridgeton, NJ to tour the **Rutgers Food Innovation Center (FIC)**. This is a unique business incubation and economic development accelerator program, which provides business and technology expertise to small and mid-sized food companies in New Jersey, and utilizes its outreach capacity to reach food and agribusinesses throughout the nation. The FIC provides assistance in business development, market research, product and process development, workforce development and training, regulations and compliance support, and quality assurance and food safety systems. This center is 23,000 sq. ft. business and functioning food product incubator facility and has been nationally recognized for its innovative work and new product development for our family farms. FIC also has a food safety lab and testing facilities for microbiological contamination with staff overseeing all facets of product development to ensure cleanliness and food safety of the commercial and pilot products coming out of their food manufacturing facility which operates at a subsidized price for those seeking small scale toll manufacturing. Our local hosts will be Lou Cooperhouse, Director of FIC and Peggy Brennan, Associate Director, New Jersey Agricultural Experiment Station, Director of Economic Growth and Development, New Jersey Agricultural Experiment Station.

For detailed information/activities, driving directions and more see: <http://www.foodinnovation.rutgers.edu>

**Stop 3:** Travelling back northwest, the third stop will be at the **Philip E. Marucci Center for Blueberry and Cranberry Research and Extension** is a substation of the New Jersey Agricultural Experiment Station (NJAES) of Rutgers University located in Chatsworth, NJ (Burlington County). Local Hosts will be: Plant breeders Prof. Nick Vorsa and Dr. Jennifer Johnson-Cicalese. Several presentations during the conference will have focused on the health properties and bioactive compounds in blueberry and cranberry as well as current breeding strategies to improve the health and nutrition of cranberries. This stop provides a close-up on the breeding on these berries with a possible visit to a commercial cranberry bog.

For detailed information/activities, driving directions and more see: <http://pemaruccicenter.rutgers.edu/>

**Stop 4:** Continuing further northwest into Hunterdon County, our last stop will be at the **Clifford E. and Melda C. Snyder Research and Extension Farm**. Local Host: Ed Dager, Research Farm Manager. This Rutgers' center for sustainable agriculture, promotes different agricultural systems (including organic) and initiates and disseminates research applicable to the production of a variety of food and fiber products, including grain crops, tree and small fruits, turfgrass, ornamentals, ethnic crops, culinary herbs, aromatic and medicinal plants. You'll see the medicinal and aromatic plant trials including studies with basil (for chilling injury, Fusarium resistance, and downy mildew), new varietal development lines (*Artemisia annua*, oregano and catnip), culinary variety trials (basil, chives, and hot/specialty peppers); Chinese vegetables and medicinals, and specialized walk-in drier unit (converted older Powell Tobacco dryer).

For detailed information, driving directions and more see: <http://snyderfarm.rutgers.edu/index.html>

Afterwards, we will be returning to New Brunswick directly from the Snyder Farm, estimated time one hour and return each of you to your respective hotels.

**CONFERENCE LOGISTICS:**

**For Tuesday evening, July 20, the reception will be held at the Log Cabin and Pavilion at the Rutgers Garden.** For detailed information, driving directions to the Rutgers Gardens see: <http://rutgersgardens.rutgers.edu/>

**(Transportation is on your own)**

**Meeting Location: All lectures, posters and discussion sessions on Wednesday and Thursday (July 21-22) including the evening banquet will be held in the first floor of the Cook College Campus Center (CCC; 59 Biel Road New Brunswick, NJ 08901).** The CCC is part of the School of Environmental and Biological Sciences, Cook College Campus (one of five campuses at Rutgers University). The CCC is across the street from the Recreation Center, and within a few minutes to the heart of the SEBS where the Department of Plant Biology and Plant Pathology is housed (59 Dudley Road), Food Science and much more.

From your hotel to the Cook Campus Center, transportation is on your own. Rutgers does have a free bus system which would operate effectively for those staying at the Heldrich, those at the Hyatt with a bit more walking to reach the bus stops, but not functional for those staying at the Ramada Inn. Please check the online schedule as the buses do not run frequently during the summer. For the Rutgers summer bus schedule see: <http://parktran.rutgers.edu/>

For detailed information, driving directions to the Cook Campus Center see:

<http://getinvolved.rutgers.edu/centers/center-listing/2>

**Parking at the Cook College Campus Center** (where all sessions will be held): If you are driving to the conference please let us know upon registering. We will then provide you with a parking permit. To do so, we need your license plate number and state; and type of vehicle. Parking without a permit will likely result in a parking ticket. We have arranged space for you to park in the parking lots 99C and 99D. These lots are both across the street from the Cook Campus Center, very close to the CCC, just past the recreation center and adjacent to a set of apartments. There is no additional cost for parking.

**ACMAP Board meetings:**

**Tuesday**, July 20, 2010 2:00- 5:00PM, Heldrich Hotel Lobby

**Thursday**, July 22, 2010, 7:00-8:30 AM Breakfast meeting, Heldrich Hotel

**Speaker Preparation room and ACPMAP Board Room** in the Cook Campus Center (CCC): A room has been reserved throughout all day Wednesday and Thursday (8am- 6pm) that will be locked but available to be used for private meetings (upon request and subject to availability) and ACPMAP BOD and other official business. Stop and see the person at the registration table or another member of the Rutgers local host team for access.

**Computer and Wireless Access:** We have requested wireless access to everyone that has registered during the two days at the Cook Campus Center. If you need access, ask at the registration table.

## Abstracts

Abstracts from the oral presentations are first listed in order of their presentation and by thematic session.

**Day 1: Wednesday, July 21, 2010**

**DAY 1 AM Plenary Session I:**

### **Characterization of Grape-Derived Polyphenols in the Prevention of Alzheimer's Disease and Promotion of Healthy Aging**

Giulio Maria Pasinetti, M.D., Ph.D.

*Geriatric Research, Education and Clinical Center, James J. Peters Veteran Affairs Medical Center, Bronx, NY, USA; Department of Neurology, Mount Sinai School of Medicine, New York, NY, USA*

A high consumer demand exists for foods and dietary supplements with the ability to help preserve cognitive functions by preventing processes involved in age-related cognitive decline. Bioactive dietary botanicals in food and supplements are complex mixtures of numerous chemical entities, and the identity of these compounds is often not well established. Ongoing studies on our laboratory are underway to characterize compounds derived from grape seed polyphenolic extract (GSPE) to investigate their potential role in Alzheimer's disease (AD) and healthy aging. We previously showed that GSPE exerts beneficial AD disease-modifying activities *in vivo*, including anti-amyloid aggregation activity and improved cognitive function in animal models of AD. GSPE is comprised of catechin and epicatechin in monomeric, oligomeric, and polymeric forms. While both monomeric and polymeric GSPE components can interfere with b-amyloid (A $\beta$ ) aggregation in the brain, we found that only monomeric GSPE components are bioavailable. This evidence indicates that monomeric GSPE may be primarily responsible for its beneficial bioactivity. Moreover, electrophysiological studies further demonstrated that monomeric GSPE promotes basal synaptic transmission and long term potentiation – both of which are key mechanisms involved in learning and memory – in hippocampal slices from treated mice. Collectively, these studies demonstrate that monomeric GSPE components, rather than polymeric or oligomeric, may promote healthy brain aging and may benefit AD phenotypes through mechanisms involving the amelioration of A $\beta$ -mediated neurotoxicity and improved synaptic plasticity in the brain.

### **DAY 1 AM Session: Human Health, Phytochemistry and Bioactives**

#### **Cranberries and Blueberries: Functional Food Superstars!**

Amy Howell

*Marucci Center for Blueberry Cranberry Research, Rutgers University, Chatsworth, NJ 08019. E-mail: ahowell@aesop.rutgers.edu*

Cranberries and blueberries are native to North America and have a rich folklore history of medicinal uses by the Native American Indians. Cranberry juice consumption has been shown clinically to prevent urinary tract infections. Research suggests that specific compounds in cranberry called proanthocyanidins act to inhibit bacterial adherence to the uroepithelium, preventing subsequent colonization and urinary tract infection. Elucidation of the chemical structures of the cranberry proanthocyanidins reveals the presence of less common

A-type, double interflavanoid linkages which may be important in eliciting anti-adhesion bioactivity. The role of cranberry proanthocyanidins in preventing bacterial adhesion will be reviewed, as well as the emerging research into the benefits of cranberry on markers for heart disease and cancer. Research on blueberries includes antioxidant, anti-inflammatory, and cell signaling. Many studies focus on age-related mental decline, including cognitive and motor functions. Increases in functionality have been observed in both animal and human trials following consumption of blueberries. Blueberries are known for their broad array of phytochemicals, especially flavonoids. Although attention has been focused on the anthocyanin pigments, the compounds responsible for the various medicinal activities have not been fully elucidated, and could include several different classes of polyphenolics.

### **Cranberry and Grape juices reduce rotavirus infectivity in cell culture, and viral protein capsid integrity in cell-free suspension**

Steven M. Lipson

*Dept. of Biology, St. Francis College, Brooklyn Heights, NY 12201. E-mail: [slipson@stfranciscollege.edu](mailto:slipson@stfranciscollege.edu)*

Cranberry (CJ), grape juices (GJ) and these species' proanthocyanidins (PACs) have an ameliorative effect on host cells following rotavirus infection of epithelial cells. Tight junction (TJ) integrity was measured by changes in transepithelial electrical resistance (TEER) and immunostaining of alpha-claudin. These analytes may protect TJ integrity from the destructive effects of rotavirus infection. After four days, virus infected monolayers pretreated with GJs exhibited TEER readings similar to uninfected controls. CJ imparted a significant [TJ] protective effect ( $p < .05$ ), but to a lesser extent than GJs. Disorganization of TJ integrity commenced at ca. 24-h post-inoculation; this effect was reduced upon CJ and GJ pretreatment of monolayer cultures. Electron micrographs showed a juice-associated sequestering of viral particles within cellular cisternae. At low rotavirus input, amplicon detection was markedly reduced following CJ and GJ monolayer treatment. Rotavirus capsid antigen (i.e., VP6) was reduced >90% following incubation for 30 min in 50% CJ or GJ. Cranberry and grape PACs at 500 ug/ml reduced VP6 detection by >1  $\log_{10}$  after 60 min at 23°C. Anthocyanidins had no effect on VP6 integrity. CJ and GJ reduce/inhibit rotavirus infectivity titers, have a protective effect on TJ function/structure, and compromise viral integrity in cell-free suspension.

### **Development of simple and rapid bioassays for screening natural products against infection and inflammation**

Slavko Komarnytsky

*Biotech Center, Rutgers University, 59 Dudley Rd, New Brunswick, NJ 08901*

Emerging health risk factors such as development of antimicrobial resistance, increasing vaccination fatigue, and long-distance travel promote the spread of infectious diseases and inflammation associated with the body's response to infection. Additionally, many allergic disorders and chronic inflammatory and autoimmune diseases constitute interactions of polygenic traits with environmental triggers such as bacterial infection. Natural products represent precious resources from which bioactive compounds can be isolated and developed into novel therapeutic agents to address these issues. Two simple, rapid, and cost-effective bioassays were developed to screen extracts, fractions, as well as purified compounds for their antibacterial and anti-inflammatory activity to enable low-cost bioexploratory and conservation efforts in the developing

countries and science education settings. The present talk provides an overview of their basic methodology, capacities and limitations. By using and implementing these screens we seek to achieve globally what is taken for granted in the US academic settings: the continuous expansion of knowledge and innovation that lead to greater scientific education and prosperity.

## **Plant polyphenols for chronic obstructive pulmonary disease**

Edward J. Kennelly, Ph.D.

