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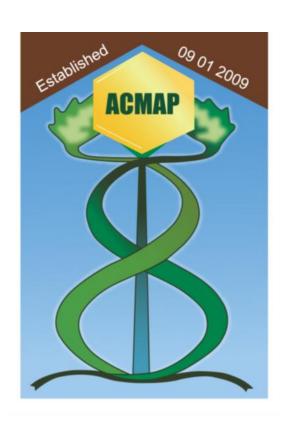
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4th Annual Conference American Council for Medicinally Active Plants University of Massachusetts, Amherst, MA, USA June 2-5, 2013



Welcome and Greetings

Room 163C, Campus Center

Lyle Craker, Program Chair and Host, University of Massachusetts, Amherst, MA Steve Goodwin, Dean, College of Natural Sciences, University of Massachusetts, Amherst, MA Agnes Rimando, President, Board of Directors, ACMAP

Plenary Session

Dr. Kalidas Shetty

Associate Vice President for Global Outreach
Professor of Plant Sciences
Plant Metabolism and Food Security
North Dakota State University
Fargo, ND

Dr. Shetty's research interests focus on the critical role of cellular and metabolic basis of oxygen biology for advancing new innovations in life sciences, especially agricultural and food innovations that advance global food security and health in a sustainable environment. This focus is contributing to very innovative advances in the areas of crops for health, post-harvest biology, nutraceuticals, functional foods, and food antimicrobial strategies as critical innovations to advance food security and attack malnutrition and hunger challenges. In particular, he has developed an innovative "crops for health" research platform to counter diet-linked chronic diseases. The rationale of this platform has relevance for managing malnutrition challenges from maternal to child health needs globally.

Dr. Shetty's current passion and commitment are to develop "sustainable and ecological basis for healthy food systems, food choices, and global food security" based on local crop and food diversity, traditional food systems, and effective food imports that incorporate an understanding of comparative cellular biochemistry of plant and animal systems and their interactions with prokaryotes (bacterial systems). This approach has potential to transform global agriculture with healthy food systems and innovative rural development strategies that are systems based with sustainable long term solutions combined with better habitat and environment, sanitation and diversified bioenergy options.

In keeping with the above focus on new innovations to advance global food security on a "crops for health and sustainability" platform, Dr. Shetty is developing close collaborations between North Dakota State University and institutions in several regions of the world from Asia, Africa, and Europe to the Americas.

Education: PhD 1989, Microbiology, University of Idaho, Moscow, ID
MS 1985, Bacteriology, University of Idaho, Moscow, ID
BS 1983, Agricultural Science, University of Agricultural Sciences, Bangalore, India

MONDAY

Panel Discussion on Bioproduction

Chair: Fabricio Medina-Bolivar (Arkansas State University)

Susan Roberts (University of Massachusetts) Culture-Based Bioproduction

Susan Roberts, Director, Institute for Cellular Engineering, University of Massachusetts, Amherst, MA

This panel session will highlight advances in producing medicinally active compounds using plant suspension and organ cultures including alkaloids, taxanes, and artemisinin. Topics to be discussed include bioprocess engineering and reactor design, cellular aggregation, yield enhancement via environmental manipulations, gene transformation methods, and metabolic pathway engineering strategies.

Pamela J. Weathers (Worcester Polytechnic Institute) Production from Differentiated Plant Cultures: the Tough Stuff to Scale-up!

<u>Pamela J. Weathers</u>, Dept. Biology Biotechnology, Worcester Polytechnic Institute, Worcester, MA 01609. E-mail: <u>weathers@wpi.edu</u>

Plants are a rich reservoir of valuable metabolites that if properly harnessed could tremendously benefit human and animals. Although the most cost effective method of production is nearly always from field crops, use of bioreactors offer well known significant benefits including year round controlled production. However, cultivation of plant organs and tissues beyond cells suspensions offers particular challenges. Here a short review of the different types of plant tissues and organs that have been considered for cultivation using bioreactors will be provided along with some pros and cons of some of the progress being made towards their scale-up.

Carolyn Lee-Parsons (Northeastern University)

Transcriptional Regulation of Jasmonate-Induced Biosynthesis of Medicinal Alkaloids from *Catharanthus roseus* Hairy Root Cultures

<u>Carolyn W.T. Lee-Parsons</u>^{1,2}, Erin J. Cram³, Noreen Rizvi¹, Jessica Weaver³ Dept. of Chemical Engineering, ² Dept. of Chemistry and Chemical Biology, ³ Dept. of Biology, Northeastern University, Boston MA 02115; E-mail: ca.lee@neu.edu

The overall vision of our research is to enhance the production of critical plant-derived pharmaceutical compounds through plant cell and tissue cultures, specifically using the production of alkaloids from cultures of the *Catharanthus roseus* plant as a model system. The *C. roseus* plant produces several highly-valued pharmaceuticals, including the anti-cancer drugs vincristine and vinblastine. The high cost (\$4 – 20 million/kg) and need for these anti-cancer compounds motivate our research to better understand their biosynthesis and ultimately overproduce these valuable alkaloids economically using cultures of *C. roseus*.

C. roseus produces alkaloids in response to environmental factors (i.e. UV light, wounding, etc.) and as defense chemicals against pathogens and insects. For example, when cells perceive the presence of pathogens in their environment, chemical signals known as jasmonates (JA) are generated, activating signaling cascades in the cell that lead to the expression and activation of molecular switches, known as transcription factors. These transcription factors then coordinately alter the expression of genes, including genes that code for enzymes involved in alkaloid biosynthesis. Application of exogenous JA or its methyl ester, methyl jasmonate, also increased alkaloid production in whole plants and cultures derived from *C. roseus*. The goals of this

research are to investigate and engineer the transcriptional regulation of alkaloid biosynthesis in *C. roseus* hairy root cultures towards further increases in alkaloid production.

Tour – Integrated Science Building

Session 1: Sustainable Production

Chair: Lyle Craker (University of Massachusetts Amherst)

Tim Blakely (Frontier Herbs) Sustainability of Medicinal Herbs and Essential Oils

<u>Tim Blakely</u> Frontier Natural Products Co-op, Norway, IA E-mail: <u>tim.blakley@frontiercoop.com</u>

There are many issues that relate to the sustainability of medicinal plants, from understanding the status of the particular species, harvesting techniques and even production issues. Understanding these issues, and then responding to them in appropriate ways can have a significant long term influence on our industry. United Plant Savers (UpS), in conjunction with the University of Kansas, has developed a tool for determining the At Risk status of a plant species. Using this tool we can determine whether a species is most likely threatened or not. Key species at this time include Sandalwood, Santalum species, Rosewood, Aniba roseaodora, Jatamansi, Nardostachys jatamansi, Black Cohosh, Actaea racemosa, Atlas Cedarwood, Cedrus atlantica, American Ginseng, Panax quinquefolius, and Goldenseal, Hydrastis Canadensis. Sandalwood, of which there are 17 species, appears to be especially threatened at this time and adulteration of sandalwood products worldwide is quite common. One particular species that appears of immediate concern is Santalum paniculatum, native to the Big Island of Hawaii. After not being harvested for 180 years, harvesting commenced recently in what appears to be the single largest concentration of Sandalwood left in the world. No population studies have been done and the forest is being harvested at a rapid pace. In running Sandalwood on the At Risk Tool it scored the highest of any species so far. United Plant Savers proposed it for CITES status as well as promoting a bill in the state legislature to create some sort of bill that would promote a population study to determine how much sandalwood exists as well as creating a management plan for the remaining Sandalwood. At present the United States is the only sandalwood producing country in the world that does not have a management plan.

Jean Giblette (High Falls Garden) Chinese Medicinal Herbs as an Organizing Principle in Local Agriculture

Jean Giblette, High Falls Gardens, P.O. Box 125, Philmont NY 12565, info@highfallsgardens.net

Chinese herbal medicine is based on a philosophy, model of health, methods of diagnosis and treatment distinctly different from western medicine. Because the herbs are accepted as agricultural rather than manufactured products, the locus of quality is the agricultural setting or landscape. Because the herbs are dispensed in empirically tested combinations or formulas, a poll of practitioners can be used as data to forecast agricultural production needs. Because practitioners, growers and patients can cooperate and exchange services, local economic gain can be realized even in the absence of outside funding. Licensed Chinese herbal practitioners have become established and distributed throughout the U.S. and are now realizing that they must act to ensure their future access to high-quality herbs. Therefore, despite obstacles, ecological domestic production is proceeding. A model for production and development in the U.S. is outlined, based on recent examples of local cooperation. Concurrent developments in China are described, showing that economic hardship and climate change are prompting a worldwide recognition of the importance of ecological agriculture. Included in this revaluation, highly

distributed local production of Chinese medicinal herbs can have a role in building resilience in the landscape as well as safeguarding public health.



Session 2: Artemisia

Chair: Jorge Ferreira (USDA-ARS)

Co-Chair: Fabricio Medina-Bolivar (Arkansas State University)

Jorge Ferreira (USDA-ARS)

Cultivar and Dihydroartemisinic Acid Levels may Affect Artemisinin and Antioxidant Capacity of *Artemisia annua* in Response to Antibiotic Stresses

<u>Jorge F.S. Ferreira</u>¹, ¹US Salinity Laboratory, 450 W. Big Springs Rd., Riverside, CA 92507 E-mail: jorge.ferreira@ars.usda.gov

The worldwide demand of artemisinin continues to increase due to to its role in treating multidrug resistant malaria. Artemisia annua is the only commercial source of artemisinin, and a higher artemisinin production per cultivated area would bring a considerable decrease in artemisinin-based drugs. Our past research showed that abiotic stresses such as drought and potassium deficiency were linked to a significant increase in artemisinin in A. annua, while others showed the same trend for salinity stress. Recently, the leaf flavonoid antioxidants have been suggested to have a synergistic effect on the pharmacological effect of artemisinin. Our research shows that drying procedures can increase artemisinin significantly, compared to freeze dried samples, while potassium-deficient soil increased artemisinin in 75% (w/w) by promoting the conversion of dihydroartemisinic acid (DHAA) into artemisinin. Also, appropriate drying can preserve most of the antioxidant activity in the leaves. Thus, a combination of abiotic stresses and the right drying procedure can lead to a significant increase in artemisinin, through a reaction between DHAA and reactive oxygen species (ROS), while preserving most of the plant antioxidant activity. Our research showed that leaf extracts, rich in both artemisinin and antioxidants, have proved beneficial in curbing parasitic infection in livestock, and also had anticancer effect in vitro. Learning how abiotic stresses increase bioactive plant secondary metabolites can lead to understanding how these compounds are involved in protecting plants against stress, while harnessing these plants to become more effective in preserving health and neutralizing excessive ROS in livestock and humans afflicted by parasitic diseases.

Pamela J. Weathers (Worcester Polytechnic Institute) Developing a Whole Plant *Artemisia annua* Antimalarial Therapeutic, pACT

<u>Pamela J. Weathers</u>, Professor of Biology & Biotechnology and Professor of Chemistry & Biochemistry, Worcester Polytechnic Institute, BB Department at Gateway, Worcester, MA 01609

Artemisia annua produces artemisinin (AN). The current therapy for malaria, is artemisinin + an older drug: artemisinin combination therapy (ACT). In Plasmodium chabaudi-infected mice, dried leaves of A. annua are more potent than equal amounts of pure AN. Pharmacokinetics also show that more AN enters mouse blood when plant material is present. Dried leaf tablets given to Kenyan malaria patients were also efficacious. This whole plant therapy is termed pACT: plant-based artemisinin combination therapy. Flavonoids and possibly other compounds in the plant may affect its efficacy when delivered as pACT, so we conducted an ex vitro simulated digestion study to measure the effects of diet and conditions affecting palatability on AN and flavonoid release into the intestine. We compared two types of capsules, sucrose, oil, and 3 staple grains (rice, corn, and millet). Compared to pACT alone, both sucrose and oil enhanced release of flavonoids by 100%, but did not affect release of AN. In contrast the 2 capsule types, corn and millet meal, all significantly reduced the amount of AN, but had no effect on flavonoids. Together these data show that diet plays an important role in developing the use of pACT as an orally delivered therapeutic for treating malaria and other artemisinin susceptible diseases.

