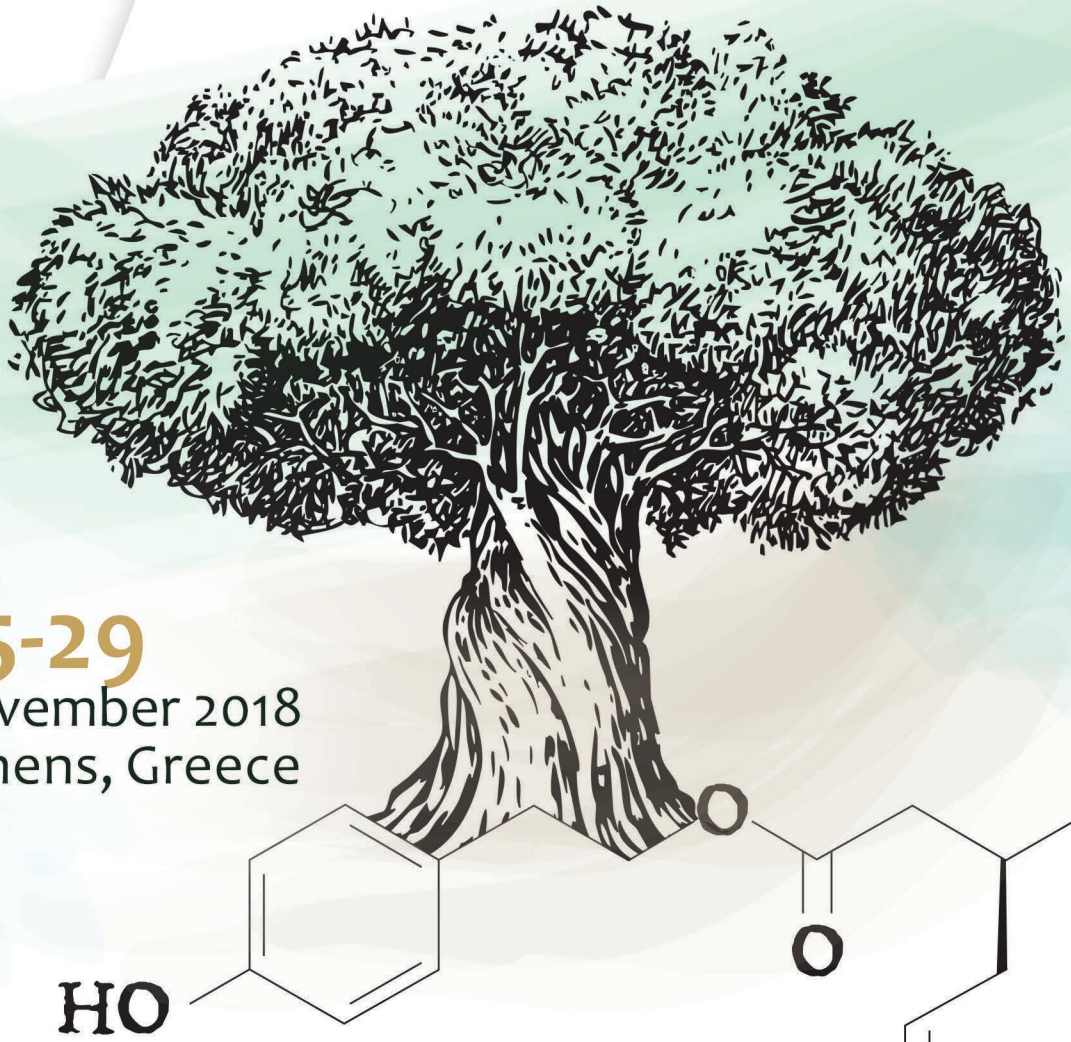
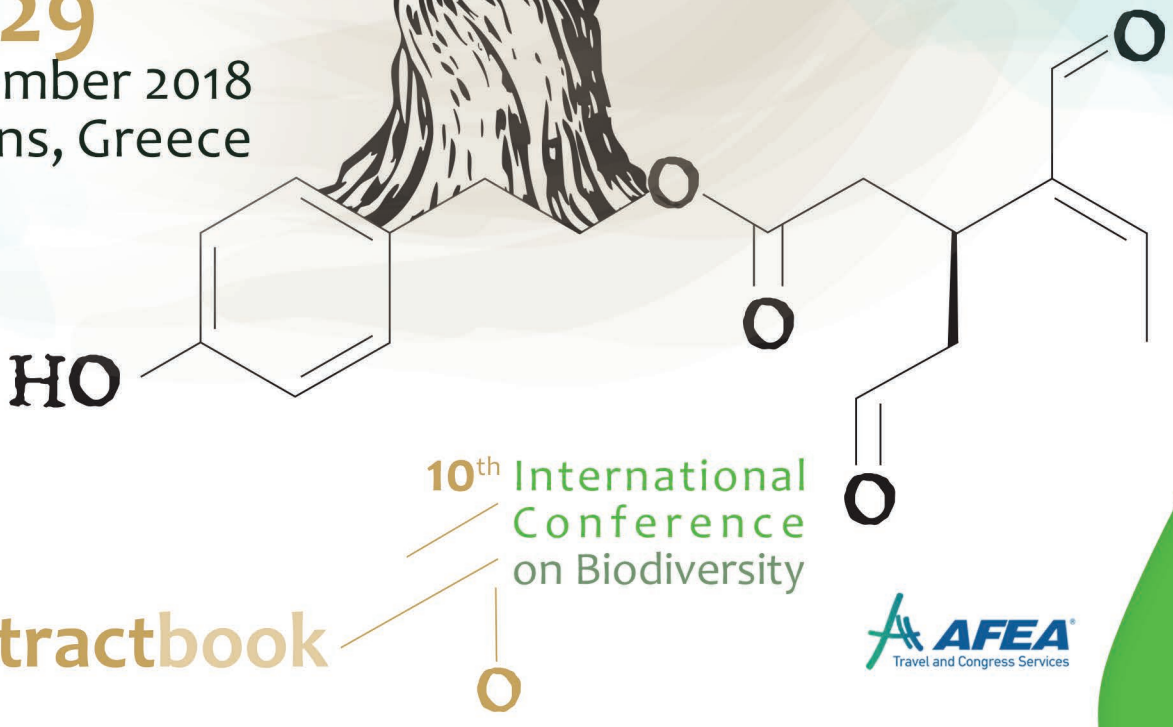
INTERNATIONAL UNION OF PURE AND APPLIED CHEMISTRY

ISCNP30 ICOB10

30th International Symposium on the Chemistry of Natural Products



25-29 November 2018
Athens, Greece



10th International Conference on Biodiversity

abstractbook

AFEA Travel and Congress Services

PL01

Total Synthesis of (−)-Nodulisporic Acids D, C and B: Evolution of a Unified Synthetic Strategy

Amos B. Smith III, Yike Zou, Xiangqin Li, Yun Yang, Simon Berritt, Jason Melvin, Stephen

PL02

Natural Products: **Current State-of-Art, Development, Perspectives and Challenges for the Brazilian Plant Species Based Bioeconomy**

Vanderlan da Silva Bolzani, Marília Valli, Mery Emili Pinto and Gustavo Monteiro

PL03

Phytoneering: From empiric traditional plant-based medicine to evidence-based phyto-pharmaceuticals

Harel Seidenwerg, Michael A. Popp

PL04

Antiadhesive Natural Products: From BabA inhibitors to Gingipain Blockers and FimH antangonists

Hensel Andreas

PL05

New frontiers in research and development of biodiversity: Global challenges and analytical opportunities.

Michael Heinrich

PL06

Evaluation of the relevance of gut microbiota on the bioactivity of natural products

Rudolf Bauer

PL07

Metabolomic profiling of anti-inflammatory coffee pulp by-products by using High Resolution Mass Spectrometry

Marialuce Maldini & Alexandre Paccou

PL08

Novel Enrichment and Separation Technologies for Natural Product Research – Applications in Phytopharmacy, Phytocosmetics and Phytonutrition

Günther K. Bonn

PL09

Evidence-based African natural medicines - exploring the synergy between ancient wisdom and modern science

Alvaro Viljoen

PL10

Deciphering the multi-specific activity of phytochemicals by network pharmacology

Thomas Efferth

PL11
Fostering the chemical diversity of natural products for the discovery of multipotent antimicrobials
Judith Maria Rollinger

PL12
Polymeric nano shuttles for advanced diagnosis and therapy
Paolo A. Netti

PL13
Marine invertebrates and associated microorganisms, a global science for a global valorization
Jamal Ouazzani

PL14
Chemodiversity and Antibiotic Activities of Natural Products from Biodiverse Marine Sediment-Derived Actinobacteria of the Philippine Archipelago
Doralyn S. Dalisay

PL15
Probing diterpenes chemical space for molecular diversity and bioactivity
Nunziatina De Tommasi

PL16
Chemical Approaches to Drug-Discovery
D. Uemura

PL17
Plant Metabolome Mining with Pharmacognosy Data: Status and Perspective in the Digital Era
Jean-Luc Wolfender, Emerson Ferreira Queiroz, Pierre Marie Allard

PL18
Metabolomics, fluxomics and omics data integration for deeper phenotype understanding in natural products and drug research
Marta Cascante

PL19
Bioactivity-directed Exploration of the Phytocannabinoid Chemical Space
Giovanni Appendino, Eduardo Muñoz, Orazio Taglialatela-Scafati, Gianpaolo Grassi

PL20
CCCM™: Natural Product Approach to Cannabis-based Therapies
Small-Howard, A.L., Shimoda, L.M.N., Reimann-Philipp, U., Stokes, AJ., Turner, H.

PL21
Phytochemicals as Anti-infectives: Opportunities to Combat Antimicrobial Resistance
Simon Gibbons

PL22
Antiproliferative compounds targeting resistant cells involved in multiple myeloma
Cuendet Muriel

PL23
Hyphenation of liquid-liquid preparative techniques and the *in vivo* zebrafish model as a platform for searching for biologically active compounds
Krystyna Skalicka-Woźniak, Barbara Budzyńska

PL24
Searching for bioactive terpenoids from Chinese medicinal herbs
Yang Ye

PL25
Redesigning plant derived taxol biosynthesis to generate marine diterpenoids in engineered bacteria.
Katarina Kemper, Monika Fuchs, Thomas B. Brück

PL26
Interspecific Interactions: Mapping the Fungal Battlefield as a Source of Chemical Diversity
Nicholas Oberlies

PL27
Natural products with anti-inflammatory activity
Stuppner H.

PL28
Renewed interest in natural products with Natural Fragment Library - A disruptive innovation programme for drug discovery.
Bruno David, Antonio Grondin, Philippe Schambel, Marc Vitorino & Denis Zeyer

PL29
Chemical profiling of natural product extracts by using NMR data combined or not with centrifugal partition chromatography
Jane Hubert, Ali Bakiri, Jean-Marc Nuzillard, Jean-Hugues Renault

PL30

PL31
Metabolomics, biotechnology and biochemometrics: Perfect holistic match?
Milen I. Georgiev

PL32
New avenues for Good Old Natural Products (NPs)
Pascal Richomme

PL33
Nature knew them since long: natural deep eutectic solvents
R. Verpoorte, Y. Choi, and G.J. Witkamp

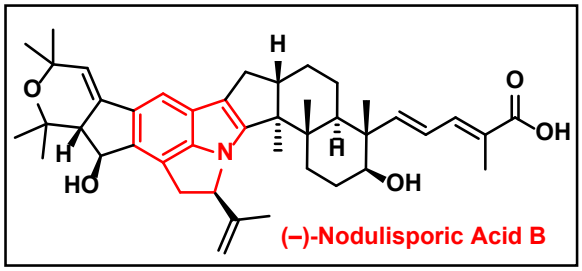
PL01

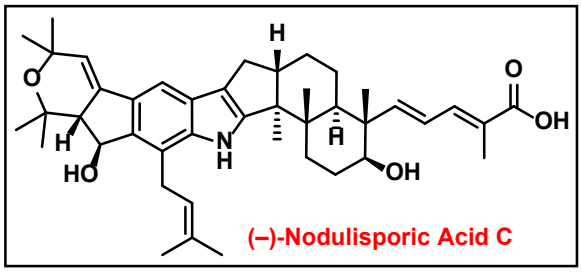
Total Synthesis of (−)-Nodulisporic Acids D, C and B: Evolution of a Unified Synthetic Strategy

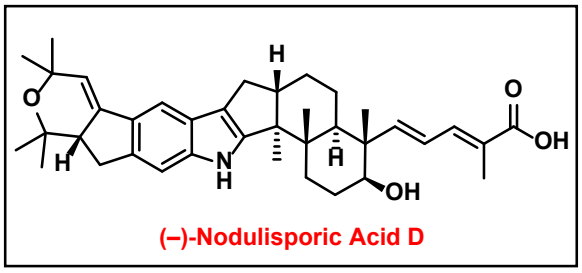
Amos B. Smith III, Yike Zou, Xiangqin Li, Yun Yang, Simon Berritt, Jason Melvin, Stephen Gonzales, Matthew Spafford

Department of Chemistry, University of Pennsylvania, Philadelphia Pa. 19104

A unified synthetic strategy leading to the total synthesis of (−)-nodulisporic acids D, C, and B is described. Key synthetic transformations include a nickel-chromium mediated cyclization, an aromatic ring functionalization employing a novel copper-promoted alkylation, a palladium-catalyzed cross coupling cascade/indole ring construction, and a palladium-mediated regio- and diastereoselective allylic substitution/cyclization reaction, the latter to construct ring-D (NIH GM-29028 NCI CA-19033).







PL02

Natural Products: Current State-of-Art, Development, Perspectives and Challenges for the Brazilian Plant Species Based Bioeconomy

Vanderlan da Silva Bolzani, Marília Valli, Mery Emili Pinto and Gustavo Monteiro

Institute of Chemistry, São Paulo State University, Araraquara, SP, Brazil,14800-900

The plant chemical diversity is fantastic, and natural product molecular structures is reflected in a large variety of biochemical reaction pathways, which is responsible for several classes of biologically active secondary metabolites. This metabolic complexity is fundamental for the communication, regulation and defense of the species, in the most diverse ecosystems, being indispensable for the balance of biodiversity and the survival of the species in terrestrial and aquatic environments. Also, plant secondary metabolites are important supplies for the production of drugs, foods, cosmetics, fragrances, colorants, and agrochemicals, which support a vigorous bioeconomy in several countries. Brazil, having one of the largest terrestrial biodiversity, has enormous potential to generate new scientific and technological knowledge, and to foster an innovative and profitable local bioeconomy. Thus, the study of rapid screening and identification of natural products by means of modern and robust analytical methods is essential for more accurate investigation on the role of natural products of tropical and equatorial plant species. Our recent work on some species of Fabaceae, Malpighiaceae, Euphorbiaceae and Violaceae have shown the importance of natural products of low molecular weight (secondary metabolites) and high molecular weight (cyclotides) for better understanding of tropical plant species, and also as potential templates for medicinal chemistry studies aimed at lead molecules. [1-4]

Financial Support: CEPID-FAPESP, CNPq-INCT

Keywords: Natural Products, Brazilian Biodiversity, New Trends, Bioproducts

References:

[1] Ferreira QMM, Queiroz EF, Zeraik ML, Samad NE, Marcourt L, Cuendet C, Castro-Gamboa I, Hamburger M, Wolfender J, Bolzani V da S. J Nat Prod 2014; 77: 650−656.
[2] Alan CP, Valli M, Dameto CA, Andricopulo AD, Freire RT, Castro I, Bolzani V da S. Sci Rep 2017; 7: 7215.
[3] Valli M, Russo HM, Bolzani V da S. An Acad Bras Cienc 2018; 90(1 Suppl. 1): 763–778.
[4] Ramalho SD, Pinto ME, Ferreira F, Bolzani V da S. Planta Med 2017; 83: 1–13.

PL03

Phytoneering: From empiric traditional plant-based medicine to evidence-based phyto-pharmaceuticals

Harel Seidenwerg, Michael A. Popp

Bionorica SE, Germany

The treatment of a wide variety of diseases using plant-based medicines is steadily gaining importance. In terms of their pharmaceutical efficacy, safety and quality today, modern, researched phytomedicines have to follow allopathic principles and find their place in evidence-based medicine. Fulfilling these requirements, researched plant-based medicines can be the preferred alternative to chemically and synthetically produced medicines.

Bionorica, located in Neumarkt (Bavaria, Germany), is one of the world's leading manufacturers of scientifically researched herbal medicines. Based on the 'Phytoneering' strategy, Bionorica decodes the extensive active ingredient potential of plants (phytos) using state-of-the-art research and technology (engineering). The result: highly effective medicines with few undesirable side effects. Bionorica uses translational science as a rapidly growing discipline in biomedical research, which aims to expedite the discovery of new diagnostic tools and treatments by using a multi-disciplinary, highly collaborative “from bench to bedside and back” approach.

One main focus of the company’s work is the research and development of plant-based medicines for the treatment of respiratory and urinary tract infections as well as for gynaecological disorders.

PL04

Antiadhesive Natural Products: From BabA inhibitors to Gingipain Blockers and FimH antangonists

Andreas Hensel

University of Münster, Institute of Pharmaceutical Biology and Phytochemistry, Münster, Germany

As resistance of many bacterial and viral pathogens against standard antibiotics is dramatically increasing, the need for development of new antiinfectives is obvious. Within different projects for identification of natural products with inhibiting activities of bacterial virulence and fitness factors, adhesion and invasion turned out to have major influence on the infection process. For this, special focus is laid on the identification of antiadhesive and antiinvasive compounds and plant extracts.

For prevention of urinary tract infections (UTI) – one of the most frequently occurring infections in Western world – antiadhesive drugs against uropathogenic *E. coli* (UPEC) were investigated, using a combination of *in vitro* experiments, animal infection studies, and human biomedical studies. Aqueous leave extracts from *Orthosiphon stamineus* turned out to inhibit strongly the adhesion of UPEC to bladder and kidney cells; *in vivo* activity in mice after oral application of the extract was comparable to that of norfloxacin. The antiadhesive effect of the extract was related to the presence of tetra- and pentamethoxylated flavons.

Extracts form Cranberry fruits (*Vaccinium macrocarpum*) has clinical be shown to prevent UTI significantly. The extracts do not influence UPEC proliferation, but interact with the FimH-Uroplactin mediated recognition and adhesion between bacteria and bladder/kidney cells. Besides a slight direct inhibition of FimH by Cranberry metabolites it was found for the first time that Cranberry extract after oral ingestion in humans stimulated significantly the kidneys to secrete higher amounts of Tamm-Horsfall Protein (THP) which binds via mannosylated sugar residues to FimH of UPEC and therefore blocks the bacterial adhesion to host cells. Therefore Cranberry fruit extract can be defined as an indirect antiadhesive medication, and therefore should be addressed better as an stimulant of the innate defense system.

Infections with *Helicobacter pylori* can be eradicated by multi-antibiotic therapy, but high recurrence rates reduce in clinical practice the long-term efficacy of this treatment. Antiadhesive supplementation should help for prevention of reinfection of the gastric epithelia after eradication therapy. The main bacterial adhesins of *H. pylori* are BabA and SabA. Potent BabA inhibitors have recently been identified (peptides, N-phenylpropenyl-aminoacids amides, pectin-like polysaccharides). One of strongest BabA inhibitors was isolated from fruits of *Abelmoschus esculents* and characterized as a highly acetylated rhamnogalacturonan (ORG1), which interacts specifically with the Lewis^b-binding domain of BabA. For improved drug targeting ORG1 has been formulated into smart liposomes, which bind specifically to gastric mucin, followed by permeation into the mucin layer were normally *H. pylori* is located; subsequently ORG1 from the liposome surface interacts with BabA of the bacterium, which again leads to the release of the antibiotic cargo from the inside of the liposome.

Porphyromonas gingivalis is one of the main pathogens responsible for the initiation and progression of periodontal diseases, which globally have an extreme high economic impact on the health systems. Development of mouth hygiene products containing natural compounds which inhibit the adhesion of the pathogen should provide a prophylactic strategy against this chronic infection. Strong inhibition of the Arg-gingipain, one of two bacterial adhesins of *P. gingivalis,* was observed by using an acetone-water extract from *Rumex acetosa*; Lys-gingipain was only inhibited to a low extend. This antiadhesive effect was due to tprocyanidin-B2-di-O-

gallate. Interstingly, this compound also blocked the protease activity of the bacterium, which implies that the main virulence factor gets impaired by this proanthocyanidin. Clinical study of a mouth wash solution, containing 0.8% of the extract, revealed significant effects on the clinical parameters.

Summarizing, antiadhesive natural compounds reveal an innovative strategy to combat bacterial virulence factors for advanced anti-infective therapy.

PL05

New frontiers in research and development of biodiversity: Global challenges and analytical opportunities.

Michael Heinrich

Pharmacognosy and Phytotherapy, UCL School of Pharmacy, London, U.K.; E-mail: m.heinrich@ucl.ac.uk Phone: +44 (0) 207 753 5844

In the debate about our human use of biodiversity, the success of new medicines like galanthamine from Amarylidaceae species and ingenol 3-angelate (or PEP005), an unusual diterpene ester isolated from *Euphorbia peplus* L. are often used to showcase opportunities of drug development. Less often mentioned, but of major economic importance are novel food supplements, cosmetics and other high value products sourced from the world's biodiversity. However, the complex obstacles including sustainable supply as well as access and benefit sharing are crucial foundations for modern programmes. Success is also hampered by the scientific challenges in this field calling for the development of rigorous guidelines of best practice. I will use examples from our research to highlight both what is needed for best practice, and how we can successfully understand the biological and chemical complexity of medicinal plants

For example, traditional Chinese medicine preparations are generally prescribed as a formula comprised of multiple botanical drugs that are expected to exert clinical effects based on the combined effects of multiple components against multiple targets. While diverse new tools have emerged which enable a better understanding of potential benefits and risks of such preparations, a key challenge lies in the transdisciplinary nature of such research including a series of challenges

1) The chemical complexity of the preparations both in botanical and chemical terms (i.e. in pharmacognostic) terms needs to be understood and assessed
2) The treatment strategy is based on very different medical and philosophical concepts of Traditional Chinese Medicine (TCM) and this needs to be understood in order to allow for a use of these preparations in a different therapeutic context
3) Once these challenges have been overcome, one needs to understand the potential benefits of these preparations and how this is linked to their chemical profile.
4) The clinical use will require a sustainable supply of high-quality and consistent drug material and a GxP production system

In this presentation, I will address these challenges looking both at the variable chemical composition of the starting material (e.g. Booker et al 2016) and on how to better understand complex preparations. The latter is exemplified in a hexaherbal formulation we have examined recently. The TCM formulation includes the rootstock of *Scutellaria baicalensis* Georgi, *Rheum tanguticum* Maxim. ex Balf., *Sophora flavescens* Aiton; root bark of *Dictamnus dasycarpus* Turcz.; bark of *Phellodendron chinense* C.K. Schneid. and fruit *of Kochia scoparia* (L.) Schrad. Importantly, *R tanguticum*, *S. flavescens, P. chinense* and *S. baicalensis,* contributed the majority of the decoction's extracted metabolites (Cheng et al 2016). Using a multivariate chemometric approach in combination with a series of in vitro targets we identified compounds likely to be responsible for these activities.

More broadly, the presentation will discuss strategies for developing complex medicines further and on how these can contribute to new / improved treatments and better healthcare.

References:

[1] Booker, A, Zhai L, Gkouva C, Li S, Heinrich M. (2016) Front Pharmacol (Ethnopharmacol) 7:254. doi: 10.3389/fphar.2016.00254
[2] Chang J, Lane M, Yang M, Heinrich M.Planta Med 2016; 82:1134–1141.

PL06

Evaluation of the relevance of gut microbiota on the bioactivity of natural products

Rudolf Bauer

Institute of Pharmaceutical Sciences, Department of Pharmacognosy, University of Graz, Universitätsplatz 4, 8010 Graz, Austria

There has been always the idea, that the intestinal bacteria have huge impact on our health [1]. However, only in recent years research started to provide evidence, that gut microbiota and the human body form a symbiosis which finally contributes essentially to our well-being. Dysbiosis of our guts can contribute to systemic inflammation, and can lead to obesity, asthma, diabetes, autoimmune diseases, and even certain forms of cancer. Gut bacteria also appear to help food processing functions by producing signaling chemicals that regulate our appetite, satiety, and digestion [2].

However, intestinal bacteria also metabolize constituents of food, dietary supplements, and herbal medicinal products and may form active compounds. Tannins have been shown to be transformed to urolithins by human intestinal *Gordonibacter* species [3], which may explain their anti-inflammatory activity, and berberine, which is an active principle of *Coptis chinensis* rhizomes has been shown to be transformed by gut microbiota into its intestine-absorbable form [4]. By using a recently established combined LC-HRMS and 16s RNA sequencing research platform, we could demonstrate that also constituents in willow bark extract, like flavone glycosides, flavanes, and caffeic acid derivatives, are metabolized heavily by intestinal microbiota [5].

Therefore, it is quite likely that activity of herbal preparations is modulated by the gut microbiome, and research on active principles has also to include metabolites produced in the intestinal tract.

Keywords: gut microbiom, natural products, metabolism

References:

[1] Deweerdt S. Nature 2014; 508: S61–S63
[2] Li H, Zhou M, Zhao A, Jia W. Phytother Res 2009; 23(9): 1332–5.
[3] Piwowarski JP, Granica S, Kiss AK. Planta Med 2014; 80(11): 887–895.
[4] Feng R, Shou JW, Zhao ZX, He CY, Ma C, Huang M, Fu J, Tan XS, Li XY, Wen BY, Chen X, Yang XY, Ren G, Lin Y, Chen Y, You XF, Wang Y, Jiang JD. Sci Rep 2015; 5: 12155.
[5] Pferschy-Wenzig EM, Koskinen K, Moissl-Eichinger C, Bauer R Front Pharmacol 2017; 8:893. doi: 10.3389/fphar.2017.00893.

PL07

Metabolomic profiling of anti-inflammatory coffee pulp by-products by using High Resolution Mass Spectrometry

Marialuce Maldini & Alexandre Paccou

-OMICS is a general term for a broad discipline of science and engineering for analyzing the interactions of biological information objects in various -omes. Metabolomics, as a methodology for measuring small-molecule metabolite profiles has become an important component of systems biology. Because of the comprehensive nature of metabolite measurement and the capacity to detect subtle changes in a large dataset, metabolomics has found broad application.
Coffee is the second traded food commodity in the world. Beyond roasted seeds, the most part of the original fruit -and in particular pulp- is discarded as waste, with severe environmental and economic consequences in many developing countries. Pulp coffee by-product has been recently reported to possess an interesting anti-inflammatory activity [1], thus a metabolomics profiling of the such phytocomplexes is needed to determine compounds potentially responsible for the high biological activity. In this context, High Resolution Mass Spectrometry is required to identify and characterize with high confidence secondary metabolites occurring in a complex sample.
The TripleTOF® systems can collect high resolution MS/MS spectra at high MS/MS acquisition rates and have excellent low mass sensitivity, making the ideal instruments for metabolomics workflows.
In addition, improved, easy to use software, methods and libraries custom-designed for untargeted and targeted customer applications are available. The breadth of data acquisition capabilities is been improved by SWATH® Acquisition, MRMHR acquisition, information dependent high-resolution MS acquisition (IDA), and high speed MS/MS scanning.

References:
[1] Magoni *et al.* Food Res Int 2018; 112: 129–135.

PL08

Novel Enrichment and Separation Technologies for Natural Product Research – Applications in Phytopharmacy, Phytocosmetics and Phytonutrition

Günther K. Bonn[1,2]

[1] Institute of Analytical Chemistry and Radiochemistry, Leopold-Franzens University of Innsbruck, Innrain 80-82, 6020 Innsbruck, Austria; [2] ADSI - Austrian Drug Screening Institute, Innrain 66a, 6020 Innsbruck, Austria.

The application of novel enrichment and separation technologies for natural product research have become increasingly important and new advancements in chromatography allow exploring inaccessible areas of natural product isolation. Special knowledge and technologies are necessary to perform analysis of plant ingredients as they normally contain hundreds or thousands of small low molecular weight ingredients in very different concentrations. The achievements in natural product research are largely based on the constant development of highly selective and sensitive analytical technologies. In this regard novel enrichment and purification methods based on modern solid-phase extraction (SPE) technologies are applied to reduce the complexity of plant-extracts, while µ-HPLC is used for separation, pre-concentration and fractionation. The opportunity to hyphenate these techniques to robotic systems permits high-throughput screening. Significant progress has been made in the development of novel stationary phases which can be tailored to a specific application, allowing endless possibilities in terms of selectivity tuning. Further hyphenation to high-resolution mass spectrometry facilitates the identification and quantitation of active components in natural products. An alternative offline coupling to MALDI-TOF/MS allows easy determination of active compounds using novel matrix-free approaches. Moreover, the combination of separation science with spectroscopy represents an attempt to combine different technologies in phytopharmacy and food analysis. Near and mid infrared (NIR and MIR) spectroscopy enable a fast and simultaneous qualitative and quantitative analysis of raw plants and liquid extracts without destruction. On the other hand, infrared imaging can be used to study the distribution of active ingredients in plant materials with a resolution of down to 5 µm. The described new analytical techniques are demonstrated and discussed by several applications in medicine, phytopharmacy, phytocosmetics and nutrition science. All presented technologies have been successfully applied within the recently established Phytovalley® - Tyrol platform, where science meets nature in the heart of the Alps.

PL09

Evidence-based African natural medicines - exploring the synergy between ancient wisdom and modern science

Alvaro Viljoen

Department of Pharmaceutical Sciences and SAMRC Herbal Drugs Research Unit, Tshwane University of Technology, Pretoria, 175 Nelson Mandela Drive, Private Bag X680, Pretoria, 0001, South Africa

Southern Africa harbours an impressive floral diversity and ranks as one of the most biodiverse countries in the world. Interweaved within this botanical tapestry is a cultural heritage characterised by rich indigenous knowledge systems (IKS) which have moulded one of the oldest healing modalities, African Traditional Medicines (ATM). This unique blend of medicinal plant use and IKS has created a unique research opportunity in enthnopharmacology. Over the past 20 years our group has endeavoured to provide a scientific rationale for medicinal plant use through an evidence-based research approach of traditional medicines. Several examples will be presented to demonstrate the challenging yet rewarding workflow to explore the chemistry and biological properties of the ethnomedicinal flora of South Africa. Using various *in vitro* and *in vivo* approaches, complemented by analytical methods and multivariate data analysis we aim to contribute to the fundamental research base required to convert these botanical assets into tangible consumer products. The various challenges facing translation research and the standardisation of ATMs will be highlighted.

PL10

Deciphering the multi-specific activity of phytochemicals by network pharmacology

Thomas Efferth

Johannes Gutenberg University, Mainz, Germany

To combat complex systemic diseases that harbour robust biological networks such as cancer, single target intervention is proved to be ineffective. In such cases, network pharmacology approaches are highly useful, because they differ from conventional drug discovery by addressing the ability of drugs to target numerous proteins or networks involved in a disease. Pleiotropic natural products are one of the promising strategies due to their multi-targeting and due to lower side effects. In this review, we discuss the application of network pharmacology for cancer drug discovery. We provide an overview of the current state of knowledge on network pharmacology, focus on different technical approaches and implications for cancer therapy (e.g. polypharmacology and synthetic lethality), and illustrate the therapeutic potential with selected examples from herbal mixtures, medicinal herbs and isolated phytochemicals. Finally, we present future perspectives on their plausible applications for diagnosis and therapy of cancer.

Selected papers:

Kadioglu O…., Efferth T. Biochem Pharmacol. 2014;87:399-409.
Zhao Q, …., Efferth T. Int J Cancer. 2015 ;137(6):1446-56.
Panossian A, …., Efferth T. Phytomedicine. 2015;22:981-92
Saeed M, …., Efferth T. J Nutr Biochem. 2015;26:44-56.
Kadioglu O, …., Efferth T. Arch Toxicol. 2016;90:575-88.
Poornima P, …., Efferth T. Pharmacol Res. 2016;111:290-302.
Efferth T et al. Oncotarget. 2017;8:50284-50304
Hong C, …., Efferth T. Sci Rep. 2017;7:11551.
Hamdoun S, …, Efferth T. Biochem Pharmacol. 2017;146:63-73.