*Lehman College, City University of New York*

Chronic obstructive pulmonary disease (COPD) is a respiratory condition characterized by irreversible airflow obstruction due to chronic inflammation. Cigarette smoke is the main etiologic factor associated with the development of COPD, for which there is no cure. Although bronchodilators and steroids are used in the treatment of COPD, only oxygen has been demonstrated to prolong survival of patients. Antioxidants and anti-inflammatory agents may be useful in the treatment of COPD. Many edible fruits are rich sources of polyphenolic antioxidants and anti-inflammatory compounds, and diets rich in fruits and vegetables are correlated with a reduced risk of COPD in men. We have demonstrated that two plant-derived polyphenols, the novel deposite jaboticabin and the anthocyanin delphinidin 3-glucoside, both from the Brazilian fruit *Myrciaria cauliflora*, have antioxidant and anti-inflammatory activities. Jaboticabin and delphinidin 3-glucoside both inhibit interleukin-8 induction in cells exposed to cigarette-smoke extract. We hypothesize that polyphenols, like jaboticabin and delphinidin 3-glucoside, will modulate the cigarette-induced inflammatory, oxidative, and proteolytic responses of the lung. We are testing this by isolating a group of polyphenols from four edible fruits in the Ericaceae (blueberry) and Myrtaceae (myrtle), plant families rich in polyphenols. The purified polyphenols are being tested in vitro using antioxidant assays including 2,2-diphenyl-1-picrylhydrazyl and oxygen radical absorbance capacity, and anti-inflammatory assays for IL-8. The polyphenols will be prioritized for further study based on the activity in these assays as well as their structural uniqueness to test in an animal model for COPD.

## **DAY 1 PM Session: Human Health, Phytochemistry and Bioactives**

### **Modulation of cognition and stress/survival pathways by resveratrol and pterostilbene in the Age-accelerated SAMP8 mouse**

Jaewon Chang<sup>1</sup>, Barbara Hale<sup>2</sup>, James A. Joseph<sup>2</sup>, Merce Pallas<sup>3</sup>, Agnes M. Rimando<sup>4</sup> and Gemma Casadesus<sup>1</sup>

<sup>1</sup>*Department of Neurosciences, Case Western Reserve University, Cleveland, OH;* <sup>2</sup>*USDA-HNRCA at Tufts University, Boston, MA;* <sup>3</sup>*Pharmacology Department, Universitat de Barcelona, Barcelona, Spain;* <sup>4</sup>*USDA Agricultural Research Service, University, MS. E-mail: gxc40@case.edu*

To date there are no mouse lines that model late-onset/age-related Alzheimer's Disease (AD), the feature which accounts for the vast majority of AD cases. The senescence-accelerated mouse (SAMP8), a model of aging, displays many features that are known to occur early in the pathogenesis of AD. Therefore, SAMP8 mice may be an excellent model for studying the earliest neurodegenerative changes associated with AD and

determine the effectiveness of therapies. Recent studies have implicated resveratrol in the protection against age-related diseases, such as cancer, diabetes, and neurodegenerative diseases in addition to age-related conditions such as cognitive decline, a risk factor for AD. The purpose of this study was to determine if equivalent doses of resveratrol and pterostilbene, a less known resveratrol derivative, can reverse age-related cognitive deficits and modulate pathways associated with aging and AD protection in the SAMP8 mouse. Two months of diet significantly improved memory function in the passive avoidance task and the Radial arm water maze task compared to control-fed animals. In addition, markers of aging such as FoxO1 and SirT1 and oxidative stress, and related signaling pathways were also positively altered by these diets. Interestingly, pterostilbene was more potent both at a behavior and cellular level implicating higher biological activity. Since substitution of hydroxy with methoxy group increases lipophilicity one potential explanation is possible better bioavailability of pterostilbene.

### **Development of brain-targeting grape-derived polyphenolics for Alzheimer's disease prevention and therapy**

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We recently demonstrated that dietary polyphenolic compounds may beneficially modulate cellular and molecular mechanisms known to contribute to Alzheimer's disease (AD) dementia. Our recent evidence suggests that select bioactive grape-derived polyphenols may protect against AD-type cognitive deterioration, in part, by interfering with beta-amyloid (A $\beta$ )-mediated neuropathologic mechanisms. To gather insights on specific bioactive polyphenolics that might exert beneficial disease-modifying activity in vivo, we explored the pharmacokinetic and bioavailability of red wine and grape juice polyphenolics. We identified a number of polyphenolic derivatives that are bioavailable in the blood and/or in the brain following oral administrations of total polyphenolic extract from red wine and grape juice. Our ongoing studies exploring the potential bioactivities of these brain-targeting polyphenols revealed specific polyphenolic capable of significantly reducing the generation of A $\beta$  peptides, in vitro, in primary cortico-hippocampal neuron cultures. Moreover, we have tentatively identified another brain-targeting polyphenolic that may promote activation of cAMP responsive element binding protein (CREB), a transcription factor implicated in neuronal synaptic plasticity. Results from our studies suggest that dietary polyphenolics may benefit AD by modulating multiple disease-modifying modalities, including A $\beta$ -dependent and independent mechanisms in the brain, and provides the impetus to develop selective polyphenolic compounds for AD prevention and/or therapy.

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## Effects of Bioactive Isoflavone Daidzein on Adipocyte Differentiation and Insulin Sensitivity

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Recent studies have shown that plant-derived isoflavones including soy isoflavones favorably affect glucose and lipid metabolism associated with type 2 diabetes. However, the underlying mechanisms by which a specific isoflavone exert anti-diabetic effects are not clearly understood. We examined the modes of action of an isoflavone, daidzein, on adipocyte differentiation, gene expression, and glucose uptake using 3T3-L1 cells, a preadipocyte cell line that undergoes differentiation into mature adipocytes. Daidzein treatment markedly enhanced differentiation of 3T3-L1 preadipocytes in a dose-dependent manner. Daidzein also increased expression of PPAR $\gamma$  mRNA, a key regulator of adipocyte differentiation and also a nuclear receptor for synthetic anti-diabetic agents. This correlated with increased expression of aP2 mRNA, a PPAR $\gamma$ -regulated gene. Moreover, daidzein treatment up-regulated the expression of mRNAs for glucose transporter 4 (GLUT4) and insulin receptor substrate-1, the key molecules for insulin signaling and action. In mature adipocytes, daidzein significantly increased insulin-stimulated glucose uptake and the translocation of GLUT4 to plasma membrane, which were abrogated in the presence of a PPAR $\gamma$  antagonist. These results indicate that the insulin-sensitizing effects of daidzein in adipocytes are mediated by increasing the expression and translocation of GLUT4 through a mechanism of PPAR $\gamma$  activation.

## Colon cancer preventive activity and bioavailability of pterostilbene analogs in severe combined immunodeficiency mice

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Twenty-five *cis*- and *trans*-pterostilbene analogs varying only in substitution at the 4'-position were synthesized and tested against the colon cancer cell lines HT-29 and Caco-2. Three analogs: (*Z*)-4-(3,5-dimethoxystyryl)aniline (DA), (*Z*)-methyl-4-(3,5-dimethoxystyryl)benzoate (MDB), and (*Z*)-1,3-dimethoxy-5-(4-methoxystyryl)benzene (DMB) showed strong inhibitory activity against these cell lines. (*Z*)-DA, (*Z*)-MDB, and (*Z*)-DMB, together with their corresponding *trans*- isomers, were tested *in vivo* using HT-29 xenografts in severe combined immunodeficiency (SCID) mice at a dose of 10 mg/kg body weight. Mice fed (*E*)-DA, (*Z*)-DA, and (*E*)-DMB showed significantly lower tumor volume. (*E*)-DA and (*Z*)-DA both showed 40% decrease of tumor growth. Analysis of the serum revealed that (*Z*)-DA isomerized to (*E*)-DA, which could explain the similar effects observed in SCID mice. (*Z*)-MDB and (*Z*)-DMB had no suppressive effect; this may be due to their low serum levels (18.8 and 15.5 ng/mL, respectively). (*E*)-DMB was found at higher levels (69.9 ng/mL) in the serum than its *cis*-isomer (*Z*)-DMB; thus, while only weakly active *in vitro* better bioavailability could explain its activity *in vivo*. Our studies have shown that *cis*-to-*trans* isomerization occurs *in vivo*, depending on the substituent at the 4'-position of the analogs. Our study identified a naturally-occurring analog, i.e., (*E*)-DMB, to have good anti-tumorigenic potential, and demonstrated its bioavailability in SCID mice.

## Scutellaria: Anti-tumor efficacy and molecular mechanisms

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Plants of the genus *Scutellaria* constitute one of the common components of Eastern as well as traditional American medicine against various human diseases, including cancer. Earlier, we have reported a comprehensive analysis of the leaf, stem and root extracts obtained from thirteen different species of *Scutellaria* for their flavonoid content as well as for their mechanism of anti-cancer activity. We further examined the *in vivo* anti-glioma efficacy of a leaf extract of *Scutellaria ocmulgee* (SocL) while also exploring its potential molecular mechanisms of action. SocL extract delayed the growth of F98 glioma in F344 rats, both in intracranial and subcutaneous tumor models, which was associated with an inhibition of Akt, GSK-3 $\alpha/\beta$  and NF- $\kappa$ B phosphorylation. Pharmacologic inhibitors of PI3K and NF- $\kappa$ B also significantly inhibited the *in vitro* proliferation of F98 glioma cells, indicating the key role of these signaling molecules in the growth of malignant gliomas. *In vitro* Akt kinase assay demonstrated that the SocL extract or constituent flavonoid wogonin could indeed bind to Akt and inhibit its kinase activity. These studies provide the first *in vivo* evidence and mechanistic support for anti-glioma activity of *Scutellaria* flavonoids and have implications in potential usage of *Scutellaria* in adjuvant therapy for malignant tumors.

## Identification and characterization of Se-responsive genes in the selenium-hyperaccumulator *Astragalus racemosus*

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Selenocompounds have been demonstrated to confer chemoprotective effects in cancers. The most effective form of selenocompounds is Se-methylselenocysteine, mainly producing in Se-accumulators and some vegetable plants. Plants with high Se-accumulating capacities are desirable for cancer prevention. Natural Se-hyperaccumulator plants, such as *A. racemosus*, cannot be directly used because they are slow growth, low biomass and not edible. Also, it is impossible to cross Se-hyperaccumulating traits into any food crop due to the genetic distance. Our goal is to produce Se-enriched functional foods or sufficiently produce selenocompounds through the understanding of the mechanisms of how natural Se-hyperaccumulators gather and tolerate a high level of Se which are still unclear. To reach our goal, we used differential display technique to identify a group of differentially expressed genes from Se-treated *A. racemosus* seedlings. qRT-PCR was employed to validate expression levels of these candidate genes under different Se treatments. Isolated genes will be further characterized by reverse genetic technique with our established *A. racemosus* transformation system. Our present study provides a starting point for a genetic analysis of Se-hyperaccumulation and help to create Se-enriched plants in future.

## **Significance of Sample Preparation for the Assay of Bioactive Phytochemicals**

Devanand Luthria

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Increased interest in bioactive phytochemicals has arisen from numerous epidemiological studies that suggest that certain phytochemicals can reduce risk of chronic diseases. Accurate analysis of bioactive phytochemicals pose a significant challenge due to the complexity of sample matrices, coexistence of multiple forms of bioactive phytochemicals, and their interaction with other cellular components. This paper will illustrate the importance of a sample preparation procedure for accurate quantification of bioactive phytochemicals from foods, herbs, and dietary supplements. Over 8000 phenolic compounds belonging to different classes such as flavonoids, tannins, phenolic acids, anthocyanins, and isoflavones have been reported in literature. Phenolic compounds were selected as the model substrate as these compounds are known to exist in nature as free aglycones or as conjugates with sugars and/or acids. This paper reviews influences of multiple variables, namely extraction solvent composition, physical extraction techniques, and parameters (temperature, pressure, number of cycles, solid-solvent ratio, particle size) that influence the quantitative extraction of phenolic phytochemicals from different matrices. Accurate quantification of bioactive phytochemicals in foods, plants, and dietary supplements will allow researchers and regulators to provide more precise guidelines on dietary intake and safety levels necessary to achieve optimum health.