Stephen M. Rich (University of Massachusetts) Whole plant *Artemisia annua* Abrogates Malaria Drug Resistance

Mostafa A. Elfawal^a, Melissa J. Towler^b, Nicholas G. Reich^c, Douglas Golenbock^d, Pamela J. Weathers^b, Stephen M. Rich^a,

^aLaboratory of Medical Zoology, Department of Microbiology, University of Massachusetts, Amherst, MA 01003; ^bDepartment of Biology and Biotechnology, Worcester Polytechnic Institute, Worcester MA 01609; ^c Division of Biostatistics and Epidemiology, School of Public Health and Health Sciences, University of Massachusetts, Amherst, MA 01003; ^d Infectious Diseases and Immunology, University of Massachusetts Medical School, Worcester, MA 01605.

The fight against malaria predates the discovery of its causative agent. For centuries malaria-associated fever was treated using herbal remedies, such as the Cinchona bark and the annual wormwood Artemisia annua. In the west, quinine (Cinchona bark extract) was the only affordable treatment against malaria until thousands of synthetic compounds were tested against malaria parasites. In the 1930s chloroquine rose to ascendancy as a miracle cure for malaria. In the late 1950s, chloroquine was the main weapon when the World Health Organization launched the Global Malaria Eradication Program (GMEP). Irresponsible use of the drug, however, led to development of drug resistant parasites and concomitant failure of chloroquine as the drug of choice and the eventual demise of GMEP by the close of the 1960s. In the 1970s, artemisinin (AN) was isolated as a pure drug extract from A. annua. AN has potent antimalarial activity, but poor pharmacokinetic properties and hence is most effective when used in combinations which became known as Artemisinin Combination Therapies (ACT). In 2012, we demonstrated that Whole Plant (WP) A. annua is more effective than a comparable dose of pure AN in a rodent malaria model. This finding facilitates greater availability and affordability of an antimalarial WP appears more effective than AN because WP constitutes a naturally occurring combination therapy. A plant Artemisinin Combination Therapy (pACT) would be a refinement to incorporate secondary metabolic products into a resilient defense system. We hypothesize the WP-based pACT may: (1) overcome existing resistance to pure AN, and (2) increase the longevity of this therapy by delaying the onset of parasite resistance among wild types. We tested these two hypotheses in the mouse model, AN resistant Plasmodium yoelii (ART) and AN sensitive *Plasmodium chabudi chabaudi* (ASS).



Session 3: Bioactives, Functional Foods and Human Health

Chair: Jeffrey W. Adelberg (Clemson University)

Trygve Tollefsbol (University of Alabama-Birmingham) The Epigenetics Diet: Can it Slow Aging and Cancer development

Trygve Tollefsbol, Biology Department, University of Alabama at Birmingham, Birmingham, AL

The single most important risk factor for developing cancer is age; therefore, dietary approaches that influence both cancer and aging are of intense interest. A major molecular process that is influenced by dietary factors is epigenetics that encompasses the changes in phenotype due to molecular changes that arise independent of primary DNA sequence. Key genes (such as telomerase that plays major roles in both aging and cancer and maintains the telomeric ends of chromosomes) are controlled through epigenetic mechanisms and are influenced by dietary changes. We have found that widely consumed dietary components such as sulforaphane (SFN) from cruciferous vegetables and epigallocatechin-3-gallate (EGCG) from green tea inhibit telomerase gene (hTERT) activity in breast cancer cells through epigenetic mechanisms such as DNA methylation and histone modification. The telomerase gene inhibition leads to specific impairment of cancer cell growth which may have major implications in cancer prevention through epigenetic effects of dietary compounds. The amount of SFN needed for these effects is equivalent to about one cup of broccoli sprouts a day. Our recent studies also indicate that the quantity of nutrients provided to a human cell can influence its lifespan and cancerous potential through epigenetic effects on genes such as telomerase and p16, a well known anti-cancer protein. These studies show for the first time that reduced sugar in the form of glucose provided to normal human cells in vitro extends their lifespan but leads to cell death of precancerous cells. In normal human lung cells that received less glucose, telomerase was increased and p16 was decreased through epigenetic effects which allowed the normal cells to live longer. Opposite effects were found for these two genes in precancerous cells which may explain why decreased glucose results in impaired growth and eventual death of these precancerous cells. This novel model for assessing the impact of nutrient quantities on specific human cells may have a major impact on our understanding of the mechanisms for dietary effects on human aging and cancer at the molecular and cellular levels. Collectively, these studies indicate that a diet rich in cruciferous vegetables may confer many health benefits as well as preventing cancer and that reducing sugar in the diet could increase the healthy lifespan and also contribute to cancer prevention through epigenetic effects.

Sallie Schneider (University of Massachusetts) Rhodiola: A Natural Immunomodulator & Regulator

Sallie Schneider, ,Institute for Cellular Engineering, University of Massachusetts, Amherst, MA

Rhodiola is a high altitude perennial plant that has been used for many years in eastern traditional medicine. It is considered a plant-derived "adaptogen", which is capable of maintaining physiological homeostasis upon exposure to stress. We have been examining the extract of Rhodiola crenulata for its potential use to prevent or treat cancer, as well as improve glucose homeostasis on a high fat diet. Our studies have identified several key pathways and cell types that are altered by treatment with Rhodiola, which likely explain these protective effects. They include signaling pathways that will affect stem cell maintenance, cellular migration, bioenergetics and immunosurveillance. Studies are underway, to identify the active components with in the extract responsible for these effects.

Anait Levenson (University of Mississippi Medical Center) Can Dietary Stilbenes Prevent Prostate Cancer Progression and Metastasis?

Swati Dhar¹, Kun Li¹, Steven J Dias¹, Liangfen Zhang¹, Agnes M Rimando³ and <u>Anait S Levenson</u>^{1,2. 1}Cancer Institute, ²Department of Pathology, University of Mississippi Medical Center, Jackson, MS 39216; ³ Natural Products Utilization Unit, Agricultural Research Services, US Department of Agriculture and University of Mississippi, Oxford, MS 38677. E-mail: alevenson@umc.edu

Dietary stilbenes with anticancer properties are of great interest as potentially safe chemopreventive and therapeutic agents. We reported earlier that resveratrol (Res) and its analogues acted as HDAC inhibitors by hindering the activity of MTA1/HDAC/NuRD corepressor complex and causing apoptosis in prostate cancer (PCa) cells in vitro. Here, we compared the in vivo efficacy of Res and its natural analogues, pterostilbene (PTER), trimethoxy-resveratrol (3M-Res) and picetannol (PIC) in various PCa xenografts models. Our studies demonstrated that Res and more so PTER inhibited the MTA1-mediated growth, progression and metastasis of orthotopically implanted tumor cells in nude mice. MTA1knockdown tumors treated with stilbenes showed reduced mitotic activity (Ki67) and angiogenesis (CD31) while exhibiting significantly increased levels of Ac-p53 and apoptosis (M30) compared to vehicle-treated EV-tumors. Res and PTER inhibited metastasis developed in kidney, liver and lungs/heart in EV-control group, and MTA1-knockdown tumors developed lesser metastasis and in a fewer organs, indicating MTA1 contribution to invasion and metastasis. Further, since several lines of evidence suggest cooperation of MTA1 overexpression with Pten loss for development of advanced PCa, we initiated preclinical studies on the chemopreventive and therapeutic efficacy of stilbenes using autochthonous prostate-specific Pten heterozygous (Pten+/-) and homozygous (Pten -/-) mice models. In summary, we propose that dietary stilbenes are potent and effective regulators of MTA1/NuRD complex holding great promise as target-specific epigenetic therapeutic agents for advanced PCa.

Session 4: Bioactives, Functional Foods and Technology

Chair: Alaa Helaly Co-Chair: James Simon

Fabricio Medina-Bolivar (Arkansas State University) Biotechnological Approaches to Discover and Produce Health Beneficial Compounds from Hairy Roots.

<u>Fabricio Medina-Bolivar</u>¹, Anna Radominska-Pandya², Stacie Bratton², Kyle Smith³, Anastasios Karydis³, Franklin Garcia-Godoy³, Luis Nopo-Olazabal¹, Linda Ogutu¹, Tianhong Yang¹, Zachary Marsh¹, Nirmal Joshee⁴, Ananddeep Patel¹, Christopher A. Tollett¹, Cesar Nopo-Olazabal¹. ¹Arkansas State University, ²University of Arkansas for Medical Sciences, ³University of Tennessee Health Science Center; ⁴Fort Valley State University. E-mail: fmedinabolivar@astate.edu

Our group has developed hairy root cultures of peanut and Scutellaria lateriflora to study the biosynthesis of selective stilbenoids and flavonoids and elucidate their biological properties. In order to increase their levels in these plant tissue culture systems, biotransformation, elicitation and metabolic engineering strategies were investigated. The treatment of hairy root cultures with the elicitors methyl jasmonate with cyclodextrin was the most effective to induce the biosynthesis of the arachidin-1 and arachidin-3 in peanut. The accumulation of these stilbenoids was increased several fold when the elicitors were combined with metabolic precursors. S. lateriflora hairy roots produced several of the known anticancer flavonoids including wogonin and baicalein. Interestingly, upon treatment with methyl jasmonate with cyclodextrin several putative phenolics were induced and accumulated in the culture medium. The stilbenoids were extracted from the culture medium of the peanut hairy root cultures with ethyl acetate and purified by high performance counter current chromatography. The cytotoxicity of the purified stilbenoids was studied in breast, pancreatic and oral cancer cell lines. We demonstrated that the hairy root cultures and these bioproduction strategies provide a valuable tool for increasing the levels of bioactive compounds in hairy roots and discovering plant-derived compounds with potential applications in human health.

Oliver Chen (Human Research Center on Aging, Tufts University) The Approaches to Evaluating Antioxidant Capacity of Bioactive Plants.

Oliver Chen, Antioxidants Resaerch Laboratory, Jean Mayer UDA HNRCA, Tufts University, Boston, MA

Reactive oxidant species (ROS) with unpaired electrons contribute to development and progression of many diseases, including cancer, metabolic diseases, cognition decline, cardiovascular diseases, neurodegenerative diseases (e.g. Alzheimer's disease, Parkinson's disease, multiple sclerosis, and Downs syndrome) and aging. Antioxidants present in plants are a molecule capable of scavenging ROS, inhibiting oxidation of labile compounds or modulating endogenous antioxidant defenses. Thus, antioxidants through either oral consumption or topical application appear to be an appealing approach to decreasing their risk of the diseases. Thus, characterization of antioxidant constituents in plant related products will help inform consumers in selection of products and manufacturers in formulating products with high antioxidant contents. However, quantification of all antioxidants in a plant is not a straightforward, easy task as a great diverse group of antioxidants is present in plants. The challenge is further aggravated by the fact that antioxidants display a wide range of reactivity toward ROS. In order to overcome this complexity, an array of antioxidant capacity assays with different principles and settings, e.g., ORAC, FRAP, DPPH, LDL oxidation, has been developed to determine antioxidant capacity of plants or their parts as a whole, with values being a sum of action of individual constituents and antagonistic and synergistic interactions between constituents. In this presentation, pros and cons of in vitro assays that evaluate antioxidant capacity is discussed.

Sudhersan Chellan (Kuwait Institute for Scientific Research) Development of a Simple Protocol and Culture Media for the Micropropagation of Medicinal Plants.