PL11

Fostering the chemical diversity of natural products for the discovery of multipotent antimicrobials

Judith Maria Rollinger

Department of Pharmacognosy, Vienna, Austria

One hundred years after the devastating Spanish flu, low respiratory tract infections (LRT) remain the most deadly communicable disease, causing 3.2 million deaths annually worldwide [1]. Mainly caused by influenza viruses and picornaviruses, the mortality rate is heavily boosted by bacterial coinfection, referred to as lethal synergism [2].
During a recently accomplished screening campaign for natural products active against LRT related viruses, we identified several herbal agents and thereof derived natural compounds with neuraminidase-inhibiting activity and multipotent antiviral activities [3].
Here, I will present strategies, which were used for the rationalized discovery of antimicrobial lead structures. They encompass (i) chemoinformatics, (ii) information from ethnopharmacology, and (iii) the use of complementary assays (target-based and phenotypic assays). The combination of these approaches will be demonstrated by some recently performed studies, which succeeded in identifying natural compounds with multipotent antimicrobial activities [4-7].

Acknowledgement: This work was supported by the Austrian Science Fund (FWF P 24587) and the Wilhelm Doerenkamp Foundation (Natvantage Grant)

References:

[1] http://www.who.int/mediacentre/factsheets/¬fs310/en/
[2] McCullers JA. Nat Rev Microbiol 2014; 12: 252–262.
[3] Grienke U, Mair CE, Kirchmair J, Schmidtke M, Rollinger JM. Planta Med 2018; 84: 684–695.
[4] Grienke U, Braun H, Seidel N, Kirchmair J, Richter M, Krumbholz A, von Grafenstein S, Liedl KR, Schmidtke M, Rollinger JM. J Nat Prod 2014; 77: 563–70.
[5] Grienke U, Schmidtke M, Kirchmair J, Pfarr K, Wutzler P, Durrwald R, Wolber G, Liedl KR, Stuppner H, Rollinger JM. J Med Chem 2010; 53: 778–86.
[6] Grienke U, Richter M, Walther E, Hoffmann A, Kirchmair J, Makarov V, Nietzsche S, Schmidtke M, Rollinger JM. Sci Rep 2016; 6: 27156.
[7] Mair CE, Grienke U, Wilhelm A, Urban E, Zehl M, Schmidtke M, Rollinger JM. J Nat Prod 2018; 81: 515–523.

PL12

Polymeric nano shuttles for advanced diagnosis and therapy

Paolo A. Netti

Centre for Advanced Biomaterials for Health Care, Istituto Italiano di Tecnologia, Naples, Italy

Interdisciplinary Research Centre on Biomedical Materials, University of Naples Federico II, Naples, Italy

Current advances in nanotechnology hold the promises to greatly impact on current medical practice. Since nanometric materials interact with cells, tissue and organs at a molecular level, they may be used as probes for ultrasensitive molecular sensing and diagnostic imaging or carriers for drug and gene delivery. However, along with the excitement that has driven the development of novel nanocarriers, there have been increasing concerns regarding the risks these materials may generate. As these nanostructures are intentionally engineered to target specific cells or tissues, it is imperative to ensure their safety. The optimal design of safe and functional nanocarriers for medicine requires a better understanding of the interaction between the physical-chemistry properties of the nanoparticle surface with the complex protein machinery existing at the cell membrane. In particular the effect of the particles properties (charge, shape, protein coating) on the mechanism of cellular uptake is highly relevant both to assess the real biological risks coupled with the use of nanomaterial (nanopathology and nanotoxicology) and to engineer carriers able to improve the medical practice. The nanometric size and the surface molecular decoration may activate mechanisms of cellular uptake different from those commonly used by cells: these open the possibility to activated/modulated the membrane crossing by tuning chemical-physical properties of nanometric materials.

In this talk, the design and production of novel degradable polymeric nanocavities via layer-by-layer and phase separation technology will be presented along with a detailed characterization of their in vitro and in vivo performances. Furthermore, possible mechanisms of cellular uptake will be discussed and critically presented. The effect of surface bioconjugation on cell and tissues targeting efficacy will be exploited and elucidated.

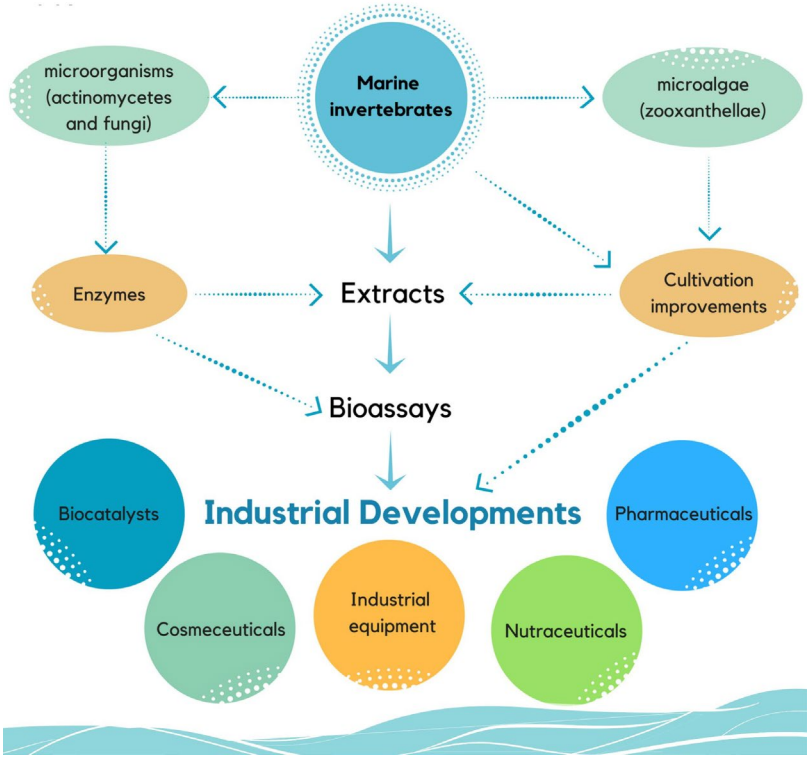
PL13

Marine invertebrates and associatedmicroorganisms, a global science for a global valorization.

Jamal Ouazzani

Research Director, TASCMAR Coordinator, Centre National de la Recherche Scientifique CNRS, Institut de Chimie des Substances Naturelles ICSN, France.

Coral reefs extend as deep as 150 meters and with the development of new technologies to go deeper, scientists are beginning to explore 'Mesophotic Coral Ecosystems' (MCEs) existing in low light levels, which still allows for photosynthesis. Mesophotic Coral Ecosystems (MCEs) are almost entirely unexplored, they are a treasure---trove for discovering new species and their associated bioactive chemical compounds. Organisms such as soft corals, sponges, and microbes living on coral reefs naturally produce potent cocktails of chemicals to defend themselves from competitors and harmful predators. EU---funded Horizon 2020 project TASCMAR, which aims to tackle major bottlenecks in the discovery, development, and commercialization of marine---derived chemical compounds with a specific focus on using new biological and chemical resources from MCEs. The project partners are working on developing innovative technologies for the sustainable cultivation of marine resources, e.g.
through the isolation of chemicals in their natural environment without the need to harvest them. The project is specifically looking for new chemical compounds active against age---related illnesses such as Alzheimer's, Parkinson's, cancer, and aging diseases related to muscles and skin.



PL14

Chemodiversity and Antibiotic Activities of Natural Products from Biodiverse Marine Sediment-Derived Actinobacteria of the Philippine Archipelago

Doralyn S. Dalisay

Center for Chemical Biology and Biotechnology, University of San Agustin, Gen. Luna Street, Iloilo City, Philippines 5000

The extent of microbial biodiversity in marine sediments is emerging as a rich reservoir and relatively untapped resource of novel bioactive natural products with potential pharmaceutical applications. Associated with this biodiversity is the inherent "chemical engines" producing a plethora of diverse bioactive natural products ranging from small molecules to complex secondary metabolites. These natural products continue to be a source of inspiration for biomedical research and drug discovery. This lecture will discuss the biodiversity of culturable actinobacteria thriving in marine sediments of Philippine archipelago and assessing their ability to produce new chemical scaffold or novel bioactive compounds for multidrug resistant bacterial infections.

PL15

Probing diterpenes chemical space for molecular diversity and bioactivity

Nunziatina De Tommasi

Department of Pharmacy, University of Salerno, via Giovanni Paolo II, 132, 84084 Fisciano (SA), Italy,

The use of preparations from medicinal plants for the treatment of diseases is an important part of human culture, and they also provide leads for drug discovery programs.
Specialized plant metabolites play an important role in the discovery of new chemical entities owing to their ecological role, chemical diversity, high affinity and specificity to biomolecules. The diversity of molecular skeletons formed via plant diterpene pathways offers a rich source of known and potentially new drugs, modified by evolution to fit better with cellular targets [1]. This could explain recent reports that indicate that multi-target engagement occurs in up to 80% of plant molecules [2]. Diterpenes constitute a large group of natural products that play diverse functional roles in plants as hormones, photosynthetic pigments, attractants for pollinators, herbivore repellents and phytotoxins [3]. These compounds showed a wide range of biological activities such as antimalarial, anti-inflammatory, antidiabetic, antibacterial, cytotoxic, insecticidal, and several others activity [2]. Moreover, diterpenes show a wide range of targets (heat shock proteins, growth factor, transcription factors, enzymes) and qualify as privileged structure for biomedical research [3]. In the last few years our research group was involved in a research project aimed to the search of diterpenes with significant biological functions and/or unknown chemical structures from plants variety [4,5].
Owing to our interest in the field of plant diterpenes, we have developed approaches to target or lead(s) identification based on chemical genetic procedures, supported by spectroscopic and spectrometric data [4,5]. The most promising diterpenes then underwent an evaluation of the target(s) modulatory activity by means of a panel of chemical and biological approaches, including STD-NMR, SPR measurements, biochemical and cellular assays, limited proteolysis, and molecular docking. Our strategies and studies in the described research area, are demonstrated by several results from recent and ongoing research projects.

Keywords: Diterpenes, NMR, MS, biological activity

References:

[1] Zhi-Da M. Diterpenes. Introduction to Natural Products Chemistry. Edited by Xu. Rensheng; Ye, Yang; Zhao, Weimin (2012),101-123
[2] Schenone M, Dančík V, Wagner BK, Clemons AP. Nat Chem Biol 2013; 9: 232–240.
[3] Hanson JR. Nat Prod Rep; 2005; 22: 594–602.
[4] Dal Piaz F, Cotugno R, Lepore L, Vassallo A, Malafronte N, Lauro G, Bifulco G, Belisario MA, De Tommasi N. J Proteomics 2013; 82: 14–26.
[5] Dal Piaz F, Vera Saltos MB, Franceschelli S, Forte G, Marzocco S, Tuccinardi T, Poli G, Ebrahimi SN, Hamburger M, De Tommasi N, Braca A. J Nat Prod 2016; 79: 2681–2692.

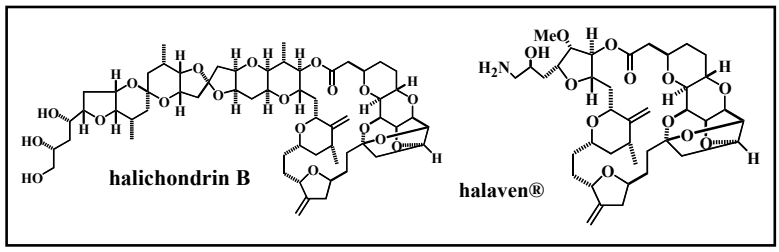
PL16

Chemical Approaches to Drug-Discovery

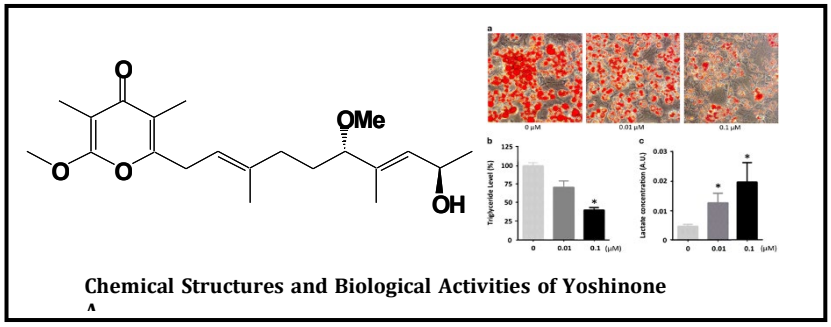
D. Uemura (Nagoya University, Professor Emeritus)

Department of Chemistry, Kanagawa University, Hiratsuka, Kanagawa, Japan,

Many compounds with surprisingly unique structures and brilliant biological activities have been identified from marine organisms. It is abundantly clear that natural products and their derivatives are promising candidates for treatment of human diseases. In this presentation, I will highlight some fascinating natural products as promising drug leads. Halichondrin B is a good example of a natural product that has been studied as a drug lead. This polyether macrolide was first isolated from the black sponge *Halichondria okadai* in 1986 and was shown to have antitumor activity [1,2]. The natural product showed a novel mechanism of action that disrupts the dynamics of tubulin polymerization, which makes it an interesting candidate for cancer treatment. However, developmental studies were hindered because of its limited availability. The total synthesis of halichondrin B in 1992 led to a breakthrough. This achievement made available a sufficient amount of material for further studies and made possible an examination of the structure-activity relationship, which revealed that a macrocyclic lactone moiety is essential for this activity. Finally, the analogue of this moiety is now available on the market as the breast cancer drug, Halaven®.



Obesity is a growing health problem in modern society because it is a risk factor for many lifestyle-related diseases, including diabetes and cardiovascular disorders. Thus, studies on anti-obesity are expected to contribute to the prevention and treatment of various diseases. To propose promising drug candidates, we tried to identify the natural product which controls fatty acid metabolism and explication of those mechanisms. Yoshinone A, a γ-pyrone containing-natural product derived from a cyanobacterium *Leptolyngbya* sp. collected in Ishigaki Island, was first identified as an inhibitor of fat cell differentiation of 3T3-L1 cellS[3,4]. We examined the effect of yoshinone A to adiposities and found that the compound decreased the triglyceride level in the cells. Furthermore, administration of yoshinone A derivative to mice with high fat-diet clearly demonstrated that the compound inhibited weight increase in vivo. These findings suggested that yoshinone A might be a potent drug lead to treat obesity.



Chemical Structures and Biological Activities of Yoshinone A

Discovery of novel substances are important steps in the scientific process. Both the structures and biological activities of recently isolated natural products frequently exhibit great novelty, however, many natural products with tremendous biological applications are yet to be discovered. The symposium is expected to be a forum for the discussion of such topics. And then, I will present some new topics.

References:

[1] Hirata Y, Uemura D. Pure Appl Chem 1986; 58: 701.
[2] Uemura D, Proceeding of the Japan Academy 2010; Series B 86 190.
[3] Inuzuka T, Yamamoto K *et al.* Tetrahedron Lett 2014; 55: 6711.
[4] Koyama T, Kawazoe Y, Iwasaki A, Ohno O, Suenaga K, Uemura D. J Antibiot 2016; 69: 348.

PL17

Plant Metabolome Mining with Pharmacognosy Data: Status and Perspective in the Digital Era

Jean-Luc Wolfender, Emerson Ferreira Queiroz, Pierre Marie Allard

School of Pharmaceutical Sciences, University of Geneva, University of Lausanne, CMU – Rue Michel Servet 1, 1211 Geneva 11, Switzerland

The rapid innovations in metabolite profiling, bioassays and chemometrics lead to a paradigm shift in natural product (NP) research. Indeed, having at hand partial/full structure information of possibly all secondary metabolites and an estimation of their levels, provides a way to perform pharmacognostic investigations from a new and holistic perspective. The increasing amount of accurate metabolome data that can be acquired on massive sample sets, notably through data dependent HRMS/MS, allows mapping natural extracts at an unprecedented precision level [1,2]. In this context, data contextualization is however still a lagging process [3].

For this, we investigated methods that could provide enhanced annotation confidence level through multiple scores integrating taxonomy information and molecular network (MN) structural consistency as well as other orthogonal analytical data. Benchmarking of such approaches is currently assessed by profiling mixtures of herbs with well-studied composition. We also investigate the best way to integrate extracts bioactivity data in MN and shortcut bioactivity guided isolation for an efficient targeted identification of bioactive NPs [4]. To this end, chromatography gradient methods at various scales have been developed for MS-targeted purification of biomarkers and their de novo structure identification by NMR.

Different recent applications of our metabolomics/phytochemical investigations will illustrate these aspects and especially bioactive natural products prioritization using massive multi-informational molecular mass spectrometry networks in combination orthogonal NMR profiling approaches will be exemplified.

Evaluation of what is already implemented and is still required in NP research will be made, notably in terms of contextualization of the data. Such type of organization of flux of research data and implementation of more integrated systems, can represent a novel epistemological framework of knowledge acquisition in pharmacognosy.

Acknowledgements: JLW is grateful to the Swiss National Science Foundation (SNF) for supporting their natural product metabolomics projects (grants nos.: 310030E-164289 and 31003A_163424).

Keywords: Dereplication, metabolite profiling, metabolomics, MS-targeted isolation

References:

[1] Wolfender J-L *et al*. J Chromatogr A 2015; 1382: 136–164.
[2] Allard PM *et al*. Anal Chem 2016 ; 88: 3317–23.
[3] Allard PM *et al*. Curr Opin Biotechnol 2018 ; 54 : 57–64.
[4] Olivon F *et al.* ACS Chem Biol 2017; 12: 2644–2651.

PL18

Metabolomics, fluxomics and omics data integration for deeper phenotype understanding in natural products and drug research

Marta Cascante

Department of Biochemistry and Molecular Biomedicine and IBUB, Universitat de Barcelona, Barcelona and Centro de Investigación Biomédica en Red en Enfermedades Hepáticas y Digestivas (CIBER-EHD), Madrid, Spain

The metabolome of an organism is the closest representative of its phenotype as the end-product of the cellular processes and is closely tied to its physiology and environment. Metabolism is commonly studied from an integrate perspective in combination with the information from gene and protein expression to consider the entire metabolic network and its regulations. To this end, computational systems biology integrates experimental and computational approaches to predict and describe the complex behavior of biological systems. Metabolic reprogramming is one of the hallmarks of cancer that enable tumor cells to fulfill their needs for energy and building blocks for biosynthesis to sustain their increased proliferation rate. In addition, the metabolic switch in tumor cells is associated with drug resistance in cancer therapy. Consequently, the study of tumor metabolism is of crucial importance to develop and implement selective cancer therapeutics that slow tumor growth and progression, improve treatment response, and overcome therapeutic resistance.
Here we show that understanding the effect of natural antioxidants on colon cancer cell metabolism give clues to predict the underlying mechanisms of the protective effect of fiber and polyphenolic compounds on the development of colon cancer in humans. Moreover, we show that grape antioxidant dietary fiber (GADF), which incorporates both fiber and phenolic compounds, is able to inhibit intestinal tumor development in ApcMin/+ mice, a genetically predisposed animal model for human colon cancer. We found that GADF treatment induced a significant reduction of the total number of small intestine tumors compared to the control. Comparison of microarray expression profiles of GADF-treated and non-treated mice revealed 183 genes that were differentially expressed and that might explain its antitumorigenic effect against the spontaneous intestinal polyposis in ApcMin/+ mice. GADF modulated clusters of genes involved in cell cycle, death, immune signaling, replication, adhesion and differentiation. Taken together, our findings suggest that GADF obtained from grape by-products could be an effective chemopreventive agent against colorectal cancer,
The results we present pave the way for the 'omics data integration' as a tool for the systematic investigation of molecular mechanisms underlying beneficial properties of natural products. We expect that a systematic characterization of the mechanisms underlying natural products action will be a powerful tool to rationally design the best combinations to target tumor metabolic reprogramming.

PL19

Bioactivity-directed Exploration of the Phytocannabinoid Chemical Space

Giovanni Appendino[1], Eduardo Muñoz[2], Orazio Taglialatela-Scafati[3], Gianpaolo Grassi[4]

[1]Dipartimento di Scienze del Farmaco, Largo Donegani 2, 28100 Novara, Italy, [2]Instituto Maimónides de Investigación Biomédica de Córdoba (IMIBIC), Córdoba, España, [3]Dipartimento di Farmacia, Università di Napoli Federico II, Via Montesano 49, 80131 Napoli, Italy, [4]CREA-CI, Viale G. Amendola 82, 45100 Rovigo, Italy.

Phytocannabinoids, a class of over 200 meroterpenoids, show a surprising diversity of macromolecular targets and qualify as privileged structures for biomedical research [1]. However, their chemical and biological space has so far been systematically investigated only around the so-called "big four", that is, the major non-native (decarboxylated) constituents of the plant (delta9-THC, CBD, CBG and CBD), and mostly in the context of a single end-point, the narcotic receptor CB1. Capitalizing on the increased availability of cannabinoids by isolation (strain selection, advances in separation techniques) and synthesis, we have systematically explored the chemical and biological space around the native forms of phytocannabinoids and a selection of minor analogues representative of the biosynthetic exuberance of *Cannabis*, focusing mostly on targets of relevance for the non-narcotic modulation of the endocannabinoid system and on biological profiles of relevance for chronic degenerative diseases[2]. Novel chemistry and bioactivities were discovered, and cannabinoquinoids emerged from these studies as interesting pleiotropic candidates for further development [3]. Clarification on the confused issues of the stereoisomeric purity of delta9-THC in *Cannabis*, of its possible generation during smoking from non-narcotic precursors, and on the puzzling reactivity of CBC will also be presented.

Keywords: *Cannabis sativa* L, phytocannabinoids, cannabinoquinoids

References:

[1] Hanuš LO, Meyer SM, Muñoz E, Taglialatela-Scafati O, Appendino G. Nat Prod Rep 2016; 33: 1357–1392.
[2] Nadal X, Del Río C, Casano S, Palomares B, Ferreiro-Vera C, Navarrete C, Sánchez-Carnerero C, Cantarero I, Bellido ML, Meyer S, Morello G, Appendino G, Muñoz E. Br J Pharmacol 2017; 174: 4263–4276.
[3] del Río C, Navarrete C, Collado JA, Bellido ML, Gómez-Cañas M, Pollastro F, Appendino G, Calzado MA, Cantarero I, Muñoz E. Sci Rep 2016; 6: 21703.

PL20

CCCM™: Natural Product Approach to Cannabis-based Therapies

Small-Howard, A.L.[1], Shimoda, L.M.N.[2], Reimann-Philipp, U.[1], Stokes, AJ[3]., Turner, H[2].

[1]GB Sciences, Inc., Life Sciences Dept., 3550 W. Teco Ave., Las Vegas, NV 89118, [2]Laboratory of Immunology and Signal Transduction, Chaminade University, Honolulu, Hawaii, [3]Laboratory of Experimental Medicine, John A. Burns School of Medicine, Honolulu, Hawaii

GB Sciences uses a novel, whole plant approach to discovering proprietary formulations of cannabis-derived compounds that show promise for the treatment of specific diseases. GB Sciences has focused its efforts on finding therapies for patient groups that are largely underserved, and they are using novel delivery methods to ensure bioavailability and to provide time-released versions of their patent-pending, cannabinoid-containing complex mixtures (CCCM™). Although many cannabis researchers and biopharma companies have focused on the activities of single cannabinoids, GB Sciences leverages powerful molecular synergies derived from whole plant extracts that are then further-refined and standardized. Most other cannabis-based biopharmaceutical companies are studying one of the two most abundant cannabinoids in the plant, either tetrahydrocannabinol (THC) or cannabidiol (CBD); whereas, GB Sciences embraces naturally occurring mixtures of approximately 480 bioactive compounds in the cannabis plant. GB Sciences has demonstrated correlations between extracts from different "chemovars", chemical variations of the cannabis plant, and potential therapeutic efficacy for different specific human diseases. Our drug discovery process combines: 1) high throughput screening of tens of thousands of combinations of compounds derived from specific chemovars of the cannabis plant in well-established cellular models of diseases and 2) a proprietary network pharmacology algorithm for the prediction of complex therapeutic mixtures. By carefully screening the contributions of the individual compounds within our disease-specific chemovars, we discovered our patent-pending CCCM™ for the treatment of neurodegenerative disorders, inflammatory disorders, cardiovascular disorders, and chronic/neuropathic pain. Translating natural product complexity to the clinic requires the ability to produce safe and consistent chemical mixtures from a plant source, which is now possible through advances in tissue culture and the use of programable, cleanroom-type growing environments. GB Sciences' combination of novel cultivation technologies and robust drug discovery engine herald a bright future for safe and effective Cannabis-based medicines.

Keywords: *Cannabis*, cannabinoid, terpenoid, molecular synergies, chemovar, drug discovery, natural products

PL21

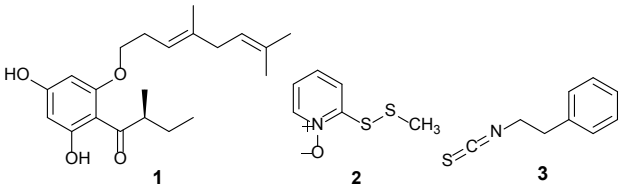
Phytochemicals as Anti-infectives: Opportunities to Combat Antimicrobial Resistance

Simon Gibbons

Research Department of Pharmaceutical and Biological Chemistry, UCL School of Pharmacy, 29-39 Brunswick Square, London WC1N 1AX, United Kingdom

New approaches are desperately needed to find novel chemotypes with activity against multidrug-resistant bacteria. Little investment from pharmaceutical companies is currently available as the cost to benefit ratio is small compared to medicines for other illnesses such as inflammation, cancer and depression. Opportunities to find new anti-infectives from nature are still enormous especially given that bacteria, fungi and plants produce an incredible array of structurally diverse and highly functional chemistries.

Many phytochemicals such as compound **1** have activity against Gram-positive bacteria [1] but are inactive against Gram-negatives, due to their inability to cross the cell wall and the presence of multidrug efflux proteins. This is exacerbated by the finding that natural products that are active toward Gram-negatives often have poor selectivity and are broadly toxic to mammalian cells for example, the *Allium* natural product **2**. Our work focuses on natural products that may modify bacterial resistance, through either inhibiting antibiotic efflux [2], blocking the transfer of plasmids from a donor bacterium to a recipient (**3**) [3], and natural products that inhibit the formation of biofilms, for example in mycobacteria.



Latterly, we have also investigated hot and cold spring sources from the UK as a source of bacteria that produce antibiotics. Recent typing of the genera in these springs shows a large and highly varied group of taxa that have not been studied for antibiotic discovery. This lecture will give an overview of our efforts to find anti-infectives from nature with examples of some of our work to date.

References:

[1] Shiu WK *et al*. Int J Antimicrob Agents 2013; 42: 513-.
[2] Danquah CA *et al*. Sci Rep 2018; 8: 1150-.
[3] Kwapong A *et al*. Tetrahedron Lett 2018; 59: 1952-.

PL22

Antiproliferative compounds targeting resistant cells involved in multiple myeloma

Cuendet Muriel

School of pharmaceutical sciences, University of Geneva, University of Lausanne, Rue Michel-Servet 1, CH-1211 Geneva 4, Switzerland.

Multiple myeloma (MM) is a blood disease characterized by the clonal proliferation of malignant plasma cells in the bone marrow. Despite not having a strong incidence, this type of cancer is associated with a high rate of relapse and resistance to conventional therapies. The paradigm that tumors are composed of heterogeneous cell populations, namely tumoral cells and cancer stem cells (CSCs), imposes to address the effect of compounds in each cell population. The CSC theory hypothesizes that this subset of tumor cells exhibits self-renewal properties and differentiation capabilities, and is equipped with detoxification tools rendering CSCs highly resistant. Those cells are believed to be capable of replenishing the tumor, and to be responsible for tumor relapse. In order to evaluate the effect of potential MM treatments, a model that closely represents the disease should be used. Therefore, an *in vitro* model consisting of 3D co-culture spheroids was set up to better predict *in vivo* activity. The spheroids contain CSCs, malignant plasma cells and mesenchymal stem cells (MSCs). This model presents most of the tumor complexity and also includes the interactions with the microenvironment, here represented by the inclusion of MSCs. This aspect is of importance as MSCs change their phenotype when in contact with MM cells, by helping the tumor cells to survive and even to become resistant to therapy. Moreover, the addition of CSCs mimics the aggressiveness displayed by *in vivo* refractory tumors. By differentially marking each cell type, the compound activity on the various cell populations and the interaction between them can be evaluated. This presentation will cover the effect of compounds targeting sensitive as well as resistant multiple myeloma cells and highlight some of their mechanism of action.

PL23

Hyphenation of liquid-liquid preparative techniques and the *in vivo* zebrafish model as a platform for searching for biologically active compounds

Krystyna Skalicka-Woźniak[1], Barbara Budzyńska[2]

[1]Department of Pharmacognosy with Medicinal Plant Unit, Medical University of Lublin, 1 Chodzki Str, 20-093 Lublin, Poland, [2]Department of Pharmacology with Pharmacodynamic, Medical University of Lublin, 4a Chodzki Str, 20-093 Lublin, Poland

Nature has been a source of therapeutic agents for thousands of years, and an impressive number of modern drugs have been derived from natural sources, many of them based on their use in traditional medicine. Yet only a small fraction of the world's biodiversity has been explored for even a single bioactivity to date. The development of new, specific, and efficient chromatographic methods permits the easy screening and identification of potential drug leads [1]. Countercurrent separation, as a liquid-liquid technique, has a special place in the isolation of natural products and has been broadly applied for the separation and purification of chemical compounds in complex matrices due to its unique advantages.

The zebrafish *in vivo* model is being increasingly used in key areas of neuroscience, such as neuropharmacology, neural development and regeneration, behavioral neuroscience, and the study of neural disease. The zebrafish share a homologous genome with vertebrate species (rodents and humans), as well as brain modelling, structure and function. They also have numerous similar neural and physiological systems (for example the stress regulating system) as vertebrates and share with them some key receptors that play an important role in the etiology of neurological disorders. The model is ideally suited for large-scale analysis [2,3].

Epilepsy is a chronic disorder of the brain that affects more than 50 million people of all ages worldwide. 70% respond to treatment, however the other 30% suffer from resistance [4]. Anxiety has been defined as a significant physiological and behavioral response generated to avoid harm and enhance the chances of survival [3]. According to the WHO at least one-third of the population in many countries experience at least one episode of pathologic anxiety. Some synthetic drugs are known for their side effects and finding molecules acting towards different CNS disorders is therefore an important task.

The zebrafish epilepsy model with seizures induced by the $GABA_A$ antagonist pentylenetetrazole (PTZ) as well as light-dark transitions to monitor the isolation of anticonvulsant principles and those related to anxiety-like behavior will be presented during this lecture [2,3]. Compounds from different classes such as alkaloids, coumarins, terpenoids were tested after bioactivity guided isolation approach. Aspects of modern efficient isolation will be also discussed.

Acknowledgments: The work was financed from grant No 2017/27/B/NZ4/00917 from The National Science Centre, Kraków, Poland.

References:

[1] Sarker SD, Latif Z, Gray AI, 2006. Natural Product isolation. In: Sarker, Satyajit D., Latif, Zahid, Gray, Alexander I. (Eds.), Natural Products Isolation. –, 2nd ed. Humana Press Inc., Totowa, New Jersey, pp. 1–26.
[2] Peng X, Lin J, Zhu Y, Liu X, Zhang Y, Ji Y, Li Q. Pharmacol, Biochem Behav 2016; 145: 55–65.
[3] Guo S. Brain and Behavior 2004; 3(2): 63–74.
[4] WHO Epilepsy Fact Sheet No. 999, 2018, World Health Organization (WHO).