## **DAY 1: AM: Controlled Environmental Horticulture and Propagation**

### **Increasing the concentration of bioactive constituents of medicinal and nutraceutical plants using controlled environments**

Gary W. Stutte

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Controlled environments (CE) allow conditions to be optimized for growth, development, and productivity of many crops. Environmental factors affecting the concentration and quality in CE include light intensity, photoperiod, light quality, atmospheric composition, temperature and relative humidity. Light intensity and quality have been shown to optimize both productivity of a crop and the nutraceutical value of many crops. Lettuce grown in CE under light emitting diodes (LEDs) has demonstrated that the concentration of bioactive anthocyanins is enhanced through timing, duration and dose of blue light. These treatments have increased both biomass and anthocyanin concentration without additional inputs of energy. Atmospheric enrichment with CO<sub>2</sub> has been shown to increase the yield of both crop and medicinal plants. In addition to increasing yield, the concentration bioactive flavonoids have been enhanced. This change in composition may or may not affect the bioactivity of the plant. There is a need to understand on the role of environment on the regulation of flavonoid metabolism in medicinal and nutraceutical plants and develop protocols to optimize the growth and composition. CE provides a tool to better understand the regulation of metabolism and exploit them for commercial production.

## Challenges and Future Prospects for Specialty Plant Biotechnology Research

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In a world where sturdy population growth is outstripping the food supply, agricultural biotechnology and more specifically plant biotechnology needs to be swiftly implemented in all walks of life. The current achievements in plant biotechnology have already surpassed all its previous expectations, and the future outlook appears to be even brighter and more promising than before. The comprehensive realization of the agricultural biotechnology revolution obviously depends on its continued success in innovative research and development activities as well as on a favorable regulatory climate and the public acceptance. Biotechnology should be fully integrated with classical physiology and breeding: (1) as an aid to classical plant breeding, (2) for developing engineered organisms, and (3) for integrating microorganisms into agricultural production systems. Biotechnology at the present time is changing the agricultural and specialty plant outlook in three major areas: (a) growth and development control (vegetative, generative and reproductive), (b) plant protection against the ever-increasing threats of abiotic and biotic stresses, and (c) expanding the horizons by producing specialty foods, biochemicals and pharmaceuticals. We have been introducing specialty plants in the Central Georgia ambiance for the past thirty years and this presentation will elaborate on our research efforts on the compounds imparting health benefits, development of *in vitro* micropropagation protocols, and scaling up for commercial production.

## In Vitro culture of Medicinal and Aromatic Plants

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In the past decades there has been an increased interest in herbal medicines. Most of the herbal preparations are obtained directly from wild populations or from field-grown plants with high plant to plant variability, consequently the raw materials are highly heterogeneous. *Minthostachys mollis* (Lamiaceae) is a native aromatic plant used in popular medicine in South America. A protocol for the *in vitro* propagation of *M. mollis* was successfully developed to obtain homogenous plants with the same bioactive composition as of the parent plants. *E. purpurea* (Asteraceae) is the most widespread and widely cultivated species of the genus. An efficient protocol for *in vitro* regeneration of *E. purpurea* from leaves was developed. Leaves of *E. purpurea* have a great organogenic potential for shoot formation, however the response is highly sensitive and directly related to the combination of exogenous growth regulators in the culture medium. These studies have shown that *in vitro* culture technologies can be applied to aromatic and medicinal plants for mass propagation to conserve endangered species or when conventional vegetative propagation is slow. *In vitro* culture technologies can produce elite genotypes and provide resources for continuing investigations of the biochemistry and pharmacology of aromatic and medicinal plants.

## **In vitro rhizomes of turmeric as a model for bioactive biomass**

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Turmeric is a sterile triploid that produces over a 100 distinct secondary metabolites with anti-cancer activities associated to its major anti-oxidants, the curcuminoids. Micro-rhizomes are produced in vitro in liquid media that promote enhanced uptake of sucrose. Fresh micro-rhizomes have greater anti-oxidant activity than commercially available dried rhizome powder (DPPH and Fe chelation assays). Dry mass in conventional small-vessel, batch culture (6-weeks) and fed-batch, bioreactor culture (23-weeks) showed congruent linear relationships with 1.8 g sucrose becoming 1 g dry mass. Rhizome mass was less 40% of the plant mass in 6-weeks and 60% in fed-batch bioreactor vessels after 23 weeks where 33 g of sucrose and 435 ml of media “bioreacted” to become 10 g of dry rhizome. When sucrose concentration in media is kept constant for 5 weeks, % sucrose is directly related to % dry mass of rhizome. Sucrose and phosphate become limiting in batch culture with MS 2x-P medium. High inorganic N concentrations increase dry mass and plant multiplication but down-regulates anti-oxidant activity. Response surface methodology was used to minimize  $\text{NO}_3^-$  and  $\text{NH}_4^+$  with the least effect on multiplication and mass. Micro-rhizomes are preferred to leafy plantlets for propagation stock. In vitro rhizomes formed secondary “fingers” in high sucrose media and yielded over 98% success during greenhouse acclimatization. This model is useful to scale-up propagation of other geophytes with medicinally active principles.

### **DAY 1 PM: Production and Post-Harvest Processing**

#### **Strategies in domestic production of Chinese medicinal plants**

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The challenge of producing Asian medicinal botanicals for the U.S. licensed practitioner market segment calls for a systems approach. The traditional practice of combining herbs into formulas creates opportunities for a diversity of complementary products. Growers interested in this market gain an advantage by risk management analysis, planning and cooperation to choose among over 150 species of potential crops. Crops are characterized by variables related to production including: suitability to the region, climate zone, ecosystem and farm setting; the harvestable portion (leaf, root, bark, seed, and fruit) which determines post-harvest handling and equipment needed; length of time until harvest. Other characteristics affect marketability, such as the grade of the crop (whether food-grade or strictly medicinal), the extent to which the herb is used in popular formulas and, ultimately, the acceptance of the product, which is measured by sales tracking data. Small interdisciplinary task forces have begun to work within the regions to select and plant crops, harvest and evaluate samples, and track sales. The list of potential species is winnowed to a “Top Fifty” priorities for each region. A Top Fifty list for the Northeast, with production and marketing variables, is appended as an illustration.

## Production of Chinese medicinal plants in the northeastern United States

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With an increase in consumer demand for integrative healthcare and the prescription of herbal formulas being included in the scope of practice of many acupuncturists and traditional Chinese medicine practitioners in the United States, the demand for Chinese medicinal plants has been increasing. In the past several years, however, quality concerns have been raised about medicinal plant products imported from China. To assure the safe and efficacious care for patients, practitioners need good quality plant material produced in accordance with good agricultural practices. The objectives of this research were (1) to determine whether quality plant material of selected species of Chinese medicinal plants could be cultivated in the northeastern United States and (2) to evaluate the effects of soil nutrient levels on secondary metabolite production. For these reasons, *Agastache rugosa* (Fisch. & C.A. Mey.) Kuntze, *Leonurus heterophyllus* Sweet, *L. sibiricus* L., and *Schizonepeta tenuifolia* Briq. were field grown in a randomized complete block design using 0, 100, and 200 kg ha<sup>-1</sup> of nitrogen supplied as soybean meal. The nitrogen treatments resulted in dose-related increases in yield in all species. Preliminary organoleptic evaluation suggests the cultivated Chinese medicinal plants are of higher quality than commercially available imported material.

## Downy mildew of basil

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Basil is the most important herb in the United States and perhaps, worldwide. Until 1992, when Fusarium wilt of basil was first reported in the U.S., basil production was not hampered by destructive diseases. Now a second destructive disease of basil, downy mildew, has become established in the U.S. Downy mildew was first reported in Uganda in 1930. The disease did not attract international attention until it recently appeared in several disparate locations; Italy (2004), France (2005) South Africa (2006), Iran (2007) United States (2007) and Argentina (2008). Downy mildew was first reported in the U.S. in Florida. During 2008 and 2009, the disease occurred throughout the east coast in epidemic proportions both in the field and in greenhouses. This member of the Oomycota was not described until 2009 when Thines *et al.* named it *Peronospora belbahrii*. It can become seed-borne and we believe that is how it spread quickly throughout the world and the U.S. Few fungicides are registered to control this disease. The Basidiomycete yeast, *Pseudozyma* colonizes *P. belbahrii* and may help reduce release of sporangia.

## Pre- and post-harvesting procedures effects on *Artemisia annua* concentration of artemisinin and leaf antioxidant capacity

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*Artemisia annua* is the only commercial source of artemisinin. Artemisinin has been established in the past 5-7 years as an effective anti-parasitic and anthelmintic agent of potential use for livestock and man. *A. annua* is currently explored as a medicinal crop in Asia and Africa, but there is little research on the effects of pre- and post-harvest procedures on its biologically-active secondary metabolites, the antioxidant activity, and its potential for animal feeding. We will present our results on the effects of pre-harvest water stress and of post-harvest drying procedures on the concentration of artemisinin and on the antioxidant capacity of the leaves. Our data indicated that the stresses imposed on the plant before and after harvest can significantly increase artemisinin concentration (based on HPLC analysis) and that improper drying procedures can severely reduce the antioxidant capacity (based on the oxygen radical absorbance capacity test) of the plant. Improper management post harvest can significantly reduce the crop value as an animal feed additive and as a medicinal crop. Our HPLC analyses also show that *A. annua* of different chemotypes (high in artemisinin or in artemisinic acid) can be currently found in the medicinal plant market, without mention to the concentration of its active metabolites. This points to the difficulty of testing commercial extracts for biological activity. Our present results, added to past results on the nutritional characteristics of *A. annua*, can benefit farmers who grow *A. annua* as a medicinal crop; could potentially decrease drying costs and increase artemisinin yield profit margins; and highlight the dire need for quality control in the booming medicinal plant market.

## Biomedicine production: Degradation and ecotoxicity of Artemisinin

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*Artemisia annua* synthesizes and accumulates artemisinin. *A. annua* is cropped on large scale in several countries for medicinal purposes and artemisinin is available commercially as an efficient drug against drug-resistant strains of the malarial *Plasmodium* parasite. Artemisinin is unlikely to be produced economically by chemical synthesis or by in vitro production. The toxicity of artemisinin to the malaria parasite is well documented and toxicity to both plants and insects has been reported. When cultivating *A. annua*, artemisinin can be transferred to the soil, either passively from the roots, by rain off or through soil tillage. Maximum artemisinin content measured in Danish *A. annua* field is 11 mg/kg DW. Although the half lives are short, artemisinin stayed for 35 and 60 days in loamy and sandy soil. In soil artemisinin affected the avoidance behavior of earthworm, *Eisenia fetida* and EC50 was determined to 21.6 mg/kg DW. In a growth test with lettuce, *Latuca sativa* L., the EC50 growth inhibition 21 days after germination was 2.48 mg/kg DW. In water artemisinin showed phytotoxic properties towards duckweed, *Lemna minor* and algae, *Pseudokirchneriella subcapitata* with EC50 values of 0.19 mg/l and 0.24 mg/l. Cultivation of *A. annua* may affect soil and water living organisms.

## Optimum date of planting and phytochemical content of *Ocimum tenuiflorum* (Basil) in North Alabama

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*Ocimum tenuiflorum*, belonging to mint family, Lamiaceae (Labiatae), is a popular herb containing essential oil with medicinal, antioxidant and antimicrobial properties. In Asian countries, basil, particularly *O. tenuiflorum* is better known as a medicinal herb used for treating ailments ranging from colds to complex diseases such as cancers and diabetes. A field trial was therefore, conducted to determine optimum date of planting and changes with time in the phytochemical content of *O. tenuiflorum* in Alabama. Three planting dates at monthly intervals beginning from April were the main plots and three *Ocimum* accessions, PI 652056, PI 652057 and PI 288779 were sub-plot treatments. The accessions were harvested monthly and compared for essential oil content, composition and phytochemical profiles of leaves and stems. The total eugenol percentage increased with increased leaf maturity for accessions PI 652056 increasing from 25.3% in very young leaves and stems to 51.5% in the mature leaves and the decreasing to 30.4% in senescing leaves, while  $\beta$ -caryophyllene was the major component of PI 652057 and PI 288779 ranging from 21 to 50%. Among planting dates, third (June) date of planting appeared to be optimum as all accessions produced a significantly greater percentage of relative oil than May or April planting dates. Among harvest stages the second harvest (peak flower) stage showed a greater accumulation of eugenol and  $\beta$ -caryophyllene in leaves and stems than first, third or fourth stages among all accessions. Of the 26 essential oil components identified in the oil, eugenol,  $\beta$ -caryophyllene, E-methyl cinnamate and (trans)- $\beta$ -guaiene were most abundant at all harvest stages. This chemotype of *O. tenuiflorum* may have potential as an alternative source of medicinal herb in Alabama and possibly in the southeastern US.