<u>C. Sudhersan, Environment and Life Science Center, Kuwait Institute for Scientific Research, P.O.Box 24885, Safat 13109, Kuwait. Email: schellan@kisr.edu.kw/sudher_san@yahoo.com</u>

Majority of the life saving drugs are produced from herbs. Herbal medicine given importance during the ancient time was neglected later on due to the development of synthetic drugs. Several medicinal plants were extinct due to the negligence of herbal medicine. Recently, once again medicinal plants are given importance after the realization of the side effects of synthetic drugs. Therefore, pharmaceutical industries started producing life saving drugs from herbs. This situation created demand for medicinal herbs. Sudden demand of medicinal herbs for the commercial drug production caused threatened nature of several medicinal plants due to over harvest. Adulteration in herbs has also been practiced by the suppliers to the pharmaceutical industries. In order to meet the demand for high quality medicinal plants without any adulteration and to protect the medicinal plants from extinction, biotechnological tool plant tissue culture was given importance worldwide. Several complicated culture media and different protocols were reported in the literature for medicinal plant clonal micropropagation which caused medicinal plant production via micropropagation technique difficult and more expensive. Therefore, a study was undertaken in our laboratory to develop a simple media and protocol common to several medicinal plants to make much easier for the medicinal plant propagators via tissue culture method. Through experimental trials using several medicinal plants, a simple culture media and protocol has been developed, and experimented with 15 medicinal plants successfully. This simple system will provide the basis for the future medicinal plant production much easier for the drug industry and also for the conservation purpose.

Tuesday Sessions

Session 5: Sourcing Medicinal Plant Material

Chair: Dipayan Sarkar

Zoe Gardner (Traditional Medicinals) Adding Quality and Value to the Botanical Supply Chain

Zoe Gardner, Research & Development Manager, Traditional Medicinals, Sebastopol, CA

Beyond the basic measures of good agricultural and collection practices, a number of tools may be used to add quality to the supply chain for medicinal plants. Social and ecological sustainability standards such as FairTrade and FairWild can add value not only to ingredients but also the quality of life for the people cultivating or collecting the plants.

Chris Kilham (Medicine Hunter/University of Massachusetts) Medicine Hunt-The Search for Cures

<u>Chris Kilham</u>, Medicine Hunter, Medicine Hunter, Inc., Levertt, MA & Stockbridge School of Agriculture, University of MA, Amherst, MA

No Abstract

Wudeneh Letchamo (Rutgers University) Omega-3 rich *Camelina sativa* with Erucic Acid under 1% has been Developed for Ethiopian Health Nutrition and Other Commercial Applications

<u>Wudeneh Letchamo</u>¹, Thomas Hartman¹, and André Gosselin²

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Camelina sativa (L.), Crantz from Brassicaceae family is an ancient, but a newly emerging multi-purpose crop attracting attention from various industries. Camelina is not known in Africa in cultivation or in the wild. Due to the high content of omega-3 EFA and phytosterols, the use of cold pressed, unrefined camelina oil as a food ingredient is approved by Health Canada for Canada, European Union in Europe, Russia, and Central Asia. Camelina oil is used in folk medicine for the treatment of burns, wounds, eye, and respiratory inflammations, stomach ulcers, and as a tonic in Central Asia, Eastern Europe, Russia, and Ethiopia. The objective of our program is to identify, develop and introduce high yielding superior camelina cvs with desirable chemical composition of oil for Ethiopia. We designed and conducted chemical analyses of the oil, and meal using TD P&T, HS, GC/MS, shelf stability, aroma/flavor, and taste tests. The results revealed variations in the content of omega-3 fatty acid (-linolenic acid), erucic acid, glucosinolates, shelf stability, and flavor. Two most promising cvs with less than 1% of erucic acid were further developed with subsequent breeding and field introductory program in Ethiopia for the 1st time (2006 to present) were conducted in multiple sites, during dry and rainy seasons, with the collaboration of local community producers. Seed yields with 34% - 41% oil varied from 2400 to 4000 kg/ha depending on location and growing season. The plant height varied from 48 to 56 cm, with 99d to 127d from seed emergence to ripeness. In temperate zone, camelina matures in 85 to 100d. Currently, in Ethiopia, breeders' rights and variety registration are not recognized for camelina cultivars. Hence, camelina cvs from our research program have been available for free to farming graduates of food insecurity households, companies, growers associations, EARI, and others.

Session 6: Medicinal Plants and Health

Chair: Lorraine Cordero (University of Massachusetts Amherst)

Dayong Wu (Tufts University) Mushrooms, Immune Function, and Host Resistance to Microbial Infection

Nutritional Immunology Laboratory, Jean Mayer USDA Human Nutrition Research Center on Aging at Tufts University, Boston, MA.

Mushrooms are suggested to positively impact multiple bodily functions including immune response. Several strains of mushrooms have been shown to favorably modulate immune cell function, which is suggested as underlying mushrooms' anti-tumor, anti-viral, and anti-bacterial properties. A majority of these studies evaluate the effect of administering extracts of exotic mushrooms through parenteral routes or in the cell-based studies. In contrast, less is known about white button mushrooms (WBM), a strain predominantly consumed in the US and other Western countries, but evidence has been accumulating about their immuno-modulating effects. We have conducted a series of studies to determine the effect of white button mushrooms on a host's immune response and resistance to infection. Our results indicate that dietary WBM enhance the activity of natural killer (NK) cells, an important component in innate immune system responsible for anti-viral and anti-tumor defense. Our results also suggest that WBM may

promote maturation and function of dendritic cells (DC), the most important antigen-presenting cells that link innate to adaptive immunity. Next, we used animal models to determine whether WBM-induced changes in immune function have any clinical relevance toward improving the defense against microbial infection. We found that although consumption of WBM enhances NK activity, it does not provide the host adequate protection from a primary influenza infection. Similarly, WBM consumption does not improve gastroenteritis and systemic (typhoid) salmonellosis after a primary infection with a virulent strain (SL1344) of Salmonella typhimurium. However, when the mice were immunized with an attenuated strain (SL1479 Δaro A) of Salmonella and then infected with the virulent strain, the vaccination-induced protection (lower weight loss and higher survival rate) was further enhanced by WBM supplementation. Consistent with these results, the mice fed WBM had higher levels of antigen-specific antibody (both serum IgG and fecal IgA) and higher antigen-specific CD4⁺ T cell subset Th1 and Th17 response, compared to the mice fed the control diet. We also found that WBM-fed mice had upregulated numbers and function of DC, which may contribute to the improved efficacy of the Salmonella vaccine. These results suggest that WBM may potentially enhance pathogen exposure-induced adaptive immunity and thus reduce the risk of the future infection. The work was supported by the Mushroom Council and mushroom industry funding partners, and the USDA contract #58-1950-0-014.

Shalesh Kaushal (University of Massachusetts Medical School) Nutritional Modulation of the Retina.

Shalesh Kaushal. Retina Speciality Institute, Gainesville, FL 32653. E-mail: skaushal108@gmail.com

The neurosensory retina is a thin diaphanous structure, no thicker than tissue paper attached to the inner wall of the back of the eye. It serves as the photographic film of the eye and is a laminated integrated circuit that subserves all aspects of visual perception including high resolution central acuity, motion detection, edge identification, contrast sensitivity, color identification and peripheral vision. Accordingly, the retina is the most metabolically active tissue in the body per unit weight and the eye has the largest blood supply per unit weight in the human body. Given the unusual metabolic demands of retina, it's easy to imagine the role of nutrition and nutritional supplements and retinal health. In this presentation, the biology of retinal activity will be briefly reviewed. Additionally, the critical, canonical pathogenetic events that cause retina diseases like diabetic retinopathy and age-related macular degeneration (AMD) including oxidative stress, inflammation, microglial activation and cell death will also be reviewed and studies presented that demonstrate how they can be mitigated by nutrition and nutritional supplements.

Mohammed Kamil (Zayed Complex for Herbal Research and Traditional Medicine-UAE) Novel Natural Products from Medicinally Active Plants and Their Biological Activities in Search of Therapeutic Agents.

Head, CAM Research, Zayed Complex for Herbal Research & Traditional Medicine, DPHR, HAAD

In the last few decades the Chemistry of Natural Products has touched a new horizon and developed as a separate discipline in itself. The importance of this field is that on one hand it is Organic Chemistry associated with plants, animals and marine products and on the other hand it is associated with plant biochemistry and utility in pharmaceutical industry. The plant kingdom contains many types of organic compounds of different classes. Natural Products in general and flavonoids in particular are a major source of biological activity. The broad spectrum of biological activities of natural products within the group and the multiplicity of actions displayed by certain individual members make the flavonoids one of the most intriguing class of biologically active compounds, termed as biflavonoids. Their occurrence in nature and the chemical & biological relationships between them have made them the subject of advance

researches. The present paper deals with development of many novel & rare bioactive flavonoids & biflavonoids isolated for the first time from plants and their characterization using recent techniques and possible biological implications.

Session 7a: Metabolomics

Chair: Oliver Chen (Tufts University)

Speaker: Edward D. Karoly (Metabolon, Durham, NC) Global Biochemical Profiling for Understanding Biology and Disease

Metabolomics is defined as "the non-biased quantification and identification of all metabolites present in a biological system" but in practice the term metabolomics is used in a rather broad sense and covers many different analytical methodologies. To address the challenges associated with metabolomics, a comprehensive integrated analytical and informatics platform was developed to provide chemo-centric global metabolomics analyses. The analytical platform incorporates two separate UHPLC/MS/MS2 methods and a GC/MS method, which increases the overall coverage of small molecules in a biological sample. The resulting MS and/or MS2 data are then searched against an in-house generated authentic standard library. This library then enables the rapid and accurate identification of the experimentally detected small molecules based on a multi-parameter match without need for time consuming further analysis. Importantly, non matching small molecules are tracked as well for analysis of statistical significance. This integrated platform will be overviewed along with case study examples highlighting the use of these biochemical data sets for identifying sarcosine as a biomarker of prostate cancer aggressivity as well as the mode of action of an anti-cancer drug.

Metabolon was founded in 2000 and has been issued the first pioneering patents covering metabolomic technologies. In addition, Metabolon is the leading research technology company studying metabolomics and has over 300 clients involved in pharmaceutical/biotech research, consumer goods, academic and government research, bioprocessing, and agriculture. Metabolon is based in the Research Triangle Park of North Carolina and has over 110 employees.

Session 7b: Emerging Research

Chair: Agnes Rimando (USDA, Oxford, MS)

Dipayan Sarkar (North Dakota State University) Improvement of Bioactive Functionality in Medicinal and Traditional Food Crops of Northern Plains for Type 2 Diabetes Management

Dipayan Sarkar, Department of Plant Sciences, North Dakota State University, Fargo, ND-58108. E-mail: dipayan.sarkar@ndsu.edu

The occurrence of non-communicable chronic diseases (NCD) including obesity-linked type 2 diabetes is prevalent among indigenous population and minority communities worldwide. To address such health disparities it is important to understand and recognize significant changes, what has happened in socio-economic, and ecological domains in these communities particularly during last two centuries. Changing dietary pattern is most critical factor contributed in the development of NCD epidemic in indigenous populations. Evolution of dietary patterns and food

in different communities and cultures has strong root and association with origin and domestication of plants specific to certain geographical areas. Traditional wisdom, invention and continuous observation guided early humans to adopt and incorporate plant-based food in diet and for other medicinal uses depending on their availability and health benefit properties. Such plant-based diet with rich source of bioactive compounds and plant phytochemicals played most valuable role in providing protection against many diseases in these native communities for centuries. Traditional food crops and plants associated with native communities of the Northern plains were corn, squash, turnip, chokecherries, spearmint, berries, wild rice etc. These plant-based foods are rich source of natural antioxidants and protect animal cells including humans.

Zhenhua Liu (University of Massachusetts Amherst) Bioactives: Obesity-Associated Inflammation and Gastrointestinal Health

Zhenhua Liu, School of Public Health and Health Sciences, University of Massachusetts, Amherst, MA and Jean Mayer USDA Human Nutrition Research Center on Aging, Tufts University, Boston, MA

Overweight and obesity have reached an epidemic level in the United States. Epidemiological studies indicate that obesity is associated with a range of cancer types, particularly colorectal cancer. However, the mechanism(s) mediating this effect remain unclear. My animal studies have shown that obesity induces an increase of colonic tumor necrosis factor alpha (TNF- α), a key inflammatory cytokine, and this occurs in parallel with alterations of several components within the Wnt pathway, one of the most critical cellular pathways in colorectal tumorigenesis, whereas these effects are attenuated when TNF- α is genetically blockaded. These data collectively suggested that obesity elevates colonic TNF- α which may activate Wnt-signaling and consequently promote intestinal tumorigenesis. Obesity is a chronic problem and long-term application of chemical agents is not practical. Instead, dietary interventions with anti-inflammatory bioactives are more attractive approaches to attenuate obesity-induced colonic inflammation and thereby prevent intestinal tumorigenesis.