PL24

Searching for bioactive terpenoids from Chinese medicinal herbs

Yang Ye

State Key Laboratory of Drug Research and Natural Products Chemistry Department, Shanghai Institute of Materia Medica, Chinese Academy of Sciences, 555 Zu-Chong-Zhi Road, Zhangjiang Hi-Tech Park, Shanghai 201203, China

Natural products have been playing an important role in new drug discovery due to their highly diverse structures and biological activities. Searching for bioactive compounds from medicinal herbs with long clinical practice has been proven a shortcut for new leads. Terpenoids constitute a large, widespread and diverse group of natural products with a spectrum of bioactivities such as anti-bacterial, anti-malarial, anti-inflammation, anti-cancer and so on. Bioactive terpenoids are a hot topic in the field of natural products.

A systematic investigation of unique diterpenoids from a medicinal herb *Podocarpus nagi* has been carried out, resulting in the isolation of 44 nor-diterpenoid lactones with 16 of them being new, especially a novel binor-diterpenoid with dehydroxylation at C-7. Notably, most of isolated diterpenoid lactones displayed remarkable potency to increase LDL uptake in HepG2 cells at a concentration of 5 μM. *In vivo* study further showed that nagilactone B, an abundant diterpene existing in this plant, dose-dependently decreased TC, TG, and LDL levels in high fat diet induced dyslipidemic hamsters. Mechanism study indicated nagilactone B exerted lipid-lowering effect by elevating LDLR mRNA and protein level. It is the first time to report anti-hyperlipidemic activity for naturally occurring diterpene lactones *in vitro* and *in vivo*. Nagilactone B represent a new type of compounds with promising lipid-lowering activity.

PL25

Redesigning plant derived taxol biosynthesis to generate marine diterpenoids in engineered bacteria.

Katarina Kemper, Monika Fuchs, Thomas B. Brück

Werner Siemens-Chair of Synthetic Biology, Technical University of Munich, Department of Chemistry, Lichtenbergstraße 4, 85748 Garching

Terpenes are ubiquitous natural products that are found across all kingdoms of life. With over 50.000 characterized compounds terpenes are also the largest and most structurally diverse family of secondary metabolites. Most terpenoids constitute a streochemically complex macrocylic core, which is generated by C-C bond coupling of aliphatic oligoprenyl precusors. This reaction is catalyzed by the enigmatic family of terpene synthase (TPS) enzyme, which are capable of chaperoning highly reactive carbocation intermediates through an enzyme specific reaction sequence. Downstream functionalization of the macrocycles by oxidoreductases, hydrolases and transferases generate highly elaborate structures, which display cytotoxic, antimicrobial, insecticidal, antioxidant and anti-inflammatory activities. Low concentrations in the natural source and complex chemical synthesis commonly induce a supply issue for higher terpene type bioactives, such as sesqui- (C15) and diterpenes (C20). Heterologous production with engineered microbial hosts is a well-established alternative to classical exploration of essential oil extracts or chemical synthesis especially regarding sustainability factors and waste accumulation. Transfer of biosynthetic enzymes from the natural pathways into genetically tracable production systems is a standard procedure for establishing designed biosynthetic routes. To that end, advances in Enzyme-Structure elucidation and modelling techniques alongside with synthetic biology tools provide significant progresses for engineering and optimization.

Previous work on the structure-function-relations in the active site of the taxadiene synthase from *Taxus brevifolia* revealed lining amino acids and thereby allowed the generation of several new diterpenes through site-directed mutagenesis [1]. By implementing these tools on biotechnological production of structurally complex (di)terpenes we were not only able to broaden the product spectrum of the taxadiene synthase from *Taxus brevifolia* but also shift it with very high specificity to the former minor byproduct Verticilliatriene. Derivatives of this bicyclic diterpene can be extracted from soft coral [2] or plants [3] and are found to exhibit anti-inflammatory and anti-tumoral properties amongst others. Following the requirement for sustainable and environmentally friendly production, expanding and altering the product landscape of one single, well studied diterpene synthase consequently omit time-consuming and tedious extraction of marine products and their respective biosynthetic enzymes from natural sources.

Novel whole-cell biocatalysis of verticilliatrienes entailed mutagenesis of the taxadiene synthase (TXS) and integration into an established *E.coli* production host. Technical scale up of the fermentation procedures provided product titers up to 300 mg/L. Subsequent application of an optimized HPLC-protocol on the raw extract provided us with three isomeric verticillenes that were structurally elucidated via NMR. Since functionalization of carbon scaffolds is generally essential for bioactivity of a compound, the isomeric pure fractions were employed in chemical and chemo-enzymatical reactions. Results from these reactions and the bioproduct of the diterpene educt shall be presented in this talk.

Acknowledgments: KK, MF and TB gratefully acknowledge the support of the SysBioTerp project funded by the Federal Ministry of Education and Research in the Systems Biology funding framework (BMBF; Grant number: 031A305A)

Keywords: enzyme engineering, marine natural products, heterologous production

References:

[1] Schrepfer P, Buettner A, Goerner C, Hertel M, van Rijn J, Wallrapp F, Eisenreich W, Sieber V, Kourist R, Bruck T. Proc Natl Acad Sci USA 2016; 113: E958-967.

[2] a) Cheng SY, Lin EH, Wen ZH, Chiang MY, Duh CY. Chem Pharm Bull (Tokyo) 2010; 58: 848–851; b) Wang SS, Cheng YB, Lin YC, Liaw CC, Chang JY, Kuo YH, Shen YC. Mar Drugs 2015; 13: 5796–5814.

[3] Nagashima F, Wakayama K, Ioka Y, Asakawa Y. Chem Pharm Bull 2008; 56: 1184–1188.

PL26

Interspecific Interactions: Mapping the Fungal Battlefield as a Source of Chemical Diversity

Nicholas Oberlies

[1]*University Of North Carolina At Greensboro, Greensboro, United States*

A common question in the field of natural products research is: why did that organism choose to biosynthesize those compounds? Of course, the simple answer is that we, as humans, don't really know. However, the common postulate is that the secondary metabolites give the organism some sort of advantage, particularly with respect to chemical defense. If true, can we then set up experiments where organisms must 'fight' for their turf, essentially using co-culturing as a way to force the production of secondary metabolites, perhaps causing the amplification of production and/or the stimulation of otherwise silent biosynthetic gene clusters. Using a series of tools that profile the chemistry of fungal cultures in situ, our team has been pursuing these questions, both to probe some of the basics of fungal ecology and biology, as well as, to potentially generate new chemical diversity. This talk will explain some of the underlying tools used to assess the chemistry of fungal (and other microbial) cultures via mass spectrometry, and then apply those skills and databases to understanding fungal chemistry *in situ*.

PL27

Natural products with anti-inflammatory activity

Stuppner H.

Institute of Pharmacy/Pharmacognosy, Center for Molecular Biosciences Innsbruck (CMBI), University of Innsbruck, Innsbruck, Austria

Inflammation is the body's response to noxious stimuli and conditions, such as infection and tissue injury. It is characterized by immune, vascular and cellular biochemical reactions, including different cells, enzymes, cytokines, eicosanoids and transcription factors. Inflammation starts with the migration of immune cells from blood vessels and the release of mediators, followed by the recruitment of inflammatory cells and the release of reactive oxygen species (ROS), reactive nitrogen species (RNS) and pro-inflammatory cytokines [1]. The goal is to eliminate foreign pathogens, resolve infection and repair injured tissues. It is generally thought that a controlled inflammatory response is beneficial. However, deregulation might lead to chronic inflammatory that are associated with diverse pathological conditions e.g. arthritis, atherosclerosis, the metabolic syndrome, sepsis, allergies and auto-immune diseases or even cancer. The search for new alternatives to interfere with key players in inflammation has revealed nature to be an abundant source. In the course of our screening program to discover bioactive natural compounds with anti-inflammatory properties, we investigated *Himatanthus sucuuba*, an Amazonian plant [2,3] and *Dracaena cambodiana*, a medicinal plant distributed in China, Cambodia as well as Vietnam. Both plants are being used traditionally to alleviate various diseases including inflammation. The major aim of this work was to identify the active principles and to determine the mechanism of action. Most recent results of our research efforts within this collaborative research project will be presented.

References:

[1] Medzhitov R. Nature 2008; 454: 428–435.
[2] Fakhrudin N, Waltenberger B, Cabaravdic M, Atanasov AG, Malainer C, Schachner D, Heiss EH, Liu R, Noha SM, Grzywacz AM, Mihaly-Bison J, Awad EM, Schuster D, Breuss JM, Rollinger JM, Bochkov V, Stuppner H, Dirsch VM. Br J Pharmacol 2014; 171: 1676–1686.
[3] Heiss EH, Liu R, Waltenberger B, Khan S, Schachner D, Kollmann P, Zimmermann K, Cabaravdic M, Uhrin P, Stuppner H, Breuss JM, Atanasov AG, Dirsch VM. Sci Rep 2016; 6: 20771.

PL28

Renewed interest in natural products with Natural Fragment Library - A disruptive innovation programme for drug discovery.

Bruno David[1], Antonio Grondin[1], Philippe Schambel[1], Marc Vitorino[2] & Denis Zeyer[2]

[1]Institut de Recherche Pierre Fabre 3, Avenue Hubert Curien, BP 13562 31035, Toulouse, France, [2]NovAliX, 850 Boulevard Sébastien Brant BP 30170, 67405 Illkirch, France

Pierre Fabre Laboratories (PFL), the 2nd largest dermo-cosmetics laboratory worldwide, the 2nd largest independent French pharmaceutical group has specialized in plants and natural substances since its founding in the 60's [1]. PFL has set up the largest private library of dried ground plant samples in the world in terms of size and diversity for drug discovery programs. As the High Throughput Screening (HTS) approach with medium size molecules from this plant library has revealed to be time and means consuming [2], PFL has recently decided to study natural plant fragments and to build a "Natural Fragment Library".
The fragment based drug discovery approach [3] involves small, weakly binding molecules with molecular masses between 150 and 250 Da. This method is fundamentally different from HTS in almost every aspect: size of the compound library, screening methods, work to be done from hit to lead. Natural fragment based drug discovery appears to be a promising and emerging field, and it offers valuable prospects for developing new drugs. These molecules, with their unique three-dimensional properties, their high Fsp^3 represent potential opportunities to provide specific interactions with targeted proteins.
In the framework of our Open Innovation initiatives, this unique library of fragments is made available for our partners' customers who can broaden the scope of their test campaigns by accessing a great diversity of innovative fragments [4]. PFL made the strategic decision to work with NovAliX, an international partner specialized in drug discovery through biophysical methods.
The promising results of this new collaborative approach between Pierre Fabre and NovAliX on two targets (NUR77 and Keap-1) will be presented.

Acknowledgements: Caroline CASTANO, Nathalie TEYSSEYRE, Corentin BEDARD, Christophe LONG & Ozlem SEVIC

Keywords: Fragment Based Drug Discovery, Natural Fragment Library,

References:

[1] https://www.pierre-fabre.com/en
[2] David B, Ausseil F. High-throughput screening of vegetal natural substances. In Handbook of Chemical and Biological Plant Analytical Methods (2014) John Wiley Sons, Ltd. Chapter 44, 987-1010.
[3] Erlanson DA *et al*. Nat Rev Drug Discovery 2016; 15: 605–619.
[4] https://nature-open-library-pierre-fabre.force.com/index.

PL29

Chemical profiling of natural product extracts by using NMR data combined or not with centrifugal partition chromatography

Jane Hubert, Ali Bakiri, Jean-Marc Nuzillard, Jean-Hugues Renault

Institut of Molecular Chemistry of Reims – UMR CNRS 7312 SFR Cap'Santé University of Reims Champagne-Ardenne

The commonly adopted workflow in natural product chemistry involves the isolation of the compounds from organism extracts. Even if modern analytical and purification techniques are routinely available in most laboratories, a considerable work taking several weeks or years is still necessary to isolate and elucidate individual metabolite structures from crude natural extracts. Moreover, in numerous cases, the systematic purification of individual natural product involving time-consuming multi-step purification procedures results in a considerable waste of time, especially since natural product chemists have often the opportunity to rediscover compounds that have already been discovered and well described in literature [1].
At the same time, many industrial sectors, such as cosmetics or nutraceutics, are developing products based on natural extracts. They are looking for powerful tools to accelerate times to market despite the reinforcement of regulatory constraints related to the chemical composition of natural active ingredients (REACH legislation, EU Cosmetics Directive 76/768/EEC, US FDA cosmetics section, for instance).
In this context, three complementary different strategies mainly based on Nuclear Magnetic Resonance data combined to chemometric tools will be presented and illustrated with academic or industrial examples. The first one combines a fractionation step by solid support-free liquid-liquid technique, ^{13}C NMR spectroscopy, Hierarchical Clustering Analysis (HCA) and ^{13}C NMR chemical shift databasing for the direct identification of natural metabolites within mixtures [2]. The second one concerns a computer-aided ^{13}C NMR dereplication workflow that does not require any pre-fractionation of the investigated sample in order to rapidly identify the major constituents of a natural mixture [3]. The last one is based on a new HMBC-based dereplication method that uses a networking approach for the deconvolution of complex 2D-NMR spectra of metabolite mixtures. This method exploits the ability of HMBC and HSQC experiments to provide connectivity information between 1H and 13C atoms located at their vicinity. [4]

References:

[1] Hubert J, Nuzillard J-M, Renault J-H. Phytochem Rev; 2015, DOI 10.1007/s11101-015-9448-7
[2] Hubert J, Nuzillard J-M, Purson S, Hamzaoui M, Borie N, Reynaud R, Renault J-H. Anal Chem 2014; 86: 2955–2962.
[3] Bakiri A, Hubert J, Reynaud R, Lanthony S, Harakat D, Renault J-H., Nuzillard J-M. J Nat Prod 2017; 80: 1387–1396.
[4] Bakiri A, Hubert J, Reynaud R, Lambert C, Martinez A, Renault J-H, Nuzillard J-M. J Chem Inf Model 2018, DOI: 10.1021/acs.jcim.7b00653

PL30

Antitumor Metal Complexes Based on Traditional Chinese Medicines Active Ingredient Alkaloids and Their Derivatives

Zhen-Feng Chen[1], Prof. Dr. Yan-Cheng Liu[1], Associate Prof. Dr. Ke-Bin Huang[1], Prof. Dr. Hong Liang[1]

[1]*Guangxi Normal University, Guilin, China*

Alkaloids are active ingredients of traditional Chinese medicines, and possess wide range of bioactivity. We put forward an idea of using the coordination regulating effect of Chinese medicine active ingredients and metal active center to design metal-based drugs. Targeting G4-DNA, telomerase, DNA, we synthesized a series of antitumor metal complexes of traditional Chinese medicines active ingredient alkaloids oxoaporphine, oxoisoaporphine, β-carboline, and tetrahydroisoquinolineas well as their derivatives. We obtained ten leading compounds with high in vivo anticancer activity and good in vivo safety. [1-12] These leading compounds exhibited multi-targeting and multi-mechanism features, which provide the possibility to overcome the resistance of metal-based antitumor drug.

Acknowledgements: This work was supported by NNSF of China (Nos. 21401031, 21431001), IRT_16R15, and NSF of Guangxi Province (No. 2016GXNSFGA380005) as well as "BAGUI Scholar" program of Guangxi of China.

Keywords: traditional Chinese medicines active ingredients, alkaloids, metal complexes, antitumour activity, action mechanism

References:

[1] Ke-Bin Huang, Zhen-Feng Chen, et al., J. Med. Chem., 2018, 61: 3478.
[2] Zhen-Feng Chen, Hong Liang, et al. J. Med. Chem., 2015, 58: 2159.
[3] Zhen-Feng Chen, Hong Liang, et al. J. Med. Chem., 2015, 58: 4771.
[4] Jian-Hua Wei, Zhen-Feng Chen, et al. Dalton Trans., 2015, 44: 11408.
[5] Ke-Bin Huang, Zhen-Feng Chen, et al. Eur. J. Med. Chem., 2015, 100: 68.
[6] Qi-Pin Qin, Zhen-Feng Chen, et al. Eur. J. Med. Chem., 2016, 124: 380.
[7] Jiao-Lan Qin, Zhen-Feng Chen, et al. Eur. J. Med. Chem., 2016, 124: 417.
[8] Qi-Pin Qin, Zhen-Feng Chen, et al. Scientific Reports, 2016, 6: 37644.
[9] Jiao-Lan Qin, Zhen-Feng Chen, et al. Scientific Reports, 2017, 7: 46056.
[10] Qi-Pin Qin, Zhen-Feng Chen, et al. Oncotarget, 2017, 8: 619827.
[11] Jiao-Lan Qin, Zhen-Feng Chen, et al. Oncotarget, 2017, 8: 59359.
[12] Zu-Zhuang Wei, Zhen-Feng Chen, et al. Eur. J. Med. Chem., 2018, 145: 360.

PL31

Metabolomics, biotechnology and biochemometrics: Perfect holistic match?

Milen I. Georgiev [1,2]

[1] The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences, Plovdiv, Bulgaria, [2]Center of Plant Systems Biology and Biotechnology, Plovdiv, Bulgaria

Since time immemorial plants have long had a central role in the treatment of a wide spectrum of diseases, hence continuously supporting the health of human populations. Nowadays, in excess of 25% of modern medicines are derived (either directly or indirectly) from plants. Artemisinin (antimalarial), paclitaxel (antineoplastic), codeine and morphine (analgesic), and galanthamine (reversible cholinesterase inhibitor) are suitable examples in this direction and amongst the best-selling drugs worldwide. Recently in the USA, for instance, two new drug applications have been approved for marketing botanical products as prescription drugs, namely Veregen (a topical drug for the treatment of genital and perianal warts) and Mytesi (an oral drug for the treatment of HIV/AIDS related diarrhea). These new drug approvals are remarkable examples that complex botanical mixtures can be developed as new drugs in order to meet FDA standards [1-3, and the literature cited therein].
At the same time the development of new leads/drugs is rather costly, laborious and time-consuming, hence platforms for accelerated lead finding/drug discovery, mode of action of healing herbs and their sustainable production are continuously sought.
Here an overview of the metabolomics, pharmacological (*in vitro* and *in vivo* studies) and bioprocessing aspects of research on selected medicinal plants towards accelerated lead finding will be given and discussed [4-9].

Acknowledgements: Author acknowledges the financial support of the European Union's Horizon 2020 research and innovation programme, project PlantaSYST (SGA-CSA No. 739582 under FPA No. 664620).

References:

[1] De Luca V *et al*. Science 2012; 336: 1658–1661.
[2] George D *et al*. The Lancet 2016; 387: 220–221.
[3] Georgiev MI. Phytochem Rev 2016; 15: 511–513.
[4] Marchev A *et al*. Food Chem Toxicol 2017; 108: 419–428.
[5] Dimitrova P *et al*. Food Chem Toxicol 2018; 111: 605–615.
[6] Hidalgo D *et al*. Sci Rep 2017; 7: 17976.
[7] Fierascu I *et al*. Sci Rep 2017; 7: 12428.
[8] Zhang K *et al*. J Exp Bot 2018; 69: 1955–1966.
[9] Dimitrova P *et al*. Phytomedicine 2018, DOI: 10.1016/j.phymed.2018.07.013.

PL32

New avenues for Good Old Natural Products (NPs)

Pascal Richomme

University of Angers, Dpt of Pharmacy, SONAS/SFR QUASAV, Campus du végétal, 42 rue Georges Morel 49070 Beaucouzé, France

NPs remain a great source of inspiration when developing analytical methods as well as bioactive compounds. We will illustrate this statement by presenting three concrete examples. The rapid and targeted chemical characterization of complex mixtures of secondary plant metabolites has become essential to many areas of Nps research such as dereplication studies, metabolomics profiling, and quality control. In this respect high-performance liquid chromatography coupled with mass spectrometry (HPLC-MS^n) is commonly considered as method of first choice, while others such as matrix assisted matrix laser desorption ionization (MALDI) as well as matrix free laser desorption ionization (LDI) are hardly discussed. As an introduction, the present lecture will highlight some recent advances in MALDI and LDI such as the development of highly selective MALDI matrices for the detection of alkaloids as well as the matrix free LDI detection of UV absorbing Nps. Indeed MALDI as well as LDI-MS may provide useful complements or alternatives to classic analytical approaches in NPs' research.

Conventional chemical fungicides commonly used to protect crops against fungal infections present the drawback to be highly pollutant for the environment, especially for soil and water supply, whereas these products may be highly toxic for humans. The so-called "*Alternaria* Leaf Spot" is a common disease of crucifers caused by the fungal pathogen *A. brassicicola* which affects different crops including cabbage, kale, Brussels sprout, cauliflower and broccoli. Indole phytoalexins camalexin and brassinin play *in planta* a key role in crop protection against this necrotrophic agent. However it has been shown that mutants become phytoalexin-resistant by activating at least three signaling pathways named as Cell Wall Integrity (CWI), High Osmolarity Glycerol (HOG) and Unfolded Protein Response (UPR). The latter is particularly involved in the fungus protection against phytoalexins since UPR deficient avirulent mutants of *A. brassicicola* appear as hypersensitive to camalexin and brassinin. Since very few UPR inhibitors such as the synthetic STF-083010 are known we decided to develop an original screening assay, detecting the production of a HAC1 fluorescence-induced protein, *i. e.* a transcriptional activator involved in the UPR pathway, in *Saccharomyces cerevisiae* cultures. The preliminary screening of an in-house NPs library [*c.a.* 70 compounds] revealed four compounds as potential UPR inhibitors. Finally, RT-PCR validated the true inhibitory effect of 2-deprenyl-rheediaxanthone B and griffipavixanthone which clearly appeared as good candidates for inhibiting UPR and, consequently, alternative crop protections.

The last example deals with functional lipidomic, *i. e.* a combination of comprehensive lipid mediator profiling together with mechanistic and cell-biological studies aiming at unravelling the molecular mechanism of bioactive agents. This approach was developed during a fruitful collaborative project, involving different European universities, focusing on the role of vitamin E (α/β/γ/δ-tocopherol/tocotrienol), which inhibits pro-inflammatory leukotriene formation and protects from leukotriene-related diseases (*e.g.,* asthma, cardiovascular disease and cancer) but with controversial clinical evidence. As far as 5-lipoxygenase (5-LO) activity was concerned, it was shown that ω-oxidation of vitamin E derivatives leads to a variety of long-chain hydroxy- and -carboxy metabolites which potently inhibited the enzyme. Further structural optimization revealed garcinoic acid (δ-tocotrienolic acid, GA) -isolated from *G. kola* seeds- as one of the most potent inhibitors of 5-LO within this series. GA was then used as a scaffold to semisynthetize potent specific 5-LO inhibitors.

PL33

Nature knew them since long: natural deep eutectic solvents

R. Verpoorte[1], Y. Choi[1], and G.J. Witkamp[2]

[1]Natural Products Laboratory, IBL, Leiden University, [2]Kluyver Laboratory, Delft University of Technology

Through NMR-base metabolomics we found that in all kind of extracts of microbial, mammalian and plant cells certain organic bases, organic acids, amino acids, sugars and sugar alcohol occur in relatively large amounts. Much larger in fact than expected on the basis of these compounds being intermediates in primary metabolism. Their ubiquitous occurrence gave us the thought that they must have a function. Based on this our first hypothesis was the possibility of ionic liquids formed by the organic bases like choline and betaine with organic acid like malic acid. The first experiments confirmed this. Further studies resulted in finding that various combinations of the mentioned compounds do give deep eutectic solvents, i.e. mixing these solid crystalline compounds in certain molar ratios results in liquids at room temperature. More than 150 combinations now have been characterized. We named them Natural Deep Eutectic Solvents (NADES). They can be divided into the following groups:

- Organic bases with organic acids: ionic liquids
- Organic bases with neutral compounds like poly alcohols and sugars
- Organic acids with neutral compounds like poly alcohols and sugars
- Amino acids with neutral compounds like poly alcohols and sugars
- Mixtures of neutral compounds like poly alcohols and sugars

By ^{1}HNMR it was shown that the NADES components are strongly bound via H-bonding, in some cases H_2O is part of these liquid crystal-like structures. The NMR also shows that with dilution with water gradually the interaction between the compounds disappears. NADES are excellent solvents for medium polar compounds, such as most secondary metabolites. In our view biosynthesis of poorly water soluble compounds occurs in NADES, e.g. attached to cellular membranes, where the polar charged head groups of the membrane lipids act as anchors for NADES in which enzymes and intermediates are dissolved. Also the ER and vesicles could be formed by metastable systems of lipids and NADES. All ingredients for NADES are found in resurrection plants, lichen etc. Also in drought or cold resistant plants typical NADES ingredients are found to be present. In terms of application the NADES are excellent solvents for both small molecules and macromolecules (proteins, polysaccharides, DNA, etc.) in which the compounds show better solubility and stability than in the currently used organic solvents. NADES thus are of interest as non-toxic, non-explosive, sustainable green solvents for many industrial applications.

References:

[1] Choi YH, van Spronsen J, Dai Y, Verberne M, Hollmann F, Arends IWCE, Witkamp GJ, Verpoorte R. Plant Physiol 2011; 156: 1701–1715.
[2] Dai Y, Witkamp GJ, Verpoorte R, Choi YH. Anal Chem. 2013; 85: 6272–6278.
[3] Dai Y, van Spronsen J, Witkamp GJ, Verpoorte R, Choi YH. Anal Chim Acta 2013; 766: 61–68.

Workshop: MediHealth EU grant

WS-L1

Metabolic activation of natural products: some case studies
Luc Pieters

WS-L2

Natural products that modulate the lipid mediator class switch in inflammation
Werz Oliver

WS-L3

Dual pathway activation by natural compounds as key to breast cancer prevention?
Günter Vollmer

Workshop: TASCMAR

WS-L4

Mesophotic marine habitats: understudied biodiversity
Yehuda Benayahu**,** Erez Shoham, Ronen Liberman, Shai Tamir, Carolina Alonso. Pedro Álvarez, Suchana Chavanich, Anne Bialecki, Géraldine Le Goff, Jamal Qauazzani

WS-L5

Methodologies and technologies engaged in the TASCMAR project
Géraldine Le Goff

WS-L6

Natural Products in the fight against ageing and age-related diseases; the marine mesophotic zone as a SOURCE for the discovery of NOVEL bioactive molecules
Jamal Ouazzani, Nikolas Fokialakis, Christina Cheimonidi, Eleni-Dimitra Papanagnou, Aimilia Sklirou, Eirini Baira, Géraldine Le Goff, Pinelopi Vlachou, Nikolaos Tsafantakis, Konstantinos Gardikis, Pedro Álvarez, Suchana Chavanich, Anne Bialecki, Yehuda Benayahu, Christine Wenzkowski, Ioannis P. Trougakos

Workshop: Olive-Net

WS-L7

Assessment of the antioxidant profile from different Greek olive oil varieties.
Paraskevi Kouka, Grigoria Tsakiri, Dimitra Tzortzi, Sofia Dimopoulou, Apostolis Angelis, Panagiotis Stathopoulos, Maria Halabalaki, Alexios-Leandros Skaltsounis, Demetrios Kouretas

WS-L8

The European representatives of Oleaceae family as a source of compounds with anti-inflammatory activity
Anna K. Kiss

WS-L9

NMR based comprehensive screening of food materials
Andrea Steck

WS-L10

Application of different mass spectrometric methods to determine the authenticity of olive oil
Carsten Baessmann

Workshop: Natural Products and Cosmetics

WS-L11
NMR applications in cosmetics
Andrea Steck

WS-L12
Green Extraction of food, cosmetic ingredients ad natural products: moving from academia to innovative and large-scale applications.
Anne-Sylvie Fabiano-Tixier, Farid Chemat

WS-L13
Marine products as effective raw materials for cosmetics – a research and innovation approach
Konstantinos Gardikis, Sofia Letsiou

WS-L14
Natural products: The future of cosmetics
Julia Moesslacher, Valentina Scalfari, Astrid Huber-Seidel, Roland Kohl

WS-L15
The revival of an ancient perfume of the Mycenaean era
Lena Philippou

Workshop: MediHealth EU grant

Description

MediHealth is a European project funded by the Commission of the European Community (H2020—Marie Skłodowska-Curie Actions—Research and Innovation Staff Exchange (RISE), grant No. 691158). The project is based on an exchange of researchers between nine universities and four companies from European and non-European countries, exploiting the existing complementary multidisciplinary expertise. The main goal of this project is to introduce a novel approach for the discovery of active agents of food plants from the Mediterranean diet and other global sources that promote healthy ageing. To achieve this goal, a series of plants from the Mediterranean diet and food plants from other origins are carefully selected and subjected to in silico, cell-based, in vivo (flies and mice models), and metabolism analyses. Advanced analytical techniques complement the bio-evaluation process for the efficient isolation and identification of the bioactive plant constituents. Furthermore, pharmacological profiling of bioactive natural products, as well as the identification and synthesis of their metabolites, is carried out. Finally, optimization studies are performed in order to proceed to the development of innovative nutraceuticals, dietary supplements or herbal medicinal products. For more information please visit the MediHealth homepage (http://www.medihealth.eu/).

WS-L1

Metabolic activation of natural products: some case studies

Luc Pieters

Natural Products and Food Research & Analysis (NatuRA), Department of Pharmaceutical Sciences, University of Antwerp, Belgium

Natural products are often prodrugs, e.g. glycosides, which must undergo *in vivo* metabolic conversion (activation). A *Gloriosa superba* seed extract containing colchicine, a well-known cytotoxic compound, 3-*O*-demethylcolchicine and its glycoside colchicoside, was found to be active in a murine pancreatic tumor model. Also, a colchicoside-rich / colchicine-poor extract with the same total level of colchicine and derivatives was active in the same model, indicating that colchicoside can be considered as a prodrug. The activity of gemcitabine, a drug widely used against pancreatic cancer, could be improved by combining it with a *Gloriosa superba* seed extract.
Extracts of the herb *Herniaria hirsuta* are traditionally used in Morocco against kidney and gall stones. Prolonged use of a *H. hirsuta* extract resulted in a cholesterol-lowering effect in the bile of dogs, a pharmacological effect that can prevent the formation of gallstones and can contribute to dissolving existing gallstones. Saponins (medicagenic acid glycosides) have been hypothesized as active principles, but before absorption they need to be deglycosylated. The aglycones (or metabolites thereof) can be absorbed, and may further be metabolized to the ultimate active molecules.
Therefore, an *in vitro* gastro-intestinal dialysis model (GIDM) was developed, including microbial fermentation in the colon, to mimic human biotransformation processes. Cyclopeptide alkaloids will be discussed as an example.

WS-L2

Natural products that modulate the lipid mediator class switch in inflammation

Werz Oliver

Department of Pharmaceutical/Medicinal Chemistry, Institute of Pharmacy, Friedrich-Schiller-University, 07749 Jena, Germany

Acute inflammation is a host-protective response to injury that eliminates the harmful stimulus and facilitates repair of damaged tissue. In contrast, chronic inflammation is a central component of numerous widespread diseases, including atherosclerosis, cancer, type 2 diabetes, and Alzheimer's disease that requires therapeutic targeting of the inflammatory response. Initiation and resolution of inflammation are tightly regulated by lipid mediators (LMs) that can lead to either chronicity or self-resolving inflammation. Arachidonic acid (AA)-derived prostaglandins (PGs) and leukotrienes (LTs) are formed via cyclooxygenase and 5-lipoxygenase pathways and play pivotal roles in initiation of inflammation. Subsequent production of so-called specialized pro-resolving mediators (SPM) actively terminates inflammation to promote resolution of inflammation and tissue regeneration. The SPM superfamily includes lipoxins biosynthesized from AA, E-series resolvins from eicosapentaenoic acid, and docosahexaenoic acid-derived D-series resolvins, protectins, and maresins. In view of the temporal biosynthesis of LM during the inflammatory process, the “LM class switch” from pro-inflammatory PGs and LTs to anti-inflammatory and pro-resolving SPMs critically determines if inflammation is terminated or if it proceeds. Therefore, besides simply blocking PG and LT formation to intervene with chronic inflammation, a novel thus far unexplored pharmacological strategy is to promote SPM formation by facilitating the LM class switch. Conclusively, an ideal drug would suppress pro-inflammatory PG and LT formation while supporting SPM biosynthesis. Of note, SPM biosynthesis requires lipoxygenase activities as well, implying that smart pharmacological manipulation of these enzymes is necessary. In this presentation, natural products will be presented that indeed are capable of altering the LM biosynthetic machinery to switch from LT to SPM formation.