## Antimalarial Plants Used in Niger Republic's Traditional Medicine

Dr. Idrissa Moussa

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Malaria is one of the most important infectious diseases worldwide and remains a problem of public health in Niger". Besides clean environments and the rational use of the existing therapeutic arsenal, the number of deaths linked to the disease continues to increase. The chimioresistance of *Plasmodium falciparum* to classical antimalarial (anti-malaria) further complicates the situation. To resolve this problem, much research is being carried out on products of the local pharmacopoeia. The present work concerns the nigérienne bio-directed study on some antimalarial plants used in Niger Republic's traditional medicine.

**Day 2: Thursday, July 22, 2010****DAY 2 AM Plenary session II:****Elucidating and manipulating alkaloid biosynthesis in the opium poppy**

Toni M. Kutchan

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Plant natural products are the principle components of many pharmaceutical, food additive and cosmetic preparations. The worldwide market volume of medicinal and nutraceutical plant materials alone is enormous and growing annually. Specifically in our research, we investigate how selected plant systems synthesize medically relevant natural products at the enzyme and gene levels. One of the ultimate goals of this work is to use natural product biosynthetic genes to metabolically engineer medicinal plants with tailored drug profiles to optimize the plant as a source of pharmaceuticals. In particular, the isoquinoline alkaloids are a class of plant natural products with 2500 members derived from the amino acid L-tyrosine. The study of the biosynthesis of plant isoquinoline alkaloids at the enzyme and gene level has greatly advanced in recent years. We now have a number of genes available from the (S)-reticuline-derived tetrahydrobenzylisoquinoline alkaloid biosynthetic pathways. We are now just beginning to meaningfully engineer alkaloid metabolism in plants culture. Multicellular compartmentation of alkaloid pathways must also be considered if meaningful metabolic engineering experiments are to be designed. An update will be given on what we understand today about enzymes, genes and spatial organization in the isoquinoline alkaloid biosynthesis field.

**DAY 2 AM Session: Commercialization****Alternative Sourcing of Omega-3 Fatty Acids: The Role of Biotechnology**

Peter J Gillies

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Omega-3 fatty acids are an essential part a balanced diet. Of the major omega-3 fatty acids, the longer chain polyunsaturated fatty acids EPA and DHA are particularly important for heart health. The most common dietary source of EPA and DHA is fish which in turn gets these fatty acids from algae. As demand for fish and fish oil begins to exceed supply, there is growing concern about the sustainability and biodiversity of fish as a primary source of omega-3 fatty acids. Additionally, changes in aquaculture practices are moving the composition of fish oil away from historical norms raising questions about the epidemiological basis of health benefits ascribed to fish and creating problems for the development of dietary guidelines. Biotechnology offers a way to create new and sustainable sources of omega-3 fatty acids of defined and reproducible composition. Before these new oils reach the market, it is important to prove rather than assume their safety and to characterize their health benefits. This is essential in light of the emerging research showing that individual fatty acids and mixtures thereof can possess their own differential nutritional pharmacology. A novel EPA-enriched oil derived from yeast will be presented as a case study.

## Botanicals for metabolic syndrome and inflammation

### Ilya Raskin

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Pharmacologically active phytochemicals (botanical therapeutics) have been used historically to treat and prevent diseases. Conventional approaches to drug discovery from plants, based on ethical bioprospecting, high throughput screening, and computer-assisted drug design have limitations that have restricted botanical therapeutics research in the pharmaceutical and food industries. In addition, biopiracy concerns, extract standardization, batch-to-batch consistency, proper botanical vouchering, and identification of actives have curtailed the development and sales of multi-component botanical therapeutics. Fortunately, novel enabling technologies and favorable regulatory changes are making the discovery and development of botanical therapeutics more efficient and less controversial. These technologies are leading to the development of novel, safe, and efficacious botanical therapeutics for major human diseases such as diabetes and inflammation. The past, present, and future of botanical therapeutic research will be discussed as well as specific examples of botanicals studied in our laboratory.

## Linking Education, Research and Development: The Chemistry and Quality of Spices and Medicinal Plants from Ghana and Liberia

H. Rodolfo Juliani<sup>1</sup>, Amna Ali, Juliana Asante-Dartey<sup>2</sup>, Dan Acquaye<sup>2</sup>, Larry Amukese<sup>2</sup> and James E. Simon<sup>1,2</sup>

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The safety and effectiveness and natural products are key components to assure their commercialization, particularly for African botanicals which are less known in the international markets. The objective of this work was to test for quality (sensory evaluation and sieve analysis, moisture analysis, and ashes) and proximate analysis of various spices from Ghana and Liberia. Two sets of samples were from Ghana, the first included *Griffonia simplicifolia*, *Aframomum melegueta*, *Xylopiya aethiopica* and *Mondia whitei* while the second *Voacanga africana*, *A. melegueta*, *X. aethiopica* and *G. simplicifolia* with two samples were from Liberia (*Piper nigrum* and *X. aethiopica*). The foreign matter analysis showed all the samples were clean, with low contamination of sand and earth. Though, the black pepper samples showed the highest amounts of foreign materials (5%). *Griffonia* contained 4.7 – 7.1% of moisture, while *A. melegueta* showed the highest levels (14.5%), *X. aethiopica* 5 – 11% and *M. whitei* 7%. In Set 2, *V. africana* showed the highest amount of moisture (17 %) with the rest of the samples showing lower levels (<12%). The development of standards for West African products is a key strategy to contribute to their commercialization and provide the base to contribute to improve their quality. While standardization in the chemistry is key, often we overlook that an initial focus in many developing countries can be on food product safety, cleanliness and consistency (color, aroma/ flavor, plant part, preparation or minimally processed materials) as this can facilitate and strengthen initial trade and product supply.

## **Business incubation resources: Supporting technology commercialization: Case study of the Rutgers Food Innovation Center**

Peggy Brennan-Tonetta

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This presentation provides an overview of the Rutgers Food Innovation Center, a business incubator that offers targeted support to the food and agricultural industries in the New Jersey region. This Center is an excellent example of a program that embraces the principles of entrepreneurship, enhances regional clusters, and leverages and links technology innovators and local universities to the private sector to create the conditions for greater productivity, innovation, and job creation. The Food Innovation Center is the recipient of numerous state, national and international awards for excellence in economic development.

### **DAY 2 AM Session: Ethnobotany and Botanical Sourcing**

#### **On the importance of being vouchered, identified, classified, and properly documented in botanical bioprospecting**

Lena Struwe<sup>1</sup> and Sasha Eisenman<sup>1</sup>

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Natural products research necessitates sources from the wild or from cultivated plants originally obtained from the wild. With the large amount of plant biodiversity (250 000 species and counting), vouchering of the original source for extracts, herbals, and other products are crucial for proper identification. An overview will be given of classification and naming of plants historically and contemporary, including the implications of new evolutionary findings based on DNA data that have led to recent major reclassifications of families, genera, and species. The importance of naming accuracy and best documentation practices for successful publication of new data, including methods for identification (DNA barcoding, floras, keys, taxonomic experts) will be discussed. During the last 10-15 years, plant systematics has gone through a revolution enhanced by new molecular-based techniques, but unfortunately literature in phytochemistry, bioprospecting, and ethnobotany is lagging behind and do not always use updated scientific names, often due to lack of understanding of botanical nomenclature or the implications and importance of name changes. The value and danger of common names and ethnobotanical information in local languages will also be discussed. Examples will be given of best practices as well as pitfalls and common mistakes.

## ***Hypericum perforata* L. (St. John's wort) in Renaissance herbal medicine: assessing the historical evidence for use of hypericum as an anti-depressant in early modern Europe**

Karen Reeds

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Species of the genus *Hypericum* have been used medicinally in Europe for at least two thousand years, as attested by the classical authorities on materia medica (Dioscorides, Galen, and Pliny). In 1996-7, *Hypericum perforatum* L., (hypericum, St. John's wort) suddenly came to Americans' attention as an alternative treatment for depression. Popular and medical interest in St. John's wort was enhanced by appeals to its long history. Manuscript annotations and a pressed specimen of *Hypericum perforatum* in a Columbia University Library copy of the 1546 herbal by Hieronymus Bock, *Kreütter Buoch*, bear witness to interest in the plant in the Renaissance as well. However, Bock's text, other early printed herbals, and the interwoven traditions of Galenic medicine, folk medicine, and Paracelsan medicine before the 17<sup>th</sup> century did *not* recommend hypericum as an ingested remedy for melancholy or any other condition comparable to depression. The dangers of using hypericum in pregnancy were recognized in the pre-Linnaean literature on hypericum, but only belatedly in modern labeling of St. John's wort products.

### **African American use of herbs in health care**

Glenda L. Smith

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The literature related to the use of plant therapies in the African-American community is limited and there is little documentation that describes the historical and present day use of plant remedies, specifically for health promotion and illness in African-Americans. The specific aim of this study was to describe the traditional health practices and types of plant therapies used by rural African-Americans. The practice and knowledge of using herbal remedies has been passed from early slavery into today through the use of oral tradition and African-Americans have incorporated herbs from Native American and European origin to their pharmacopoeia. A typology of 15 plant remedies was developed with Latin and folk name, herb place of origin, known scientific properties, and informant folk usage and dosage information. Several of the herbal remedies used by this population for illnesses and health promotion had well documented therapeutic properties. Further study is needed to investigate the efficacy of the plant remedies used by this and other African-American populations for illness and health promotion.

### **Rose Lore: Essays in Cultural History and Semiotics.**

Frankie Hutton

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The rose is an exquisite flower that blooms beautifully in many varieties throughout the earth for thousands of years. Little known is the history of this primordial flower that is associated with many medicinal, metaphysical and even cosmic manifestations. This presentation will give an overview of little known facts, esotericism and semiotics of the rose.

## DAY 2 PM: Session on Sustainability and Economics

### Enterprise development in rural areas: The case for natural products as sustainable income generating activities using a market-first science-driven model

J.E. Simon<sup>1</sup>, R. Juliani<sup>1</sup>, D. Acquaye<sup>2</sup>, E. Jefthas<sup>3</sup>, J. Asante-Dartey<sup>2</sup>, Bismarck Diawuo<sup>4</sup>, M. Diatta<sup>5</sup>, B. Diouf<sup>5</sup>, P. Langenhoven<sup>3</sup>, N. Hitimana<sup>6</sup>, Ramu Govindasamy<sup>7</sup> and Jerry Brown<sup>8</sup>

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The development of sustainable agricultural businesses in high-value niche sectors can stimulate agricultural growth that could improve the livelihoods of rural Africa and have a multiplier effect on the rest of the economy. The natural plant product (NP) industry is one that can offer higher income and niche markets for resource limited farmers in developing countries compared to traditional agronomic crops. However, the NP industry is beset with challenges hindering the realization of its full contribution to economic development and poverty reduction. Among these challenges are: (1) limited appropriate germplasm; (2) lack of quality control and quality assurance systems for production and processing; (3) Lack of knowledge and understanding of international markets and markets channels; (4) Limited processing infrastructure which constrains value-added opportunities; (5) variable market prices, and (6) weak enterprises with low technical and managerial skills to meet the requirements of buyers. In 1999, we began to develop models for the sustainable commercialization of NP in sub-Saharan Africa using a market-first and scientific-driven approach. This program implemented under ASNAPP ([www.asnapp.org](http://www.asnapp.org)) is conducted in partnership with the public and private sector to facilitate diversification of agricultural commodities and marketing channels. Sustainable development incorporates good environmental stewardship from the bush to final product, GAP and robust quality assurance and quality control systems for collection or cultivation to ensure high quality and food safety. This evolving model consists of a multi step value-addition process through the commodity chain to provide safe and high quality NPs and economic opportunities to those rural communities. The models developed can be replicated in other regions and countries.