Jeffrey Adelberg (Clemson University) Laboratory Micropropagation Environment Affects Acclimatization of *Veratrum californicum*

Ju Yeon Song¹, Jacqueline Naylor-Adelberg¹, Sarah A. White¹, David A. Mann², <u>Jeffrey Adelberg^{1,*}</u> School of Agricultural, Forest, and Environmental Sciences, Clemson University, Clemson, SC 29634 ²Infinity Pharmaceuticals, Inc., Cambridge MA 02139. Email: jadlbrg@clemson.edu

Temperature and light quality during laboratory micropropagation was observed to effect survival of Veratrum californicum during acclimatization in the greenhouse. The temperature study used 7 stage II clones placed in chambers at 10, 16, and 24°C, with 20 µM cool white fluorescent light and a 16 hr photoperiod. Light effects were evaluated using 5 stage II clones placed at 16°C under 20 µM light emitting diode (LED) monochromatic blue, red, red+blue, and 40 µM red+blue light. After 3-5 subculture periods, stage III plantlets obtained from temperature and light quality treatments were transferred into 36 cell trays (21" x 10.5" x 2.25") filled with Fafard 3B mix and were placed in a mist bed with plastic dome tray (11" x 21" x 2.5") for 10 days. After ten days the plastic domes were removed, and the trays remained in the mist bed for 20 additional days. Survival data were collected after 30 days. Temperature induced differential survival within individual clones was observed. Under fluorescent light, no plants from any clone at 24°C survived acclimatization, whereas two of the 7 clones at 16°C had an 80% - 100% survival ratio, and there was 50% survival of plantlets from the other 5 clones at 10 and 16°C. Survival of 4 of the 5 clones exhibiting 50% survival at 16°C, increased to 70% -80% when micropropagated under 40 uM red+blue or 20 uM blue LED light. The growth stage of tissue culture (TC) plants were compared with seedlings grown in the greenhouse. Most of the TC plants (65%) after 30 days of acclimatization were as massive as three-year-old seedlings. Although individual clones responded differently to environmental conditions, survival of plantlets cultured in the laboratory at 16° C under 20 μ M blue or 40 μ M red+blue LED light treatments had the best overall survival during acclimatization.

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Session 8: Herbal Markets and Marketing

Chair: Rodolfo Juliani (Rutgers University)

James Wilson (Pine Street Investments)

James Wilson, Managing Partner, Pine Street Investments, Medford, MA No Abstract

Cynthia Barstow (University of Massachusetts) Market Trends for the Herbal Industry

<u>Cynthia Barstow.</u> University of Massachusetts/Amherst Isenberg School of Management cbarstow@isenberg.umass.edu

Real-time market data provides much needed guidance to industry members for market strategy development. Sales figures and projected growth are critical. According to Mintel market research firm, "sales of herbal remedies have increased 13% since 2007 and are expected to continue growing to reach \$6.2 billion by 2017." Growth is anticipated due to the Patient Protection and Affordable Care Act (Obamacare). Consumer behavior and opinions about herbal efficacy has changed over the past decade. How current market trends will affect herbal sales and opportunities in the near future will be shared.

Session 9: Industry Needs

Chair: Gary Stutte (Kennedy Space Center)

James Tillotson (Tufts University)
From Success in the Lab to Profit in the Market - a Strategically Approach for Commercializing Plant Materials

James Tillotson, Food Policy & International Business, Friedman School, Tufts University, Boston, MA

We know too well that often success in the laboratory does not guarantee success in the market place. Market failures of new products based on attractive new scientific findings happen far too often, with loss of significant investment. This short presentation presents a strategy for initial evaluating of laboratory discoveries for their potential for ultimate market success. Market simulations of this type have become common practice today in the consumer product industries (foods, personal care and household care products). In recent years, such primary

screening techniques are increasingly be used for potential new functional foods and beverages and dietary supplements as well as medically active plant substances.

Stephen Gafner (Tom's of Maine)

Culture-Based Bioproduction

Stefan Gafner¹, Chantal Bergeron¹, Jacquelyn R. Villinski¹, Markus Godejohann², Pavel Kessler², John H. Cardellina II³, Daneel Ferreira⁴, Jacynthe Desjardins⁵, Karine Feghali⁵, Daniel Grenier⁵

¹Tom's of Maine, 302 Lafayette Center, Kennebunk, ME 04043; ²Bruker-Biospin, Silberstreifen 4, 76287 Rheinstetten, Germany; ³ReevesGroup, 9374 Highlander Boulevard, Walkersville, MD 21793; ⁴Department of Pharmacognosy and National Center for Natural Products Research, Research Institute of Pharmaceutical Sciences, School of Pharmacy, The University of Mississippi, University, Mississippi 38677; ⁵Groupe de Recherche en Écologie Buccale, Faculté de Médecine Dentaire, Université Laval, Quebec City, Quebec, G1V 0A6, Canada

The phytochemical investigation of a supercritical fluid extract of *Glycyrrhiza uralensis* led to the isolation of 20 known isoflavonoids and coumarins, as well as glycycarpan, a new pterocarpan. The presence of two isoflavan-quinones, licoriquinone A and licoriquinone B in a fraction subjected to gel filtration on Sephadex LH-20 is believed to be due to metal-catalyzed oxidative degradation of licoricidin (1) and licorisoflavan A (2). The licorice extract as well as 1 and 2 were able to reduce VSC production by *Porphyromonas gingivalis*, *Prevotella intermedia*, and *Solobacterium moorei* as well as in a human saliva model. Although the extract and isolates did not inhibit the proteolytic activity of bacteria, they blocked the conversion of cysteine into VSCs by *P. intermedia*. Compounds 1 and 2 also showed potent antibacterial activities, causing a marked growth inhibition of the cariogenic species *Streptococcus mutans* and *Streptococcus sobrinus* at 10 μg/mL and the periodontopathogenic species *P. gingivalis* (at 5 g/mL) and *P. intermedia* (at 5 g/mL for 1 and 2.5 g/mL for 2). Only 1 moderately inhibited growth of *Fusobacterium nucleatum* at the highest concentration tested (10 g/mL).

Session 10a: Contributed Talks

Chair: Mohammed Kamil (Zayed Complex for Herbal Research and Traditional Medicine-UAE)

Sanjeev Banerjee (Wayne State University School of Medicine) Anti-tumor Activity and Improved Bioavailability of a Novel Curcumin Analog - CDF against Pancreatic Tumors.

<u>Sanjeev Banerjee</u>, Subhash Padhye and Fazlul H Sarkar, Department of Pathology, Karmanos Cancer Institute, Wayne State University School of Medicine, Detroit, MI 48201. E-mail: sbanerjee@wayne.edu

Purpose: The current study was aimed to assess the antitumor effect as well as, the pharmacokinetics and tissue distribution of a novel curcumin analog- curcumin-difluorinated (CDF) in mice compared to curcumin. **Methods:** Serum pharmacokinetics and tissue distribution studies were carried out using the validated HPLC with LC-MS/MS methods. For antitumor studies, ICR-SCID mice were orthotopically implanted with pancreatic cancer cells and randomized into treatment groups comprising (*n*=5-7): (*a*) untreated control; (b) Curcumin (250 mg/kg b.wt/day orally by gavage for 21 days; and (c) CDF only (250 mg/kg body weight/day orally by gavage) for 21 days. **Results:** Molecular docking showed that CDF do not introduce any major steric changes compared to the parent curcumin molecule, which was consistent with down-regulation of NF- κB and reduced PGE₂ levels in cells treated with CDF. Pharmacokinetic parameters revealed that CDF had better retention and bioavailability and that the concentration of CDF in the pancreas tissue was 10-fold higher compared to curcumin. Analysis of average

pancreatic tumor weight in curcumin and CDF treatment groups revealed significant reduction in tumor weight in CDF group relative to untreated control and curcumin (p<0.05). **Conclusion:** Our observations clearly suggest that the bioavailability of CDF is much superior compared to curcumin, suggesting that CDF would be clinically useful in the future, and thus further development is warranted.

Yan Dong (Tulane University School of Medicine) Methyl-Selenium compound Enhances MDV3100 Efficacy for Treatment of Castration-Resistant Prostate Cancer

Yang Zhan¹, Bo Cao¹, Yanfeng Qi¹, Qi Zhang², Hua Lu², and <u>Yan Dong¹</u>. ¹Depts. of Structural and Cellular Biology, ²Biochemistry, Tulane University School of Medicine, New Orleans, LA 70112. E-mail: ydong@tulane.edu

The next-generation anti-androgen MDV3100 prolongs overall survival of patients with metastatic castration-resistant prostate cancer. However, patient responses are variable, and survival benefit remains relatively small. Developing effective modality to improve MDV3100 efficacy is urgently needed. Recent evidence suggests that androgen receptor splice variants (AR-Vs) drive resistance to MDV3100. Here, we tested the hypothesis that methyl-selenium compounds downregulate AR-Vs and enhance MDV3100 efficacy. Methyl-selenium compounds are bioactive food components. We found that methyl-selenium compound downregulates the expression of both the full-length AR (AR-FL) and AR-Vs as well as the activity of AR. The downregulation is independent of androgen and could be attributable to repressed transcription of the AR gene. Co-treatment with methyl-selenium compound and MDV3100 suppresses AR signaling more dramatically than either agent alone, and synergistically inhibits cell growth in vitro. Downregulating AR-FL and AR-V7 is importantly involved in mediating this synergistic effect. Significantly, methyl-selenium compound also downregulates AR-FL and AR-Vs in vivo and substantially improves the antitumor efficacy of These findings provide a rationale for evaluating the clinical application of combining methyl-selenium compound with MDV3100 for the treatment of castration-resistant prostate cancer.

Jaisa Baig (University of Houston-Victoria) Phytochemical Food Pyramid for Cancer Prevention

Richard S. Gunasekera, Ph.D.¹; <u>Jasia Baig¹</u>; and Siva Somasundaram, Ph.D.¹ ¹Department of Biological Sciences, University of Houston-Victoria, UH Sugar Land, Sugar Land, TX 77479 USA. E-mail: <u>GunasekeraR@uhv.edu</u>

Prostate cancer is the most frequently diagnosed form of cancer among men in the United States. Its pathogenesis includes multiple genetic and epigenetic mechanisms and additional environmental factors. Studies have shown that dietary phytochemicals play a crucial role in prostate carcinogenesis by influencing biological processes such as cell-cycle control, apoptosis, inflammation, and DNA repair. Dietary phytochemicals can act as blocking agents by obstructing the initiation phase of carcinogenesis or act as suppressing agents by hindering the promotion and progression phases of carcinogenesis. The primary focus of this research is to design a phytochemical bioactive molecule (PBAM) based pyramid which will provide cancer patients, survivors, and the average consumers with information on bioactive foods consisting of PBAM from dietary sources for cancer prevention. Ranked via a hierarchy based system the pyramid will inform users of healthy foods with phytochemicals that have cancer/disease preventive properties. The pyramid is created electronically linking to data mining information from scientific literature, epidemiological databases, and medical information on diet, nutrition, and cancer-preventive phytochemicals, which includes the individual's age and available genetic

information. It is structured to help an average consumer make an informed choice of foods that is based on good nutrition practice specific their subjective needs. Consumers will be able to buy foods and get rankings ranging from most beneficial for cancer prevention. Each food will be ranked via a hierarchy based point system based on an algorithm that includes the concentration of the phytochemical and the number of PBAM it provides. This phytochemical pyramid will be particularly useful for people who are genetically prone to certain cancers and chronic diseases. Thus, it will provide consumers with the confidence to choose foods from grocery stores and restaurants that will help fight cancer and certain diseases.