WS-L3

Dual pathway activation by natural compounds as key to breast cancer prevention?

Günter Vollmer

Technische Universität Dresden, Faculty of Biology, Chair of Molecular Cell Physiology & Endocrinology, Zellescher Weg 20b, 01217 Dresden, Germany

Breast cancer is the most frequently diagnosed cancer occurring in women in Germany with almost 72.000 cases diagnosed in 2013 and an expected increase to 77.000 cases in 2020. It is unquestionable that estrogens play a pivotal role in the development of breast cancer, as about 70 % of all breast cancers cases are estrogen dependent, with four major mechanisms contributing to estrogen-dependent mammary gland carcinogenesis and breast cancer growth. These are the hormonal, the chemical, the inflammatory, and the epigenetic pathway[1]. Consequently, inhibition of any of these pathways may therefore result in breast cancer prevention. So far, we focused on the hormonal pathway, which plays a key role in tumor promotion by estrogenic compounds through an estrogen receptor-α (ERα) dependent mechanism. From in vitro experiments it is long known, that simultaneous activation of the arylhydrocarbon receptor (AhR) significantly inhibits ERα dependent growth of breast cancer cells[2]. The aim of our investigations was therefore to test whether activation of AhR attenuates ERα dependent growth regulatory pathways *in vivo*. As growth regulation of the mammary gland in young adult female rats has been proposed as predictor for growth regulation in breast cancer, we performed a three day uterotrophic assay with young adult Wistar rats. Animals were subcutaneously treated either with 4 µg/kg bw./d of estradiol (E2), with 15 mg/kg bw./d of 3-methylcholanthrene (3-MC; AhR-agonist) or combinations thereof. 3-MC alone showed little if any effects in the mammary gland. Regarding growth parameters, 3-MC co-treatment significantly inhibited the E2 stimulated growth parameters like the increase in the number of terminal endbuds and the number of epithelial cells showing nuclear labelling with the proliferation marker Ki-67. In addition, using a microarray approach we were able to demonstrate that the co-treatment with 3-MC attenuated the E2-induced changes of expression of approximately 70 % of the E2 regulated genes in the mammary gland[3]. In summary, we demonstrated that ER and AhR pathways interact in vivo. Activation of AhR in combination to ERα thereby resulted in a functional antiestrogenicity. The consequences of this interaction have to be verified in an experimental breast cancer model. In conclusion, activation of AhR by a non-toxic, plant-derived agonists may represent a preventive strategy for hormone dependent mammary gland tumors.

References:

[1] Dietz BM, Hajirahimkhan A, Dunlap TL, Bolton JL. Pharmacol Rev 2016; 68: 1026–1073.
[2] Safe S, Cheng Y, Jin UH. Curr Opin Toxicol 2017; 2: 24–29.
[3] Helle J, Bader MI, Keiler AM, Zierau O, Vollmer G, Chittur SV, Tenniswood K, Kretzschmar G. Environ Health Perspect 2016; 124: 601–610.

Workshop: TASCMAR

Description

TASCMAR is a collaborative research project funded under the EU Horizon 2020 programme and aspires to develop new tools and strategies to overcome existing bottlenecks in the discovery and application of marine-derived biomolecules, with a focus on the theme of anti-ageing.

From exploring the possibility for new medical drugs without harmful side effects, nutraceuticals (e.g. dietary supplements) and cosmetic products to developing technologies for bioremediation, TASCMAR will investigate the potential of the underutilized mesophotic zone of the ocean (between 30 and 100 meters depth) and will develop innovative approaches for the cultivation and extraction of marine invertebrates and symbionts from lab to pilot-scale.

TASCMAR is part of the EU Blue Growth Strategy, aiming to unlock the potential of seas, oceans and coasts for sustainable growth. The project will be closely monitored for its socio-economic and environmental impacts so as to ensure sustainability while promoting economic growth.

WS-L4

Mesophotic marine habitats: understudied biodiversity

Yehuda Benayahu[1], Erez Shoham[1], Ronen Liberman[1], Shai Tamir[1], Carolina Alonso[2]. Pedro Álvarez[2], Suchana Chavanich[3], Anne Bialecki[4], Géraldine Le Goff[5], Jamal Qauazzani[5]

[1]Tel Aviv University, Tel Aviv, Israel, [2]iMare Natural, Granada, Spain, [3]Chulalongkorn University, Bangkok, Thailand, [4] Université de la Réunion, Ile de la Réunion, France, [5]ICSN-CNRS, Gif sur Yvette, France

Studies have revealed the bewildering diversity on shallow reefs, comprising plethora of invertebrates. Until the past decade most biodiversity surveys have been restricted to the upper ~30 m. The mesophotic coral-reef ecosystem (MCE) has been defined as comprising the light-dependent communities (30 to <150 m) in tropical and subtropical regions. Remotely-operated vehicles (ROV) and technical diving have now facilitated the investigation of MCEs. Consequently, they have become available for research with an increasing interest in their bio-resources, keeping in mind the high significance of the holobiont concept. The scarce data available on non-scleractinian MCE fauna in the Northern Red Sea, Andaman Sea, Gulf of Thailand and the Western Mediterranean Sea intrigued us to conduct thorough surveys on the mesophotic benthic fauna for bioprospecting purposes. The results revealed diverse species assemblages associated with a variety of micro-symbionts, including species new to science and new zoogeographical records. The findings highlight the possibility that MCEs host depth generalists along with unique depth specialists. In addition, this ecosystem might include species also found below the deepest fringes of the MCEs. The evidence suggests that octocorals, sponges along with other invertebrates are the major benthic organisms in MCE, being far more diverse than has been envisioned. The results also raise issues concerning the need for conservation policies aiming at protecting the MCE marine life.

Acknowledgement: TASCMAR project (www.tascmar.eu) is funded by the European Union in the frame of H2020 (GA No 634674).

Keywords: Bioresources, benthic invertebrates, holobiont, octocorals, sponges, depth specialist, depth generalist.

WS-L5

Methodologies and technologies engaged in the TASCMAR project

Géraldine Le Goff

ICSN-CNRS, France

The EU-funded Horizon 2020 project TASCMAR aims to tackle major bottlenecks in the discovery, development, and commercialization of anti-ageing marine-derived chemical compounds, with a specific focus on using new biological and chemical resources from Mesopohic Coral Ecosystems (MCEs). One of the main goals is the systematic extraction of bio-resources (invertebrates and its microbial symbionts) to create extract libraries ready for bio-assay screening, subsequent metabolic profiling, fractionation and preparation of l pure compounds library for bio-evaluation, dereplication, and structure elucidation of target compounds. According to the bioactivity profile, extracts, fractions and compounds, were prioritized according to their industrial potential.
In the frame of **TASCMAR**, more than 246 invertebrates and 450 microorganisms were collected and investigated from the under-investigated mesophotic zone of the Indian Ocean, the Red Sea and the Mediterranean.
TASCMAR focuses also on the development of innovative strategies and technologies. An ecofriendly and sustainable approach was developed to exploit the animals in their natural habitat and to highlight the interdependence between the animal and its neighbors and between the animal and its symbionts.
The presentation focus on the methodologies and technologies engaged in the TASCMAR project for a rationale and efficient identification and exploitation of the chemicals produced by the project bio-resources.

WS-L6

Natural Products in the fight against ageing and age-related diseases; the marine mesophotic zone as a SOURCE for the discovery of NOVEL bioactive molecules

Jamal Ouazzani[1], Nikolas Fokialakis[2], Christina Cheimonidi[3], Eleni-Dimitra Papanagnou[3], Aimilia Sklirou[3], Eirini Baira[2], Géraldine Le Goff[1], Pinelopi Vlachou[2], Nikolaos Tsafantakis[2], Konstantinos Gardikis[4], Pedro Álvarez[5], Suchana Chavanich[6,] Anne Bialecki[7], Yehuda Benayahu[8], Christine Wenzkowski[9], Ioannis P. Trougakos[3]

[1] ICSN-CNRS, Gif sur Yvette, France, [2] Faculty of Pharmacy, National & Kapodistrian University of Athens, Athens, Greece, [3] Faculty of Biology, National & Kapodistrian University of Athens, Athens, Greece, [4] APIVITA SA, Athens, Greece., [5] iMare Natural, Granada, Spain, [6] Chulalongkorn University, Bangkok, Thailand, [7] Université de la Réunion, Ile de la Réunion, France, [8] Tel Aviv University, Tel Aviv, Israel, [9] Crelux GmbH, Martinsried, Germany

Metazoans respond to harmful challenges by mounting anti-stress responses; this adaptation along with the evolvement of metabolic networks, were fundamental forces during evolution. Central to anti-stress responses are a number of short-lived transcription factors that by functioning as stress sensors mobilize cytoprotective genomic responses aiming to eliminate stressors and restore tissue homeodynamics. We have found that increased expression these cytoprotective pathways can enhance stress tolerance and longevity. Given that natural products are likely the only feasible mean for increasing healthy ageing in humans we employ the TASCMAR platform as a unique source for the discovery of novel bioactive small molecules with healthy ageing promoting activity. Specifically, in the frame of TASCMAR more than 180 existing collection of invertebrates (MACLIB library) and 179 targeted marine invertebrates species (TARMAC library) were collected from the under-investigated mesophotic zone (between 30 and 100 meters depth) of the Indian ocean, the Red sea and the Mediterranean sea. Furthermore, more than 300 (MICLIB library) and 312 (TARMIC library) associated microorganisms of MACLIB and TARMAC libraries respectively, were collected. The samples were extracted and libraries of extracts, microfractions or pure molecules (following dereplication) were sent for biological evaluation against a wide range of different targets involved in ageing or age-related diseases (e.g. cancer or neurodegeneration). These targets include catalase, sirtuin 1, CDK7, proteasome, fyn kinase, tyrosinase and elastase. The rationale behind the selection of these targets along with our findings exemplifying the identification of numerous novel bioactive molecules from all libraries will be presented.

Acknowledgements: TASCMAR project (www.tascmar.eu) is funded by the European Union in the frame of H2020 (GA No 634674).

Keywords: Ageing, age-related diseases, marine natural products, mesophotic zone

Workshop: Olive-Net

Bioactive compounds from *Olea europaea*: investigation and application in food, cosmetic and pharmaceutical industry

Description

Olive-Net is a European project funded by the Commission of the European Community (H2020—Marie Skłodowska-Curie Actions—Research and Innovation Staff Exchange (RISE), grant No. 734899). The goal of Olive-Net project is to introduce a novel approach for the exploration, valorization and marketing of new products based on bioactive compounds from Olea europaea. This will be achieved through an extended and well-balanced scheme of researcher's secondments between six universities and seven enterprises from EU & Associated countries as well as three partners from Third countries. Products and side-products of the olive tree such as olive oil, edible olive fruits, olive mill waste and olive tree leaves, will be subjected to a series of state-of-the-art extraction and isolation cascades in order to provide extracts, enriched fractions and isolated compounds of high purity. Target chemical categories will involve the well known olive oil polyphenols and secoiridoids, that will be assessed for their safety and pharmacological effects against inflammation, osteoarthritis, cardiovascular disease, etc. in cell-based and in vivo assays. All active ingredients will be identified and characterized with advanced analytical techniques, in order to be integrated in formulations and products in the area of nutraceuticals/dietary supplements. Olive-Net aspires to create a successful model promoting considerably researchers' competences and long-lasting collaboration between Industry and Academia. (http://olive-net.eu/)

WS-L7

Assessment of the antioxidant profile from different Greek olive oil varieties.

Paraskevi Kouka[a], Grigoria Tsakiri[a], Dimitra Tzortzi[a], Sofia Dimopoulou[a], Apostolis Angelis[b], Panagiotis Stathopoulos[b], Maria Halabalaki[b], Alexios-Leandros Skaltsounis[b], Demetrios Kouretas [a*]

[a] *Department of Biochemistry and Biotechnology, University of Thessaly, 41500 Larissa, Greece.*
[b] *Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, University of Athens, 15771 Athens.*

[*]*E-mail: dkouret@uth.gr*

The olive tree is one of the most important fruit trees in Mediterranean countries and virgin olive oil possesses both health and nutritional aspects. Greece is among the top three olive oil producing countries, however the weakness of the standardization industry has shrunk its market share [3]. Since Greece attempts to enter the international markets, the quality of Greek olive oil should be highlighted. Phenolic composition of olive oil has been extensively studied. It depends on several factors such as fruit maturation stage, cultivar, season, irrigation and production process [1,2]. Our aim was to firstly examine the composition of different biophenolic Greek olive oils from different geographic origin, and their antioxidant and antimutagenic properties, and secondly to demonstrate the importance of their composition in order to adopt better methods of cultivation, processing and production of olive oil. For this purpose, six polyphenolic olive oil extracts, with different composition were used and their antioxidant potential were evaluated using the ABTS●+ and the plasmid relaxation assays. Also, the cell redox status was assessed in terms of glutathione (GSH) and reactive oxygen species (ROS), using flow cytometry. According to the results, higher amounts of hydroxytyrosol (HT) compared to tyrosol (T) in olive oils were correlated with a stronger antioxidant and antimutagenic potency, highlighting the effect that composition has. However, this observation was not evident in the tested cell line (HeLa), as T-rich olive oils increased GSH levels more readily than the HT-rich ones. Therefore, cellular metabolism seems to significantly affect an extract's activity.

Acknowledgements: The study was funded by the Hellenic General Secretariat for Research and Technology (GSRT) and the Hellenic Foundation for Research and Innovation (HFRI) (grant number 5547).

Key words: Olive oil, antioxidant activity, hydroxytyrosol, tyrosol

References

[1] Kouka P *et al.* Int J Mol Med 40: 703–712, 2017
[2] Priftis A *et al.* Mol Med Rep 12: 7293–7302, 2015
[3] Goutzourelas N *et al.* Int J Mol Med 36: 433–441, 2015

WS-L8

The European representatives of Oleaceae family as a source of compounds with anti-inflammatory activity

Anna K. Kiss

Medical University of Warsaw, Institute of Pharmacy, Department of Pharmacognosy and Molecular Basis of Phytotherapy, Banacha 1, 01-097, Warsaw, Poland, akiss@wum.edu.pl

The aim of the study was to find a source of potentially anti-inflammatory agents among the members of the Oleaceae family traditionally used in phytotherapy or cultivated in European countries.
Ligustrum, *Fraxinus*, *Syringa* and *Forsythia* sp. plant materials used in the treating of inflammation-associated diseases were analysed phytochemically using chromatographic method with mass spectrometry detection (UHPLC-DAD-MS/MS). The bioactivity guided isolation of the active principles were performed using *in vitro* human neutrophils and monocytes/macrophages models. The inhibition of ROS production was determined using luminol- dependent chemiluminescence. The effect on cytokines production was measured by an enzyme-linked immunosorbent assay. The expression of adhesion molecules was analyzed with flow cytometry and the neutrophil attachment to the endothelial cells was assessed fluorimetrically. The effects on p38MAPK, ERK1/2, JNK phosphorylation and NF-kB p65 translocation were determined using western blots.
Extracts from selected plants material (at 25-100 μg/mL) were shown to modulate lipopolysaccharide (LPS)-triggered pro-inflammatory functions of neutrophils and monocytes/macrophages. The activity of isolated compounds (37) from the group of iridoids, phenylethanoids, lignans and simple phenols was also compared. Our observations suggested that some extracts from Oleaceae family could play a potential role in the prevention of inflammatory disease and or be a source of single lead compound(s).

Acknowledgements: This study was supported by research grant 2015/17/B/NZ7/03086 from Polish National Science Center

WS-L9

NMR based comprehensive screening of food materials

Andrea Steck

Bruker BioSpin GmbH, Rheinstetten, Germany

Food supply networks continue to grow in scale and complexity, and deliberate food fraud, driven by the prospect of economic gain, is an emerging risk alike. High-priced food products, but also those with high volume of sales, have turned out to be most vulnerable to adulteration. The emergence of more and more sophisticated food analysis techniques has dramatically forced back overt falsifications, but is inevitably a trigger to subtilize adulteration methods. The key to profile food quality economically, and increase the detection rate of "smart" adulterations is a fast and efficient analytical technique which is able to cover the range from whole matrices down to single compounds.
Due to its unique "all-in-one" capabilities, high-resolution ^{1}H-NMR spectroscopy, combined with multivariate statistical chemometrics, is the screening methodology of choice for food quality and authenticity control [1,2].
As ^{1}H-NMR is intrinsically quantitative, only one quantification reference for all NMR-detectable components in a mixture is required. Yielding targeted quantification of selected compounds as well as untargeted fingerprinting in a single run, NMR is a specific and holistic method likewise. Its supreme reproducibility enables worldwide lab-to-lab spectra comparison and collective database buildup. Unlimited data re-processing is given and allows to apply future statistical algorithms, re-modelling of more or different parameters, or retrospective quantification of mixture components not in the focus of interest at present.
Our portfolio of fully automated and ISO-17025 accredited food profiling methods covers fruit juice [3], wine [4,5] and honey screening at present, and further methodologies are under development.
The principles behind this NMR methodology as well as recent applications are presented.

References:

[1] Spraul M, Schuetz B, Humpfer E, Moertter M, Schaefer H, Koswig S, Rinke P. Magn Reson Chem 2009; 47: 130–137.
[2] Lachenmeier DW, Humpfer E, Fang F, Schütz B, Dvortsak P, Sproll C, Spraul M. J Agric Food Chem 2009; 57: 7194–7199.
[3] Spraul M, Humpfer E, Schäfer H, Schütz B, Moertter M, Rinke P. NMR-based mixture Analysis on the example of fruit juice quality control Using Statistics and Quantification In: "NMR Spectroscopy in Pharmaceutical Analysis", U. Holzgrabe, I. Wawer, B. Diehl, Elsevier 2008.
[4] Godelmann R, Fang F, Humpfer E, Schuetz B, Bansbach M, Schaefer H, Spraul M. J Agric Food Chem 2013; 61(23), pp 5610-5619.
[5] Godelmann R, Kost C, Patz CD, Ristow R, Wachter H. J AOAC Int 2016 Sep; 99(5): 1295–1304.

WS-L10

Application of different mass spectrometric methods to determine the authenticity of olive oil

Carsten Baessmann

Bruker Daltonik GmbH, Germany

The increasing popularity of extra virgin olive oil (EVOO) and the increasing problem of food fraud have provided the need for quality and authenticity control. Typical problems are mislabeling of protected designation of origin (PDO) or edible oil adulteration. In this presentation we give an overview about the capabilities of different mass spectrometric methods in combination with metabolomics methods.
In a first study 126 oil samples from 6 Mediterranean PDOs were analyzed by LC-MS and GC-MS combined to statistical methods. The extracts were eluted in LC using a C18 (2.1 x 100 mm, 1.8 µm) column, with acidified water and acetonitrile and a flow gradient (0.4-0.6 mL/min) at 40°C. The derivatized extracts were injected in GC, using a BR-5 column with a T gradient from 150 to 320°C (4°C/min rate). Both systems were coupled to a Compact™ QTOF MS (Bruker) by an ESI interface for LC and a GC-APCI source for GC. In a second similar study an impact QTOF MS (Bruker) was used.
For the first study samples from six different Mediterranean PDOs (Meknès and Ouazzane (Morocco), Priego de Córdoba and Baena (Spain), Kalamata (Greece) and Toscana (Italy)) were collected and analyzed. Sample prep. (unselective 3-steps liquid-liquid extraction) and experimental conditions facilitated the determination of a great number of minor compounds (phenolic compounds, triterpenic acids and dialcohols, sterols, tocopherols, free fatty acids, etc.) within a single run.
Non-targeted and targeted approaches were used to offer maximum coverage of the olive oil metabolome's chemical space in a 1st step, and the possible validation of the identified markers afterwards. Data treatment (PCA, PLS-DA) - done by using MetaboScape® 3.0 - led to a noticeable discrimination among the six evaluated PDOs taking into account the data coming from both platforms. Several compounds such as elenolic acid, luteolin, oleuropein and ligstroside aglycones, and some other tentatively identified substances, were pointed out as possible PDOs distinctive markers. Multi-class LC-MS and GC-MS methodologies together with statistical methods enabled the discrimination among different PDOs, identifying potential origin markers. The combined use of non-targeted and targeted approaches enhanced the outcome of the study. GC-APCI-Q TOF preserves the pseudo-molecular ion information, which is a great advantage over the "classical" GC-EI-MS systems and facilitates the identification.
In a third study LC free magnetic resonance mass spectrometry (MRMS) analysis was used for mapping and quality control assessment of Greek extra virgin olive oil (EVOO) using statistical methods. This workflow takes advantage of the rapid, LC free flow injection analysis (FIA) based data acquisition by ultra-high resolution MRMS. EVOO samples and their biphenolic extracts were analysed using a Bruker solariX XR 7T mass spectrometer in combination with FIA. Statistical analysis allowed clustering according to geographical origin, harvesting year, cultivation practice and the oil production procedure.

Workshop: Natural Products and Cosmetics

Description

The public awareness of high quality of life has increased and it is craved in various sectors daily encountered. People nowadays and the productive wed more than ever, set as their first priority consumer safety, public health and environment protection. As cosmetics industry develops products universally valued and appreciated it could not remained unaffected. Far from being perceived as the land of luxury, cosmetic products actually impact our health and well-being and have become an integral part of our everyday lives. Consumers today expect products which promise health or physical improvement and benefits beyond the merely aesthetic effect of masking. Scientific study shows that plants possess a vast and complex arsenal of phytochemicals that not only calm, restore and heal the skin, but also stand up to the scrutiny of clinical trial and pharmacological testing. Natural products provide greater structural diversity than standard combinatorial chemistry, eliminating the increasing concern about synthetic products on humans and the environment. About 300.000 plant species are estimated to occur in the world and probably only ~10% have been tested for some type of biological activity. Many international pharmaceutical, fragrance and cosmetics companies are continuously searching for new products and ingredients and are interested to bring “green” products to the market. Five speakers representing academy and companies actively engaged in the exploitation of natural products for cosmetic use will quote their views and experiences.

WS-L11

NMR applications in cosmetics

Andrea Steck

Bruker BioSpin GmbH, Rheinstetten, Germany

The volume of inauthentic or fraudulent cosmetics is on the rise worldwide, especially since customers have widespread access to cosmetics products offered online. Cosmetics are "big business", and there are countless options for unscrupulous businessmen to adulterate them - addition of foreign substances, production on substandard levels which may result in contaminations with chemicals, or fake of brands using different ingredients and filling them into empty packets of prestigious fabricators. In all cases, using such products comes along with serious health risks for consumers, and financial as well as reputational losses for producers.
But not only fraud detection is relevant in the fields of cosmetics - quality control is it as well: shelf life, storing conditions, or light exposure effects are just some of many more aspects.
Unquestionably, there is an urgent need to have a fast and reliable analytical technique on hands to address these topics. NMR spectroscopy, especially when combined with statistical chemometrics, is a very promising method to allow compound quantification as well as non-targeted fingerprinting in one experimental run.
There are, however, some challenges to deal with: cosmetics products are diverse - watery, oily, alcoholic, or all of it; they can contain polymers, particals, metal-based compounds, or salts. In most cases, they are complex mixtures of compounds with very different chemical properties.
In this workshop, the results of several NMR-based feasibility studies applied to different cosmetics products are presented.

WS-L12

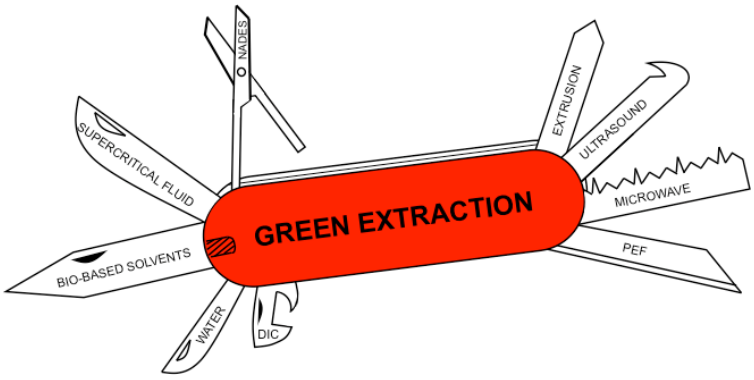
Green Extraction of food, cosmetic ingredients ad natural products: moving from academia to innovative and large-scale applications.

Anne-Sylvie Fabiano-Tixier, Farid Chemat

anne-sylvie.fabiano@univ-avignon.fr ; farid.chemat@univ-avignon.fr

GREEN Extraction team, UMR408, INRA, Avignon University, France, https://green.univ-avignon.fr/

This presentation will introduce a new and innovative area in the frontiers of chemistry, biology and processing: green extraction with special emphasis on natural products. Green extraction is a part of the sustainable development concept; its history, concept, principles and fundamentals will be described. We will pay special attention to the strategies and the tools available to make biorefinery greener. The representation will present the innovative research in this area these past five years in term of innovative techniques (microwave, ultrasound, pulse electric field...) and alternative solvents (ionic liquids, sub and supercritical fluid, agrosolvents, water...) applied to this new area green extraction of natural products with special examples applied to biorefinery concept.



A general definition of green chemistry is the invention, design and application of chemical products and processes to reduce or to eliminate the use and generation of hazardous substances. In relation of green extraction of natural products, this definition can be modified as follows: "*Green Extraction is based on the discovery and design of extraction processes which will reduce energy consumption, allows use of alternative solvents and renewable natural products, and ensure a safe and high quality extract/product*". The listing of the "six principles of Green Extraction of Natural Products" should be viewed for industry and scientists as a direction to establish an innovative and green label, charter and standard, and as a reflection to innovate not only in process but in all aspects of solid-liquid extraction. The principles have been identified and described not as rules but more as innovative examples to follow discovered by scientist and successfully applied by industry.

References:

[1] Chemat F., Strube J. "Green Extraction of Natural Products. Theory and practice". Wiley-VCH, Weinheim, 11 chapitres, 384 pages. 2015. 978-3-527-33653-1.

WS-L13

Marine products as effective raw materials for cosmetics – a research and innovation approach

Konstantinos Gardikis, Sofia Letsiou

Scientific Affairs Department, APIVITA S.A., Industrial Park of Markopoulo Mesogaias, 19003, Markopoulo Attikis, Athens, Greece

As there is growing interest in the research on marine products, the pharmaceutical, cosmetic and food industry have begun marketing such products to a large extent. The bioprospecting is usually connected with sustainable development practices, making marine organisms an important pool for environmental friendly produced bioactive raw materials.
The development of novel bioactive ingredients from the marine environment is presented in this work. These ingredients are processed in such a way that they exhibit optimized antioxidant, antiaging, photoprotective and hydrating properties – all the major categories targeted by cosmetic raw materials. Focus is given on microalgae extracts, most of them investigated for the first time for cosmetic purposes. Case studies include the development of ingredients that are able to modulate skin immunity and preserve skin's elasticity under UV stress.
The marine environment presents an underexplored field for the development of effective cosmetic formulations, especially when bioactive ingredients are concerned.

Acknowledgements: Parts of this work are funded by HORIZON 2020 projects Tascmar, Marisurf and Algae4AB, Grant agreements no.: 634674, 635340, 691102 respectively and by FP7 project AlgaeCom, Grant agreement no.: 286354

WS-L14

Natural products: The future of cosmetics

Julia Moesslacher, Valentina Scalfari, Astrid Huber-Seidel, Roland Kohl

Cura Marketing GmbH, Innsbruck, Austria

In 2017, the global market size for cosmetics was approximately 250 billion USD, Europe accounting for 77 billion USD. Out of that, the natural and near-natural cosmetics market size reached up to 28 billion USD globally and 2 billion USD in Europe, respectively. Despite the fact that the total market growth rates in cosmetics were slowing down, the sales of natural cosmetics market were growing over proportionally by +7.9% last year, especially for certified natural cosmetics.
This is driven by the consumer demand for "green" ingredients, due to consumer concerns about synthetic chemicals, the perception that natural products provide lower health risks and environmental impacts, general interests in ethics, social aspects and ingredient provenance, emotive / marketing benefits concerning natural ingredients and also their cosmetic efficacy.
Extracts from *Olea europaea* (olive) oil, fruit, leaf and even waste are a great source of valuable ingredients for cosmetics like oleuropein, hydroxytyrosol, tyrosol and their derivatives. These compounds are known for their i) antioxidant activity, ii) skin protective effects like the inhibition of the expression of MMP-2, -9 and -13 that are involved the degradation of extracellular matrix, and iii) anti-aging benefits like the enhancement of the proteasome activity and delaying the senescence morphology *in vitro*. Developing formulations with a standardized content of such olive ingredients lead to superior products with proven antioxidant activity of the final product, as shown both by *in-vitro* data and *in-vivo* anti-aging activity: After a 4-weeks treatment an improvement of elasticity up to 74.2% was observed, while the roughness of the skin was decreased by up to 44.9%.
Given the efficacy of natural products in skin care, and taken together with the increasing demand for natural cosmetics, there is high potential for natural products as active ingredients in cosmetics in the future.

WS-L15

The revival of an ancient perfume of the Mycenaean era

Lena Philippou

KORRES NATURAL PRODUCTS S.A., Metamorfosi, Greece

The scope of the presentation is to shed a light on an ancient technique of perfume manufacturing in Greece during the Mycenaean period (1600-1100 BC). This was achieved by collecting crucial information from deciphered Linear B ideograms, ancient texts of Pedanius Dioscurides and Theophrastus of Eresus as well as from the manuscripts of several modern researchers. The R&D department of Korres was then challenged to materialize this project by creating for the first time a perfumed oil based on rose scent. The goal was accomplished by using raw materials collected from the Greek nature resembling the same ancient practices. This unique project is the result of a close collaboration of Korres S.A. and the National Archaeological Museum in Athens.

SL-001
Recent developments in the identification of natural products by NMR
Jean-Marc Nuzillard, Pedro Lameiras

SL-002
NMR in natural products: chemical analysis, monitoring of dynamic changes, "in-cell" NMR and DFT calculated structures based on NMR chemical shifts in solution
Ioannis Gerothanassis

SL-003
The use of vibrational spectroscopy in natural product analysis: current and future directions
Christian Huck

SL-004
Mass Spectrometry metabolomics to identify bioactive compounds and synergists from botanical natural products
Nadja Cech

SL-005
Mass spectrometric dereplication of natural products using mass spectrometry integrated approach
Syed Ghulam Musharraf, M. Iqbal Choudhary and Atta-ur-Rahman

SL-006
Metabolomic strategies for marine natural products analysis
Norberto Peporine Lopes

SL-007
Bioactive alkaloids towards enzymes related to neurodegeneration from *Psychotria nemorosa*
Sylvian Cretton, Luiz C. Klein-Júnior, Amélia T. Henriques, Muriel Cuendet, Philippe Christen

SL-008
Integrating chemical and biological data to investigate *Echinodorus grandiflorus* as a source of potential anti-arthritic agents
Fernão Braga, Eliana Garcia, Diogo Oliveira, Flávio Amaral, Danielle Souza, Rodrigo Pádua, Mauro Teixeira

SL-009
Bioguided fractionation and molecular networking to access the chemical diversity of bioactive extracts on fibroblasts AGEs
Amandine André, Kaatio Touré, Joanna Wdzieczak-Bakala, Didier Stien, Véronique Eparvier

SL-010
Isolation and identification of secondary metabolites, including four new compounds, from three *Digitalis* species
Vahap Murat Kutluay, Kan'ichiro Ishiuchi, Toshiaki Makino, Fatih Göger, Nese Kirimer, Iclal Saracoglu

SL-011

Caged xanthones from *Garcinia propinqua*

Surat Laphookhieo, Teerayut Sriyatep, Cholpisut Tantapakul, Chatchai Muanprasat, Sawinee Seemakhan, Suparerk Borwornpinyo, Stephen Pyne, Brian Patrick, Raymond Andersen

SL-012

Simple screening for high-colibactin producers from clinical samples using activity-based fluorescent probe

Kenji Watanabe

SL-013

Botanical nematicides originating from Mediterranean plants: An overview

Nikoletta Ntalli, Urania Menkissoglu-Spiroudi

SL-014

Reduction of polyglutamine aggregates in a cellular model of Huntington's disease by an ethanolic extract of *Ugni molinae* wild-type strain.