### Increasing Marker Constituent Levels of Medicinal Plant Species Ex Situ to Enhance Market Value: Two case studies: *Echinacea purpurea* L. and *Hydrastis canadensis* L.

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The primary focus in a vast majority of crops cultivated for the medicinal industry is on increasing biomass production; however there is a vital need to emphasize the importance of improving marker constituent levels in raw materials. With an increasing demand for superior quality in both *Echinacea purpurea* L. and *Hydrastis canadensis* L., our focus was placed upon increasing the quality and consequently the market value of these two medicinal crops. Through utilizing selective breeding, certain cultural techniques and post harvest handling

practices that influence the growth and development of each genus, we increased the total phenols, echinacosides, and chicoric acid in *Echinacea purpurea* and total alkaloids, i.e., berberine, and hydrastine in *Hydrastis canadensis*. Providing raw material with these improved levels of marker constituents also increases the market value of these botanicals from 7% to 12%. These techniques impact the plants during their growth cycles, harvesting and post harvest, thus the techniques shared may be applied to other medicinal crops. We improved the *Echinacea purpurea* constituents along with an increased biomass by 23% and increased the total alkaloids in *Hydrastis canadensis* by 40% through continuously monitoring the plants throughout their annual growing cycle(s), allowing us to determine the amount of stress we were imposing along with improving post harvest handling techniques. Our findings prove that with proper monitoring and management of each species, the dollar per pound value of the raw material can be enhanced resulting in better returns for the farmer/grower in today's market.

### **Amendment of Low-Fertility Soils for Fairywand Culture in Appalachia**

Mario R. Morales

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Modern agriculture is a profitable business due mainly to the use of synthetic fertilizers, chemical pesticides, soil tillage, irrigation, and large extensions of land. However, these factors have had a negative impact on the environment and rural way of life. Currently, rural America faces serious problems with soil erosion, air pollution, water contamination, soil salinization, plant diversity loss, and general environment deterioration. These problems can be reversed with the application of sustainable agricultural practices, which include no-till planting, cover cropping, crop rotation, multiple cropping, organic matter incorporation, mulching and ecological pest management. Appalachia with its hilly topography, steep terrain, and shallow rocky soil is a fragile ecosystem that needs special management practices in order to improve and conserve the soil, water and air quality and promote and maintain plant diversity. To assess the potential for improving the poor soils of southern West Virginia, fairywand [*Chamaelirium luteum* (L.) Gray], an Appalachian native medicinal plant, was planted in a replicated trial with 3 rates of humus (0, 20, 40 lb) and 3 rates of bonemeal (0, 40, 80 oz) Results indicated that bonemeal significantly increased plant width and leaf length in fairywand and pH, phosphorous, calcium and magnesium in the soil. Humus significantly increased potassium, calcium, and magnesium in the soil.

### **The Role of Medicinal Plants in Food Security and Nutrition**

Lorraine Cordeiro<sup>1</sup>, David Nyachuba<sup>1</sup>, and Lyle Craker<sup>2</sup>

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The use of medicinal plants for improving health in Africa has an extensive history. In the past decade, considerable concern has been raised about loss of biodiversity due to population growth and the use of marginal/fragile lands. Medicinal and other traditional plants are derived from areas of rich biodiversity. There is evidence that food insecure households in Sub Saharan Africa commonly harvest and consume wild plants

during periods of food insecurity and often rely on medicinal plants to treat common ailments. The Coping Strategies Index, an indicator of household food security, considers this as one of many coping strategies utilized during food shortages. Reliance on medicinal plants in Africa is based on their accessibility, availability, benefits, as well as inadequate access to health care and conventional medicine. Few studies have explored the role of medicinal plants in improving food security and nutrition. However, pharmaceutical companies have made headway in advancing the livelihoods of local communities through research and harvesting of medicinal plants for biomedical research. Integration of research on food security, nutrition, and their links use of medicinal plants are necessary for improved and innovative strategies aimed at alleviating food security and improving nutritional health in developing countries.

## DAY 2 PM: Session on Biotechnology of Medicinally Active Plants

### Artemisinin: *in planta* production and therapeutic options

PJ Weathers<sup>1,3</sup>, PR Arsenault<sup>1,3</sup>, DR Vail<sup>1,3</sup>, K Teoh<sup>3</sup>, KK Wobbe<sup>2</sup>

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*Artemisia annua* L. produces the sesquiterpene lactone, artemisinin, a potent antimalarial drug that is also effective in treating other parasitic diseases, some viral infections and various neoplasms. Artemisinin is also an allelopathic herbicide that can inhibit the growth of other plants. Unfortunately, the compound is in short supply and thus, studies on its production in the plant are of interest as are low cost methods for drug delivery. Here we review our recent studies on artemisinin production in *A. annua* during development of the plant as it moves from the vegetative to reproductive stage (flower budding and full flower formation), in response to sugars, and in concert with the production of and regulation by ROS (reactive oxygen species). Also provided are results from animal experiments measuring the potential of using the dried plant directly as a therapeutic. Together these results provide a synopsis of a more global view of regulation of artemisinin biosynthesis in *A. annua* than previously available. We further suggest an alternative low cost method of drug delivery to treat malaria and other neglected tropical diseases.

### Molecular characterization of peach [*Prunus persica* (L.) Batsch] germplasm in the United States using microsatellite markers

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Peach [*Prunus persica* (L.) Batsch] is an important medicinal fruit with immense health benefits and antioxidant activity. In this study, microsatellite markers were used as DNA fingerprinting tools for the identification and characterization of peach germplasm in the United States. Eleven microsatellite primers amplifying twenty-two loci were used to characterize 31 peach accessions. Alleles were detected using fluorescent-labeled primers and capillary electrophoresis. Microsatellite alleles were scored using the CEQ 8000 Fragment Analysis

software and edited based on the bin list using a SAS program. Polymorphic information content, allele frequencies and heterozygosity were calculated using the program GenAlex 6.0 and Powermarker v. 3.0. Ten primers revealed a high level of diversity and detected an average of 6.2 alleles per locus. The mean expected heterozygosity was 0.77. The probability of identity was lower than 0.001 demonstrating the high efficacy and reliability of these primers in genotype identification. Principle coordinates analysis revealed significant variation among the accessions. A minimum of 5 loci was sufficient to discriminate all 31 accessions. This study indicates that microsatellite markers can serve as a DNA fingerprinting tool for peach cultivar identification, parentage and sibship analysis, and diversity assessment. These potential applications could be significant for genetic improvement of peach.

### **Engineering Taxol biosynthesis in *Taxus* suspension cell culture**

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Plant cell culture provides an environmentally friendly, renewable alternative for supply of plant derived pharmaceuticals. A notable example of the success of plant cell culture technology is the commercial synthesis of the anti-cancer agent paclitaxel (Taxol™). One defining characteristic of plant cell suspension cultures is the tendency of cells to grow in aggregates, which introduces significant cellular heterogeneity which affects secondary metabolite accumulation. Different microenvironments in an aggregate induce differentiation, resulting in changes in cellular gene expression and metabolic function. Our laboratory focuses on characterizing such cellular heterogeneity and production variability in *Taxus* suspension cell cultures through the use of novel flow cytometric and metabolic engineering techniques. By comparing expression profiles of known paclitaxel biosynthetic pathway genes in methyl jasmonate-elicited and unelicited *Taxus* suspension cultures to total taxane accumulation, potential bottlenecks in the pathway have been identified. To understand system-wide cellular heterogeneity and to determine effective strategies for metabolic engineering, we are examining expression of pathway genes in both aggregates of different sizes and accumulating and nonpaclitaxel-accumulating populations isolated through flow cytometry. These analyses will enhance our understanding of paclitaxel metabolism at the cellular and molecular level and will provide a basis for more targeted metabolic engineering and bio-process design.

## Natural resveratrol analogs from root cultures of peanut and muscadine grape: bioproduction, biotransformation and bioactivity

Fabricio Medina-Bolivar<sup>1,2,5</sup>, Jose Condori<sup>1</sup>, Julie Carrier<sup>3</sup>, Malathi Srivatsan<sup>2</sup>, Anna Radomska-Pandya<sup>4</sup>, Ganapathy Sivakumar<sup>1</sup>, Vipin Nair<sup>1</sup>, Maureen Dolan<sup>1,5</sup>.

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Resveratrol and its natural analogs are stilbenoids that have been associated with a multitude of health benefits. To study the biosynthesis of these compounds and establish a sustainable bioproduction system, we developed hairy root cultures of peanut (*Arachis hypogaea*) and muscadine grape (*Vitis rotundifolia*). The biosynthesis of these polyphenols was induced upon medium exchange in combination with elicitors (sodium acetate and methyl jasmonate). In peanut, most of the stilbenoids included resveratrol and prenylated analogs such as arachidin-1 and arachidin-3, whereas in muscadine grape, prenylated stilbenoids were not found. In muscadine grape, the main stilbenoids included resveratrol, piceid and viniferins. Centrifugal partition chromatography was used to purify the stilbenoids from the culture medium. Arachidin-1 showed strong neuroprotection *in vitro* and higher antioxidant activity than resveratrol in a lipid peroxidation assay. Furthermore, human UDP-glucuronosyltransferases involved in the metabolism of arachidin-1 and arachidin-3 were determined *in vitro* to study the biotransformation of these prenylated resveratrol analogs and potential bioactivity of their conjugated glucuronidated metabolites. Our results indicate that the hairy root cultures provide a valuable tool for studying the biosynthesis of stilbenoids and discovering bioactive polyphenolic compounds with potential applications in human health.

### DAY 2 PM: Session Quality Control

#### Quality Assurance and Quality Control: Making it Relevant to Growers and Processors

H. Rodolfo Juliani

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In the last years there has been an increased interest in natural plant products, which is driving a multibillion dollar market and generating commercial opportunities all over the world. The production and processing of safe and effective botanical products are challenging activities, since product quality is affected by all the steps of the production chain from selection of germplasm all the way to the final consumer. The objective of this presentation is to provide selected examples on the production and processing of botanical products (medicinal plants, spices and herbal teas) to illustrate the factors can affect product quality (safety and effectiveness) relevant not only to growers and processors but also to consumers alike. The lack of information on new plant products particularly on their effectiveness and safety sometimes can limit their commercialization. Thus, the implementation of quality systems and studies that can identify the physical, chemical and biological activities of plants and their products, can ultimate assist growers and processors,

particularly in the developing countries, to generate high quality products for their increased the price of the products of facilitate their market access.

## **Using natural products chemistry for QC and botanical standardization**

Qingli Wu

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Natural products chemistry has played a very important role in human health covering the areas of medicine and public health, agriculture and food, and personal care. In this presentation, an overview on natural products chemistry application, accomplishments and highlights, and classification is provided. Practicum on natural products chemistry including extraction, purification, structure determination and the quality control on natural products is discussed. Quality control on natural products chemistry is the basis for the application, and is also a challenging and dynamic area. Different analytical approaches on quality control of natural products are reviewed, and case studies on polyphenols in wide range of botanicals carried out in NUAPP are presented.

## **Regulating herbal products: A North American perspective**

Dennis V.C. Awang

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Lack of assurance of plant species identity is arguably the most serious deficiency of commercial herbal products. Product vitiation is almost invariably revealed by reports of adverse health effects. With the usual destruction of morphological features, which attends processing of plant material: comminution, extraction, and other formulation treatment, and without a reliable chemical basis for unequivocally determining botanical identity, the only sensible and reliable immediate route to such assurance would appear to be institution of a system for independent botanical certification. At present, reliance on identification of plant morphological features and/or phytochemical profiles does not ensure correct species identity. However, the advent of DNA-based approaches, proceeding with now steadily decreasing time demand and reducing cost, give promise of overcoming most of the shortcomings of the two currently dominant methodologies — particularly respecting blended plant preparations; a comprehensive DNA database should be urgently promoted. Along with a registry of certified growers and a raw material tracking system, there ought also to be instituted a registry of chemical analytical laboratories, qualified to test both raw material and finished products, as well as a schedule of rotational random testing of marketed products.