Session 10b: Contributed Talks

Chair: Rao Mentreddy

Rodolfo Juliani (Rutgers University) An Ethno-Botanical Survey to Identify Non-Timber Forest Products in Liberia

<u>H. Rodolfo Juliani¹</u>, James E. Simon, Ramu Govindasamy and Larry Amekuse² ¹New Use Agriculture and Natural Plant Products Program, Plant Biology and Pathology Dept., Rutgers University, 59 Dudley Rd., New Brunswick, NJ 08901. ² ASNAPP – Ghana. Accra Ghana. E-mail: hjuliani@rci.rutgers.edu

Forests have been increasingly recognized as reservoirs of valuable biological resources other than timber. The term non-timber (or non-wood) forest products (NTFP) emerged as a group of products derived from these various forest resources. In Liberia, NTFP are used by Community Forest dwellers and they include living animals, bush meat, rattan, bamboo, nuts, seeds, roots, mushrooms, and medicinal plants, among others. The objective of this work was to collect traditional knowledge on Liberian NTFP related to their uses and identify new or emerging NTFP. Of the 51 NTFP, bush pepper (Piper guineense) was the most cited and used by the interviewees (12% out of the total 159), followed by bitter cola (Garcinia kola, 9%), country spice (Xylopia sp., 7%), bush cherry (Maesobotrya barteri, 6%), bush yam (Dioscorea sp., 5%), bush peanut (Ricinidondron heudelotii, 4%), walnut (Cola edulis, 4%), among many others. NTFP were categorized into clusters according to their origin and/or processing or applications. NTFP were categorized into clusters according to their origin and/or processing or applications. These clusters include: spices (6% of the 51 NTFP identified in this study), medicinals (48%), colas and edible fruits (12%), nuts and edible oils (18%), indigenous vegetables and mushrooms (12%) and building materials (14%). In the survey, NTFP with medicinal purposes were the most cited by interviewees. Results from this study allow us to conclude that many Liberian NTFP are currently used by local communities in different applications suggesting NTFP can provide income generating opportunities.

Tomas Zavada (University of Massachusetts Boston) Genetic Diversity in Natural Populations of Medicinal Plants Cichorium intybus and Cichorium pumilum (Asteraceae)

<u>Tomáš Závada</u>¹, Rondy Malik¹ and Rick Kesseli¹. ¹Biology Department, University of Massachusetts Boston, 100 Morrissey Blvd., Boston, MA 02125, USA Email: tomas.zavada@umb.edu

Cichorium intybus (chicory) is a perennial herb with bright blue flowers. As a tea or extract, chicory root has been administered as a digestive, liver tonic, anti-rheumatic and mild laxative. Chicory is native to Europe and temperate regions in Asia. It has been naturalized to the United States. Chicory was cultivated as early as 5,000 years ago by Egyptians as a medicinal plant. Cichorium pumilum is an annual herb, shorter than chicory, native to Mediterranean. It has been

used in traditional Arabic medicine and recent pharmacological investigation of *C. pumilum* root extract revealed immunomodulator, antitumor and anticancer properties. In order to study genetic variation in chicory populations, twelve polymorphic microsatellite markers were developed and used for genotyping 510 chicory plants from 28 Eurasian and North American wild populations, and 30 *C. pumilum* plants coming from Turkey and Italy. Allelic profiles of all individuals were analyzed using programs Arlequin, HP-Rare. The genetic relationships based on marker similarities were visualized in Structure and AMOVA. Four cpDNA haplotypes revealed a strong geographic differentiation in the chicory native range, as well as the cultivar origin of North American chicory. The highest private allelic richness was detected among North American populations, suggesting local adaptation. Genetic clustering showed intraspecific hybridization in chicory, and a presence of novel genotypes in North America. Microsatellite markers crossamplified in other species in the genus *Cichorium* and can be used for the future DNA fingerprinting of genotypes most suitable for the medicinal use.

Agnes Rimando (USDA, Oxford, MS) Resveratrol and Piceatannol Inhibit Mammalian α-Glucosidase Activity

Albert J. Zhang¹, <u>Agnes M. Rimando</u>², Cassia S. Mizuno², Suresh T. Mathews¹. ¹ Department of Nutrition and Dietetics, Boshell Diabetes and Metabolic Diseases Research Program, Auburn University, Auburn, AL 36849; ² U.S. Department of Agriculture, Agricultural Research Service, Natural Products Utilization Research Unit, P.O. Box 8048, University, MS 38677

Dietary polyphenols have been shown to inhibit α -glucosidase, an enzyme target of some anti-diabetic drugs. Resveratrol, a polyphenol found in grapes, has been reported to inhibit the activity of yeast α -glucosidase. This triggered our interest to determine the effect of resveratrol analogs on mammalian α -glucosidase (EC 3.2.1.20) activity. Of the analogs tested, piceatannol showed the strongest inhibition. Piceatannol and resveratrol (0.5 mg/ml) inhibited mammalian α -glucosidase (IC50: 14-84 and 111-120 µg/ml, respectively) to a lesser extent than acarbose (IC50 = 3-13 µg/ml). We investigated the effect of resveratrol or piceatannol on post-prandial blood glucose response *in vivo*. High-fat fed C57Bl/6 mice administered resveratrol (30 mg/kg BW) or piceatannol (14 mg/kg BW) 60 min prior to sucrose- or starch-loading had a delayed absorption of carbohydrates, resulting in significant lowering of post-prandial blood glucose concentrations, similar to the antidiabetic drug acarbose. Our studies demonstrate that the dietary polyphenols resveratrol and piceatannol lower post-prandial hyperglycemia, and indicate that inhibition of intestinal α -glucosidase activity is a potential mechanism contributing to their anti-diabetic property.

Poster Presentations

Monday

Daniel M. Motlhanka (Bostwana)

Phytochemical and antioxidant analysis of wild and Ex Situ cultivated shoots and tubers of Harpagophytum Procumbens (Burch) DC Ex.Meisn from Botswana

Comparative phytochemical analysis [TLC method] and antioxidant activity of wild and ex *situ* cultivated shoots and tubers of *Harpagophytum procumbens* were done. Total phenolic content [Folin-Ciocalteu method] and free *radical* scavenging activity [1,1-diphenyl-2-picryl-hydrazyl assay] of both chloroform and methanol extracts were determined. Analysis of ex *situ* cultivated plant material showed presence of phytochemicals comparable with those found in the wild plants. The total phenolic contents (mg GAE/L) of methanolic tuber extracts from wild plants

(3366±22.68) were comparable to the methanolic tuber extracts (3297.00±54.56) from ex *situ* cultivated plants. The total phenolic contents

Daniel M. Motlhanka (Bostwana)

Analysis of nutrients, total phenols and antioxidant activity of Ficus Sansibarica Warb.

Fruits from Eastern Botswana Motlhanka, Daniel¹, Ebineng, Topo¹

¹ Botswana College of Agriculture, Medicinal Plants Research Laboratories, Department of Basic Sciences, Private Bag 0027, Gaborone, Botswana

Ripe fruits of *Ficus sansabanca* were assessed for *in vitro* antioxidant activity using the Free radical scavenging assay (FRSA). The scavenging activity of the fruits was evaluated spectrophotometrically as maximum fading power of 1,1-Diphenyl-2-picrylhydrazyl (DPPH) at 525nm. The total phenolic content of water and methanol extracts was determined using the Folin-Ciocalteau method. The mineral composition of the fruits was analyzed using the optical emission spectrophotometer. At all tested concentrations the scavenging power of methanolic extract was higher than that of water extract. Between 100 and 200ug/ml, both extracts exhibited scavenging power of s85% comparable to the standard compound quercetin. The total phenolic contents of water and methanolic extracts were 3145 and 3584 mg/L

Daniel M. Motlhanka (Bostwana)

Polyphenic content and antioxidant analysis of *Pseudolachnostylis Maprouneifolia Pax Var Dikindtii* used as livestock feed by farmers from Eastern Botswana Motlhanka D.M.T

Botswana College of Agriculture, Medicinal Plants Research Laboratories, Department of Basic Sciences, Private Bag 0027, Gaborone, Botswana.

In the present work, fresh fruits(FF), sun dried fruits(SDF), fresh leaves(FL) and sun dried leaves (SDL) of *Pseudolachnostylis maprouneifolia* var *dikindtii* were analyzed for total phenolic, total flavonoid content, presence of reducing sugars, proteins and lipids as well as free radical scavenging power. *P.maprouneifolia* is used as a dry season supplementary feed by livestock farmers in Eastern Botswana. The fruits and leaves of the studied plant tested positive for presence of reducing sugars and proteins. Lipids were detected only in fresh fruit samples. The order of total phenolic content (mg gallic acid equivalents/L) was $SDF(1240.3\pm200.5)>FL(1097.9\pm154.6)>SDL(952.7\pm86.8)>FF(838.6\pm5.7)$. The order of the flavonoid content (mg Quercetin equivalent/L) was $FL(384.9\pm5.2)>SDL(256.7\pm3.9)>SDF(159.9\pm8.3)>FF(139.1\pm2.6)$.

Daniel M. Motlhanka (Bostwana)

Phytochemical screening and antioxidant activity of Cardiospermum Corindum L Faux Persil From Botswana

Motlhanka D.M.T*1., Nthoiwa, K.K2

¹Botswana College of Agriculture, Medicinal Plants Research Laboratory, Department of Basic Sciences, Private Bag 0027, Gaborone, Botswana. ²Rochester Institute of Technology, Slaughter Hall. 78 Lomb Memorial Drive, Rochester, NY 14623-5604, USA.

The aim of this study was to compare the phytochemical composition and antioxidant activity of roots and shoots of *Cardiospermum corindum* collected from two geographically distant regions of Botswana (Tswapong Hills and Kgale Hills). Qualitative phytochemical analysis revealed presence of alkaloids, reducing sugars, saponins, phytosterols, phenols, flavonoids and terpenoids. Analysis by thin layer chromatography, revealed that both shoots and roots of plant collected from the two respective regions showed no differences in phytochemical constituents. Total phenol and flavonoid contents were quantitatively estimated. Total phenolic content

measured by Folin-Ciocalteu method varied from 164.4±2.2 to 364.2±3.1mg/L (GAE) Gallic Acid Equivalents. The order of total phenol contents were

Fabricio Medina-Bolivar (United States)

Expression of the AtMYB12 transcription factor in Scutellaria lateriflora hairy roots and its effect on the biosynthesis of the anticancer flavone wogonin

Hemnath Elango (India)

Beneficial Assessment Of Herb Drug Interaction- A Pharmacokinetic Approach

Jeong-Hoon Lee

The Classification and Characteristics of Korean Artemisia Species

<u>Jeong-Hoon Lee</u>¹, Chun-Geon Park¹, Ae-Jin Choi¹, Chung-Berm Park¹ and Sung-Gi Moon², department of Herbal Crop Research, Nat'l Ins. of Hoticultural & Herbal Science, RDA, Eumseong 369-873, Korea. ²Dept. of Biology, Kyungsung University. E-mail: artemisia@korea.kr

Artemisia spp. have been used in various fields, such as pharmacology, foods and ornamentals in Korea. In particular, diverse pharmaceutical effects have been reported anti-malarial, anti-cancer and anti-diabetic, etc. In order to development genus Artemisia into herb resources, Artemisia taxa distributed in Korea were researched origin of medicinal herbal, morphological characteristics, molecular phylogenetic analysis. 9 taxa of Artemisia plants are used as important medicinal material in traditional Korea medicine, such as 'Aeyup', 'Injin', 'Chengho', 'Moho', 'Amryeo'. The origin of wormwood was called 'Aeyup' were identified as A. indica, A. montana, A. argyi. On the hand, A. capillaries, A. gmeliniiwere called 'Injin' which were classified as 'Injinho' and 'Hanjin', respectively. In addition, molecular phylogenetic analysis was conducted using trnLF region of cpDNA and molecular authentication on trnLF region was carried out using PCR-RFLP technique. The results, A. caruifolia, A. keiskeana and A. sieversiana have specific banding patterns.