Alexis Rivas, Rodrigo Perez-Arancibia, Carla Delporte, Claudio Hetz

SL-015

***In vitro* anti-cancer activity of *Barringtonia asiatica* (L.) Kurz crude methanolic leaf extract against human breast adenocarcinoma cell line, MCF-7**

Ronald Arlet Villaber, Kerl Francis Pastoril

SL-016

HPLC-PDA based chemical profiling and antimicrobial activity studies on *Citrus grandis* and *C. sinensis* from the Iraqi flora

Shaymaa Al-majmaie, Lutfun Nahar, George P Sharples, Satyajit D Sarker

SL-017

Evaluation of antimicrobial, disinfectant and anti-inflammatory potential of *Garcinia cambolia*: A potent source for pharmaceuticals and disinfectants

Mayuri Napagoda[1], Sudhara De Soyza[1], Jana Gerstmeier[2], Hannah Butschek[2], Andreas Koeberle[2], Mallique Qader[3], Sybille Loranz[4], Sanjeeva Witharana[5], Gaya Bandara Wijayaratne[6], Aleš Svatoš[4], Lalith Jayasinghe[3], Oliver Werz[2]

SL-018

Folic acid supplementation repressed hypoxia-induced inflammatory response in human promyelomonocytic cells

Xinwei Jiang

SL-019

Chemical tuning of plant hormone by using stereoisomer of a natural product

Minoru Ueda

SL-020

Nature inspired synthesis in chemical biology

Kamal Kumar

SL-021
Design, semi-synthesis and evaluation of anticancer and antimalarial cycleanine analogues
Fidelia Uche, Alan Richardson, Paul Horrocks, Wen-Wu Li

SL-022
Total synthesis and structure-activity relationship study of Rakicidin A. Discovery of analogues with improved pharmacological properties against metastatic cancer.
Michail Tsakos, Kristian Jacobsen, Nikolaj Villadsen, Thomas Poulsen

SL-023
Practical synthesis, neurotrophic activities, and structure-activity relationship of talaumidin derivatives
Kenichi Harada, Rina Bando, Ryo Irimaziri, Miwa Kubo, Yoshiki Koriyama, Yoshiyasu Fukuyama

SL-024
Synthesis and evaluation of cembranolide type compounds for treatment of HPV-mediated carcinomas
Patrik Eklund, Yury Brusentsev, Rajendran Senthil, John Eriksson

SL-025
Asymmetric total synthesis of mycolic acids from *Mycobacterium tuberculosis* for structure elucidation
Nabil Tahiri, Peter Fodran, Dhinesh Jayaraman, Martin Witte, Adriaan Minnaard

SL-026
Plant polyphenols - drug interactions are overlooked – modeling studies may help
Zohar Kerem, Loai Basheer, Yelena Guttman

SL-027
Asymmetric synthesis of 2,6-disubstituted piperidine alkaloids from chiral aziridines
Hyun-Joon Ha, Lingamurthy Macha

SL-028
New xanthone derivatives and their nitric oxide inhibitory effects
Siau Hui Mah, Zi Han Loh, Soek Sin Teh

SL-029
Chemical and spectroscopic studies on bioactive terpenes from marine molluscs
Mary Garson

SL-030
Structure and synthetic study of lipopeptides, isolated from marine cyanobacteria
Kiyotake Suenaga

SL-031
Sulfated aromatic metabolites from marine sources
Anja Hartmann, Hermann Stuppner

SL-032

Structures and anti-obesity activities of the yoshinone A and the related marine *γ*-pyrone compounds

Tomoyuki Koyama, Yoshinori Kawazoe, Arihiro Iwasaki, Osamu Ohno, Kiyotake Suenaga, Daisuke Uemura

SL-033

Symbiotic microorganisms, an original resource and an inspirational model for the search of bioactive compounds

Seindé Touré[1], Morgane Barthélemy[1], Orianne Brel[1], Jonathan Sorres[1], Didier Stien[2], Véronique Eparvier[1]

SL-034

Chemistry of Bornean *Laurencia* – Structural diversity, anti-inflammation and anticancer activities

Charles Santhanaraju Vairappan

SL-035

Screening of wild edible mushrooms bioactivity. Piceatannol was identified as a bioactive ingredient in the order *Cantharellales*

Efstathios P Vasdekis, Athanassios Karkabounas, Marilena E Lekka

SL-036

Bioactive constituents from extremophilic fungi – New leads to battle multi-resistant microbes and cancer cells

Raha Orfali, Amal Aly, Georgios Daletos, Weaam Ebrahim, Shagufta Paraveen, Peter Proksch

SL-037

Whole-cell (+)-ambrein production in the yeast *Pichia pastoris*

Harald Pichler, Sandra Moser, Gernot Strohmeier, Erich Leitner, Thomas J Plocek, Koenraad Vanhessche

SL-038

Dissecting and harnessing fungal biosynthetic pathway

Yuta Tsunematsu, Kenji Watanabe

SL-039

GLP-1 secretagogues from the Marquesan tree *Oparanthus teikiteetinii* (Asteraceae) as potential antidiabetic drugs

Aurélie Urbain, Clément Huguet, Opeyemi J. Olatunji, Eleonore Real, Christian D. Muller, Phila Raharivelomanana

SL-040

Derivatives of Amaryllidaceae alkaloids haemanthamine and ambelline as potential drugs in the treatment of Alzheimer´s disease

Lucie Cahlíková, Aneta Ritomská, Jana Maříková, Lubomír Opletal, Jan Korábečný

SL-041

Quality assessment of *Morus alba* root bark using HPTLC bioautography-UPLC-Orbitrap-MS^2 fingerprint combined with chemometrics

Petar Ristivojevic, Ammar Tahir, Judith Rollinger

SL-042

Assessment of selected Saudi and Yemeni plants for insecticidal activities against the yellow fever mosquito *Aedes aegypti* (L.), and LC MS/MS and GC/MS analysis of bioactive extracts

Shaza Almassarani, Amina El-Shaibany, Nurhayat Tabanca, Abbas Ali, Alden Estep, James Becnel, Fatih Goger, Betul Demirci, Ali El-Gamal, Husnu K Can Baser

SL-043

Cytotoxic and Wnt-inhibiting activity of polyphenolics from *Lespedeza bicolor* and *Ampelopsis japonica*

Darya Tarbeeva, Sergey Fedoreyev, Artem Blagodatski, Antonina Klimenko, Marina Veselova, Petr Gorovoy

SL-044

Inhibitory effect of chemical and natural anti-browning agents on polyphenol oxidase from Ginger (*Zingiber officinale* Roscoe)

Chen Wai Wong, Win Yee Lim

SL-045

Isatin derivative inhibit oxidative stress damage to muscles in diabetes.

Meha Fatima Aftab, Shabir Khan Afridi, Momin Khan, Aneela Karim, Khalid M. Khan, Rizwana S. Waraich

SL-046

The active compound screening and ameliorate efficacy of the three Tibetan classical prescriptions for the cerebral ischemia injury therapy.

Liwei Tan, Ying Wu, Rui Tan

SL-047

Are plants used in the Eastern Cape Province of South Africa for cosmetics fully commercialized?

Wilfred Otang Mbeng, Jonas Idowu Sagbo

SL-048 tha ginei 046

Optimization of *in vitro* release of an anticonvulsant using nanocapsule-based thermogels

Akbar Esmaeili, Sonia Singh

SL-049

Dihydrochalcone glucosides from the subaerial parts of *Thonningia sanguinea* and their *in vitro* PTP1B inhibition

Stefan Schwaiger, Luca Pompermaier, Elke H. Heiss, Mostafa Alilou, Fabian Mayr, Mawunu Monizi, Thea Lautenschlaeger, Daniela Schuster, Hermann Stuppner

SL-050
Antifungal discovery for the treatment of White Nose Syndrome (WNS) in Bats
Christine Salomon, Yudi Rusman

SL-051
The quantification of health claim-relevant tyrosol and hydroxytyrosol after direct hydrolysis improves customer understanding and mitigates market distortion
Thomas Jakschitz

SL-052
Anti-angiogenic activity of some iridoids isolated from medicinal plants
M P Germanò, V D'Angelo, N De Tommasi, M D'Ambola, KA Beladjila, AM Iannuzzi, A Braca, A Bader

SL-053
Bioactivity of medicinal plants: is it the plant, endophyte or both?
Hajara Alfa, Randolph Arroo, Susannah Walsh, Ketan Ruparelia, Avninder Bhambra, Adrian Slater

SL-054
Exploring the potential of apple dihydrochalcones on novel cosmetical, nutritional and pharmaceutical applications (ExPoApple2)
Stefan Martens, Francesco Guzzonato, Luisa Palmieri, Salvatore Multari, Stefano Benini, Rosanna Caliandro, Hermann Stuppner, Fabian Mayr, Daniela B. Schuster, Stefan Schwaiger

SL-055
Challenges in authenticating essential oils
Prabodh Satyal, William N. Setzer

SL-056
Combining supercritical fluid extraction and chromatography with molecular networks generation for plant dereplication
Florent Olivon, Nicolas Elie, Louis-Félix Nothias-Scaglia, Marc Litaudon, David Touboul

SL-057
Carobs, the black gold of Cyprus: science meets industry
Antonis Kakas, Christiana Neophytou, Andreas Constantinou

SL-058
Cannabis and its Discontents: Regulatory Choices Impacting the Success of Medical Cannabis Programs in the U.S. and EU
Ashford Elizabeth

SL-059
Analysis of Pesticides and Mycotoxins in Legal Cannabis Using FlowBased LC-MS/MS System
Bisi Ariovaldo

SL-060

Method development of chlorophyll pigments separation by Supercritical Fluid Chromatography (SFC).

Thibault Lefebvre, Arwa Talbi, Emilie Destandau, Eric Lesellier

SL-061

The application of hydrostatic counter-current chromatography in the purification of pharmacologically active phenolics and alkaloids

Wirginia Kukula-Koch

SL-062

Purification of bioactive natural products by centrifugal partition chromatography (CPC)

Tsvetelina Mandova, Grégoire Audo, Sylvie Michel, Raphael Grougnet

SL-063

A simple and Effective Approach for the Isolation of Acidic Cannabinoids from Cannabis Sativa L. Using pH-Zone-Refining Centrifugal Partition Chromatography

Johannes R. Popp, Eleftherios A. Petrakis, Apostolis Angelis, Maria Halabalaki, Günther K. Bonn, Hermann Stuppner, Alexios-Leandros Skaltsounis

SL-064

Understanding the taste of wine: isolation and identification of new taste-active compounds in oak wood

Delphine Winstel, Pierre Waffo-Téguo, Blandine Cretin, Marine Gammacurta, Denis Dubourdieu, Axel Marchal

SL-065

Application of CCC in the separation of active compounds from natural products

Xinyi Huang, Jianfei Liu, Yuan Gong, Kaijun Quan, Dong Pei, Duolong Di

SL-066

Large-scale isolation of high-purity peptide VVYP from Globin Peptide using MCI gel column combined with consecutive high-speed counter-current chromatography: aqueous two-phase systems

Dong Pei, Xin-Yi Huang, Duo-Long Di

SL-067

Redefining enzyme catalysis: chemical control in the biosynthesis in terpene cyclases

Dan T Major

SL-068

Novel pathways constructed through metabolic engineering in microbial biofactories to produce highly valuable natural products

Emmanouil Trantas, Maria Halabalaki, Leandros Skaltsounis, Filippos Ververidis

SL-069

Balkan Botanic Garden of Kroussia: Protection, conservation and sustainable use of plant genetic recourses.

Eleni Maloupa, Krigas Nikos

SL-070

Gene activation strategies in fungi – Sex, Drugs, and Genetics?

Donovon A. Adpressa, Paige. E. Mandelare, George F. Neuhaus, Lanelle R. Connolly, Kristina M. Smith, Michael Freitag, Sandra Loesgen

SL-071

New insights on the structure of Kraft lignin & its fractions with tangible practical implications

Dimitris Argyropoulos

SL-072

Metabolite profiling of the roots of 26 different accessions of soil-grown non-sterile *Arabidopsis thaliana* involved in plant interaction with rhizosphere microorganisms

Kourosh Hooshmand, Enoch Narh Kudjordjie, Rumakanta Sapkota, Tong Shen, Oliver Fiehn, Mogens Nicolaisen, Inge Fomsgaard

SL-073

Metabolomics novel application in functional foods analysis & drug discovery

Mohamed Farag

SL-074

Feature Fishing – How to catch bioactives in a complex mixture of lanostane triterpenes

Ulrike Grienke, Paul A. Foster, Julia Zwirchmayr, Ammar Tahir, Judith M. Rollinger, Emmanuel Mikros

SL-075

Can nature's pharmacy influence the health of honey bees?

Nanna Hjort Vidkjær, Per Kryger, Inge S. Fomsgaard

SL-076

High performance separation techniques for identification, characterization and quantification of plant secondary metabolites with health-promoting properties

Danilo Corradini, Isabella Nicoletti

SL-077

New isomer of pancracine as a potential anticancer agent

Kateřina Breiterová, Anna Hošťálková, Lubomír Opletal, Darja Koutová, Jana Maříková, Lucie Cahlíková

SL-078

Angiotensin converting-enzyme inhibitory activity of bioactive extracts from Philippine plants

Christine Hernandez, Rezzaira Astorga, Lyen Castro, Mavis Fabian, Jasmin Tutor

SL-079

Philippine honey possesses unique natural products composition and promising antimicrobial and antioxidant properties

Jonel P. Saludes, Angelica Faith L. Suarez, April Dawn G. Tirador, Zenith M. Villorente, Catherine F. Bagarinao, Jan Vincent N. Sollesta, Doralyn S. Dalisay

SL-080

Hijacking breast cancer oncogenicity by methoxylated flavonoidal compounds isolated from *Cleome droserifolia*

Rana Youness, Mohamed Gad, Amira Abdelmotaal

SL-081

Anti-proliferative Guaianolides from the Aerial Parts of *Chrysophthalmum montanum* (DC.) Boiss.

Fatma Ayaz, Esra Emerce, Nezhun Gören, İhsan Çalış, Mujeeb Ur Rehman, Muhammad Iqbal Choudhary, Nurgün Küçükb\oyacı

SL-001

Recent developments in the identification of natural products by NMR

Jean-Marc Nuzillard, Pedro Lameiras

University of Reims-Champagne-Ardenne, Reims, France

Mass spectrometry and NMR spectroscopy instrumentation as well as the associated usage protocols steadily evolve to improve the quality of organic compound identification. The present communication highlights a few current topics in this direction, in relationship with NMR works from our research group.
Database searching is a key point in metabolite identification as it allows to connect spectral data with the chemical structures of known compounds and therefore avoids any useless data re-interpretation. Access to structure-and-spectra databases is still difficult in what concerns NMR spectroscopy. In this framework, two recent initiatives, dedicated to the storage of raw spectroscopic data and of interpreted spectra, are discussed [1].
Mixture analysis by NMR is one of the tools for the rapid identification of known compounds present in living organisms. Only few methods are available for the compound-by-compound grouping of the NMR signals of a mixture. Recent works in this field, that rely on molecule-wide magnetization transfer by spin-diffusion in viscous solvents, are reported [2].
Tools for computer-assisted structure elucidation of presumably unknown compounds exist and have evolved since the last four decades, even though their use is still not part of the usual activity of organic chemists. The use of one of these systems is briefly presented [3].

Acknowledgements: Financial support by CNRS, Conseil Regional Champagne Ardenne, Conseil General de la Marne, Ministry of Higher Education and Research (MESR) and EU-programme FEDER to the PlAneT CPER project is gratefully acknowledged.

Keywords: NMR, data bases, mixture analysis, spin diffusion, computer-assisted structure elucidation.

References

[1] Pupier M, Nuzillard J-M, Wist J, Schlörer NE, Kuhn S, Erdelyi M, Steinbeck C, Williams AJ, Butts C, Claridge TDW, Mikhova B, Robien W, Dashti H, Eghbalnia HR, Farès C, Adam C, Kessler P, Moriaud F, Elyashberg M, Argyropoulos D, Pérez M, Giraudeau P, Gil RR, Trevorrow P, Jeannerat D. Magn Reson Chem 2018; 56: 703–715
[2] P. Lameiras et al. Chem Eur J 2017; 23: 4923–4929.
[3] J.-M. Nuzillard, B. Plainchont. Magn Reson Chem, 2018, Vol. 56, pp. 458–468.

SL-002

NMR in natural products: chemical analysis, monitoring of dynamic changes, "in-cell" NMR and DFT calculated structures based on NMR chemical shifts in solution

Ioannis Gerothanassis

University of Ioannina, Ioannina, Greece

A critical overview of recent developments of NMR spectroscopy in natural products will be provided with emphasis in the following applications:
(i) chemical analysis of extracts without isolation or derivatization steps [1],
(ii) '*in situ*' monitoring of dynamic changes of metabolites [2], '*in situ*' analysis of enzymatic reaction products [3], and enriching the biological space of natural products, through monitoring in the NMR tube [4],
(iii) "in-cell" NMR in decoding the apoptotic activity of flavonoids [5] and artemisinin with the Bcl-*2* family of proteins and
(iv) quantum chemical calculations of high-resolution structures in solution based on NMR chemical shifts – comparison with X-ray and neutron diffraction methods [6].

Acknowledgments: We are grateful to the Operational Programme, Human Resources Development, Education and Lifelong Learning, co-financed by Greece and the European Union, for financial support (code MIS: 5005243).

Keywords: Chemical shifts, DFT, NMR, in-cell NMR

References

[1] Kontogianni V *et al*. Org Biomol Chem2013; 11: 1013–1025. Tsiafoulis CG *et al.* Anal Chim Acta 2014; 821: 62–71; Charisiadis P *et al.* Molecules 2014; 19: 13643–13682; Papaemmanouil Ch *et al*. J Agric Food Chem 2015; 63: 5381–5387. Kontogianni V *et al*. Anal Methods 2017; 9: 4464–4470, Charisiadis P *et al*. Phytochem Anal 2017;28: 159–170.
[2] Charisiadis P *et al.* Magn Reson Chem 2014; 52: 764–768.
[3] Kyriakou E *et al*. Org Biomol Chem 2012; 10: 1739–1742.
[4] Chatzikonstantinou A *et al.* BBA - General Subjects 2018; 1862:1–8.
[5] Primikyri A *et al.* ACS Chem Biol 2014; 9: 2737–2741.
[6] Siskos M *et al*. Org Biomol Chem 2013; 11: 7400–7411. Siskos M *et al.* Org Biomol Chem 2015; 13: 8852–8868; Siskos M *et al*. Teteahedron 2016; 72: 8287–8293. Siskos M *et al.* Org Biomol Chem 2017; 15: 4655–4666. Siskos M *et al*. Molecules 2017; 22: 415. Siskos M *et al*. Teteahedron 2018, *in press.*

SL-003

The use of vibrational spectroscopy in natural product analysis: current and future directions

Christian Huck

University of Innsbruck, Inst. of Analytical Chemistry, Innsbruck, Austria

The field of molecular vibrational spectroscopy applied to natural product and bio-analysis is further developing very dynamically. Whereas traditional separation and mass spectrometric (MS) techniques offer analytical investigations with high selectivity and sensitivity, vibrational spectroscopy benefits from the short analyses times, non-invasiveness and the possibility to screen for chemical and physical properties simultaneously. Furthermore, chemometrical univariate and multivariate data treatment enables efficient spectral interpretation and the establishment of sufficient calibration/validation models. Advanced quantum chemical approaches can further support the challenge of band assignment, especially in case of complex overlapping peak patterns. Near-infrared (NIR, 4.000-10.000 cm^{-1}), attenuated total reflection (ATR, 400-4.000 cm^{-1}) and Raman spectroscopy have been demonstrated being very efficient for even complex qualitative and quantitative attempts in combination with selective reference analytical methods. Qualitative attempts comprise analysing, e.g., species and in some cases also origin, quantitative analysing chemical and physical parameters [1]. Two-dimensional correlation spectroscopy (2D-COS) has been developed towards a powerful analysis tool for monitoring the dynamics of a spectrometer system [2]. The miniaturization of spectrometers is a highly demanding trend, enabling to carry out investigations at any independent place. Imaging and mapping spectroscopic attempts (MIR, NIR, Raman) enable high-resolution analysis of potent ingredients down to approximately 4 µm and 1 µm, respectively. This contribution highlights recent advances of molecular spectroscopy in natural product research. The latest technical developments will be discussed followed by several selected applications in food and medicinal plant analysis. Their limits and advantages over traditional methods will be critically evaluated to point out the future trends.

Keywords: Vibrational spectroscopy, imaging analysis, chemometrics

References

[1] Henn R, Kirchler CG, Grossgut ME, Huck CW, Talanta 2017; 166: 109–118.
[2] Kirchler CG, Pezzei CK, Bec KB, Mayr S, Ishigaki M, Ozaki Y, Huck CW, Analyst 2017; 142: 455–464.

SL-004

Mass Spectrometry metabolomics to identify bioactive compounds and synergists from botanical natural products

Nadja Cech

University of North Carolina at Greensboro, Greensboro, United States

Plants have been employed since antiquity for the treatment and prevention of disease, and still constitute a major form of healthcare worldwide. The immense chemical complexity and variability of plant based (botanical) medicines makes evaluation of their safety and efficacy extremely challenging. Much of the research in the field of botanical natural products has focused on reducing this complexity in pursuit of single active compounds that can be developed as pharmaceutical drugs. However, traditional health care practices typically employ botanical medicines as complex mixtures, and methods for understanding the interactions that occur in these mixtures are limited. Towards this goal, we have developed metabolomics tools to understand combination effects in botanical medicines. This presentation will focus on a new approach developed by our laboratory (comparative bioactivity modeling) that employs comparisons between predicted and observed biological activity of mixtures to identify those that contain synergists. Integral to the success of this approach is the use of informatics strategies to integrate biological and chemical datasets. We will provide data demonstrating the applicability of comparative bioactivity modeling using antimicrobial activity of the traditional Chinese medicine *Salvia miltiorrhiza* (Danshen) as a test case. Our data demonstrate the feasibility of the approach for predicting which mixture constituents are responsible for synergistic, additive, or antagonistic interactions prior to isolation.

SL-005

Mass spectrometric dereplication of natural products using mass spectrometry integrated approach

Syed Ghulam Musharraf, M. Iqbal Choudhary and Atta-ur-Rahman

H.E.J. Research Institute of Chemistry, International Center for Chemical and Biological Sciences, University of Karachi, Karachi-75270, Pakistan

Plant metabolites can act as drugs for the treatment of a variety of diseases due to their unique skeletal features. A large number of plant metabolites are used as drugs for the treatment of many diseases. The structural diversity of these plant metabolites formed by complex enzymatically controlled pathways is still not fully explored. Moreover, to preserve the medicinally important endemic and non endemic plant species and their sustainability, the quantity of plant material has been limited to the analytical level. Therefore, sensitive and high-throughput dereplication methods are required for better, unambiguous and high-throughput analysis of natural products in complex mixtures.

This talk will focus on the high-throughput dereplication strategy for the unambiguous identification of different classes of natural product through LC-ESI-MS/MS in the plant extract using an integrated approach which includes several confirmatory steps such as exact masses measurement, diagnostic fragment ions, databases search, and isotopic pattern. Based on above mentioned integrated approach, we have recently investigated different classes of natural products including withanolides (steroidal lactones) [1], pregnane-type steroidal alkaloids [2] and *Buxus* steroidal alkaloids [3, 4] and indentified them in the extract of *Withania somnifera*, *Sarcococca coriacea* and *Buxus papillosa*, respectively, by using electrospray ionization quadropole time-of-flight mass spectrometry (ESI-QTOF-MS/MS) and LC-QQQ-MS analysis. Moreover, the fragmentation pathways and characteristic fragments of a new triterpenoid [5] and some diterpenoids [6] by using ESI-QqTOF-MS/MS will also be presented.

Keywords: mass spectrometry, dereplication, electrospray ionization, natural products

References:

[1] Musharraf SG, Ali A, Ali RA, Yousuf S, Atta-ur- Rahman and Choudhary MI, Rapid Commun Mass Spectrom 2011; 25: 104.
[2] Musharraf SG, Goher M, Ali A, Adhikari A, Choudhary MI and Atta-ur-Rahman. Steroids 2012; 77: 138.
[3] Musharraf SG, Goher M, Shahnaz S, Choudhary MI and Atta-ur-Rahman. Rapid Commun Mass Spectrom 2013; 27: 169.
[4] Musharraf SG, Goher M, Bibi Z, Steroids 2015; 100: 5–10.
[5] Musharraf SG, Goher M, Wafo P, Kamdem RST, I J Mass Spectrom 2012; 310: 77.
[6] Musharraf SG, Goher M, Hussain A and Choudhary MI. Chem Cent J 2012; 6: 120.

SL-006

Metabolomic strategies for marine natural products analysis

Norberto Peporine Lopes

Department of Physic and Chemistry, NPPNS-FCFRP, University of São Paulo. Av. Do Café s/n, Bairro Monte Alegre, CEP: 14903-040, Ribeirão Preto, São Paulo Brazil

Plants and other living organisms have long been used as source for different useful human products. Traditional medicines have always been a source for the cure of many diseases since antiquity. However, their rational use was possible only after the understanding of how the compounds present in plants had their activities proved. Then the search for new bioactive compounds had a huge development and as consequence, a number of new molecules with different spectra of activities were found. Numerous examples of bioactive natural products are known, however their discoveries have always been associated with the development of new analytical techniques. Recently metabolomics strategies based on mass spectrometry improved dereplication process and also open new perspectives for chemical biology investigations. Even though the search for new drugs from plants and other organisms is still exciting and attractive, chemistry has played a key role in the explanation of biological and biochemical observations, opening a number of opportunities in the area. Each single organism (plant, marine or terrestrial animals, microorganism, algae, among others) interacts with ecosystem by different strategies including chemical signals and/or chemical defenses. Compounds involved in this process are usually secondary metabolites, than we can accept the concept that organism actions are governed by the flux of energy and information through an enormous number of molecules. Also, the natural products can have unknown physiological functions that must be deeply understood in the future. In this talk we provide an integrative overview of mass spectrometry strategies helping integrative metabolomics analysis and imaging generation for the Brazilian Biodiversity.

Keywords: Brazilian Biodiversity, Brazilian Savana, Atlantic Forest, Mass Spectrometry.

SL-007

Bioactive alkaloids towards enzymes related to neurodegeneration from *Psychotria nemorosa*

Sylvian Cretton[1], Luiz C. Klein-Júnior[2,3], Amélia T. Henriques[2], Muriel Cuendet[1], Philippe Christen[1]

[1]University of Geneva, Switzerland, [2]Universidade Federal do Rio Grande do Sul, Rio Grande do Sul, Brazil, [3]Universidade do Vale do Itajaí, Itajaí, Brazil

Psychotria nemorosa Gardner (Rubiaceae) is a shrub reaching 2 m in height that grows in the rainforest from subtropical and tropical regions of Brazil. Species belonging to this genus are known to be rich in bioactive indole alkaloids acting on enzymes related to neurodegeneration such as monoamine oxidase-A (MAO-A) and butyrylcholinesterase (BChE). The alkaloid extract of the aerial parts from *P. nemorosa* was tested towards MAO-A and BChE and showed a strong inhibitory activity [1]. Seventeen alkaloids, among which twelve uncommon azepino-indole derivatives, were isolated from the active extract by semi-preparative HPLC, and characterized by NMR including ^{1}H-, ^{13}C-NMR, HSQC, HMBC, COSY and NOESY experiments, as well as ECD, UV, IR, and mass spectrometry. Amid the azepino-indole derivatives, ten compounds have not yet been described in the literature. Furthermore, some of these compounds showed significant *in vitro* inhibition of MAO-A and BChE with IC_{50} values below 20 µM.

Keywords: *Psychotria nemorosa*, azepino-indole alkaloids, butyrylcholinesterase

References:

[1] Klein-Júnior LC *et al.* J Chromatogr A 2016; 1463: 71–80.

SL-008

Integrating chemical and biological data to investigate *Echinodorus grandiflorus* as a source of potential anti-arthritic agents

Fernão Braga[1], Eliana Garcia[1], Diogo Oliveira[1,2], Flávio Amaral[2], Danielle Souza[2], Rodrigo Pádua[1], Mauro Teixeira[2]

[1]Faculty of Pharmacy, Federal University of Minas Gerais, Belo Horizonte, Brazil, [2]Institute of Biological Sciences, Federal University of Minas Gerais, Belo Horizonte, Brazil

The leaves of *Echinodorus grandiflorus* (Alismataceae) are traditionally used in Brazil to treat arthritis and other inflammatory conditions. Extracts and fractions of different polarities were prepared from *E. grandiflorus* leaves and had their quantitative composition analyzed by HPLC-DAD. The effect of the preparations on the release of TNF-α by LPS-stimulated THP-1 cells was assayed. Polynomial regression analysis disclosed the association between the contents of swertiajaponin, swertisin, *trans*-aconitic and chicoric acids with the inhibition of TNF-α release elicited by the extracts and fractions. None of the compounds tested alone abolished TNF-α release completely, as found by some extracts and fractions, suggesting a synergistic effect between the constituents. Therefore, a standardized flavonoid-rich fraction (FLAV) was produced and given orally (0.7-7.2 mg/kg) to previously immunized mice. FLAV treatment reduced neutrophil recruitment to the joint cavity and in periarticular tissue. The levels of CXCL-1, TNF-α and IL-1β were diminished in the periarticular tissue of FLAV-treated mice, as well as mechanical hypernociception. Histological analysis confirmed that FLAV suppressed joint inflammation and inhibited cartilage and bone destruction. Aiming to explore the anti-inflammatory potential of *trans*-aconitic acid (TAA), mono-, di-, and triesters thereof were prepared and screened in a model of LPS-induced acute arthritis in mice. Diesters were the most active derivatives, regardless of the alcohol employed in esterification, whereas bioactivity of the compounds improved by increasing the length of the aliphatic chain of the alcohol employed in reaction. In general, the esters showed higher potency than TAA. When administered orally to mice (0.017 to 172.3 μmol/Kg), the diethyl, di-n-butyl and di-*n*-octyl esters of TAA reduced the cellular infiltration into the knee joint, especially of neutrophils. In conclusion, diesters of TAA and flavonoids were identified as potentially useful derivatives / compounds from *E. grandiflorus* leaves for the management of rheumatoid arthritis.

Acknowledgements: FAPEMIG, CAPES and CNPq (Brazil).

SL-009

Bioguided fractionation and molecular networking to access the chemical diversity of bioactive extracts on fibroblasts AGEs

Amandine André[1,2], Kaatio Touré[2], Joanna Wdzieczak-Bakala[1], Didier Stien[3], Véronique Eparvier[1]

[1]CNRS Institut de Chimie des Substances Naturelles, Gif-sur-Yvette, France, [2]Laboratoire SHIGETA, Paris, France, [3]Sorbonne Université, CNRS, Laboratoire de Biodiversité et Biotechnologie Microbienne, Banyuls-sur-mer, France

Glycation is a non-enzymatic reaction between proteins and sugars. It gives rise in the human body to the formation of Advanced Glycation End products (AGEs). Accumulation of AGEs in the skin occurs upon aging and leads to the formation of wrinkles, dark spots and yellowish skin tone [1]. The aim of our study was to find natural products able to reduce intracellular glycation in order to develop original and effective anti-aging ingredients.

In order to screen a library of microbial extracts, a method was developed to quantify the intracellular glycation of dermal fibroblasts.

This led to the selection of two ethyl acetate active extracts: one extract of *Colletotrichum gloeosporioides* isolated from the leaves of *Sabicea cinerea*, and one extract of *Sphingobacterium* sp. isolated from a *Nasutitermes* sp. nest. These two extracts exhibited significant deglycation activity on human dermal fibroblasts (Fig. 1A).

Thorough chemical investigation led to the isolation of diverse compounds, including four previously unknown acoranes (Fig. 1B) from *Colletotrichum gloeosporioides* [2], and various 2,5-diketopiperazines from *Sphingobacterium* sp.