## DAY 2 PM: Session Ethnobotany and Plant Improvement for MAPs

### Genetics and biotechnology of the genus *Mentha*: a model for other polyploid species with essential oils and other plant constituents?

Arthur O. Tucker

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The greatest amount of research on the genetics and biotechnology of the biochemical pathways and inheritance of essential oils has been with the model systems of the genus *Mentha*. In particular, the genetic work of Dr. Merritt Murray and the biotechnological work of Dr. Rodney Croteau stand out for the amount of good, new data, but these two directions of research have not been reconciled previously. In addition, new insights on previously published research in *Mentha* reveal that cytomixis fuels complement fractionation, which, in turn, produces transgressive segregation in *Mentha*. Assimilating over a century of different methods for breeding in *Mentha*, two approaches stand out: (1) gamma-irradiation of large populations and (2) promoting cytomixis with chemical/physical agents and raising large numbers of hybrid progeny. Are these methods applicable in other polyploid species with essential oils? Are they applicable for other plant constituents?

### Breeding the American Cranberry (*Vaccinium macrocarpon*) for Health Constituents: Genetic Variation for Flavonoid Content

Nicholi Vorsa and Jennifer Johnson-Cicalese

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The American cranberry is recognized for a number of benefits to human health. Much of the attention has focused on urinary tract infections and A-type proanthocyanidins inhibiting adhesion of *E. coli* to cells. Proanthocyanidins (PAC) are well known for their anti-microbial and anti-herbivory activities, and occur in high levels in cranberry, primarily as epicatechin polymers. Cranberry also has one of the highest flavonol contents relative to other fruit crops, with predominantly quercetin glycosides, which exhibit anti-bacterial and anti-fungal activity, and are potent antioxidants. Cranberry fruit are also abundant in a third flavonoid class, anthocyanins, largely glycosides of cyanidin and peonidin, both noted antioxidants. Through interspecific hybridization, we can alter cranberry anthocyanin glycosylation, with the potential goal of improving bioavailability. Significant genetic variation exists in cranberry for all three flavonoid classes and is largely quantitative, indicative of polygenic traits with significant environmental effects. A survey of our cranberry germplasm found a six-fold variation for PAC content. Parent-offspring regression indicates relatively high heritability for anthocyanin content and low to moderate heritability for proanthocyanidin content. The interrelationships of horticultural traits, e.g. yield, with flavonoid composition is currently being evaluated in our advanced breeding materials.

## **Breeding for active principles in aromatic plants and herbs**

Jim Simon, Rodolfo Juliani, Chung H. Park, Christian A. Wyenandt, Julia Coppin and Qingli Wu  
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Aromatic plants and herbs are popular because of their desirable aromas and flavors. The large genetic variation in content and composition of aromatic compounds lends itself to the development of new varieties. Coupled with the wide diversity in other horticultural characteristics such as growth, yield, leaf size and shape, and disease resistance, the opportunities to create new herbs are exciting. Recent interest in the health and nutritional aspects of culinary herbs suggests a focus on the bioactive compounds rather than the aromatic volatile compounds as a promising avenue to pursue. This paper will present research on the bioactive compounds in basil, catnip, mint and oregano, and specifically the polyphenols that accumulate in herbs.

## Poster Session

The following papers are presented as posters all day July 21 and 22.

### Ethnobotany and Botanical Sourcing

#### Malarial Curatives from Northeast United States

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Malaria is caused by four species of parasites in the genus *Plasmodium*: *P. vivax*, *P. falciparum*, *P. ovale*, and *P. malariae* and is transmitted among humans by female mosquitoes in the *Anopheles* genus. In 2006, there were 247 million cases of malaria and 881,000 deaths worldwide. Many currently-used treatments have lost efficacy in the face of increasing drug resistance. Plants are a proven source of antimalarial drugs, as evidenced by the development of quinine and artemisinin from the *Cinchona spp.* and *Artemisia annua* plants, respectively. To date, a great number of studies have focused on species found in Africa and Asia. Though malaria is now a non-issue in the United States, it was widespread before the eradication campaign in the 1950s. Therefore, few have considered the possibility of finding effective treatments in North American plants. However, reports of traditional medicinal systems indicate that some species may be active. The aim of this study was to evaluate the antiplasmodial and cytotoxic qualities of plants used by Native Americans and American settlers to treat malaria, specifically in the Northeastern and Mid-Atlantic regions of the United States through the Civil War. Five plant species were selected from the literature.

#### Dogwoods: a new anti-malarial drug?

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Malaria is a mosquito-borne infectious disease that kills nearly 1 million people each year. There are a number of drugs to treat malaria, but drug resistance is spreading. This problem is further complicated by cross-resistance among drugs in the same chemical family. Scientific studies based on traditional use and chemistry indicate that species of the Cornaceae family, deciduous woody plants commonly known as dogwoods, have been used as anti-malarial medicines and should be investigated further. We performed an ethnobotanical review of all 55 species, of which 24 species were found to be used as medicinal plants, and 16 of these reported to have anti-malarial properties (see table to the right) and 3 were stated to be used as quinine substitutes. Therefore, all *Cornus* species should be tested for potential anti-malarial activity. Continued research and anti-malarial screening is needed for the Cornaceae family. Given that the family is large and diverse and that taxonomy and phylogenetic relationships are still debated, additional evolutionary research is needed as well.

## Germplasm and Plant Improvement

### ***In Vitro* Germplasm Maintenance Growth, Yield and Quality of Assiut-1, Broccoli (*Brassica oleracea* var. *italica*) cultivar.**

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Broccoli is highly nutritious, and has been deemed as a vegetable with potential anti-cancer activity due to high levels of glucoraphanin, which can hydrolyse to form sulphoraphane, an isothiocyanate. Assiut-1 is a synthetic cultivar that was produced at the Department of Horticulture, Assiut University by Damarany and Aboul-Nasr (2000). The original parents of this genotype were namely Parma, Atlantic, Walthon-29 and Toro. A mass selection was conducted for twelve years to get a late flowering broccoli genotype under Assiut conditions. Several research papers were done about this cultivar concerning its yield, head quality, chemical components, storage quality, seed oil contents and quality. Assiut -1 gave the heaviest plant fresh weight, number of leaves per plant, head height, high yield, good quality oil and high quality chemical components as compared to other cultivars or hybrids. Recently we are doing in vitro study to produce haploid plants through anther culture. Pre- results about germplasm maintenance will be discussed.

### **Biochemical qualities of new sea berry (*Hippophaë rhamnoides*) hybrids developed for Latvia – an example for New Jersey berry industry**

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Sea berry or sea buckthorn (*Hippophae rhamnoides* L) (SBT) is an old medicinal plant from Northern hemisphere with new multi-perspectives. The objective of this ongoing study is to identify and develop high quality SBT hybrids/cultivars, having optimum biological activity, suitable for commercial cultivation in Baltic Coastal areas, with potential extension of cultivation towards northern boundaries. With subsequent crossing and backcrossing among different strains, including *H. rhamnoides* ssp. *mongolica*, ssp. *fluviatilis*, and ssp. *rhamnoides*, new Latvian hybrids such as 'No 3.1', 'No 3.2', 'No 1.4', 'No 1.5', 'Mary', 'Tatjana', and 'Lord' were developed. The first four hybrids along with "Podarok Sadu", and 'Trofimovskaya' were developed and recommended for commercial cultivation under maritime climate, where frequent winter thaws are a common phenomenon. The new Latvian hybrids were compared with old Russian cultivars, such as, 'Maslichnaya', 'Dar Katugni', 'Vitamnaya', 'Chuisckaya', 'Obilnaya', 'Prozrachnaya', 'Botanicheskaya Ljubitel'skaya', 'Podarok Sadu', 'Luchistaya', 'Augustinka', and 'Trofimovskaya'. The color intensity evaluated by color analyzer CIE L\*a\*b\* showed a close correlation ( $r = 0.948$ ,  $n=12$ ) between the total carotenoids in fruit juice and a\* value (characterizes the intensity of red color). We found that the content of total soluble solids, oil, vitamins C and E, carotenoids, total phenolic compounds, and total acids were higher in newly developed Latvian hybrids than in older Russian cultivars. The total acidity of the fruits in older Russian cultivars varied from 2.70 - 3.50 %, while this value for new Latvian hybrids was 2.54 - 4.16 %. A few Latvian hybrids have been distributed and planted

in test plots in Latvia, Estonia, Finland, Sweden, Poland, Canada, UK, Norway and Chile. New Jersey and other states with extended shores, ecological problems, and emerging market opportunities should capitalize on available human expertise on SBT, and enable eco-agro-industrial advancement.

## **Control Environmental Horticulture and Propagation**

### **Cytokinin Effects Multiplication and Rooting of *Aloe barbadensis* from Agar and Liquid Medium**

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*Aloe barbadensis* (syn. *Aloe vera*) was micropropagated on agar and liquid medium, at varied benzyladenine (BA) and meta-topoloin (MT) concentrations (0, 1, 3.2, and 10  $\mu\text{M}$ ) for 3 successive culture cycles, followed by transfer to greenhouse. MT did not induce multiplication unless at the highest concentration (10  $\mu\text{M}$ ) and BA produced the greatest number of plantlets at 3.2  $\mu\text{M}$  although optimal multiplication was at approximately 6  $\mu\text{M}$ . Liquid medium did not affect multiplication rate when compared with agar, but plants were 75% larger from liquid than from agar at time of greenhouse transfer. After 5 weeks of growth, plants in greenhouse from liquid culture were still larger than plants from agar, and both BA and MT reduced plant size. Cytokinin carryover inhibits rooting and plants on agar had a more severe carryover effect than plants from liquid. Cytokinin carryover reduced the *per cent* rooted from 92% (control) to 68% with either 3.2  $\mu\text{M}$  MT or 10  $\mu\text{M}$  BA; and 10  $\mu\text{M}$  MT had about 20% rooted. There appears to be a trade-off between maximum multiplication rates and best plant quality for *ex vitro* transfer. Using liquid medium gives larger plants, and lessens the cytokinin carryover effect on rooting without affecting the multiplication rate. Approximately 6  $\mu\text{M}$  in liquid medium would be optimal for multiplication and rooting of *Aloe barbadensis*.

## **Production and Post-Harvest Processing**

### **Growth and Yield of Fifteen Garlic Ecotypes.**

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The effects of three planting dates, fifteen garlic ecotypes (11 Local cultivars and 4 Chinese ecotypes) and two plant density (30 and 60 cloves in each row sides) on garlic growth, yield and some chemical component were studied. The obtained results indicated that plant height, fresh weight per plant, total yield, number of cloves per bulb and weight of cloves per bulb were increased by early planting. However late planting increased the bulb dry weight. On the other hand, bulbing ratio was not affected by planting dates. Ecotypes from Balady (local cultivars) surpassed Chinese in all studied characters. On the other hand, Chinese ecotypes surpassed Balady ecotypes in weight of cloves per plant and bulb dry weight. Increasing plant density significantly

decreased plant height, both bulb and neck diameters, bulb fresh weight, plant fresh weight and weight of cloves per bulb. However, increasing plant density significantly increased total yield and bulb dry weight.

### **The IR-4 Project – Registering Pest Control Needs for Specialty Crops Since 1963.**

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Since 1963, the IR-4 Project has been the major resource for supplying pest management tools for specialty crop growers by developing research data to support registration clearances. IR-4 is a partnership between USDA (NIFA & ARS), the State Agricultural Experiment Stations, agrichemical and biopesticide registrants, growers, commodity groups, food processors and US EPA. IR4 data is used to establish food tolerances, or the legal limit, in the U.S. and International Maximum Residue Limits (MRLs) for exporting from the U.S. The edible herbs and spices crop group was established in 1983 and this allowed basil and chive data to be used to obtain a tolerance on all herbs and spices included in the crop group. A new Herbs and Spices crop group is currently being established and it includes many more species and it breaks the herbs and spices into subgroups. Pest control tools registered for use on herbs through IR-4 include: avermectin, azoxystrobin, bifenthrin, carfentrazone-ethyl, clethodim, cryolite, cyprodinil, ethylene oxide, fludioxonil, imidacloprid, glyphosate, mefenoxam, propylene oxide, spinosad, spinetoram, and sulfuryl fluoride. Downy mildew of basil is the reason for three new projects with cyazofamid, mandipropamid, and fluopicolide.