Jim Simon (David Byrnes) (United States) Quality and Nutritional Assessment of African Indigenous Vegetables (AIVs)

<u>David Byrnes</u>¹, H. Rodolfo Juliani¹, Qingli Wu¹, James E. Simon¹. ¹New Use Agriculture and Natural Plant Products Program, Plant Biology and Pathology Dept, Rutgers University, 59 Dudley Rd., New Brunswick, NJ 08901 E-mail: iesimon123@hotmail.com.

The drying and processing of leafy green vegetables is a valuable post-harvest marketing strategy to make valuable nutrients available beyond the growing season at an affordable price. Many of these African Indigenous Vegetables (AIVs) can contribute to food security and generate income. The production of safe and nutritious AIVs is then an important aspect of their production. The objective on this study was to assess the quality and nutritional value of *Gynandropis gynandra*, black nightshade *Solanum nigrum*, and amaranth *Amaranthus retroflexus*. This report determined quality control parameters such as moisture, ashes, acid insoluble ashes, color, and nutritional value (select micro and macro nutrients, and vitamin A via (3-carotene). Black nightshades showed high levels of moisture (higher than 12%). In Amaranth, the ash content was found to be high indicating a high total mineral content that was associated with high levels of calcium, magnesium and potassium. Spider plant was found to be higher in iron than spinach (known as a high source of iron). These results showed the high nutritional value of this AIV Acid insoluble ash content was also high in amaranth indicating a potential contamination by sand. Foreign matter content was low in all samples. All samples were dark

green, with no signs of moldiness. While some of the samples showed high nutritional value, post-harvest handling practices should be improved to obtain products that are safe to consume.

Jim Simon (Robert Pyne) (United States) Variation of Essential Oil Constituents and Antioxidant Capacity in a Segregating Basil (*Ocimum* spp.) Population

Robert Pyne¹, H. Rodolfo Juliani¹, Adolfina R. Koroch^{1,2}, and James E. Simon¹. ¹New Use Agriculture and Natural Plant Products Program, Plant Biology and Pathology Dept., Rutgers University, 59 Dudley Rd., New Brunswick, NJ 08901 ²Borough of Manhattan Community College (BMCC, CUNY), New York, NY. E-mail: iesimon123@hotmail.com

Sweet basil (Ocimum basilicum L.) is the most commercially important culinary herb in the United States. Emergence of the destructive disease basil downy mildew (Peronospora belbehrii) has devastated basil crops in recent years with no effective chemical or management control available. While traditional sweet basil varieties are highly susceptible to downy mildew, two commercial varieties, 'Spice' O. americanum and 'Sweet dani' O. citriodorum, show high levels of disease tolerance in field trials. Introgression of disease tolerance to sweet basil may be possible through interspecific crosses, but not without consequences to prominent traits. Aroma quality and antioxidant capacity in basil leaves are important characteristics for commercial and health purposes. In an effort to begin to understand the inheritance patterns of these traits, a segregating F2 population was generated from a cross between inbred parents, 'Sweet dani' 0. citriodorum (SD) and sweet basil (O. basilicum) breeding line (CB10). Major volatile compounds of essential oils were identified and quantified using GC/MS and antioxidant capacity measured with the Trolox equivalent antioxidant capacity assay (TEAC). Parents varied extensively in composition of essential oil, while antioxidant capacity was only slightly higher in CB10. There was significant variation in the F2 population, which demonstrated a wide range antioxidant capacity and volatiles present in either parent. Basil breeding programs for selection of resistance to downy mildew should also consider aromatic and other bioactive components.

Jing Zhen (Qingli Wu) (United States) Polyphenols from Hibiscus leaf: A Promising New Source of Antioxidants

Polyphenols from Hibiscus leaf: A Promising New Source of Antioxidants

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A liquid chromatography-mass spectrometry method was developed for the simultaneous determination of phenolic acids and flavonoids in the leaves of the vegetable/nutraceutical plant *Hibiscus sabdariffa*. By analyzing the UV/MS data, and comparing with authenticated standards, a total of 9 polyphenols were identified including chlorogenic acid and the isomers, quercetin, kaemferol and the glycosides, as well as 5-(hydroxymethyl)furfural. Individual polyphenols were quantitated by HPLC under UV detection. The antioxidant activity of 39 Hibiscus accessions, determined using Folin-Ciocalteu's assay and ABTS assay, ranged from 19.0 ± 2.7 to 39.1 ± 3.9 mg/gram gallic acid equivalent and 101.5 ± 17.5 to 199.0 ± 20.5 mg/gram Trolox equivalent.

Mohamed Mohamed Aly Abdalla (Egypt) Effect of biofertilization on yield and yield component of three potato cultivars (Solanum tuberosum L.)

Sayed Ali Mohamad Modares Seed Germination of *Withania coagulans* under Light Regimes and Germination Media

Leila Edalatifard¹, <u>Seyed Ali Mohammad Modares</u> Sanavy² and Hossein Askari^{3 1 and 2} Department of Agronomy, Faculty of Agriculture, Tarbiat Modares University, Iran Department of Biotechnology, Faculty of Energy Engineering and New Technology, Shahid Beheshti University, Iran. E-mail: <u>modaresa@modares.ac.ir</u>

Withania coagulans is a plant of repute in the traditional medicine and has become low and scattered in the south of Iran. Self-propagation of this species is by seed. Propagation by natural re-seeding is thus no longer sufficient to guarantee the survival of this plant therefore; it has become necessary to develop rapid methods of cultivating the plant in order to ensure its sustainable utilization in tissue culture. In an effort to improve and promote the germination of this medicinal herb, the effects of media culture and light regimes on the germination of seeds were investigated. Seeds of twelve different chemo types were surface sterilized, plated on different media incubated for 25 days. Also To determine effect of light on germination, two light regimes were used through the experiment. During the experiments, some chemo types germinated faster than others. Also Germination was found to be dependent on media and light regimes. Although seed germination was varied, maximum germination was obtained in top of paper media and under complete darkness. Light treatment reduced the germination of the seeds and completely inhibited germination. So use of suitable media under complete darkness could be improved germination of the seeds.

Umang Haribhai Gajjar (India) Improvement in Safety Profile of *Semecarpus anacardium* Linn by Shodhana – An Ayurvedic Purification Method

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Semecarpus anacardium shows the presence of bioflavonoid, phenolic compounds, bhilawanols, minerals, vitamins and amino acids. Detoxified *S. anacardium* and its oils are considered to have anti-inflammatory properties and used in nervous debility, neuritis, rheumatism and leprous modules. *S. anacardium* if used without purification causes toxic skin inflammation problem because it contains toxic phenolic oil. During this Shodhana Process – An ayurvedic purification method, toxic phenolic oil was removed, have marked effect on the concentration of the phytoconsituents and antioxidant activity of *S. anacardium*. Total phenolic content decreased up to 70% (from 28.9% w/w to 8.94% w/w), while there is a negligible effect on the concentration of total flavonoid (7.51% w/w to 7.43% w/w) and total carbohydrate (0.907% w/w to 0.853% w/w) content. IC₅₀ & EC₅₀ value of extract of *S. anacardium* before and after purification are 171.7 & 314.3 whileEC₅₀ values are 280.ug/ml & 304.ug/ml, shows that antioxidant activity of *Semecarpus anacardium* is decreased but the safety profile of the drug is increased as the toxic phenolic oil was removed during Shodhana – An ayurvedic purification method.

Yerakala Anusha Prasad (India) Herbal Anti-diabetic Mouth Dissolving Film

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Diabetes mellitus, a bane of this era is one the most chronic diseases around the world. The present study aims at modernising the traditionally used herbal medicines for diabetes through a novel approach. Mouth dissolving film is an unique drug delivery system which enables better bioavailability, a convenient dosage form, bypasses the first pass metabolism, ensures better compliance. Hydroal coholic dried patient extract of the leaves of Gymnema silvestre's(Asclepidiacea) and aqueous extract of Trigonella foenicum{Fabacea} were incorporated in the mouth dissolving film comprising of hydroxypropylmethylcellulose(HPMC). These plants were selected on the basis of the ethno medicinal knowledge of the native tribes of The Nilgiris, Physicochemical evaluation of herbal extracts in the film was carried out and also compatibility studies of the herbal extracts with the additives and adjuvants were done. A combination extracts of Gymnema sy/vesfres(Asclepidiacea) and Trigonella foenicum(Fabacea) in the ratio of 40:35mg showed significant antidiabetic activity in Alloxan induced rabbit models.

Emad A. Mady ()

Assessment of Genetic Variability of Squash Landraces Using Isozymes and Protein SDS-PAGE

Maiko Inoue (University of Massachusetts)

Genetic Diversity of Wild vs. Cultivated Goldenseal (*Hydrastis canadensis* L.) Using RAPD Markers

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Goldenseal (Hydrastis canadensis L.) is a medicinal plant valued for the treatment of sore eyes and mouths. Although cultivation of the plant has helped meet growing demand, goldenseal is still considered a threatened or endangered species throughout much of its range in North America. In an effort to assess possible conservation strategies for goldenseal genetic resources, levels of genetic diversity within and among cultivated and wild populations were quantified. RAPD analysis was used to examine 6 cultivated and 11 wild populations sampled from North Carolina, Ohio, Pennsylvania, and West Virginia. The average percentage of polymorphic bands in cultivated and wild populations was low (16.8 and 15.5%, respectively), and geographic range did not predict the level of genetic diversity. Most of the genetic variation (81.2%) was within populations; only 3.6% was partitioned between cultivated and wild populations. Our results differed from a previous study which concluded that genetic differences were greater among than within populations. The results of the current study indicate that, although goldenseal grows clonally and in dense patches, a mixed mating system in which both selfing and outcrossing occur is also operating. We therefore suggest that the ex situ conservation of individual plants within populations, chosen carefully to account for clonal propagation in situ, is an appropriate strategy for sustaining the genetic diversity of goldenseal.

Tuesday

Aijaz Ahmad, Nikhat Manzoor Antioxidant Adaptation by Eugenol and Its Derivatives and Their Effect on the Expression of Virulence in *Candida Species*

Aijaz Ahmad, Nikhat Manzoor, Department of Biosciences, Jamia Millia Islamia, New Delhi, India

Present work investigates the antifungal activity and mode of action of eugenol (EUG), and its three derivatives - methyl eugenol (MEUG), eugenyl acetate, myresticin, thymol (THY) and carvacrol (CARV). EUG and its derivatives were tested for antifungal activity by standard methods of CLSI. The mean MIC of EUG was 500 mgL"1 which reduced down to 350, 100 l and 50 mgL"1 with MEUG, THY and CARV respectively. EUG and its derivatives varied in their mechanism of action depending upon the period of exposure. Short exposures of 5-15 minutes resulted in reduced H+ efflux by the H+-pump. An exposure of lh resulted in membrane leakage while prolonged exposures of 18h resulted in highly reduced ergosterol content indicating the involvement of ergosterol biosynthesis pathway in mechanism of action of test compounds. It was also observed that exposures of 8h, at very low concentrations induce oxidative stress in yeast contributing to further membrane disintegration. From our studies we conclude that EUG and its derivatives induce production of free radicals which stimulates the enzyme SOD. An increased SOD activity resulted in an increase in the concentration of H2O2 which further stimulates the peroxide eliminating enzyme, primarily GPx. It is noteworthy that the levels of GSH an essential substrate of GPx were drastically reduced by the test compounds and this reduction gets even greater as increased levels of H2O2 decrease the activity of G6PDH which provides reducing equivalents to GR, an enzyme that recycles GSH from GSSG. As a result, decreased G6PDH activity aids further in the reduction of GSH. Again, reduced availability of GSH explains decreased GPx activity. Another enzyme characterized to eliminate H2O2 is catalase, which triggers a cellular response leading to an increase in its activity. Hence increase in the activity of two important antioxidant enzymes SOD and catalase, clearly demonstrates an increase in the concentration of ROS when the Candida cell were exposed to the EUG and derivatives. However, these enzymatic responses were not enough to defend the cell completely against such a high rise in ROS and therefore did not meet the required cellular antioxidant demand. Ultimately, the outburst of free radical production led to severe lipid peroxidation. Cell death on exposure to EUG and its derivatives hence may be due to (i) decrease in the rate of H+ efflux (ii) reduced ergosterol content (iii) Induction of oxidative stress in the cell (iv) These processes impair membrane structure and function and as a result form lesions. Infection process of C albicans is characterized by crucial pathogenicity markers. The initial process of germ tube induction followed by the secretion of hydrolytic enzymes help in the invasion of the host cells. EUG and its derivatives significantly inhibited these pathogenicity markers even at sub-MIC values. The expression profile of selected genes associated with Candida virulence by RT-PCR showed a reduced expression of HWP1, SAP1 and PLB2 genes in C albicans cells treated with Eugenol and its derivatives. Virulence traits depend upon the cells antioxidant status which was impaired by the test compounds. Further studies using animal models are necessary to see the in vivo efficacy of the compounds.