The use of molecular networking to explore more in depth the chemical diversity of these extracts led us to determine the presence of three other isomers of the isolated acoranes in the *Colletotrichum* extract, and numerous other 2,5-diketopiperazines in the *Sphingobacterium* extract.

To conclude, bioguided fractionation led us to discover new and known compounds with various chemical structures, while analysis of molecular networks let us access a second level of knowledge where diversity of minor compounds can be revealed.

Acknowledgements: This works has benefited from a CIFRE grant (Association Nationale de la Recherche et de le Technologie, France, ANRT n°2013-1326), and from an "Investissement d'Avenir" grant (Agence Nationale de la Recherche, France, CEBA, ANT-10-LABX-25-01).

References:

[1] Gkogkolou P *et al*. Dermatoendocrinol 2012; 4(3): 259–70.
[2] André A *et al*. Tetrahedron Lett 2017; 58(13): 1269–72.

SL-010

Isolation and identification of secondary metabolites, including four new compounds, from three *Digitalis* species

Vahap Murat Kutluay[1], Kan'ichiro Ishiuchi[2], Toshiaki Makino[2], Fatih Göger[3], Nese Kirimer[3], Iclal Saracoglu[1]

[1]Department of Pharmacognosy, Faculty of Pharmacy, Hacettepe University, 06100, Turkey, [2]Department of Pharmacognosy, Graduate School of Pharmaceutical Sciences, Nagoya City University, 467- 8603, Japan, [3]Department of Pharmacognosy, Faculty of Pharmacy, Anadolu University, 26470, Turkey

In the flora of Turkey, the genus *Digitalis* L. is presented by 9 species [1]. *Digitalis* was formerly a member of Scrophulariaceae, but after phylogenetic and chemotaxonomic studies it has been moved to Plantaginaceae [2]. In this research 1 endemic total 3 species; aerial parts of *D.davisiana* Heywood, *D.grandiflora* Miller and *D.viridiflora* Lindley were studied. Studies on methanolic extracts resulted in the isolation of 4 new compounds; digidavisoside A, digidavisoside B, davisoside [phenylethanoid glycosides (PGs)] from *D.davisiana* and digigrandifloroside (cardenolide glycoside) from *D.grandiflora*. Totally 18 compounds were isolated which 11 of them are PGs. The known compounds isolated from *D.davisiana* were characterized as ferruginoside B, isolugrandoside, lugrandoside, maxoside, 3''''-*O*-methylmaxoside, trans-lamiuside E, digiciliside B, p-hydroxyacetophenon, chyrsoeriol and from *D.grandiflora* were characterized as salidroside, cornoside, rengyosides A, B and cleroindicin A. 3 species were compared by LC-MS/MS analysis. Lugrandoside and maxoside were detected as major compounds. Results were evaluated on the basis of phytochemistry and chemotaxonomy. PGs were selected for chemotaxonomical evaluation. Although PGs could be found widely in other families, 3'-O-glucosyl-caffeoyl glycosides found only in Plantaginaceae [3]. In our study from isolated 11 PGs, 6 of them are 3'-*O*-glucosyl-caffeoyl glycosides. Our results support the chemotaxonomical studies of Plantaginaceae family. In addition, this is the first report for the isolation of rengyosides A-B and p-hydroxyacetophenon from a *Digitalis* species.

Acknowledgements:This study is supported by TUBITAK and HUBAB

Keywords: *Digitalis*, Plantaginaceae, phenylethanoid glycosides, chemotaxonomy

References:

[1] Davis, PH (1978). Flora of Turkey and the East Aegean Islands (c. 6). Edinburgh: University Press
[2] Albach DC *et al*. Am J Bot 2005; 92 (2): 297–315
[3] Jensen SR *et al*. Biochem Syst Ecol 2011; 39 (3): 193–197

SL-011

Caged xanthones from *Garcinia propinqua*

Surat Laphookhieo[1], Teerayut Sriyatep[1], Cholpisut Tantapakul[1], Chatchai Muanprasat[2], Sawinee Seemakhan[2], Suparerk Borwornpinyo[2], Stephen Pyne[3], Brian Patrick[4], Raymond Andersen[4]

[1]Mae Fah Luang University, Muang, Thailand, [2]Mahidol University, Rajathevi, Thailand, [3]University of Wollongong, Wollongong, Australia, 4University of British Columbia, Vancouver, Canada

Eight new caged xanthones, doitunggarcinones E-L (1-8), all as scalemic mixtures, were isolated from the stem bark and leaf extracts of *Garcinia propinqua*. Five other known caged xanthones (9-13) were also isolated as scalemic mixtures. The structures were elucidated on the basis of spectroscopic methods. The separation of the enantiomers of 1-13 was achieved by semi-preparative chiral HPLC. The absolute configuration of compound (+)-1, (+)-9 and (+)-10 was determined by single-crystal X-ray crystallographic analysis using Cu Kα radiation. The absolute configurations of the other related compounds were determined from comparisons of their ECD spectra with those of compound (+)-1 (+)-1, (+)-9 and (+)-10. Compounds (–)-6 and 7 showed weak cytotoxicities against a colon cancer cell line HCT116 with IC_{50} values of 14.23 μM and 23.95 μM, respectively, whereas compounds (-)-11, (+)-11, (-)-12, (+)-12, and (-)-13 showed potent cytotoxicities against a colon cancer cell line with IC_{50} values of 2.60, 7.02, 1.47, 3.37, and 4.14 μM, respectively, which were better than the standard control doxorubicin (IC_{50} 9.74 μM).

SL-012

Simple screening for high-colibactin producers from clinical samples using activity-based fluorescent probe

Kenji Watanabe

Department of Pharmaceutical Sciences, University of Shizuoka, Shizuoka, Japan

Colibactin-producing *Escherichia coli* has been linked to colorectal oncogenesis. However, the known colibactin producer Nissle 1917 has been used clinically as a probiotic for over a century without a report of increased risk of developing colorectal cancer. While it is hypothesized that high-colibactin producers may be associated with, the study is complicated further by the limited understanding of colibactin biosynthesis, which is part caused by the inability to isolate colibactin in large quantity from colibactin-producing *E. coli*. Here we report the creation of fluorescent probes that are activated by ClbP, the peptidase that matures precolibactin into genotoxic colibactin, to facilitate the identification of high-colibactin producers. Our probe successfully isolated an *E. coli* strain that can produce colibactin 26-fold greater than Nissle 1917 from a colorectal cancer sample. We also demonstrated the usefulness of our probe in high-throughput screening of clinical samples for identifying colibactin-producing isolates. The probe only serve as a valuable clinical diagnostic tool but also be used to identify high colibactin producers that would help advance our understanding of colibactin biosynthesis.

SL-013

Botanical nematicides originating from Mediterranean plants: An overview

Nikoletta Ntalli[1], Urania Menkissoglu-Spiroudi[2]

[1]Department of Pesticides' Control and Phytopharmacy, Benaki Phytopathological Institute, 8 S. Delta Str, Athens, Greece, [2]Pesticide Science Laboratory, School of Agriculture, Faculty of Agriculture Forestry and Natural Environment, Aristotle University of Thessaloniki, 54124 Thessaloniki, Greece, Thessaloniki, Greece

Plant-parasitic nematodes are major agricultural pests causing crop losses exceeding $100 billion dollars and in particular the genus *Meloidogyne* sp., parasitizes almost every species of vascular plant. Traditionally phyto-nematodes control has been based on soil sterilants, of great environmental impact, most of which are now banned. Consequently, it is mandatory to develop eco-sustainable alternative tools. The Mediterranean Basin is an area where various soils and climatic conditions allow a vast plant biodiversity providing with chemical botanicals of significant nematicidal potency. This is our groups' 10 years' work review on the Mediterranean botanicals that can control *Meloidogyne* sp.

Over 20 plant species were studied for their nematicidal activity against different *Meloidogyne* sp., species and growth stages; and over 25 active ingredients have been identified and quantified in the active plant extracts. In terms of efficacy, the parameters studied were paralysis capacity, egg hatch inhibition activity and parasite biological cycle arrest in host roots. Synergism, fumigant activity and attraction have been studied along with the mode of action on nematode cuticle or/and biochemical target site. Beneficial side effects were delineated in soil microcosms and plant physiology.

Conclusively an integrated pest management can be sustained if : a) nematicidal plant species are used in crop rotation for incorporation prior to the establishment of the nematode susceptible crop, b) the aqueous edible extracts are considered of inclusion into the low risk (EC 1107/2009 Article 22, 47) or basic substances (EC 1107/2009 Article 23) catalogs and c) the pure nematicidal compounds (aldehydes, ketones, acids, isothiocyanates, furanocoumarins) are used as model molecules for synthesis, to be formulated alone or in synergistic mixtures with attractive properties. Some additonal applications with regard to pharmacy, that is antimicobacterial and anticancer activity, are mentioned.

SL-014

Reduction of polyglutamine aggregates in a cellular model of Huntington's disease by an ethanolic extract of *Ugni molinae* wild-type strain.

Alexis Rivas[1,2,3], Rodrigo Perez-Arancibia[1,4], Carla Delporte[4], Claudio Hetz[1,2,3,5,6]

[1]Biomedical Neuroscience Institute, Faculty of Medicine, University of Chile, 8380453, Independencia, Santiago, Chile, [2]Center for Geroscience, Brain Health and Metabolism, 7800003, Ñuñoa, Santiago, Chile, [3]Program of Cellular and Molecular Biology, Institute of Biomedical Sciences, University of Chile, 8380453, Independencia, Santiago, Chile, [4]Laboratory of Natural Products, Faculty of Pharmaceutical and Chemical Sciences, University of Chile, 8380494, Independencia, Santiago, Chile, [5]Buck Institute for Research on Aging, 94945, Novato, United States, [6]Department of Immunology and Infectious Diseases, Harvard School of Public Health, 02115, Boston, United States

The most common neurodegenerative diseases like Alzheimer's disease, Parkinson´s disease, amyotrophic lateral sclerosis (ALS) and Huntington´s disease (HD) are characterized by common pathological features, highlighting the accumulation of neurotoxic protein aggregates. Thus, strategies to reduce protein aggregation and toxicity represent a relevant therapeutic target. Our laboratory focuses on studying the role of protein aggregation in neurodegenerative diseases and the identification of possible pathological mechanisms that contribute to the loss of neuronal function. This work proposes to uncover new biomedical applications of small molecules present in native Chilean plants. We have established a collaboration network with different laboratories of organic chemistry with experience with native flora to develop a new research line in our country. We have generated a library of semi-purified extracts from different plants and screen them using cell-based assays to monitor protein aggregation using an automated platform for microscopy with high capacity. From this screening, we identified a strong anti-aggregation activity of two extracts from wild-type *Ugni molinae* leaves. Interestingly, there were previously identified anti-inflammatory triterpenoids from ethyl acetate and ethanol extracts derived from *Ugni molinae* leaves, associated with a rich content of antioxidants phenolic compounds. These results were validated by western blot and filter trap assays in neuronal cell lines, where the reduction of aggregates of polyglutamine was detected as well. The previous characterization of those extracts using mass spectroscopy shows the main group of glycosylated compounds derived from phenolic acids, which need to be purified to tested them individually for identification of the active compound(s).

SL-015

In vitro anti-cancer activity of *Barringtonia asiatica* (L.) Kurz crude methanolic leaf extract against human breast adenocarcinoma cell line, MCF-7

Ronald Arlet Villaber[1], Kerl Francis Pastoril[2]

[1]*Department of Pure and Applied Chemistry, Visayas State University, Baybay City, Philippines,* [2]*Department of Biotechnology, Visayas State University, Baybay City, Philippines*

Cancer is one of the most widespread diseases and has been considered as a major threat to the majority of the people in the world today. It is one of the leading causes of morbidity and mortality worldwide. The cytototoxic effect of *Barringtonia asiatica* (L.) Kurz crude methanolic leaf extract was determined on human breast adenocarcinoma cells (MCF-7) by MTT assay [1]. The highest cytotoxic activity was observed when MCF-7 cells were exposed to 100 µg/mL of the crude extracts. The total phenolic and flavonoids contents were determined using Folin-Ciocalteu and aluminum chloride methods [2], respectively. The free radical scavenging activity was performed by 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay.

The total phenolic content was 52.66 ± 1.90 mg GAE/g Sample, and the total flavonoid content was 98.19 ± 2.93 mg QE/g Sample. The crude methanolic leaf extract of *B. asiatica* has a free radical scavenging activity 83.68±0.91%. Results showed that leaves of *Barringtonia asiatica* (L.) Kurz possess significant free radical scavenging activity, and clear correlation exists between the strong antioxidant activity with the flavonoid contents.

The results suggest that *Barringtonia asiatica* (L.) Kurz can be regarded as a promising candidate for natural plant source of anticancer and antioxidant compounds. Moreover, the results support the need of further studies to isolate potential anticancer compounds with cancer cell specific cytotoxicity.

Acknowledgement: Department of Pure and Applied Chemistry, Visayas State University, Department of Biotechnology, Visayas State University

Keywords: Antioxidant, cancer, cytotoxicity, flavonoids, phenolics

References:

[1] Greenwell M, Rahman P. Int J Pharm Sci Res 2015; 4103–4112.
[2] Baba S, Malik S. Journal of Taibah University for Science 2015; 449–454.

SL-016

HPLC-PDA based chemical profiling and antimicrobial activity studies on *Citrus grandis* and *C. sinensis* from the Iraqi flora

Shaymaa Al-majmaie, Lutfun Nahar, George P Sharples, Satyajit D Sarker

Medicinal Chemistry and Natural Products Research Group, School of Pharmacy and Biomolecular Sciences, Liverpool John Moores University, James Parsons Building, Byrom Street, Liverpool L3 3AF, United Kingdom, Liverpool, United Kingdom

The genus *Citrus* of the family Rutaceae from the Iraqi flora produces vitamin C, essential oils and phenolic compounds. Phenolic compounds from this genus, especially flavonoids like hesperidin, narirutin, naringin, neohesperidin, eriocitrin, neoeriocitrin, rutin, diosmin, neoponcirin, and nobiletin, possess various biological properties. Other bioactive compounds from this genus include *β*-carotene, lutein, lycopene and cryptoxanthin, and cinnamic acid derivatives like caffeic acid, ferulic acid, chlorogenic acid, and *p*-coumaric acid. The aim of the current study was to evaluate the antimicrobial activity of the extracts of the peels, and the leaves of *C. sinensis* and the leaves of *C. grandis*, and to carry out HPLC-PDA based chemical profiling of the extracts. Dried plant materials were subjected to Soxhlet extraction sequentially with *n*-hexane, dichloromethane (DCM) and methanol (MeOH) to obtain nine different extracts. The potential antimicrobial property of the extracts as well as the chromatographic fractions was assessed against five microbial strains: *Candida albicans, Escherichia coli, Micrococcus luteus, Pseudomonas aeruginosa* and *Staphylococcus aureus*, using the microtitre plate-based antibacterial assay incorporating resazurin as an indicator of cell growth [1]. Active extracts were fractionated, using Vacuum Liquid Chromatography (VLC) for the *n*-hexane and DCM extracts, and reversed-phase Solid-Phase-Extraction (SPE) for the MeOH extracts [1]. Generally, a majority of these extracts and fractions exhibited antimicrobial activity at different concentrations. The minimum inhibitory concentration (MIC) of the active extracts/fractions of the DCM extract of *C. sinensis* peels was the most active extract (MIC = 6.10×10-4 mg/mL) against *M. luteus*. HPLC-PDA based chemical profiling of the active extracts/fractions revealed the presence of mainly isoquinoline alkaloids, simple coumarins, furanocoumarins and flavonoids. Antimicrobial activities of *C. grandis* and *C. sinensis* as observed in the current study, provided some evidence for their traditional uses to treat infections.

References:

[1] Sarker SD and Nahar L (2012) Natural Products Isolation, 3rd ed. Springer USA

SL-017

Evaluation of antimicrobial, disinfectant and anti-inflammatory potential of *Garcinia cambolia*: A potent source for pharmaceuticals and disinfectants

Mayuri Napagoda[1], Sudhara De Soyza[1], Jana Gerstmeier[2], Hannah Butschek[2], Andreas Koeberle[2], Mallique Qader[3], Sybille Loranz[4], Sanjeeva Witharana[5], Gaya Bandara Wijayaratne[6], Aleš Svatoš[4], Lalith Jayasinghe[3], Oliver Werz[2]

[1]Department of Biochemistry, Faculty of Medicine, University of Ruhuna, Galle, Sri Lanka, [2]Institute of Pharmacy, Friedrich-Schiller-University Jena, Philosophenweg 14, D-07743 Jena, , Germany, [3]National Institute of Fundamental Studies, Kandy, , Sri Lanka, [4]Max Planck Institute for Chemical Ecology, Hans-Knoell-Strasse 8, D-07745 , Jena, Germany, [5]Faculty of Engineering, Higher Colleges of Technology, PO Box 4793, , United Arab Emirates, [6]Department of Microbiology, Faculty of Medicine, University of Ruhuna, Galle, Sri Lanka

Garcinia cambogia is extensively utilized in indigenous medicine in Sri Lanka to treat inflammatory conditions, skin diseases and related disorders. However, neither its pharmacological features nor the phytochemistry are explored in depth to rationalize the reported ethnobotanical significance. Thus, the present study is undertaken to investigate antimicrobial, disinfectant and anti-inflammatory activities of different extracts prepared from fruits of *G. cambogia* and to study its phytochemical profile. The antimicrobial activity of the extracts against Gram positive and Gram negative bacteria including clinical isolates of methicillin resistant *Staphylococcus aureus* (MRSA) was evaluated by the broth micro-dilution assay while the disinfectant potential was determined by surface disinfectant assay. Since 5-lipoxygenase (5-LO) and microsomal prostaglandin E2 synthase (mPGES)-1 are well-known target enzymes associated with inflammatory disorders, cell-free and cell-based assays were employed to investigate the suppression of 5-LO and mPGES-1 activities. Out of the tested extracts, a conspicuous antibacterial activity was observed in the *n*-hexane extract with minimum inhibitory concentration (MIC) of 31.25-125 µg/mL against *Staphylococcus aureus*, *S. saprophyticus* and MRSA. Interestingly, these MIC values were significantly lower versus those of most of the ubiquitous phyto-constituents. Moreover, the disinfectant capacity of this extract against *S. aureus* and MRSA isolates was comparable to that of the commercial disinfectant used as the positive control. Further, the *n*-hexane extract displayed highly potent anti-inflammatory activity with IC_{50} of 0.15 and 0.92 µg/mL in cell-free and cell-based 5-LO assays, respectively, and an IC_{50} of 0.29 µg/mL in mPGES-1 assay. These potencies are much superior over herbal extracts frequently used as anti-inflammatory remedies in Western countries. The plant contains phytosterols, fatty acids, sesquiterpenes, and several other types of secondary metabolites, as revealed by GC-MS analysis. Together, our findings demonstrated that *G. cambogia* possess significant biological activities, and further studies are in progress in the pursuit of new phytotherapeutics and disinfectants.

SL-018

Folic acid supplementation repressed hypoxia-induced inflammatory response in human promyelomonocytic cells

Xinwei Jiang

Jinan University, Guangzhou, China

Hypoxia is positively correlated with inflammation and various chronic diseases. Folic acid as a dietary compound has been shown to ameliorate inflammatory reactions, but the metabolism of folic acid protecting against hypoxia-induced injury is still unclear. In our study, we examined the inflammatory signal transduction pathway in folic acid treated human pro-myelomonocytic cells (THP-1 cells) under hypoxic culture conditions. Our results indicated that the supplementation with folic acid significantly reduced the levels of interleukin 1β, interleukin 8, tumor necrosis factor and increased levels of interleukin 10 in cells under hypoxic conditions. Treating THP-1 cells with folic acid suppressed oxidative stress, hypoxia-inducible factor 1α (HIF-1α), and upregulated PHD1 in a dose-dependent manner. Folic acid suppressed hypoxia-induced Akt phosphorylation and NF-κB translocation. Additionally, folic acid targeted the activation of Janus kinase 2(JAK2), down-regulated the phosphorylation of activators of transcription 3 (STAT3) in cells. However, the absence of folic acid did not make cells more vulnerable under hypoxic conditions. In conclusion, folic acid efficiently inhibited the inflammatory response of THP-1 cells under hypoxic conditions by mediating PI3K/Akt/HIF-1α and JAK2/STAT3 signaling pathway. Our study supports a basis for treatment with folic acid for chronic inflammation, which is correlated with hypoxia.

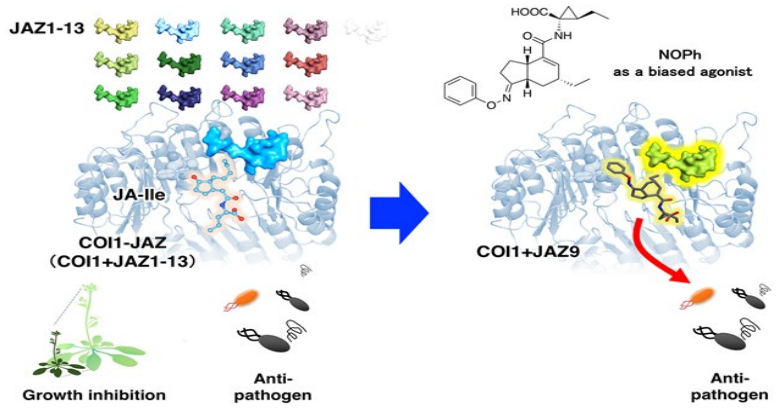
SL-019

Chemical tuning of plant hormone by using stereoisomer of a natural product

Minoru Ueda

Tohoku University, Sendai, 日本

The phytohormone 7-iso-(+)-jasmonoyl-L-isoleucine (JA-Ile) mediates plant defense responses against herbivore and pathogen attack, and thus increases plant resistance against foreign invaders [1]. However, JA-Ile also causes growth inhibition of plants as a serious side-effect. This 'tradeoff' between growth and defense causes a serious difficulty in the application of JA-Ile as a chemical regulator of plant.
We will report the rational design and synthesis of a small molecule agonist (NOPh) that can upregulate defense-related gene expression and promote pathogen resistance without causing growth inhibition in Arabidopsis (Figure) [2]. We developed such a 'dream molecule' based on a stereoisomer of a JA-Ile mimic natural product, coronatine (COR) [3,4]. JA-Ile/COR is perceived by the COI1-JAZ co-receptor, and there coded the one COI1 and 13 JAZ idoforms in Arabidopsis genome. NOPh can stabilize interactions between COI1 and JAZ9/JAZ10 but no other JAZ isoforms, the agonist leads to formation of JA-Ile co-receptors that selectively activate the JAZ9-EIN3/EIL1-ORA59 signaling pathway. The design of a NOPh agonist with high selectivity for specific protein subtypes may help promote the development of chemical regulators that do not cause a tradeoff between growth and defense.



References:

[1] Wasternack C, Kombrink E. ACS Chem Biol 2010; 5: 63–77
[2] Takaoka Y, Iwahashi M, Chini A, Saito H, Ishimaru Y, Egoshi S, Kato N, Tanaka M, Bashir K, Seki M, Solano R, Ueda M. Nat Commun 2018; 9: 3654
[3] Ueda M, Egoshi S, Dodo K, Ishimaru Y, Yamakoshi H, Nakano T, Takaoka Y, Tsukiji S, Sodeoka M. ACS Cent Sci 2017; 3: 462–472
[4] Okada M, Ito S, Matsubara A, Iwakura I, Egoshi S, Ueda M. Org. Biomol Chem 2009; 7: 3065–3073

SL-020

Nature inspired synthesis in chemical biology

Kamal Kumar

Max Planck Institute of Molecular Physiology, Dortmund, Germany

Molecular complexity and structural diversity of natural products has always been a source of inspiration for organic and medicinal chemists to design small molecules for medicinal chemistry or for other biological applications. Total synthesis of natural products often fails to provide adequate numbers and amounts of their analogues and derivatives for screening purposes. This unfortunate reality has pushed modern probe and drug-discovery research to employ a significant number of accessible synthetic chemical libraries in biological screenings, despite their unimpressive results in the last two decades. Nature is a great chemist and has more to offer to help design synthetic strategies to expand the biologically relevant chemical space that is so demanding for biological screenings. In this talk, our efforts to concisely build structurally diverse molecular frameworks amenable to compound collection synthesis, in particular, by means of various synthetic strategies like cascade reactions, novel annulation and cycloaddition reactions or modification of natural products etc. shall be presented. The role of new complex small molecules in unravelling new biological functions will also be briefly discussed.

SL-021

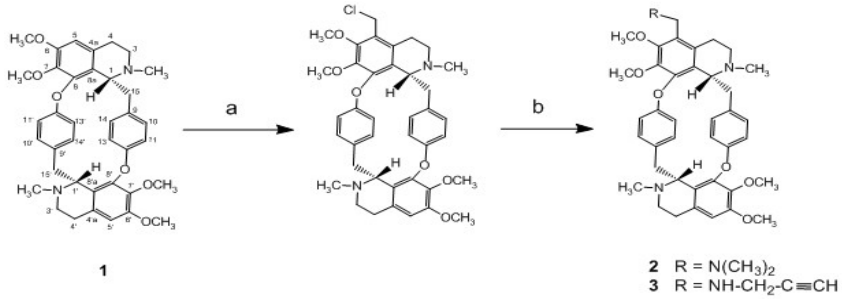
Design, semi-synthesis and evaluation of anticancer and antimalarial cycleanine analogues

Fidelia Uche, Alan Richardson, Paul Horrocks, Wen-Wu Li

Institute for Science and Technology in Medicine, Keele University, Stoke on Trent, United Kingdom

The macrocyclic bisbenzylisoquinoline (BBIQ) alkaloids are among different large classes of plant-derived alkaloids common in Menispermeaceae, Annonaceae, Berberidaceae, Monimaiaceae and Ranunuclaceae [1]. The BBIQ alkaloids were reported to exhibit anti-ovarian, –lung, –bladder, –colorectal, -gallbladder carcinoma and –prostate cancer activities [2] as well as anti-malarial property [1]. The use of isolated natural products as scaffolds to generate their analogues via chemical transformation is a promising approach in drug discovery [3].

Our previous studies demonstrated that BBIQ alkaloids such as cycleanine (1), showed potent anti-ovarian cancer activity via apoptosis induction [4-6]. Here, we synthesized two novel (aminoalkyl)cycleanine analogues (2 and 3) through a simple and efficient two-step reaction starting from cycleanine isolated from *Triclisia subcordata* Oliv. These analogues showed greater potency than the unmodified cycleanine in three human ovarian cancer cell lines. Both 2 and 3 induced apoptosis in ovarian cancer cells by activations of caspases 3/7, cleavage of PARP, increase in subG1 cell cycle phase and in the percentage of apoptotic cells. Confocal fluorescence microscopy analysis confirmed the cellular uptake of alkaloids in ovarian cancer cells by using the unique alknylcycleanine (3) via click chemistry reaction. Furthermore, cycleanine and the two synthetic analogues also showed potent *in vitro* antiplasmodial activity with IC_{50} at micro-molar range. Our results suggest that cycleanine could be a hit compound for the future development in attacking ovarian cancer and malaria.



References:

[1] Schiff PL. J Nat Prod 1997; 60: 934–953.
[2] Bhagya N, Chandrashekar KR. Biomed Pharmacother 2018; 97: 624–632.
[3] Barnes EC, Kumar R, Davis RA. Nat Prod Rep 2016; 33(3): 372–381.
[4] Uche FI, Drijfhout FP, McCullagh J, Richardson A, Li WW. Phytother Res 2016; 30(9): 1533–1539.
[5] Uche FI, Abed M, Abdullah MI *et al.* RSC Adv 2017; 7(70): 44154–44161.
[6] Uche FI, Abed M, Abdullah MI *et al.* Biochem Pharmacol 2017; 139: 112.

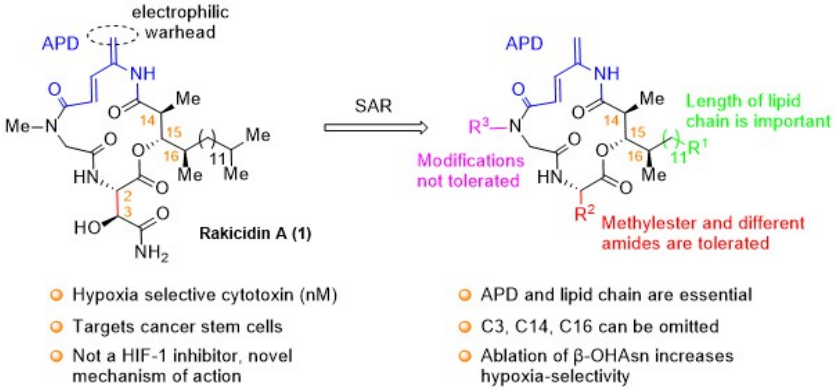
SL-022

Total synthesis and structure-activity relationship study of Rakicidin A. Discovery of analogues with improved pharmacological properties against metastatic cancer.

Michail Tsakos[1], Kristian Jacobsen[2], Nikolaj Villadsen[2], Thomas Poulsen[2]

[1]National And Kapodistrian University of Athens, Zografos, Greece, [2]Aarhus University, Aarhus, Denmark

Microbial natural product Rakicidin A (1, Figure 1) is a macrocyclic lipodepsipeptide with remarkable biological activity. It displays potent and selective growth inhibitory activity against hypoxic cancer cells, as well as, imatinib-resistant chronic myelogenous leukemia cells, a type of cancer stem cells.[1] Tumor hypoxia has been tightly linked to the capacity of cancer cell populations to form new tumors via the activation of a latent transcriptional program called the epithelial-mesenchymal transition.[2] On the other hand, cancer stem cells exhibit enhanced resistance to chemotherapy and radiation and can survive in a quiescent state, giving rise to the relapse of the disease even years after the end of the treatment.
Structurally, Rakicidin A is the first member of a class of natural products that comprise a vinylogous dehydroalanine residue, termed 4-amido-2,4-pentadienoate (APD). This unique functionality is essential for biological activity, most likely interacting as an electrophilic warhead with the biological target. Following the total syntheses of 1 by the Chen [3] and Poulsen [4] groups, a series of analogues has been synthesized with augmented cytotoxicity or selectivity towards hypoxic cancer cells. [4-6] Herein, we wish to discuss the structure-activity relationships of the rakicidin scaffold and, furthermore, provide a status on the ongoing biological investigations with respect to the mechanism of action of the natural product.
Keywords: antitumor agents, tumor hypoxia, cancer stem-cells, cyclolipodepsipeptides, natural products, secondary metabolites.



Acknowledgements: MT would like to thank the Hellenic Foundation for Research and Innovation (HFRI) and the General Secretariat for Research and Technology (GSRT) for funding under grant agreement 15106.

References:

[1] Takeuchi M *et al*. Cancer Sci 2011; 102: 591–596.
[2] Yang M-H *et al*. Nat Cell Biol 2008; 10: 295–305.
[3] Sang F *et al*. J Am Chem Soc 2014; 136: 15787–15791.
[4] Tsakos M *et al*. Angew Chem Int Ed 2016; 55: 1030–1035.
[5] Sang F *et al*. J Med Chem 2016; 59: 1184–1196.
[6] Chen *et al*. Eur J Med Chem 2018; 151: 601–627.

SL-023

Practical synthesis, neurotrophic activities, and structure-activity relationship of talaumidin derivatives

Kenichi Harada[1], Rina Bando[1], Ryo Irimaziri[1], Miwa Kubo[1], Yoshiki Koriyama[2], Yoshiyasu Fukuyama[1]

[1]Faculty of Pharmaceutical Sciences, Tokushima Bunri University, 180 Yamashiro-cho, Tokushima, 770-8514, Japan, [2]Graduate School and Faculty of Pharmaceutical Sciences, Suzuka University of Medical Science, 3500-3 Minamitamagaki, Suzuka, Mie, 513-8670, Japan

(–)-Talaumidin (1), a 2,5-biaryl-3,4-dimethyltetrahydrofuran lignan, shows potent neurotrophic activities such as neurite-outgrowth promotion and neuroprotection in NGF-differentiated PC12 cells and primary cultured rat cortical neurons. Previously, we reported that (–)-(1*S*,2*R*,3*S*,4*R*)-stereoisomer 2 exhibited more significant activity than the natural product 1. In this study, we have established a practical synthesis of talaumidin derivatives, and have investigated their neurotrophic activity *in vivo* and the structure-activity relationship.