### **Crop Grouping and the Efficient Registration of Pesticides for the Production of Herbs and Spices**

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Crop grouping in the U.S. is a well accepted and cost effective approach that facilitates the efficient establishment of pesticide tolerances for both major and minor crops. The purpose of Crop Grouping is to facilitate the establishment of Maximum Residue Limits (MRLs) on crop groups and to provide maximum crop protection with minimum effort. The US crop grouping regulation has brought great benefit to growers, researchers, agri-chemical companies as well as regulatory agencies since 1983. The crop grouping regulations (40 CFR § 180.40 and 40 CFR § 180.41) allow for the establishment of tolerances for a group or subgroup of crops based on residue data from representative crops of the crop group or subgroup. When crops which have similar morphology, cultural practices, edible portion, growing season, geography and pest problems are contained in the same crop group or subgroup, they can be expected to have similar residue profiles. Representative crops are the most economically important crops in the group and are most likely to have the highest residue and a greater dietary burden.

## Developing Strategies to Improve Resistance and Control of Basil Downy Mildew

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One of the most popular herb crops grown throughout the world is basil (*Ocimum basilicum*). A member of the Lamiaceae family, the genus *Ocimum* consists of 30 species that originated mainly from Asia and Africa. Grown throughout the world in warm, Mediterranean and temperate climates, basil is used as a flavoring, spice, health product, fragrance, medicine and religious offering. Insecticidal, nematocidal, fungistatic and antimicrobial properties have also been identified. Unfortunately, many diseases affect this crop. One of the most recent pathogens affecting this crop is Downy Mildew (D.M.) (*Peronospora belbahrii*) of basil. Moderate to warm temperatures and humid to wet conditions during the growing season are favorable for the development of this devastating disease. Yellowing, downy, purplish gray appearing sporulation, and eventual necrosis in basil affected by D.M., results in complete crop loss. There are a number of strategies to improve resistance and promote control of downy mildew of basil. In this study, breeding, chemical control, genetic screening, and plant morphology are being researched to determine strategies for control.

## Role of botanicals in management of crops diseases

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In order to maintain the productivity of various crops, xenobiotic chemicals are being added in the natural environment by the farmers. According to the WHO survey, more than 50,000 people in developing countries are annually poisoned and 5,000 die as a result of the effects of toxic agents, used in agriculture. In India 35,000 – 40,000 tons of hazardous chemicals are sprayed on the crops every year, instead of helping the poor, these chemicals are causing cancer, sterility and death. So there is an urgent need to develop sustainable methods for these horrible diseases. As plants and their product are known to possess various secondary metabolites, which showed inhibitory effect against the growth of pathogens, therefore, the plants and their product should be utilized to combat the diseases causing pathogens. In the present study experiments were carried out to evaluate the antimicrobial properties of 120 plant parts samples of 100 plants spanning over 45 families against five plant pathogenic fungi by the food poisoning method. The results are promising and some of the plants have shown inhibitory activity against one or two fungi whereas others have shown a broader spectrum of activity, some plants showing good activity against all the test fungi. Plants samples of some families such as *Apocynaceae*, *Caesalpinaceae*, *Combretaceae*, *Compositae*, *Ebenaceae*, *Liliaceae*, *Lythraceae*, *Meliaceae*, *Mimosaceae*, *Rosaceae*, *Salvadoraceae*, *Sapindaceae*, *Theaceae* and *Zingibraceae* were found to be comparatively more effective against the test fungi.

## **Afghan poppy blight of 2010**

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In February 2010, opium poppy and other plants in Southern Afghanistan started exhibiting wilting, leaf spot, and root problems. No clear cut pattern of symptoms that allowed a rapid diagnosis was apparent, suggesting the problem could be caused by a number of diseases. Laboratory testing is underway in Germany and elsewhere and until the lab reports are published, little else is available to report, except that as a result of the disease, opium prices are climbing. Rather than wait for the tedious science, The Guardian newspaper (UK) has reported the disease was caused by the Americans using biological warfare. Despite the complete absence of evidence, the article is being taken as true by a non-scientific public.

## **BioTechnology of Medicinally Active Plants**

### **Manipulation of secondary metabolism in hairy roots of tomato transformed with a mammalian polyphosphate 5-phosphatase**

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Inositol (1,4,5) trisphosphate (InsP<sub>3</sub>) is a second messenger in plants that increases upon challenge to different stimuli. Previous work has shown that tomato plants engineered with a human polyphosphate 5-phosphatase (InsP 5-ptase) show increased drought tolerance, biomass and lycopene content. To further elucidate the mechanisms underlying these observations, we established hairy roots from the wild type and transgenic tomato plants. The presence and expression of the InsP 5-ptase transgene was confirmed by PCR and RT-PCR, respectively. The transgenic root cultures exhibit significantly lower biomass in comparison to the wild type cultures. To study the impact of manipulating InsP<sub>3</sub> pathway on secondary metabolism, 15-day cultures were treated with 100  $\mu$ M methyl jasmonate (MeJA) for 24 hours. HPLC analyses showed a different profile of compounds in the medium and root tissue following elicitation. Whilst MeJA induced production of non-polar compounds that secreted in the medium, more polar putative phenolics were induced and retained in the tissue. More significantly, much higher levels of these latter compounds were found in the root tissue of the transgenic hairy roots. Our work proposes that manipulation of InsP<sub>3</sub> levels in the hairy roots has a strong effect on root growth and secondary metabolism.

## Comparison of sodium acetate- and methyl jasmonate-mediated elicitation of bioactive stilbenoids in hairy root cultures of peanut

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Stilbenoids are polyphenolic compounds that have shown a multitude of beneficial properties that impact human health. In order to study the mechanisms involved in the biosynthesis of these compounds, we established hairy root cultures of peanut (*Arachis hypogaea*) cultivar Hull. Sodium acetate (10.2 mM NaOAc) and methyl jasmonate (100  $\mu$ M MeJA) were used as elicitors. Nine-day cultures were treated with these elicitors after replacing the spent medium with fresh medium containing the elicitors. The concentrations of resveratrol, arachidin-1 and arachidin-3 were analyzed in the culture medium by HPLC in a time course study following elicitation. A more rapid production of stilbenoids was observed in the MeJA-treated cultures. However the levels of stilbenoids were at least 10 times higher in the NaOAc-treated cultures. Interestingly, when the spent medium was not replaced with fresh medium, the production of the stilbenoids upon NaOAc treatment was much lower than in the presence of fresh medium. More importantly, the simple act of replacing the spent medium with fresh medium did not elicit the production of stilbenoids. This research will help understand the mechanisms involved in the regulation of the biosynthesis of these bioactive stilbenoids in *in vitro* cultures.

## Production of antitumor flavonoids in hairy roots of *Scutellaria lateriflora*

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The genus *Scutellaria* has been widely used by many cultures to treat several medical conditions including nervous disorders and cancer. Previous studies have shown that flavonoid-containing extracts from the roots and leaves of *S. lateriflora* (American skullcap) exhibit antitumor activity *in vitro*. In order to develop a bioproduction system for these valued compounds, we established hairy root cultures of this species via *Agrobacterium rhizogenes*-mediated transformation. Several lines of *S. lateriflora* hairy roots were produced and line 4 was selected based on its growth performance liquid medium. In order to study the production of the bioactive flavonoids, the hairy roots were treated with methyl jasmonate (MeJA) for either 24 or 48 hours. HPLC analysis was performed with ethyl acetate extracts of the medium and lyophilized tissue. The known antitumor flavonoids wogonin, baicalein and baicalin were identified in the root tissue of the MeJA-treated and non-treated root cultures. Among these flavonoids, wogonin was found in higher levels. Novel putative phenolic compounds were found in the medium extracts of the MeJA-treated cultures. Our work shows that hairy root cultures of *S. lateriflora* have the biosynthetic capacity to produce antitumor flavonoids and that elicitation induces the biosynthesis of several compounds with potential biological activity.

## Human Health, Phytochemistry and Bioactives

### Resveratrol mitigates the pro-inflammatory responses in LPS-treated macrophages: Relevance for Alzheimer disease treatment

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Alzheimer disease (AD) is a neurodegenerative disorder characterized by amyloid- $\beta$  (A $\beta$ ) peptide deposition and by prominent inflammation of the brain. Our group has recently demonstrated that the natural polyphenol resveratrol modulates the progression of AD-type pathology in a mouse model for cerebral amyloid deposition by a mechanism promoting A $\beta$  clearance. Evidence is also emerging indicating that resveratrol promotes anti-inflammatory responses. To further characterize the anti-inflammatory potency of resveratrol and determine its potential beneficial effect against the inflammatory component of AD, we have assessed the effect of resveratrol treatment on the pro-inflammatory responses in macrophage and microglial cells. We aimed to determine whether resveratrol modulates distinct important pro-inflammatory pathways that are activated by lipopolysaccharide (LPS) treatments in these cells. We found that resveratrol dose-dependently prevented LPS-induced activation of iNOS and COX-2, and of STAT1, STAT3, and Akt pathways. Furthermore, resveratrol potently blocked TNF $\alpha$  production in stimulated macrophages. Recent studies have shown that increased production of A $\beta$  can directly activate the immune system to evoke AD-type pathology, indicating that inflammation is central in the neurodegenerative process of the disease. Thus, our results strongly support the notion that resveratrol has therapeutic potential against AD by modulating both cerebral amyloid deposition and the associated inflammation.

### Soxhlet Extraction of *Veratrum* with HPLC Analysis for the Assay of Cyclopamine, a Precursor for a Novel Smoothened (Smo) Inhibitor in Clinical Development

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IPI-926, a semi-synthetic molecule derived from cyclopamine (CA), is a novel Smoothened (Smo) inhibitor currently in clinical development. CA is an endogenous alkaloid in *Veratrum*, and it is critical to understand the alkaloid content in *Veratrum* collected through wild harvesting to increase yield and production. The technique of soxhlet extraction with HPLC analysis provided an efficient screening approach for consistent CA yields with low variability. A representative lot of *Veratrum* biomass was extracted, and a reverse phase HPLC method using UV and MS detection was developed using standard alkaloids such as veratramine, veratrosine, cycloposine, and muldamine. These different alkaloids were identified in the biomass using relative retention times and MS detection. Also, the use of relative response factors by UV facilitated quantitation of the different alkaloids in the biomass. A lot of *Veratrum* biomass analyzed with soxhlet extraction showed consistent

recovery of > 5 g CA/Kg biomass. Development of a new extraction and analysis method provided the capability to screen and profile alkaloid content in various samples of *Veratrum*.

### **Effect of concomitant administration of aqueous extract of leaves of *Trichosanthes Dioica* and metformin in diabetic rats.**

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The purpose of the study was to investigate the interaction of aqueous extract of leaves of *Trichosanthes dioica* (TD-aqu), an anti-hyperglycemic, with metformin on serum glucose level and oral glucose tolerance test (OGTT) in streptozotocin (40mg/kg) induced diabetic rats. TD-aqu (800mg/kg), metformin (250 and 500mg/kg) singly and concomitantly were administered orally in diabetic rats. In the acute study serum glucose levels were determined at 0, 2, 4, 6, 12, 18 and 24hrs after drug administration. The subacute study involved repeated administration of drugs for 14 days. In OGTT glucose (2.5g/kg) was administered in diabetic rats 30 min after pretreatment with TD-aqu (800mg/kg), metformin (250 and 500mg/kg) and their concomitant administration. The peak anti-hyperglycemic effect of TD-aqu (800mg/kg) and metformin (250 and 500mg/kg) given alone was at 2hrs and the duration of action was 6hrs. The peak anti-hyperglycemic effect in concomitant administration of TD-aqu (800mg/kg) with metformin (250 and 500mg/kg) was at 2hrs and the duration of action was 12hrs. Repeated (once a day for 14 days) concomitant administration of TD-aqu and metformin caused significant ( $p < 0.05$ ) synergistic reduction in serum glucose level as compared to TD-aqu or metformin alone. In OGTT, concomitant treatment significantly ( $p < 0.05$ ) increased the glucose tolerance compared to TD-aqu or metformin alone. We conclude that this combination has a synergistic effect both on reducing the blood glucose and also on prolonging the duration of action of each other; hence TD-aqu could be used as a useful adjuvant to prolong the action of metformin and also to increase the glucose tolerance in diabetic patients who are on metformin.