Catherina O'Keefe (Ireland)

Is Ragwort a Medicinally Active Plant?

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Ragwort, Senecio Jacobaea, is an invasive species which is listed as noxious and prohibited in Ireland. Ragwort has gained the interest of the scientific, horticultural and agricultural communities in the past number of decades due to the economic losses it imposes on farmers, crop growers and local and state authorities. Ragwort contains toxins known as pyrrolizidine alkaloids that have been proven to cause $\bf a$ variety of health problems when ingested. Although the toxic nature of ragwort has been widely reported in literature, research is accumulating over the past number of decades supporting the medicinally bioactive potential surrounding this plant. Ragwort contains high quantities of polyphenols compounds that exhibit an antioxidant capability and trends were seen between the different tissues examined. A total of 40 plants were examined from 4 counties in the south-west of Ireland and different tissues were examined (flowers, leaves and stems). It was found that a linear relationship exists between the phenolic compounds and the antioxidant capacity of the tissues ($\bf r^2 = > 0.56$; $\bf p = < 0.07$). It was also found that the greatest antioxidant capability was in the inflorescence and the least in the stems tissues.

Chima Cartney Igwe (Lagos, Nigeria) Antibacterial and Physico-Chemical Properties of Neem Seed, Leaf and Bark Organic-Solvent Extracts

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Several parts of Neem tree {Azadirachta indica) are used in traditional medicine in many West African countries for the treatment of various human diseases. The leaf, stem bark and seed were air dried for 8, 5 and 7 days respectively. The shells were carfully separated from the seeds, each powdered sample obtained with mechanical miller and 250 mm sieve. The Neem samples were individually subjected to extraction with acetone, n-hexane for 48hrs and 72 hrs. respectively. Physico-chemical and antibacterial evaluation were carried out using standard methods. Results of physico - chemical analyses of the extracted oil from the seed shows that it has a brownish colour, with a smell similar to garlic while the moisture content, refractive index are 0.76% and 1.47 respectively. Other vital chemical results obtained from the Neem oil such as saponification value (234.62), acid value (10.84 %), free fatty acid (5.84 %) and peroxide value (10.52%) indicated the oil extracted satisfied standard oils parameters for quality soap and cosmetics production. The antibacterial screening by disc diffusion revealed the oil demonstrated high activity against Staphylococcus aureus The preliminary results of this study may validate the medicinal value of the plant. Further studies are in progress to clarify the *in vivo* potentials of Neem extracts in the management of human communicable diseases and this is a subject of investigation in our group.

Gehad T. El-Sherbini (Egypt)

In vitro Effect of Pomegranate Peel Extract on Trichomonas tenax

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The incidence of *Trichomonas tenax* (*T. tenax*) in patients with acute ulcerative gingivitis has been demonstrated in several published reports. Metronidazole was known as the most effective drug for human trichomoniasis, however, drug resistance and toxicity appeared. This study was designed in vitro

to investigate the inhibitory activity of *Punica granatum* (*P. granatun*) ethanol extract on the growth and motility of 7 *tenax* in comparison to metronidazole. Pomegranate ethanol extract group was treated with concentrations of 12.5, 25, 50, and 100 pg/ml. Metronidazole group and blank control were included. At 12 h, 24 h 48 h and 72 hr after drug treatment, the anti-7 *tenax* effect of pomegranate ethanol extract was tested by microscope counting method. The results showed 60% motility of *Ttenax* trophozoite after treatment with 12.5fjg/100 ml of pomegranate ethanol extract group and 25 (jg/ml showed higher anti-7 *tenax* (P<0.01). The ethanol extract of pomegranate peel has a remarkable effect on 7 *tenax*, and among the groups, 60% ethanol extract shows the best anti-7. *ten ax* activity.

Gehad T. El-Sherbini (Egypt) Anthelmintic Activity of Unripe *Mangifera indica* L. (Mango) against *Strongyloides* Stercolaris

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Infections with *Strongyloides stercoralis* and other helminths represent important, yet often neglected issues in developing countries. Indeed, strongyloidiasis can be fatal, but only a few studies provide information regarding its health relevance in Africa. *Strongyloides stercoralis* is an intestinal nematode that can persist in the human host for decades after the initial infection and can progress to fulminant hyperinfection syndrome in immunocompromised hosts, and the rapid development of nematode resistance to anthelminthics has limited the success of control in several countries, stimulating the search for alternatives. In this study, extracts of immature fruits of the mango *Mangifera indica* L. were evaluated for inhibition of larval development. In the phytochemical analyses, tannins and flavonoids were the metabolites identified. Aqueous extracts of immature fruits at 100 mg ml¹¹ showed 100% inhibition of larval development. In vitro results indicate that this fruit could assist *Strongyloides stercoralis* control.

H. Rodolfo Juliani (United States)

Linking education, research and development: quality control standards of Liberian Non-Timber Forest Products (NTFP)

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Several studies have shown that Non-Timber Forest Products (NTFP) can contribute to household food security by providing food, medicines and raw materials for enterprises, as well as commodities for trade. Liberia has a long history of the use of NTFP by forest dwellers. As Liberian NTFP has the potential to generate sources of income, this work sought to conduct quality and nutritional assessments of several NTFP found in local markets. Six NTFP were used in this study they were: Bush peanut (Ricinodendron heudelotii), Bitter kola (Garcinia kola), Wolor (Beilschmiedia manii), Bush pepper (Piper guineense), Grains of Paradise (Aframomum melegueta) and country spice (Xylopia aethiopica). Quality control analysis were conducted to develop trade standards, they included moisture content, foreign materials, botanical dust content, ashes, acid insoluble ashes, crude fats, total proteins and secondary metabolites (e.g. essential oils in spices). All these NTFP showed variations in these parameters. Some of the bush pepper samples showed high levels of moisture and foreign materials. Bush peanut was characterized by high levels of crude fats, protein, calcium and phosphorus. Wolor was characterized by high levels of iron. Bitter cola showed low levels of total ashes, thus characterized by low amounts of micro and macronutrients. This initial study served to generate initial quality control standards to support the commercialization of these NTFP.

Jim Simon (Thomas S. Villani) (United States) New Clearing Agent for Quality Assessment of Commercial Herbal Products

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Light microscopic observation is still today an important tool used for authentication and quality assessment for many commercial herbal products in industry and academia. Since plant tissues are opaque, a clearing procedure is necessary to reveal internal components. There are different clearing solutions described for plant tissues, however the most commonly used one is acidified chloral hydrate. Many Pharmacopeias have published protocols for microscopic authentication analyses of herbal preparations using acidified chloral hydrate as clearing agent. Consequently, chloral hydrate has become the industry standard and an important reagent required on a daily basis for many laboratories focused on quality assessment of herbal products. However, in the U.S., chloral hydrate is a Federally Regulated Schedule IV substance, and thus a special permit is required to purchase, possess and use it. This regulation places chloral hydrate out of the reach for many laboratories. We report here the use of a new solution, VisikolTM, as a suitable, non-regulated proprietary substitute for chloral hydrate in microscopic applications for botanical and agricultural quality assessment.

Several milligrams of fine powder of authenticated botanical materials were spread on a microscope slide and mounted either with two drops acidified chloral hydrate solution (control) or mounted with two drops of VisikolTM clearing solution and a cover slip, then the slides were heated on hot plate for 30-60 seconds. VisikolTM clearing solution proved to be an effective clearing agent (i.e. resulting in transparent tissues) in all samples tested; comparable to the results observed using chloral hydrate. We have demonstrated that the new clearing agent VisikolTM can be effectively used as a replacement of chloral hydrate in botanical microscopy. VisikolTM can thus be used for clearing herbal products to yield high quality images for quality assessment, authentication and teaching.

Jim Simon (William Reichert) (United States) Identification and Quantification of Polyphenols in *Nepeta cataria*

Identification and Quantification of Polyphenols in Nepeta cataria

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In this study, we analyzed 10 different catnip (*Nepeta cataria*) lines that consisted of several commercial ones and our Rutgers breeding lines for polyphenols and to determine optimum harvest method relative to tissue accumulation. Flowers, upper leaves and lower leaves were harvested and analyzed separately. Analysis of the flavonoids and phenolic acids was carried out using HPLC/UV/MS. Oxygen radical absorption capacities were determined using the ABTS radical scavenging assay. The specific flavonoids and phenolic acids present as well as their concentration varied by tissue and harvest location. Eight different flavonoids were identified as glycosylated luteolin's and apigenin's while two phenolic acids caffeic and rosmarinic were detected and all confirmed with UV and MS data. Using linear regression clustering techniques, a heatmap was constructed using Quantitative data from the HPLC analysis to visualize chemical

Motamarri Venkata Naga Lalitha Chaitanya A Polyherbal Formulation Development and Standardization for Its Sedatie and Antianxiety: Validation of Traditional Knowledge.

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Valerian contains over 150 chemical constituents including amino acids, such as gamma- amino butyric acid, tyrosine, arginine and glutamine. Out of which the key active constituents present in valerian are iridoid valepotriates (0.5-2.0%), valeric and isovaleric acids (0.2-2.8%), volatile oil (0.2%-0.4%), alkaloids (0.01-0.05%) and lignans. Passiflora edulis contains indole alkaloids (0.01-0.09%), vitexine and isovitexine (0.8-2.5%), cyanogenic glycosides, fatty acids, gum, maltol, sterol, and a trace of volatile oil. Hypericum Perforatum contains various flavonoids (epigallocatechin, rutin, hyperoside, isoquercetin, quercitrin, quercetin), phenolic acids (chlorogenic acid, 3-O-coumaroylquinic acid), and various naphtodianthrones (hypericin, protopseudohypericin), pseudohypericin, protohypericin, phloroglucinols adhyperforin). These plants are traditionally used as sedative, relaxant and used for relieves muscle pains, anxiety and lowers blood pressure. The mechanism may be based on the inhibition of enzyme-induced break down of Gamma -Amino-Butyric Acid (GABA) in the brain, with concomitant sedation.500 gms of each crude drugs (Valeriana officinalis (rhizomes), Passiflora incarnata (aerial parts) and Hypericum perforatum (Aerial Parts)) was subjected to soxhelation individually using water as menstrum. The spray dried extracts were granulated and subjected to tablet compression. The formulations were subjected to qualitative and quantitative chemical analysis by using HPTLC .The study confirmed the presence of valeric acid in Valeriana officinalis and Isovitexine and Harmaline in Passiflora incarnata. A tablet formulation was prepared using the crude drugs and also aqueous extracts. The prepared polyherbal formulations were subjected to in-vivo pharmacological screening by using Swiss Albino mice to evaluate the sedative and anti anxiety activity of the prepared tablet formulation by using two model namely Phenobarbitone potentised sleeping time (for sedative activity) and Elevated plus maze (For anti anxiety activity). The formulation showed potent sedative and anti anxiety activity and also a pharmacological synergy was observed in case of crude drug formulation.