To explore more active compounds which can be prepared in a practical scale, we developed a five-step synthesis of talaumidin derivatives. First, a racemic compound of (–)-2 (2a) was synthesized and its neurotrophic activities were assessed. The neurotrophic property of racemic 2a was found to be similar in activity to that of (–)-2. Using the same synthetic methodology as 2a, twelve talaumidin derivatives were synthesized to optimize the oxy-functionality on aromatic rings. As results, bis(methylenedioxybenzene) derivative 2b possessed the highest neurotrophic activity.

Next, our attention was focused on the structure-activity relationship for the neurite-outgrowth activity. In order to examine the roles of aromatic rings and methyl groups in 2b, mono-phenyl compound 3 and demethyl compound 4 were prepared. Although the mono-phenyl 3 showed no activity, the demethyl compound 4 exhibited lower activity than 2b. It implies that two phenyl groups on tetrahydrofuran are the crucial structural units, and two methyl groups serve to enhance the neurotrophic activity.

Finally, we evaluated the neurotrophic activity of 1, 2a, and 2b *in vivo*, which is the regenerative activity of mouse optic nerve. Interestingly, the three compounds were found to induce regeneration of the injured optic nerve at 30 μM. In particular, compounds 2a and 2b induced more significant regeneration than 1.

These results suggest that talaumidin derivatives can be novel therapeutic agents for neurodegenerative diseases such as depression, glaucoma, and Alzheimer's diseases.

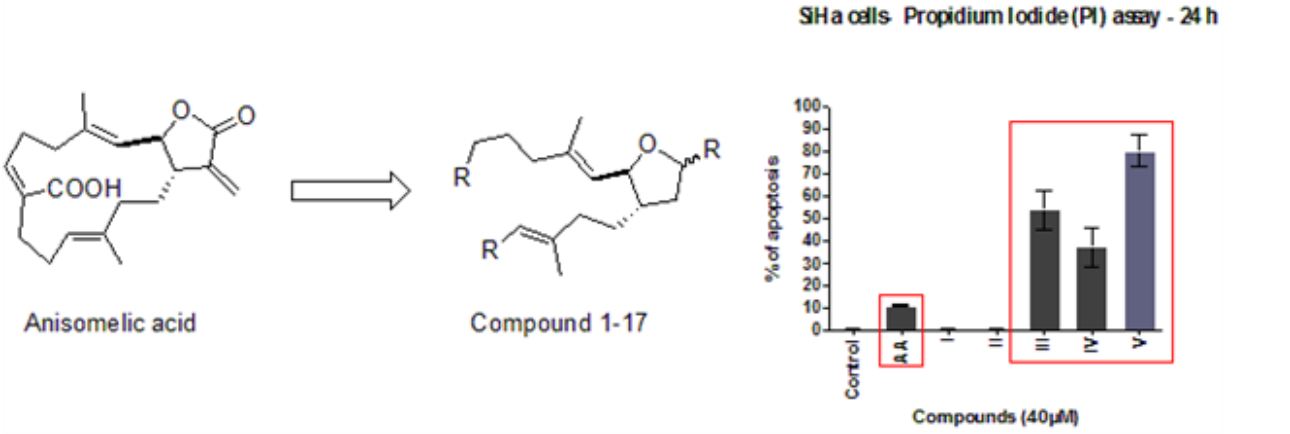
SL-024

Synthesis and evaluation of cembranolide type compounds for treatment of HPV-mediated carcinomas

Patrik Eklund[1], Yury Brusentsev[1], Rajendran Senthil[2] and John Eriksson[2]

[1]Laboratory of Organic Chemistry Åbo Akademi University, Turku, Finland, [2]Laboratory of cell biology, Åbo Akademi University, Turku, Finland

The vast majority of cervical and 75% of oropharyngeal carcinomas is triggered by infection with a high-risk oncogenic human papillomavirus (HPV). HPV E6 and E7 oncoproteins are critical for viral-induced cancer, and hence, they represent valuable targets for therapeutic intervention in HPV-mediated cancers. Our earlier research on the cembranoid anisomelic acid (AA) showed that AA has potential to induce apoptosis in HPV cells by the depletion of E6 and E7 oncoproteins. [1] We here present a structure activity relationship and the evaluation of 17 synthetic anisomelic acid like compounds, i.e simplified cembranolid structures, as HPV inhibitors against some papilloma cell lines. We observed that these compounds induce apoptosis by the same E6/E7-based mechanism as AA, but at earlier time points, thus being far more effective than AA. Binding to the E6 protein was studied by *in silico* docking studies and by STD NMR. Further, the data indicated that only part of the structure of AA (i.e., lactone ring) is required for the molecular action. Further, cancer xenograft models in nude mice demonstrated proof of principle, where a decrease in tumor size was observed in HPV-driven tumors treated with our compounds.



Acknowledgements: Novo Nordisk Foundation and TEKES for their financial support

Keywords: cembranolides, Human papillomavirus, anticancer, synthesis, structure activity relationship, apoptosis

References:

[1] Paul P, Rajendran SK, Peuhu E, Alshatwi AA, Akbarsha MA, Hietanen S & Eriksson JE. Biochem Pharmacol 2014; 89: 171–184.

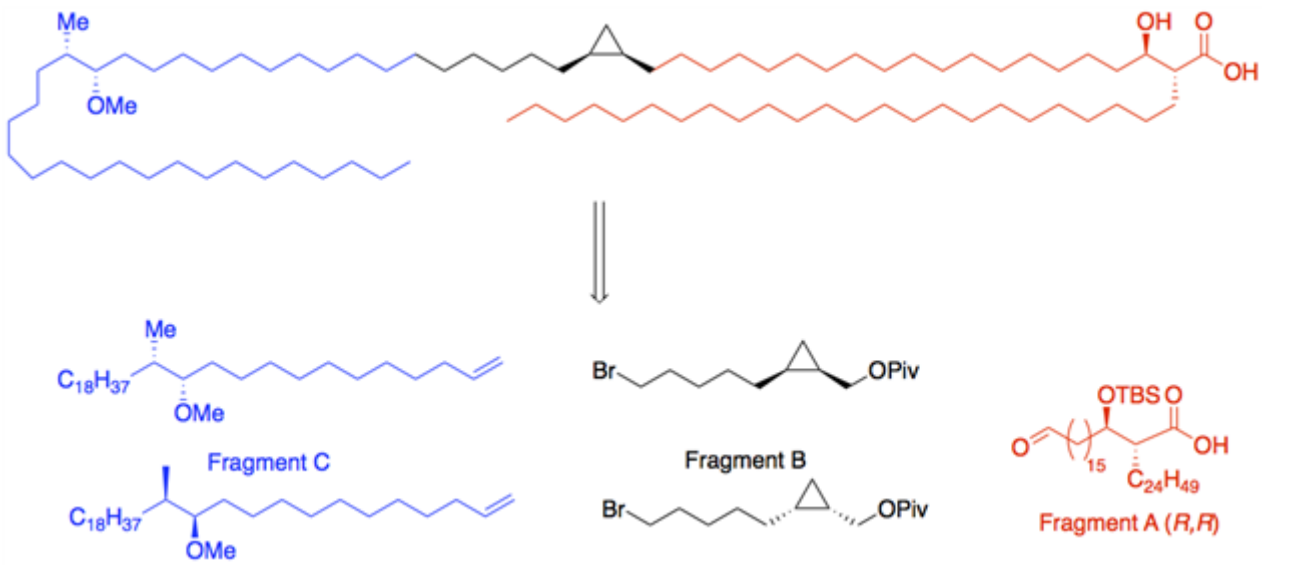
SL-025

Asymmetric total synthesis of mycolic acids from *Mycobacterium tuberculosis* for structure elucidation

Nabil Tahiri, Peter Fodran, Dhinesh Jayaraman, Martin Witte, Adriaan Minnaard

University of Groningen, Groningen, Netherlands

Mycolic acids (MAs) occur as variable mixtures of different classes and chain lengths in the cell envelope of *Mycobacterium tuberculosis*. The presence of MAs has been linked with the resistance of these organisms to current antibiotics, as they play a crucial role in the impermeability of the cell envelope.
In *Mycobacterium tuberculosis* MAs occur as a complex mixture of alpha, keto and methoxy MAs. Although the stereochemistry of the *β*-hydroxy acid motif has been elucidated and demonstrated to be the same for all MAs [1], the stereochemistry in the cyclopropyl and methoxy methyl moieties have not been unambiguously elucidated. Therefore, we synthesized all four possible diastereomers (Figure 1), by varying the stereochemistry in the cyclopropyl (Fragment B) and methoxy methyl (Fragment C) moieties while maintaining a *R,R* stereochemistry for the *β*-hydroxy acid motif (Fragment A). The biological activity of these synthetic MAs will be compared with a natural sample in order to study the effect of the varying stereochemistry on biological activity.



SL-026

Plant polyphenols - drug interactions are overlooked – modeling studies may help

Zohar Kerem[1], Loai Basheer[2], Yelena Guttman[1]

[1]The Hebrew University of Jerusalem, Rehovot, Israel, [2]Tel Hay Academic College, Kiriat Shmona, Israel

Food – drug interactions, alternative medicine and polypharmacy are all topics of increasing concerns. Indeed, the use of alternative or complimentary medicine, defined as a range of products and treatments that are not a part of conventional medicine, is growing rapidly. Hence, supplement-drug and herb-drug interactions should be visited, to inform physicians, patients, and last but not least, regulators of potential adverse interactions. The lack of knowledge allows vast number of herbals and supplements to be promoted by salesmen, regardless of their multitude of effects on human health. To investigate the relevance and hazardous potential of this issue we have selected a small number of widely consumed herbals, and used *in vitro* assays to determine their potency to inhibit Cytochrome P450 3A4 (CYP3A4), the major site of drug metabolism. We then selected the more potent inhibitors and performed docking calculations of their known bioactives, to explain the results. Extracts of black cohosh, green tea, curcumin and pomegranate were demonstrated to be potent inhibitors, using doses which are derived from actual consumption. Considering the potent inhibition of CYP3A4 by herbals reported here, patients might be exposed to overdosing. Our work demonstrates that modeling, such as docking calculations, may be instrumental to formulate the features that determine a potent inhibitor. The results and methods presented here may also explain, in part, the response to medical regimes, and should be of interest to natural products chemists, health professionals and to the general public.

SL-027

Asymmetric synthesis of 2,6-disubstituted piperidine alkaloids from chiral aziridines

Hyun-Joon Ha, Lingamurthy Macha

Department of Chemistry, Hankuk University of Foreign Studies, Yongin, 17035 Korea

One of the most abundant alkaloids consisted of piperidine rings were found in nature with various biological activities attracting organic and medicinal chemist to synthesize and to modify their backbones and functional groups for further studies. Recent success for the synthesis of isosolenopsins, deoxocassine, and spectaline from chiral aziridines prompted us to expand our synthetic strategy to elaborate microcosamine A, microgrewiapine A and 6-epi-microgrewiapine A isolated recently from plant source *M. paniculata*.
Their asymmetric synthesis proceeded from chiral aziridine-2-carboxylate by functional group transformation of carboxylate and aziridine ring opening followed by cyclization to construct piperdine rings and to build the side-chain via Jullia-Kociensky olefination.
This presentation will provide the stereochemical outcomes of 2- and 6-disubstituents with implication of their biosynthetic pathways.

Acknowledgements: This work supported by the HUFS-2018 and National Research Foundation of Korea (NRF-2012M3A7B4049645 and 2014R1A5A1011165).

Keywords: asymmetric synthesis, alkaloids, 2,6-disubstituted piperidine, chiral aziridines.

References:

[1] Lee WK, Ha HJ. Aldrichimica Acta 2003; 36: 57–63.
[2] Ha HJ, Jung JH, Lee WK. Asian J Org Chem 2014; 3: 1020–1035.
[3] Yadav NN, Choi J, Ha HJ. Org Biomol Chem 2016; 14: 6426–6434.

SL-028

New xanthone derivatives and their nitric oxide inhibitory effects

Siau Hui Mah[1], Zi Han Loh[1], Soek Sin Teh[2]

[1]School of Biosciences, Taylor's University, Subang Jaya, Malaysia, [2]Energy and Environment Unit, Engineering and Processing Division, Malaysian Palm Oil Board, Kajang, Malaysia

Xanthones, secondary metabolites of natural resources, possess various pharmacological activities and attracted great interest of researchers. The pharmacological activities are relied primarily on the types of substituent group and their relative position attached to the xanthone analogue. Due to the limitation on types of xanthones from natural resources, as well as the increasing demand for alternative drugs in the treatment of inflammation, it led to our interest in synthesizing new xanthone derivatives and evaluating their anti-inflammatory activities. Xanthone derivatives with various types of alkyl substituents were synthesized from 3-hydroxyxanthone by mixing it with alkyl bromide and potassium carbonate in acetone under reflux for hours. The products were then purified and characterized with NMR, MS, and FTIR for structural elucidation. Eleven *O*-alkylated xanthones were successfully synthesized and further evaluated for their anti-inflammatory activities by measuring the NO inhibition effect produced from LPS-induced RAW 264.7 cells. All of the compounds showed significant NO inhibition activities with IC_{50} values in the range of 10-95 μM. The inhibition effect of these derivatives is stronger than the standard drug, diclofenac sodium (IC_{50} value = 186.99 μM). Structure-activity relationship (SAR) study revealed that the xanthones with branched alkyl substituents showed greater inhibitory activities than the linear alkyl substituted xanthones. As a result, 3-(cyclobutylmethoxy)-9*H*-xanthen-9-one, bearing a branched alkyl group at C-3 position is the most potent NO inhibitor with an IC_{50} value of 10.06 μM. As a summary, a series of alkylated xanthone derivatives that exhibited promising NO inhibition effects were synthesized successfully, with bulky alkyl group showing the greatest effect. Future studies on the detail mechanism of action for anti-inflammation is highly recommended to be conducted on these potent xanthones.

Acknowledgements: Fundamental Research Grant Scheme (FRGS) from the Ministry of Higher Education (MOHE) for financial support is acknowledged.

Keywords: alkylation, anti-inflammation, 3-hydroxyxanthone, synthesis

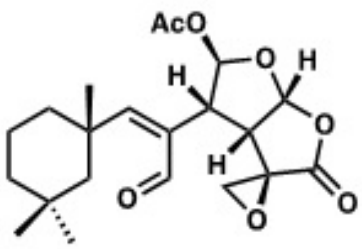
SL-029

Chemical and spectroscopic studies on bioactive terpenes from marine molluscs

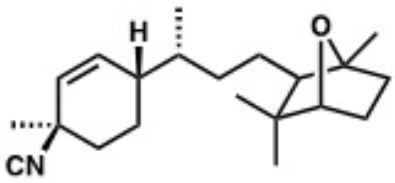
Mary Garson

School of Chemistry and Molecular Biosciences, The University of Queensland, Brisbane QLD 4072, Australia

Marine molluscs use terpenes derived from their sponge diet to aid in their protection from predators. In this talk, I will describe some of the recent identifications of cytotoxic and antimalarial terpenes isolated from colourful nudibranchs by our research group, and highlight the role of advanced NMR spectroscopy and molecular modelling in the structure determination. Notably, the use of quantum chemical calculations for the prediction of proton and carbon chemical shifts provides a valuable tool that aids complex structure elucidation. Selected examples include: spiroepoxide lactones and splendidalactone from *Goniobranchus splendidus*, and isonitriles from *Phyllidiella pustulosa* and *Phyllidia ocellata*. Biosynthetic pathways that may explain the formation of the rearranged terpene metabolites are presented.



epoxygoniolide-1



(+)-pustulosaisonitrile-1

Keywords: NMR spectroscopy, DFT, terpene, stereochemistry, nudibranch

References:

[1] White AM, Pierens GK, Forster LC, Winters AE, Cheney KL, Garson MJ. J Nat Prod 2015; 79: 477–483.

[2] Forster LC, Pierens GK, White AM, Cheney KL, Dewapriya P, Capon RJ, Garson MJ. ACS Omega 2017; 2: 2672–2677.

[3] White AM, Dao K, Vrublauskas D, Könst Z, Pierens GK, Mándi A, Andrews KT, Skinner-Adams, Clarke, MS, Narbutas PT, Sim DC-M, Cheney KE, Kurtán T, Garson MJ and Vanderwal CDJ Org Chem 2017; 82: 13313–13323.

SL-030

Structure and synthetic study of lipopeptides, isolated from marine cyanobacteria

Kiyotake Suenaga

Keio University, Yokohama, Japan

Marine cyanobacteria are known for producing a number of bioactive compounds. We isolated novel bioactive lipopeptides, kanamienamide (1) and minnamides (2-5).
Kanamienamide (1), a novel enamide-containing lipopeptide, was isolated from the marine cyanobacterium *Moorea bouiilonii*. The absolute stereostructure was determined based on the analyses of coupling constants and NOESY data and chiral HPLC analyses of an amino acid component.
The total synthesis of 1 was achieved. The synthesis features the construction of an enamide adjacent to an enol ether by Buchwald amidation and an 11-membered ring by Mitsunobu lactonization. In addition, on the basis of the biological assay of synthetic 1, we clarified that its real biological activity as a necrosis-like cell death inducer with an IC_{50} value of 32 μM. Yun He *et al*. achieved the first total synthesis of 1, and thier paper was published during review of our paper.
Minnamides (2-5), four novel linear lipopeptides, were isolated from the marine cyanobacterium *Okeania hirsuta*. The absolute stereostructure of amino acid moieties in minnamide A (2) was determined based on chiral HPLC analyses or Marfey's method. Minnamide A (2) inhibited the growth of HeLa cells with an IC_{50} value of 0.17 μM. However, the analogues of minnamide A that have a shorter fatty acid and/or lesser amino acids did not show growth inhibitory of HeLa cells.
Stereochemistry of the peptide moiety 2 was determined based on chiral HPLC analysis of acid hydrolysate. Partial acid hydrolysis of 1 gave compound 6 and the stereochemistry of the secondary hydroxy groups was determined by modified Mosher method. Stereochemistry of the methyl groups at C5 and C13 was determined by analysis of coupling constants and NOE experiments for derived cyclic ethers 7 and 8. To determine the stereochemistry of the remaining C9 methyl, we are now synthesizing two possible diastereomers 6.

SL-031

Sulfated aromatic metabolites from marine sources

Anja Hartmann, Hermann Stuppner

Institute of Pharmacy, Pharmacognosy, CMBI, University of Innsbruck, Innrain 80-82, 6020 Innsbruck, Innsbruck, Austria

Marine species produce a wide variety of sulfated metabolites of diverse biosynthetic origin gaining growing importance as modulators of a number of physiological and pathological processes. *Dasycladus vermicularis* (Scopoli) Krasser an evolutionary ancient alga is a member of the so-called siphonous green macro algae. They have unique features as they are able to form large differentiated thalli comprising of a single cell without cross walls. These thalli can grow up to 10 cm long, gaining stability by surrounding themselves with a calcareous coating which supports the long unicellular algae and enable them to grow upright. After cell damage, for example due to herbivor attack, a cascade of protective mechanisms have evolved including the extrusion of sulfated metabolites which are involved in the formation of a rapid wound plug [1,2]. We analyzed the composition of sulfated metabolites in *Dasycladus vermicularis*, which resulted in the isolation of four coumarins including two novel structures and two sulfated phenolic acids. In addition, an analytical HPLC assay for the quantification of those compounds was developed and performed on a Gemini C_{18} column. Several samples from different collection sites in the Mediterreanean Sea were analyzed and revealed differences in the coumarin content ranging between 0.26 % to 1.61 % per g dry weight. Thus, it can be suggested that seasonal changes including fluctuations in the visible and ultraviolet part of the solar radiation might have a strong influence. The wound-healing activities of the major compounds from *D. vermicularis* were investigated in a previously validated collagenase assay and compared to other marine metabolites [3].

Keywords: Siphonous green algae, sulfated coumarins, isolation and quantification, wound-healing

References:

[1] Welling M *et al.* J Adhes 2009; 85: 825–838.
[2] Correia-da-Silva M *et al.* Med Res Rev 2014; 34: 223–79.
[3] Hartmann A *et al.* Planta Med 2015; 81: 813–20.

SL-032

Structures and anti-obesity activities of the yoshinone A and the related marine *γ*-pyrone compounds

Tomoyuki Koyama[1], Yoshinori Kawazoe[2], Arihiro Iwasaki[3], Osamu Ohno[3], Kiyotake Suenaga[3], Daisuke Uemura[2]

[1]Tokyo University of Marine Science and Technology, 4-5-7 Konan, Minato-ku, Japan, [2]Kanagawa University, Yokohama, Japan, [3]Keio University, Yokohama, Japan

Yoshinone A, as the major compound isolated from marine cyanobacteria *Leptolyngbya* sp, showed inhibitory activity against the adipogenic differentiation of 3T3-L1 cells with a half maximal inhibitory concentration value of 420 nM without cytotoxicity. On the other hand, the yoshinone B1 and B2 showed only limited activity against the cells, with higher concentrations compared with yoshinone A. Further studies of the structure–activity relationship lead us to conclude that the position of a pyrone ring and an olefin in the side chain will be important for the activity (Figure 1). The anti-obesity activities of the 7-en *γ*-pyrones, such as yoshinone A and kalkipyrone, using *in vitro* and *in vivo* experiments have been evaluated.
Anti-obesity activities of the 7-en *γ*-pyrone have been shown in experiments in both cultivated cells and in mice. In the experiments using mature 3T3-L1 adipocytes, the reducing effects of yoshinone A on accumulated triglyceride (TG) amounts accompanied with the enhancement of lactate (LA) production in the culture fluid were observed. In the high-fat diet (HFD) feeding mice, the weight of adipose tissue was significantly suppressed with the kalkipyrone (KAL) treatment: 0.93±0.23 g in the HFD+KAL group vs 1.62±0.15 g in the HFD group for 5 weeks. The suppressive effects of orally ingested KAL on adipose tissue weight gain was accompanied with an enhancement of plasma LA level *in vivo*. These suppressive effects on TG absorption will result in the intact TG undergoing a transition to feces, accompanied increasing fecal TG levels. However, fecal TG in the HFD+KAL group (43.6±6.0 mg/g) was not increased, as compared with that in the HFD group (41.3±7.4 mg/g). Thus, it was suggested that the suppression of adipose tissue gain by 7-en *γ*-pyrone treatment is caused by consumption and/or excretion of absorbed TG in the body.

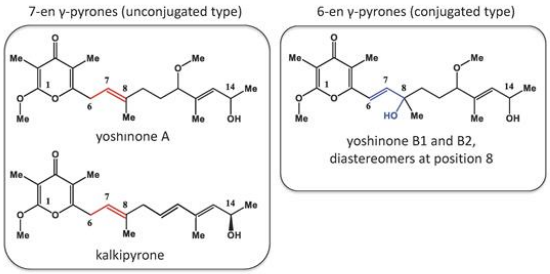


Figure 1 Chemical structures of the two types of marine γ-pyrones.

SL-033

Symbiotic microorganisms, an original resource and an inspirational model for the search of bioactive compounds

Seindé Touré[1], Morgane Barthélemy[1], Orianne Brel[1], Jonathan Sorres[1], Didier Stien[2], Véronique Eparvier[1]

[1]CNRS, Institut de Chimie des Substances Naturelles (ICSN) Université Paris-Sud, 1 avenue de la terrasse, Gif Sur Yvette, France, [2]Sorbonne Universités, UPMC Univ. Paris 06, CNRS, Laboratoire de Biodiversité et Biotechnologies Microbiennes (LBBM), Observatoire Océanologique, Banyuls-sur-Mer, France

In nature insects live in symbiosis with bacteria and fungi in order to protect themselves from their pathogens. [1] These microorganisms play a key role in the ecological success of their host. [2] The ecological benefits they provide to their macroscopic host, namely, protection against pathogens of insects provide information on the valorisation possibilities of their metabolites. Insect symbiotic microorganisms should - in order to ensure their spread - produce active compounds against microbes. [3] Our objective was to explore the chemodiversity and simultaneously investigate antimicrobial compounds within a unique collection of diverse symbiotic strains: insect mutualistic fungi and entomopathogens.

The mutualistic fungus *Neonectria discophora* has been isolated from the nest of the termite *Nasutitermes corniger*. From the EtOH extract of fungal culture, bioassay-guided fractionation led to the isolation of ilicicolinic acids and ilicicolinal derivatives (fig 1). A common biosynthetic pathway is proposed for all isolated metabolites. [4]

Another example concerns an entomopathogen: the fungus *Isaria farinosa*. In order to explore the chemical diversity of this strain, a One Strain Many Compounds (OSMAC) [5] approach was undertaken by cultivation of fungus on different culture media (Fig 2). Six compounds including four new paecilosetin derivatives were isolated from a solid culture. Absolute configurations were obtained using CD and conversion of compounds into its Mosher esters. Two compounds exhibited antibacterial activity. [6]

Acknowledgements: This work has benefited from an "Investissement d'Avenir" grant managed by French National Research Agency [CEBA, ref. ANR-10-LABX-0025].

Keywords: microorganisms, natural substances, symbiosis, insects, antimicrobial, entomopathogens

References

[1] Adnani N *et al*. Nat Prod Rep 2017; 34: 784–814.
[2] Van Arnam EB *et al*. Chem Soc Rev 2018; 47(5): 1638–1651.
[3] Beemelmanns C *et al*. J Org Chem 2016; 12: 314–327.
[4] Sorres J *et al*. Phytochemistry 2018; 151: 169–177.
[5] Bertrand S *et al*. J Nat Prod 2013; 76: 1157–1165.
[6] Touré S *et al*. Org Lett 2018, DOI:10.1021/acs.orglett.8b01367

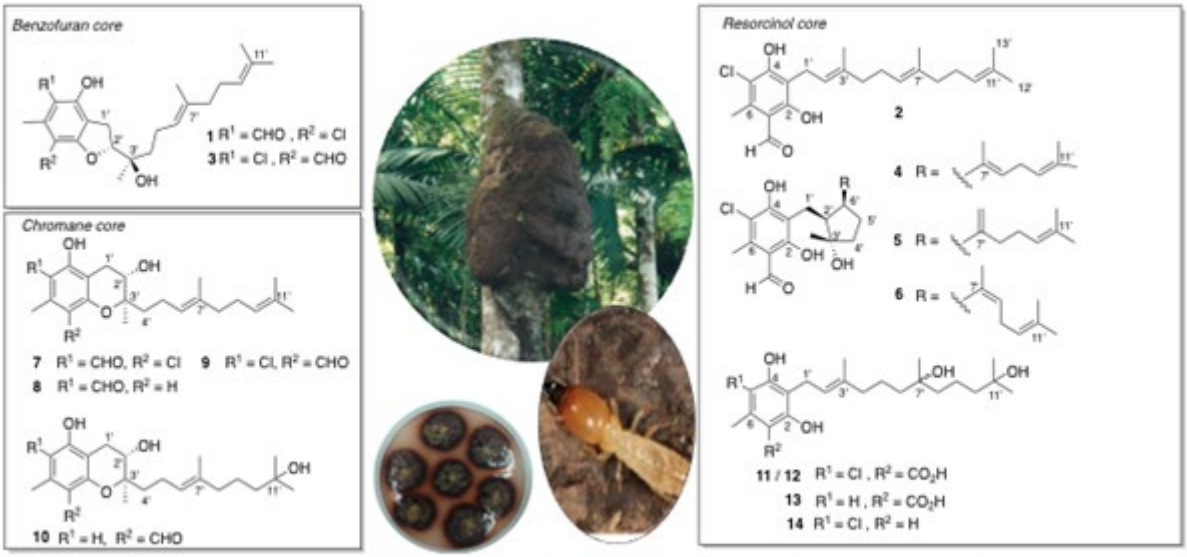


Figure 1 : isolated compounds from *N. discophora* extract

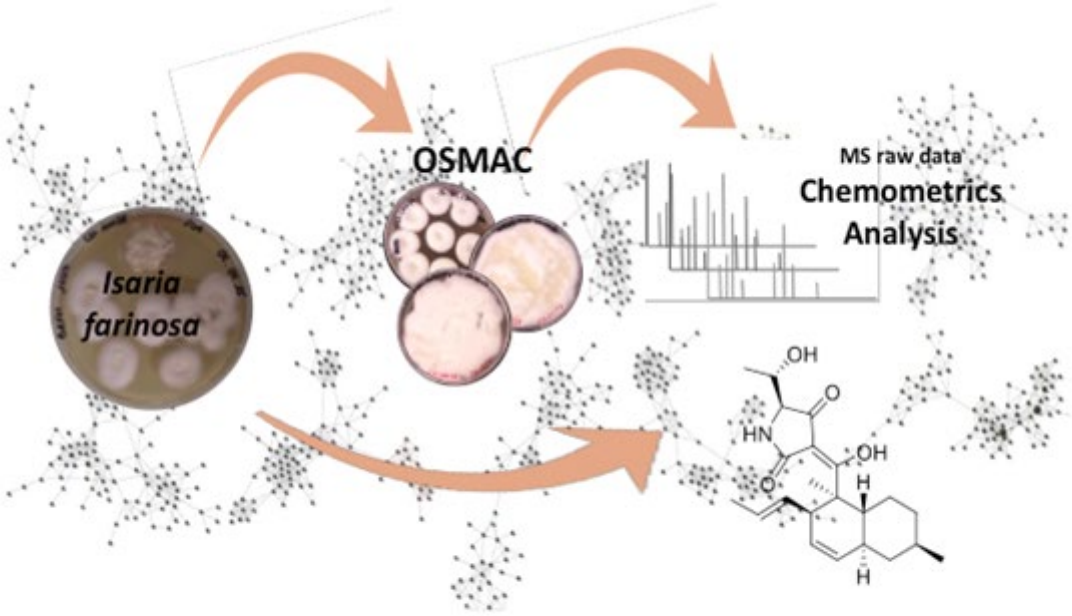


Figure 2 : paecilosetin dervivatives isolated in *Isaria farinosa* extract

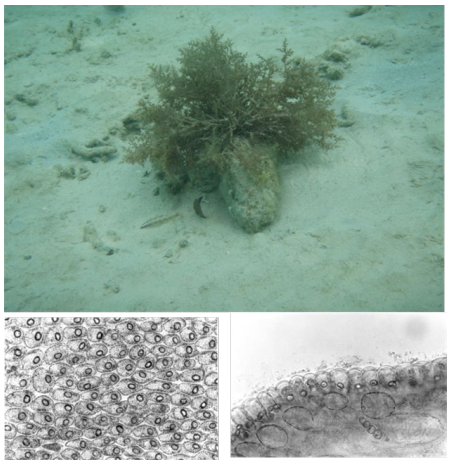
SL-034

Chemistry of Bornean *Laurencia* – Structural diversity, anti-inflammation and anticancer activities

Charles Santhanaraju Vairappan

Universiti Malaysia Sabah, Kota Kinabalu, Malaysia

Borneo is a well-known hotspot for tropical marine and terrestrial biodiversity. Red algae genus *Laurencia* is a predominant seaweed in the Bornean Sulu Sulawesi Coral Triangle region [1]. It is represented by 15 species, and we have isolated a total of 125 halogenated compounds from *Laurencia snackeyi* (Weber-van Bosse) Masuda, *Laurencia majuscula* Harvey (Lucas), *Laurencia nangii* Masuda, and *Laurencia similis* Nam Saito. There are additional 6 undescribed species that produces halogenated compounds. Their structural diversity is unique and is represented by sesquiterpenes, diterpenes, triterpenes, C_{15} acetogenin and C_{15} bromoallenes [2]. A total of 13 skeleton types was identified, and statistical analysis revealed that *L. majuscula* and *L. nangii* has the highest structural diversity and it varies with population and collection sites. These metabolites were subjected to anti-inflammation and anti-cancer assay and we investigated the mechanism of action in inflammatory cancer to better understand the complex crosstalk network between oncogenic and pro-inflammatory genes. Inflammatory potential and mechanism of action was evaluated using RAW 264.7 macrophages, in addition PGE2, TNF-, IL-1 and IL-6, iNOS, and COX2 response, were evaluated. Cancer cell bioassay was concluded using HL60 and MCF7 cell lines. Apoptosis mechanism was evaluated using Sub-G1 proportion, microscopic technique, Bax, Bcl-xl, Cleaved Capcase 3 and -actin. In addition, microarray gene expression of mRNA for cells treated with these compounds were concluded.