### **Anti-tumor properties and effects on vascular smooth muscle cell proliferation and migration of extracts of *Hibiscus sabdariffa* accessions**

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*Hibiscus* extracts are used in traditional African and Chinese medicine. Previous studies have demonstrated medicinal properties of select *Hibiscus* extracts; however information is very limited about variations among *Hibiscus sabdariffa* accessions. Accessions of *Hibiscus sabdariffa* can be grouped by calyx color into green, pink, red, and dark red. Here we examine differences among accessions in anti-tumor properties and on

vascular smooth muscle cell proliferation and migration. Cells were exposed to concentrations ranging from 0.0 to 4.0 mg/ml of Dark Red, Red, Pink, or Green extracts. Tumor cell proliferation, evaluated by Cell Titer Glo and crystal violet assays, showed a dose dependant decrease in cell proliferation after treatment with the dark red extract at 1.0mg/ml for all cell lines except A172 (2.5 mg/ml). Red extract decreased proliferation for most cell lines around 3.0 mg/ml. Pink extract results were closer to the dark red extract, significantly reducing cell proliferation between 1.0-2.5 mg/ml. Green extract was least effective, with no significant decrease in cell proliferation until 3.0-3.5 mg/ml. To determine effect on vascular cell migration and proliferation a scratch wound assay was utilized with A7r5 cells. Results showed a dose dependent decrease in cell migration after 5 and 10 days exposure.

### **Identification of Anthocyanidins in grape derived products by HPLC/UV/MSD**

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A new high-performance liquid chromatography/UV/electrospray ionization-mass spectrometric detector (HPLC/UV/ESI-MSD) method for the simultaneous identification of various anthocyanins in grape derived products was developed. Twenty anthocyanins, including glycosides and the acetyl and coumaroyl derivatives, were individually analyzed and identified by their molecular ions and characterized fragment ions and in comparison with commercial standards. The quantification of anthocyanidins was achieved by an acid assisted hydrolysis method, which was developed and optimized to simplify the various anthocyanin profiles and to degrade the anthocyanins to five major anthocyanidin aglycones of delphinidin, petunidin, cyanidin, malvidin and peonidin. The hydrolysis method provided an accurate, precise and efficient way to quantify the grape derived products with aglycone standards.

### **Thyme extract enrichment inhibits mold growth, extends shelf life and improves sensory flavor attributes of naturally fermented ethnic Ethiopian teff enjera**

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Hydroalcoholic extract (1:5) of thyme (*Thymus vulgaris*, cv. 'Laval') and common oregano (*Origanum vulgare*) leaf was blended between 15 and 25 mL) in self fermented teff (gluten free ancient grain from Ethiopian highlands being introduced to Europe, America, and Asia as a non obesity causing endurance nutrient) flour dough and compared with the control. Thyme and oregano extracts are traditionally incorporated in many

Ethiopian households in various food preparations to impart flavor, ensure safety, and extend shelf life. Total leaf extract-enrichment resulted in a clearly visible reduction on subsequent growth of mold and spoilage of enjera at room temperature. Total mold growth was inhibited by up to 99% at 25 mL extract, while this value for 15 mL was 88 % when compared with the control. At higher concentration, mold growth was significantly suppressed for five hot/humid summer days of room storage. Furthermore, incorporation of thyme oregano leaf extracts maintained the original enjera color, while the sensory attributes of the treatment was slightly enhanced and was acceptable by untrained new sensory tasters. Our study and data presented in this work suggest that the use of selected thyme-oregano whole leaf extract is an adoptive traditional knowledge, innovative, and useful tool as an alternative to the use of antibiotics, synthetic fungicides or other sanitation techniques in processing, storage or packaging of enjera. Thyme-oregano whole extract may reduce disease development with the potential to limiting the spread of pathogens by lowering the spore production in storages, transits, atmospheres while becoming an alternative natural food preservative that could be developed for control release bioactive packaging. However, for modern food industry the dependence of effectiveness on chemotype choice, target pathogen, the processing without affecting the sensory quality deserve further research.

### Quality Control

#### Physicochemical properties of shea butter from Ghana

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Shea Butter is a plant fat extracted from shea nut, a seed of shea tree (*Vitellaria paradoxa*) native to West African countries among which Ghana is one of the most important exporters of shea nuts along with Burkina Faso. In order to characterize Ghanaian shea butter, physicochemical properties important for quality control were analyzed on six Ghanaian shea butter samples. Density was measured as an average of 0.91; refractive index was measured as 1.4641 and 1.4640 in raw and filtered samples, respectively. Melting point was measured in three ways that clear, liquefying, and dropping point were measured as 49.7°, 30.45° and 31.86°, each. Moisture content and the amount of insoluble impurities were measured as 0.025% and 0.13%, respectively. The properties related to oxidation, free fatty acids and peroxide value, were measured as 4.32 % and 7.6 meq/kg, respectively. Shea butter samples from Ghana were found to consist primarily of four fatty acids: palmitic acid (3.82 %), stearic acid (42.89 %), oleic acid (42.28 %), and linoleic acid (5.99 %). The sum of the four fatty acids averaged 94.88 %. This work is part of a project aimed in developing quality control standards for in-country processed shea so that Ghana could capture value-addition rather than only serving as an exporter of the dried shea nut.

## The chemistry and quality of the Ashanti pepper (*Piper guineense*) from Liberia

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The West African black pepper (*Piper guineense*, Family: Piperaceae) is also known as Ashanti pepper and is closely related to traditional black/white pepper (*Piper nigrum*) of international commerce. The Ashanti pepper is a climbing vine commonly grown in the swamp forest areas of West Africa. The Ashanti pepper contains essential oils and the pungent principle piperine in high amounts (5-8%) and is an ingredient of many medicinal preparations. The objective of this work was to initiate studies on the chemistry and quality of the Ashanti pepper from Liberia, and to assess the whether the 'local Liberian Ashanti pepper' is or could meet international quality. As local farmers are starting to produce this unique spice for income generation activities a key question was first to assess the base-line quality to determine whether it would be appropriate for local, regional or international markets. Three Liberian samples were used in this study; the color of the seeds was dark brown and the seeds highly aromatic. Each of the samples showed accepted quality from seed size, shape, color and aroma. Each of the samples also showed low levels of moisture percent (<10%). However, samples 1 and 2 showed higher levels of foreign materials (>1%) and high levels of fine particles (~1%). The total ashes varied from 5.5 to 7%, while the acid insoluble ashes showed low contamination with sand and earth. We conclude that the local Liberian Ashanti pepper can be of international quality if strong and robust QC programs are implemented to ensure consistently clean and a well-defined products. These initial quality characters and results can serve to aid in the training of local Liberian producers and agents to achieve a higher quality and cleaner product using low cost affordable technologies that will allow meeting international specifications.

### Sustainability and Economics

#### Sustainable Production of Specialty Horticultural Crops in Ghana for Income Generation and Increased Export Value

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In Ghana, one of the major gaps in the commodity chain for horticultural crops includes the availability and quality of the product that is in market demand, establishing sustainable systems of collection and production, providing back-up technical support services, access to high quality germplasm, and ensuring close communication between the myriad of actors along the commodity chain. The objective of this project is to strengthen those weaker areas or gaps in the commodity chain of indigenous horticultural crops while

strengthening the overall product supply and quality and ensuring profitability at the rural community level. In the first six months of this new project which began in February, 2010, under the UC-Davis led International Horticultural CRSP, we have mobilized 200 collectors whom earned \$675,000 by collecting and processing *Griffonia* seeds, the source of 5-hydroxytryptophane, and who are following the WHO Guidelines on Good Agricultural and Collection Practices (2004). In addition, this project is already visibly improving rural livelihoods in the selected communities by way of increased income earnings, higher standard of living and increased property ownership. Increasing community awareness as to the value of their own natural indigenous plant products as a sustainable and viable activity capable of addressing the poor household economic situation in the six districts. This project builds upon our earlier nontimber forest products programs which brought together the collectors, researchers and industry players and with an applied quality control based program strengthened the natural products sector in Ghana under the USAID funded Partnership in Food Industry Development for Natural Products.

### **Evaluation of amino acids foliar application on essential oil variations in flaxweed (*Descurainia sophia* L.) under drought stress conditions**

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This experiment was carried out using by a split-plot design with four replications to determination of amino acids foliar application on essential oil variations in flaxweed (*Descurainia sophia* L.) under drought stress conditions at Iran in 2010. The factors including irrigation regimes (irrigation interrupted from flowering stage, irrigation interrupted from silique formation stage and irrigation interrupted from seed filling stage) in main plots and commercial amino acids (Aminol-Forte, Hyomi-Forte, Kadostim and Phosnotron) that sprayed in three stages (25, 50 and 75 days after germination) in subplots were studied. Our data showed that amino acids foliar application significantly affected essential oil yield, essential oil percentage, grain yield, thousand grain weight, silique number per plant and grain number per silique in  $P \leq 0.01$ . The results of this experiment showed that the highest essential oil yield and other characteristics were achieved by Kadostim under irrigation interrupted from seed filling stage, while the highest essential oil percentage was obtained by Hyomi-Forte under irrigation interrupted from flowering stage. The findings indicated that drought stress was reduced the essential oil yield of flaxweed but essential oil percentage was increased under this conditions. Also, amino acids foliar application was reduced the drought stress damages.

## **Sustainable Development of Horticultural Crops in Zambia for Food Security, Income Generation and in Support of the Tourism Industry**

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The potential benefits of horticulture for the developing world are numerous. Economic growth in horticultural products has far exceeded the growth of other agricultural commodities, and the demand for horticultural produce continues to accelerate in both domestic and international markets. Yet, Africa is the one region of the world where per capita supply of fresh produce has seen virtually no increase over the past 40 years. The goals of this new project which began in February, 2010, under the UC-Davis led International Horticultural CRSP, are to increase food security and generate income for rural farmers in new communities that had not previously participated in commercial horticultural production. We are conducting research on several horticultural technologies all (e.g. greenhouse production, trickle irrigation, high quality seeds, post-harvest handling, scheduling production to meet market requirements) of which are being transferred to the growers. This project is also focusing on entrepreneurship capacity building, to assist farmers to develop crop budgets for selected vegetables, proper invoicing. This project is implementing a holistic, and comprehensive market-first science-driven model that uses a participatory approach and community and private sector partnerships that cover the full commodity chain (seed, production, post-harvest handling, quality, marketing and business skills) into communities which we have not previously been engaged and that lacked experience in growing commercially or for household consumption horticultural crops. This project builds upon our earlier programs in southern Africa which brought together small-scale-growers, researchers and buyers with an applied quality control based program to locally produce and market fresh produce. This program has been funded regionally by the USAID Regional Center for Southern Africa, and in the past with support under our USAID funded Partnership in Food Industry Development for Natural Products and HED on Non-Traditional Germplasm for Southern Africa. Working with the local hotels as our private sector partners that are buying the local product, the original project has generated over \$1,000,000 in sales to new small-scale growers in southern Zambia in the Livingstone area and offers an excellent model for expansion and replication.

### **Routes of Market Access for Natural Products in the European Union: An overview**

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Natural (herbal) products can enter the European Union in various ways and categories, ranging from herbal medicines, supplements, foods and food ingredients all the way to cosmetics and cosmetic ingredients. The European Union is in the process of harmonizing the regulatory framework for each of those categories. Individual routes of access will be described and discussed with regard to their relevant regulatory requirements. Focus will be put on market authorizations of herbal medicines: for full drug status, as bibliographic registrations for well-established use and under the framework provided in the Traditional Herbal Medicinal Products directive. Information on various resources, pharmacopoeial and non-pharmacopoeial, will be provided. Market opportunities will be highlighted as well as the ubiquitous regulatory hoops and hurdles.