Rathinavel Suriliandi (India) Antimicrobial Property of Adult Leaf and *In vitro* Developed Callus Extracts of *Baliospermum Montanum*

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Baliospermum montanum (Willd.)Muell.Arg.belongs to the family Euphorbiaceae. A study was made to evaluate the antimicrobial property of leaf and in vitro developed callus extracts of B. montanum. The analyses of leaf and callus extracts of B. montanum evidenced qualitative difference in the phytochemical compounds. The minimum inhibitory concentration (MIC) of different solvent extracts of leaf and callus ranged from 20 to 40 mg/ml. The inhibitory effect of chloroform leaf extract against Klebsiella pneumonia and callus extracts of methanol against Bacillus subtilis and Escherichia coli was at significant levels. The inhibitory efficiency of methanolic leaf and callus extract against Klebsiella pneumonia and Solmonella typhi was also greater than their respective positive control. It was apparent that leaf and callus extracts of petroleum ether and chloroform had poor inhibitory effect in most of the tested organisms. It was interesting to note that the methonolic extract of callus had better inhibitory effect against

Bacillus subtilis, Escherichia coli, Klebsiella pneumonia and Solmonella typhi than methonolic extract of leaf and antibiotic streptromycin (positive control). It is concluded that the *in vitro* developed callus from leaf of selected medicinal plant can be used for pharmaceutical application.

S.K.M.Basha (India)

Medicinal Plant Recourses and Its Conservation of Seshachalam Biosphere Reserve (Eastern Ghats) – Andhra Pradesh, India

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Seshachalam biosphere reservse of Eastern ghats is the richest floristic hot spot harbouring many endemic and rare plants. This area is not properly explored due to prohibition and religious importance because the holy shrine of Lord Sri Venkateswara or Balaji situated here. Namalagadhi, Dharmagiri, Gogarbham, Srivarimettu, Microwave station and adjacent forests like Mamandur, Kukkaiadhodi, Nagapatla and Talakona to mention a few are endowed with a very rich flora and also abound in many medicinal plants of importance. Varied micro-climatological, undurlating topographical conditions prevailing at various water falls(theerthams) situated at different heights, provide a very favourable habit and support the growth of the plant species. The present study the basis of hebarium collection a short account of different plant resources is presented here. Its dry deciduous type of forest with patches of moist deciduos forest. It is famous for plant species like sandalwood, redsanders, shorea thumbergia, shorea taiura, terminalia pallida, cycas beddomei, syzygium alternifolium etc. it has about 1500 plant species belonging to 176 families.

Endangered plants:

Homalium zeylanicum, Buteamonosperma, Rhynchosia heynei, Tephrosia colophylla, Saraca asoca, Entada rheedii, Heracleum regins, Plumbago indica, Stryhnos colubrina, Cerapegia spiralis, Decalepis hamiltonii, Pectranthus barbatus, Pipemigrum, Santalum album. Vanilla wightiana and Acorus calamus.

Critically endangered plants:

Rauvolfia serpentina and Litsea glutinosa.

Vulnerable plants:

Hildegardia populifolia, Sterculia urens, Aegle marmelas, Rubia cordifolia, Gymnema sylvestre. Oroxylum indicum, Euphorbia fusiformis, Phyllanthus indofischeri. Stemona tuberosa and Gloriosa superba.

Near threatened plants:

Shorea robusta, Celastrus paniculatus, Pueraria tuberosa, Holostemma ada-kodien and Costus speciosus.

Endemic plants:

Shorea tumbaggaia (Dipterocarpaceae), Boswellia ovalifoliolata (Burseraceae), Indigofera barberi, Pteroarpus santalinus, Rhynchosia beddomei, Sophora interrapta (Fabaceae), Terminalia pallida (Combretaceae), Syzygium alternifolium. (Myrtaceae), Pimpinellat irupatlensis (Apiaeae), Leucas indica var. nagaiapuramiana (Lamiaceae) and one gymnosperrn Cycas beddomei (Cycadaceae) are endemic to this geographical area.

Conclusion:

The Eastern ghats is an important area in India and the rich biodiversity The current rate of extension demands immediate concerted efforts for conservation of biodiversity for future generation. Conservation of biodiversity could be accomplished using both in situ and ex situ methods.

Seon Yi Won (Korea)

Composition of seed oil and extracts by germination phase of Panax ginseng seed

Composition of seed oil and extracts by germination phase of *Panax ginseng* seed.

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The root part of *Panax ginseng* C. A. Meyer mainly has been used as an precious medicinal purpose. Some studies reported that the berry of P. ginseng had biological and pharmacological activities more than the root. But the bioactive constituents and pharmacological activities from P. ginseng seed oil(PGSO) and extract(PGSE) have not been well studied yet. In this studies, we investigated the change of bioactive constituents of *P. ginseng* seeds as the germination of seeds progressed. Dried seeds(DS), low temperature stratification treated seeds for 90 days(LTSS), rooting seeds from the seed coat(RS), young seedling with 2~3 leaves(YS) were analyzed for 15 ginsenoside contents using high performance liquid chromatography(HPLC), fatty acid using gas chromatography(GC), polyphenols compound, total flavonoid, organic compound, inhibition capacity of nitric oxide(NO) and 1,1 -diphenyl-2-picryl hydrazyl (DPPH) radical scavenging activity to apply the cosmetic materials. HPLC analysis showed that the main constituents in seeds of P. ginseng(DS) were ginsenoside Rb2(2.2mg/g), Rb1(1.25mg/g) and Rc(1.0mg/g) belong to protopanaxadiol type saponin. The fatty acids contained 89.7% of the unsaturated fatty acids such as oleic acid and linoleic acid. The contents of unsaturated fatty acids were increased as the stratification treatment progressed. The polyphenols compound, total flavonoid, inhibition capacity of nitric oxide(NO) and DPPH radical scavenging activity had not almost changed during the low temperature stratification treatment period, but they were significantly increased in YS only.

LaDarius Pugh (Mentreddy) (United States)

Response of two medicinal basil (Ocimum tenuiflorum) varieties to nutrient stress

Mahmoud Alazzazy (Egypt)

Effect of Intercropping Medicinal Plants on Mite Populations in Cultivated Cucumber

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This study was conducted to assess the potential of pest repellent plant (PRP) species for managing mite pests. In order to reduce the use of pesticides and to improve the quality of the product . Three PRP species (sweet basil, peppermint and spearmint) were assessed together with a control treatment (without PRP). The results showed that the cultivated cucumber intercropped with the sweet basil, peppermint and spearmint led to lower infestation rate of mites to (92.9 , 91.7%), (92.1 , 92.7%) and (87.3 , 82.03%) for Polyphagotarsonemus latus (Banks) and Tetranychus cucurbitacearum (Sayed) respectively under field condition, and produced (5.43, 4.69 and 4.29 tons/ hectare). While the control treatment produced (4.07 tons/ hectare). In the green house the plant leaves of cucumber plants intercropping with the same medicinal plants reduced infestation rate to (87.7 , 82.9%), (80.6 , 75.2%) and (57.8 , 36.1%) for P. latus , T. cucurbitacearum, and produced (4.74, 4.12 and 4.00 tons/ hectare) respectively. While the control treatment produced (3.80 tons/ hectare).

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Radioprotective Activity of Calendula and Complex Plant Extract (Calendula, Chamomile and Yarrow) Collected in Azerbaijan

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Exposure to radiation has significantly increased during the past hundred years with the development and use of x-rays and radioisotopes in medicine and through environmental pollution from nuclear weapons and power plants. Because radiation can induce mutagenic changes, interfere with the immune system, and lead to the development of cancers, agents that could protect the body from radiation effects would be of great benefit. In this study, ethanol extract of calendula (*Calendula officinalis* L.) and Complex Plant Extract (mix of calendula with chamomile (*Matricaria chamomilla* L.) and yarrow (*Achillea milliefolium* L.) reduced mutagenic effects of γ -radiation on white mice, and prolonged life of animals. Plant material was collected in Ismayilli district of Azerbaijan. Radiation was applied in a gamma-ionization chamber at the Rkhund-20000 facility located at the Institute of Radiation Problems in Baku using an average power of 0,396 Gy/sec.

Carolyn Lee-Parsons (United States)

Transcriptional regulation of Jasmonate-Induced Biodynthesis of medicinal alkaloids from Catharandthus roseus Hairy Root Cultures

Leila Edalaitfard (Iran)

Seed Germinatin Withania Coagulans under Light Conditions and Germination Media

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Withania coagulans, a traditional medicine plant scattered in Southern Iran, has become scarce due to overharvesting. Natural propagation of this species is by seed but to improve poor germination and promote the germination of this medicinal herb, the effects of media and light conditions on seed germination were investigated. During the experiments, some chemotypes germinated earlier than others. Seed germination was determined to be dependent on media and light condition. Although seed germination was varied, maximum germination was obtained in water media and under complete darkness. Seeds germination was completely inhibited by light treatment.

Expression of the AtMYB12 Transcription Factor in *Scutellaria lateriflora* Hairy Roots and Its Effect on the Biosynthesis of the Anticancer Flavone Wogonin

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American skullcap (*Scutellaria lateriflora*) produces a diversity of polyphenolic compounds that have shown various biological activities. In particular, the flavones wogonin and baicalein have shown anticancer properties *in vitro* and are included in several traditional medicines. However, the molecular mechanisms that regulate the biosynthesis of these bioactive metabolites have not been elucidated. In this study, hairy root cultures of *S. lateriflora* expressing the flavonoid-specific transcription AtMYB12 were developed under the control of the SuperP:TEV

expression system. HPLC and LC-tandem mass spectrometry analysis confirmed the presence of the phenylpropanoid compound verbascoside and the flavonoids bacailein, baicalin, wogonin and wogonoside in the hairy roots of *S. lateriflora*. Semi-quantitative RT-PCR analysis showed an increase in gene expression of selected genes associated with phenylpropanoid and flavonoid biosynthesis. Furthermore, the accumulation of wogonin was increased suggesting that the AtMYB12 transcription factor could regulate its biosynthesis in *S. lateriflora*.

Protective Effects of *Ficus Religiosa* Stem Bark Extracts against Isoniazid and Rifampicin Induced Hepatotoxicity in Albino Rabbits

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Tuberculosis (TB) is a worldwide pandemic and its death ratio is highest after HIV/AIDS. Rifampicin (RIF) and Isoniazid (INH) are the first line antituberculosis drugs that are administered for 6-9 months depending upon the severity of disease. This standardized therapy is associated with a number of problems including neuropathy, gastrointestinal and hematological disorders. As plants are used for the management of many ailments so the current study is planned to evaluate health protective effects of *Ficus religiosa* against adverse effects associated with antituberculosis therapy. A total no. of 40 rabbits were divided into 5 groups (n=8). Group 1 was kept as control without drugs. Group 2 was given anti TB drugs orally for 28 days while groups 3, 4 and 5 were given silymarin, ethanolic ficus extract, and aqueous ficus extract along with anti TB drugs orally for 28 days respectively. Blood samples were taken before drug administration and at 7th, 14th, 21^{s1}, and 28th days post treatment for hematological and biochemical analysis. Liver tissues were taken for histopathological studies. Biochemical results concluded that INH+RIF exerted synergistic effects causing hepatocellular damage thus induces the release of aminotransferases. There was statistically significant rise in ALT and

Evaluation of Medicinal Plant as a source of safer Drug: with special reference to the Middle East area

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Historical background Principles of Phytotherapy

Safety studies: Purification, identification storage, Acute toxicity, Chronic toxicity (teratogenicity, effect on

fertility, hormone like action, liver, kidneys, CNS,

Screening of therapeutic actions:IN-vitro (in laboratory- isolated organs,IN_vivo (blood pressure, sugar, cholesterol,

Preclinical studies Standardization of the identified action on animals in the same condition as in human

Each experimental animal according to the action **Pharmaceutical formulations: Ideal formula, Stability, Activity, Costs Clinical studies:** According to the authorities regulation, According to the clinical protocols **Registration:** According to FDA and authorities regulations