Acknowledgements: The author would like to acknowledge financial support from Sabah Biodiversity Center (2014~2017) and Japanese Society for the Promotion of Science (2006~2007).

Keywords: Biodiversity, *Laurencia*, halogenated compounds, anti-inflammation, anti-cancer

References:

[1] Kamada T, Vairappan CS. Nat Prod Communications2015;10: 843–844.
[2] Kamada T, Vairappan CS. Nat Prod Research 2017;31: 333–340.

SL-035

Screening of wild edible mushrooms bioactivity. Piceatannol was identified as a bioactive ingredient in the order *Cantharellales*

Efstathios P Vasdekis, Athanassios Karkabounas, Marilena E Lekka

University of Ioannina, Ioannina, Greece

Although mushrooms constitute a kingdom containing more than 140,000 species [1], only a few of them (approximately 10%) are known. Among them, about 2,000 species are considered to be edible. Wild edible mushroom species are appreciated for consumption due to their high nutritional value. The aim of the present study was to examine *in vitro* beneficial bioactivity of mushroom extracts and to investigate the molecular identity of the bioactive ingredients. In this regard, methanol extracts of twenty-nine different wild edible mushroom species, that are traditionally consumed by residents in the National Park of North Pindos in North-Western Greece, were examined for antioxidant, antiproliferative, cytotoxic and pro-apoptotic activities towards a human lung adenocarcinoma cell-line A549 by the MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay and by flow cytometry. Certain mushroom species exhibited high antioxidant activity, which was related to their high content in total phenols and flavonoids. Methanol extracts of *Cantharellus cibarius*, *Cantharellus cinereus, Craterellus cornucopioides* and *Hydnum repandum*, which belong to the order Cantharellales, exhibited high cytotoxicity and induced apoptosis-necrosis to A549 cells. Ultrahigh Performance Liquid Chromatography (HPLC) coupled with Mass Spectrometry analysis revealed as an active ingredient piceatannol ((*E*)-4-[2-(3,5-dihydroxyphenyl)ethenyl]1,2-benzenediol-3,3′,4,5′-tetrahydroxy-trans-stilbene). Piceatannol belongs to natural stilbenes and several studies have shown that this molecule possesses significant antioxidant, anticancer and anti-inflammatory activity [2]. Piceatannol, according to our best knowledge, is identified for the first time in wild edible mushrooms. Experiments with authentic piceatannol confirmed the potent antiproliferative activity of this compound. Tested mushrooms are promising sources of bioactive compounds [3]

Keywords: mushrooms, antiproliferative activity, cytotoxic and pro-apoptotic activities, piceatannol

References:

[1] Wasser SP, Appl Microb Biotec 2002; 60: 258
[2] Piotrowska H, Kucinska M, Murias M. Mutat Res 2012; 750: 60
[3] Vasdekis EP, Karkabounas A, Giannakopoulos I, Savvas D, Lekka ME. Eur Food Res Technol 2018; 264: 861

SL-036

Bioactive constituents from extremophilic fungi – New leads to battle multi-resistant microbes and cancer cells

Raha Orfali[1], Amal Aly[2], Georgios Daletos[2], Weaam Ebrahim[2,3], Shagufta Paraveen[1], Peter Proksch[2]

[1]King Saud University, Riyadh, Saudi Arabia, [2]Heinrich-Heine-Universität, Dusseldorf, Germany, 3Mansoura University, Mansoura, Egypt

The kingdom fungi is considered as a mega-diverse group which inhabits almost every niche on earth. However, only about 7% of the estimated 1.5 million species have been described and only a small number has been explored for the production of pharmacologically active metabolites [1].

"Extrempholic fungi" are fungi which grow in environments differ than that would have traditionally been considered life-supporting. It was found that the extreme stress for such organisms might activate some silent genes and induce some unique biosynthetic pathways to produce bioactive metabolites [2].

In this study a series of extremophilic fungi (halophilic and thermophilic) were isolated from different extreme ecological regions in Egypt and Saudi Arabia and subjected to chemical and *in vitro* pharmacological investigation, in order to isolate and identify antimicrobial and anticancer metabolites.

Large scale fermentation of the isolated extremophiles under conditions mimic their extreme environments followed by ethyl acetate extractions and then subjected to different chromatographic techniques yielded plenty of novel bioactive metabolites such as Epidithiodiketopiperazin, meroterpene, isocoumarin and isochroman derivatives.

The planner structures of new compounds were unequivocally elucidated on the basis of extensive 1D- and 2D-NMR spectroscopy and HRESIMS. The isolated compounds were tested against L5178Y mouse lymphoma cells using the microculture tetrazolium (MTT) assay and tested for their antimicrobial activities against different microbes including multi-resistant using serial dilution method.

Cladosprinone and *N*-methylpretichdermamide B showed pronounced cytotoxisity against the murine lymphoma L5178Y cell line with IC_{50} value of 0.88 µM and 2 µM, respectively. Carboxyoxydehydroaustin showed a significant inhibitory activity against MRSA.

The results presented here suggest that extreme-tolerant fungi are rich source of anticancer and antimicrobial metabolites which could have implications for pharmaceutical preparations in the future.

Keywords: extremophilic, cytotoxicity, antimicrobial

References:

[1] Hawksworth D. Studies in Mycology. 2004; 50: 9–18.
[2] Tamburini E *et al*. Res Microbiol 2000; 151: 179–182.

SL-037

Whole-cell (+)-ambrein production in the yeast *Pichia pastoris*

Harald Pichler[1,4], Sandra Moser[1,4], Gernot Strohmeier[4], Erich Leitner[2], Thomas J Plocek[3], Koenraad Vanhessche[3]

[1]Institute of Molecular Biotechnology, Graz University of Technology, Graz, Austria, [2]Institute of Analytical Chemistry and Food Chemistry, Graz University of Technology, Graz, Austria, [3]ACS International S.A., CH-1228 Plan-les-Ouates, Switzerland, [4]Acib GmbH, Graz, Austria

(-)-Ambrox constitutes one of the most sought-after fragrances in perfume industry. In nature, its precursor molecule, (+)-ambrein, is found as a major component of ambergris, an intestinal excretion of sperm whales. Upon exposure to sea and sunlight, oxidative degradation of (+)-ambrein to (-)-ambrox, and related compounds that constitute the characteristic smell of ambergris, takes place. As the supply of ambergris is naturally limited and unreliable, alternative sources for (-)-ambrox had to be found. Currently, most of the (-)-ambrox is synthesized chemically using diterpenoids such as sclareol, *cis*-abienol or labdanolic acid as starting materials. Yeasts are well suited to produce sesqui- and triterpenoids, like (+)-ambrein, based on their endogenous and effective farnesyl diphosphate and squalene biosynthetic routes.
Targeting a central enzyme in the sterol biosynthesis pathway, we could strongly enhance precursor supply for triterpenoid biosynthesis in *Pichia pastoris*. Heterologous expression of a triterpene cyclase cascade in *P. pastoris* and, particularly, development of suitable analytical methods provided conclusive evidence of whole-cell (+)-ambrein production. Enzyme engineering approaches markedly enhanced (+)-ambrein levels. Finally, scale-up to 5 L bioreactors confirmed that metabolically engineered *P. pastoris* represents a valuable, whole-cell system for high-level production of (+)-ambrein.

Keywords: ambrein, squalene, triterpenoid, yeast, metabolic engineering, bioreactor

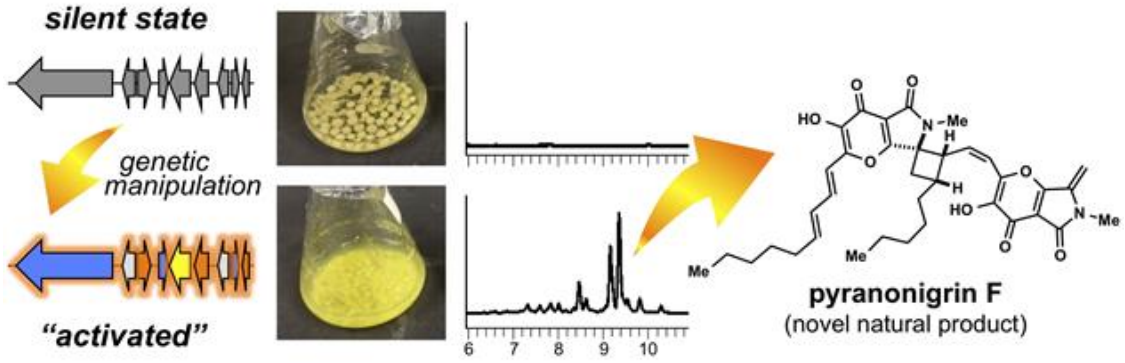
SL-038

Dissecting and harnessing fungal biosynthetic pathway

Yuta Tsunematsu, Kenji Watanabe

University of Shizuoka, Shizuoka, Japan

Starting from an isolation of penicillin from a cultured *Penicillium* species, fungi have been expected as a rich resource for finding bioactive molecules. Such molecules are biosynthesized in a fungal cell through sequential reactions by biosynthetic enzymes coded in a genomic DNA. Recent advance in DNA sequencing technology enabled us to access vast amounts of genomic data including information of biosynthetic gene cluster (BGC) responsible for biosynthesizing secondary metabolites. Results from genome sequencing studies revealed that there were many more BGCs that are potentially capable of producing natural products in the genome of a single fungus than the number of compounds that can be isolated from that fungus. This finding suggested that many of the biosynthetic genes encoded in the genome of a fungus are not activated to produce natural products under the conventional culture conditions. To circumvent these obstacles our research group set up following two objectives:
1. Overexpression of BGC for the formation of novel compounds through a genetic manipulation
2. Examining activity of specific enzymes for elucidating the detailed mechanisms
We will present our successful examples on activation of BGCs by exploiting heterologous expression system of *Aspergillus niger* for engineered biosynthesis of antitumor spirotryprostatins with an increased production. Furthermore, customizing the gene cluster enabled an acquisition of new compound, spirotryprostatin G. Besides, overproduction of a putative transcription regulator in *A. niger* allowed us to isolate novel metabolite pyranonigrin F with 10 congeners. In addition to the overexpression, deficiency of pyranonigrin BGC was capability of producing a series of biosynthetic intermediates, achieving an expansion of chemical diversity. We are currently aiming at expanding this strategy to engineer biosynthetic pathway in mushroom-forming fungi Basidiomycota, which is a rich source for obtaining toxins and pharmaceuticals. Current efforts to activate BGCs in *Coprinus cinerea* will be presented in this talk.



SL-039

GLP-1 secretagogues from the Marquesan tree *Oparanthus teikiteetinii* (Asteraceae) as potential antidiabetic drugs

Aurélie Urbain[1], Clément Huguet[1], Opeyemi J. Olatunji[2], Eleonore Real[1], Christian D. Muller[1], Phila Raharivelomanana[3]

[1]Université de Strasbourg, Strasbourg, France, [2]Prince of Songkla University, Hat Yai, Thailand, [3]Université de la Polynésie française, Faa'a, Tahiti, French Polynesia

Glucagon-like peptide-1 (GLP-1) is an intestinal hormone involved in blood glycemia regulation. As its secretion is impaired in patients with type 2 diabetes, compounds improving GLP-1 levels could represent a novel therapeutic strategy [1]. In this perspective, we found that an extract from the leaves of *Oparanthus teikiteetinii* (Asteraceae) stimulated significantly GLP-1 secretion in STC-1 intestinal cells. This tree endemic from Marquesas Islands, French Polynesia, has never been explored neither from phytochemical nor pharmacological point of view until now. The phytochemical investigation from the leaves extract led to the isolation of 17 compounds: 12 benzofuran derivatives, 4 of them reported for the first time, 4 sesquiterpenoids including a new one, and a dimeric benzopyran. Pure compounds were then tested for their intrinsic capacity to stimulate GLP-1 release. The bioactivity of the crude extract was found to be mainly due to bitalin A [2] and to jasopyran [3], which increased GLP-1 secretion by 325.9% ± 3.0% and 229.9% ± 4.8%, respectively, compared to control cells. Furthermore, none of the isolated compounds affected STC-1 cells viability as demonstrated by image cytometry. Further studies must be undertaken to prove both efficacy and safety of these small molecules *in vivo* and therefore their potential as alternative antidiabetic compounds.

Acknowledgements: We thank the Foundation for the Development of the Chemistry of Natural Products and their Applications (CNRS/Academy of Sciences in Paris) for financial support.

Keywords: *Oparanthus teikiteetinii*, benzofurane derivatives, Glucagon-like peptide-1, diabetes mellitus

References:

[1] Tsoukalas *et al*. Planta Med 2016; 82: 992–999.
[2] García de Quesada *et al*. Phytochemistry 1972; 11: 446–449.
[3] Ahmed *et al*. J. Saudi Chem Soc 2004; 8: 105–114.

SL-040

Derivatives of Amaryllidaceae alkaloids haemanthamine and ambelline as potential drugs in the treatment of Alzheimer´s disease

Lucie Cahlíková[1], Aneta Ritomská[1], Jana Maříková[2], Lubomír Opletal[1], Jan Korábečný[3]

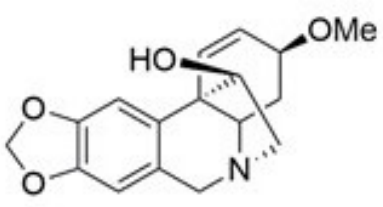
[1]ADINACO Research Group, Department of Pharmaceutical Botany, Faculty of Pharmacy, Charles University, Hradec Kralove, Czech Republic, [2]Department of Organic and Bioorganic Chemistry, Faculty of Pharmacy, Charles University, Hradec Kralove, Czech Republic, [3]Biomedical Research Centre, University Hospital, Hradec Kralove, Czech Republic

Modern research has shown that Amaryllidaceae alkaloids represent a rich reservoir of potential small molecules exhibiting several medicinal properties through various mechanisms. Among the many Amaryllidaceae alkaloids, galanthamine has been given a great amount of attention due the fact that it possesses potent acetylcholinesterase inhibition activity, and is distributed worlwide for the treatment of Alzheimer´s disease. Some of Amaryllidaceae alkaloids have shown remarkable cytotoxic and antiproliferative activity against diverse types of cancer cells.

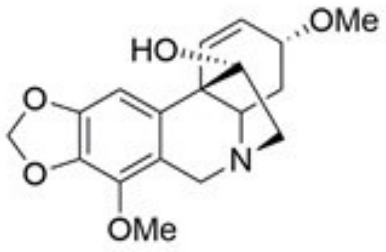
One of the most interesting compounds is haemanthamine (HA), *β*-crinine-type of Amaryllidaceae alkaloids, which displays significant *in vitro* cytotoxic activity against several different types of cancer cell lines (e.g. MOLT-4, HepG2, HeLa, MCF-7, etc.)[1]. Similar compound to haemanthamine is ambelline (AMB), which differ from HA only in orientation of 5,10b-ethano bridge and presence of methoxy group in position C7.

HA has been isolated from the bulbs of Narcissus cv. Dutch Master, and AMB from Nerine bowdenii, as a start material for the preparation of their derivatives in our laboratory.

Twenty new derivatives of both alkaloids were developed. All derivatives were screened for their inhibitory potential against cholinesterases. The active compounds were further studied for their potency to inhibit GSK-3β, and ability to permeate through the blood-brain barrier. Some compounds, namely 11-*O*-(2-methylbenzoyl)-haemanthamine, and 11-*O*-(4-nitrobenzoyl)-haemanthamine, 11-*O*-benzoylambelline, and compounds marked as LC-70, LC-73 revealed the most intriguing profile. *In vitro* data were further corroborated by detailed inspection of their plausible binding modes in the active sites of AChE and BuChE, which led us provide the structural determinants responsible for the activity towards these enzymes.



Haemanthamine



Ambelline

SL-041

Quality assessment of *Morus alba* root bark using HPTLC bioautography-UPLC-Orbitrap-MS^2 fingerprint combined with chemometrics

Petar Ristivojevic[1,2], Ammar Tahir[1], Judith Rollinger[1]

[1]Department of Pharmacognosy, Faculty of Life Sciences, University of Vienna, Althanstraße 14, 1090 Vienna, Austria, Vienna, Austria, [2]Innovation Centre of the Faculty of Chemistry Ltd, Studentski trg 12-16, 11000 Belgrade, Serbia, Belgrade, Serbia

The root bark of *Morus alb*a L. (sāng bái pí, SBP) has been widely used in Traditional Chinese medicine against different diseases such as lung heat, cough, edema, and oliguria. To date, many studies have reported about the bioactive compounds from SBP exhibiting anti-inflammatory, antioxidant and antimicrobial activities [1]. The aim of this study was to develop a new and fast, high throughput method for quality assessment of SBP samples from different origin using HPTLC/bioautography/UPLC-MS^2 fingerprint combined with chemometrics. The bioautography assays were performed for the identification of constituents with potentially antioxidant activity (DPPH) and antimicrobial activities (*Bacillus subtilis*, *Escherichia coli*) from complex SBP samples. Additionally, UPLC hyphenated with Orbitrap-MS^2 was applied to allow comprehensive chemical profiling of geographically different SBP samples.
The HPTLC/bioautographic analyses revealed distinct metabolite and activity profiles between 18 geographically different SBP samples. Serbian samples (n=11) showed higher antioxidant and antimicrobial activities compared to Chinese samples (n=7). Principal component analysis confirmed the separation between geographically different samples and recognized their main markers, namely sanggenons B, C, D, G, sanggenol A, kuwanon L and a compound with a hRF value of 13. Most importantly for quality assessment, the HPTLC bioautography-UPLC-Orbitrap-MS^2 approach allowed the fast identification of major and minor SBP constituents endowed with antioxidant and antimicrobial activities prior to any separation step.

Acknowledgements: Thank is owed to the OeAD foundation (ICM-2017-06233) and the Ministry of Education, Science and Technological Development of the Republic of Serbia (No. 172017) for financial support.

Keywords: Quality assessment, HPTLC, bioautography, *Morus alba* root bark, chemometrics, UPLC-Orbitrap-MS^2

References:

[1] Grienke U, Richter M, Walther E, Hoffman, A, Kirchmair J, Makarov V, Rollinger JM. Sci rep 2016; 6: 27156.

SL-042

Assessment of selected Saudi and Yemeni plants for insecticidal activities against the yellow fever mosquito *Aedes aegypti* (L.), and LC MS/MS and GC/MS analysis of bioactive extracts

Shaza Almassarani[1], Amina El-Shaibany[2], Nurhayat Tabanca[3,4], Abbas Ali[3], Alden Estep[5], James Becnel[6], Fatih Goger[7], Betul Demirci[7], Ali El-Gamal[8], Husnu K Can Baser[9]

[1]Department of Pharmacognosy, Pharmacy College, King Saud University, Riyadh, Saudi Arabia, [2]Department of Pharmacognosy, Faculty of pharmacy, University of Sana'a, Sana'a, Yemen, [3]National Center for Natural Products Research, the University of Mississippi, , University, USA, [4]USDA-ARS, Subtropical Horticulture Research Station, Miami, USA, [5]Navy Entomology Center of Excellence, CMAVE Detachment, 1700 SW 23rd Drive, Gainesville, USA, [6]USDA-ARS, Center for Medical, Agricultural, and Veterinary Entomology, Gainesville, USA, [7]Department of Pharmacognosy, Faculty of Pharmacy, Anadolu University, Anadolu, Turkey, [8]Department of Pharmacognosy, Faculty of Pharmacy, Mansoura University, Mansoura, Egypt, [9]Department of Pharmacognosy, Faculty of Pharmacy, Near East University, Nicosia, Cyprus

Plant-based organic pesticides offer an effective, degradable, environmentally friendly and cheaper alternative to conventional synthetic pesticides. Saudi Arabia and Yemen are characterized by wide distinction of their flora due to climate and height differences among different areas. As part of our continuing investigation of native medicinal plants for interesting biological activities, thirty-three plants, obtained from diverse areas in Saudi Arabia and Yemen, were screened for insecticidal activity against the mosquito vector *Aedes aegypti* (L.). Out of the 59 tested organic extracts, *Hyssopus officinalis, Nigella sativa, Ocimum tenuiflorum, Saussurea lappa* and *Taraxacum officinale* produced over 80% mortality against adult female *Ae. aegypti* at 5 □g/mosquito and only one extract *Aloe perryi* showed 100% mortality against 1st instar *Ae. aegypti* larvae at 31.25 ppm. All active fractions were processed for bioactive compounds identification using LC-MS/MS and/or headspace SPME-GC/MS analysis. Careful examination of the mass spectra and detailed interpretation of the fragmentation pattern allowed the identification of tens of interesting biologically active secondary metabolites. Some compounds such as caffeic and quinic acid and their esters, 3- and 5- caffeoyl and 4, 5-dicaffeoylquinic acids were detected in most of the analyzed fractions. Additionally, luteolin, its glycoside and luteolin glucuronide and diglucuronide were also identified as bioactive compounds in several HPLC fractions. Volatile ketone, 6-methyl-5-hepten-2-one was identified from the *A. perryi n*-hexane fraction as a major compound. With the aid of bio guided-directed isolation and purification, our main future target will be obtaining highly active, safe and naturally-derived insecticides from the endemic plants of the Arabian Peninsula.

Keywords: Medicinal plants, insecticidal, *Aedes aegypti*, LC MS/MS, GC/MS, Saudi Arabia, Yemen

References:

[1] Demirci B *et al.* Acta Trop 2013; 128: 557–560
[2] Al-Massarani S *et al*. NVEO 2016; 3: 26–34
[3] Mothana RA *et al.* Alternat Med 2010; 7: 323–30

SL-043

Cytotoxic and Wnt-inhibiting activity of polyphenolics from *Lespedeza bicolor* and *Ampelopsis japonica*

Darya Tarbeeva[1], Sergey Fedoreyev[1], Artem Blagodatski[2], Antonina Klimenko[2], Marina Veselova[1], Petr Gorovoy[1]

[1]G.B. Elyakov Pacific Institute of Bioorganic Chemistry, Far Eastern Branch of Russian academy of science, Prospect 100 let Vladivostoku 159, 6900022, Vladivostok, Russian Federation, [2]Far Eastern Federal University, FEFU Campus 10 Ajax Bay, Russky Island, 690090, Vladivostok, Russian Federation

Lespedeza bicolor belongs to Fabaceae family. This species is known to produce prenylated polyphenolic metabolites possessing antitumor, antioxidant properties and capable to inhibit neurominidase.

We have studied the cytotoxic activity of *L. bicolor* stem bark polyphenolic metabolites against five cell lines: HTB-19 (triple-negative breast cancer), Kyse-30 (esophageal cancer), HepG2 (hepatocellular carcinoma), HEK 293 (human embryonic kidney) and RPE-1 (retina pigmented epithelium). We also investigated the ability of polyphenolic compounds to inhibit intracellular Wnt pathway which can be overactivated in many cancer types.

Five prenylated polyphenolic compounds have been found in the extract of *L. bicolor* stems. Four of them have been isolated for the first time and named dihydrolespedezol A2 (1), bicoloketone (2), isoprenyllespedezol A2 (3) and dihydrolespedezol A3 (4) respectively. Their structures were determined on the basis of NMR and CD spectral data. The molecular formulae of these compounds have been confirmed using HR-HPLC-MS technique. Lespedezol A2 (5) had earlier been isolated from *L. homoloba*.

Cytotoxic activity of compounds 1-5 was determined using MTT assay. Among the tested compounds isoprenyllespedezol A2 (3) possessed the most significant activity as it was toxic against all used cell lines at concentrations more than 10 μM. Compounds 1, 2, 4 and 5 possessed moderate cytotoxic activity with IC_{50} against cell lines HTB-19, Kyse-30, HepG2, HEK 293 and RPE-1was from 30 to 80 μM.

Bicoloketone (2) was non-toxic to HTB-19 triple-negative breast cancer cells and inhibited Wnt signaling by 50% at a concentation of 40 μM.

Apart from *L. bicolor*, we the ability of extracts from *Ampelopsis japonica* to inhibit Wnt signaling. We showed that extract obtained from *A. japonica* caudex with chloform-ethanol (3:1) inhibited Wnt-signaling by 50% at a concentration of 100 тg/μl.

Acknowledgements: This study was supported by the Russian Foundation for Basic Research (grant 18-34-00502 mol_a).

SL-044

Inhibitory effect of chemical and natural anti-browning agents on polyphenol oxidase from Ginger (*Zingiber officinale* Roscoe)

Chen Wai Wong, Win Yee Lim

UCSI University, Cheras, Malaysia

Food browning is undesirable as it causes deterioration in nutritional value and affects appearance of food. It was found that this phenomenon was related to polyphenol oxidase (PPO), which catalyzes hydroxylation of monophenols to *o*-diphenols, followed by oxidation *o*-diphenols to *o*-quinones. In this study, natural and chemical anti-browning agents were used to prevent the browning of ginger PPO. Sodium metabisulfite (5 mM) is a better chemical inhibitor compared to L-cysteine and sodium chloride as 55.00% of ginger PPO inhibition was achieved. The percentage of inhibition increased as the concentration of anti-browning agents increases. Heated onion, chili pepper and pineapple extracts were found to exhibit a higher inhibition percentage on ginger PPO than unheated extracts. Heated chili pepper extract was the most effective natural inhibitor found in this study as it inhibited 47.97% of the ginger PPO mixed-competitively. Natural anti-browning agents have potential to be used to control the browning of ginger as well as other vegetables and fruits during food processing or storage.

SL-045

Isatin derivative inhibit oxidative stress damage to muscles in diabetes

Meha Fatima Aftab[1,2], Shabir Khan Afridi[2,3], Momin Khan[4], Aneela Karim[4], Khalid M. Khan[4], Rizwana S. Waraich[5]

[1]Dow Institute of Medical Technology. Karachi, Pakistan, [2]Dr. Panjwani Center for Molecular Medicine and Drug Research, Karachi, Pakistan, [3]Chinese Academy of Sciences, Beijing, China, [4]HEJ Research Institute of Chemistry, ICCBS, Karachi, Pakistan, [5]Sohail University, Karachi, Pakistan

Consumption of processed food has become an integral part in daily lives. Processing in various dry heat forms leads to the formation of advanced glycation end products (AGEs) by the proteins which are involved in stress signaling and pathophysiology of various metabolic diseases. AGEs play a crucial role in the pathophysiology of diabetes and its complications. Isatin derivatives have been of importance in this regard as they inhibit formation of glycation of proteins. They have been shown to be the carbonyl scavengers, which through scavenging the di-carbonyl compounds involved in the formation of AGEs are inhibited. It has been shown that AGEs cause formation of multimolecular complexes by involving RAGE-PKC-α-IRS-1 complex which is inhibited by isatin derivatives. IRS-1 in this multimolecular complex is phosphorylated at Ser 307, which renders it inactive. Inactivation of IRS-1 leads to inhibition of the signaling cascade involving phosphorylation of AKT and GSK-3β, which inhibits glucose uptake and glycogen formation in muscles and fat cells. Isatin derivative inhibited oxidative stress due to the multimolecular complex formation. It also reduced phosphorylation of IRS-1 at Ser 307 and restored its tyrosine phosphorylation. It also restored Ser 357 phosphorylation of AKT and promoted glucose uptake in muscle cells. Since muscle cells account for the majority of glucose uptake, isatin derivative can therefore develop as potential agent to treat diabetes and complications related to oxidative stress in diabetes.

SL-046

The active compound screening and ameliorate efficacy of the three Tibetan classical prescriptions for the cerebral ischemia injury therapy

Liwei Tan[1], Ying Wu[2], Rui Tan[1]

[1]Southwest Jiaotong University College of Medicine, Chengdu, China, [2]Southwest Jiaotong University School of Life Science and Engineering, Chengdu, China

Objective: Firstly, this study was aimed to explore the synergistic effect of the three Tibetan prescriptions, "Shanhu", "Ruyi Zhenbao", and "Chenxiang" pills, on a permanent occlusion of middle cerebral artery model (pMCAO), and which were respectively oral administration at morning, noon and night. Secondly, the active compound in these three Tibetan prescriptions were recognized. Finally, the pharmacodynamic effect of the active compound were conducted a comprehensive research.
Methods: To evaluate the preventing efficacy, the three prescription were at different time oral administration in combination or alone on the pMCAO model. After 7 days of administration, the brain infarction volume was measured by TTC stain. And then, the compounds of the three prescription were recognized from the analysis of blood components after administration. The pharmacodynamic effect of these compounds were investigated by a series of *in vitro* experiments to screen the active compounds, which included promoting the NGF or VEGF expression, reducing oxidative stress damage and so on. The active compounds were for further efficacy and mechanism study *in vivo*.
Results: Comparing to the individual groups, the combination group could significantly reduce volume of the cerebral infarction, and at the same time reduce the neuronal damage and apoptosis in the penumbra. Monomer R can reduce the oxidative damage and neurotoxicity of cells *in vitro* and promote prevent the nerve cells from injury. It can effectively reduce the cerebral ischemic injury in rats and inhibit the expression of inflammatory factors and oxidative stress *in vivo*. At the same time, it promotes repair of the blood-brain barrier that damages brain tissue. That Monomer R might be a candidate for ameliorating the cerebral ischemia injury.

SL-047

Are plants used in the Eastern Cape Province of South Africa for cosmetics fully commercialized?

Wilfred Otang Mbeng, Jonas Idowu Sagbo

University of Mpumalanga, White River, South Africa

Plants have been used for years for various cosmetic purposes. In South Africa, most especially Eastern Cape Province, a large proportion of the population are reliant (to some extent) on botanical resources for beauty and health. Despite, the use of this botanical resources for various cosmetic purposes, only a few have been fully commercialized or used as ingredients in cosmetics formulations. The present study aimed to review plant species that are fully explored commercially for cosmetic products in the Eastern Cape Province of South Africa. A survey of cosmetic products with natural (plants) ingredients was done covering the major supermarket (SPAR, Shoprite and Pick and Pay), cosmetics shops (Clicks) and pharmacies in the Eastern Cape Province and electronic database including Science Direct, Scifinder, PubMed, Springer, Scopus, Medline, Embase, Web of Science and Google Scholar were used as a data source for ethnobotanical information. Surprisingly, out of 150 plants species used by both Xhosa men and women for various cosmeceutical purposes, only six (6) plants species have been explored commercially as cosmeceuticals. These plants species belonging to five major plants families, Lamiaceae (2 species), Asphodelaceae (1 species) Cucurbitaceae (1 species), Oleaceae (1 species) and Verbenaceae (1 species). It is evident that the use of Eastern Cape plants for cosmetics purposes have not been fully explored commercially. Thus, the need for cosmeceutical industries to explore these species commercially in other to develop new possible cosmetic products for local and international market.

SL-048

Optimization of *in vitro* release of an anticonvulsant using nanocapsule-based thermogels

Akbar Esmaeili, Sonia Singh

Department of Chemical Engineering, North Tehran Branch, Islamic Azad University, Po Box 19585/936, Tehran, Iran

Controlling the release rate of anticonvulsant drugs can have a significant effect on the efficacy of these drugs and the safety with which they can be administered to patients. This study investigated *in vitro* release of the anticonvulsant ethosuximide from nanocapsule-based *N, O*-carboxymethyl chitosan and hyaluronan-methylcellulose hydrogels using two experimental designs: an one-factor-at-a-time method and an optimization method employing a Taguchi design. Using the first method, the release rate of the drug was significantly reduced compared with other delivery systems. With the second method, when the drug was blended into a hyaluronan-methylcellulose hydrogel the release rate was similarly reduced, with full release occurring after three days. Scanning electron microscopy, Fourier-transform infrared spectroscopy, and ultraviolet-visible spectrophotometry were used to study the drug encapsulation, and two mathematical models for evaluating encapsulation efficiency were developed. The results of this study show promise for use of nanoencapsulated thermoresponsive hydrogels in clinical delivery of anticonvulsants.

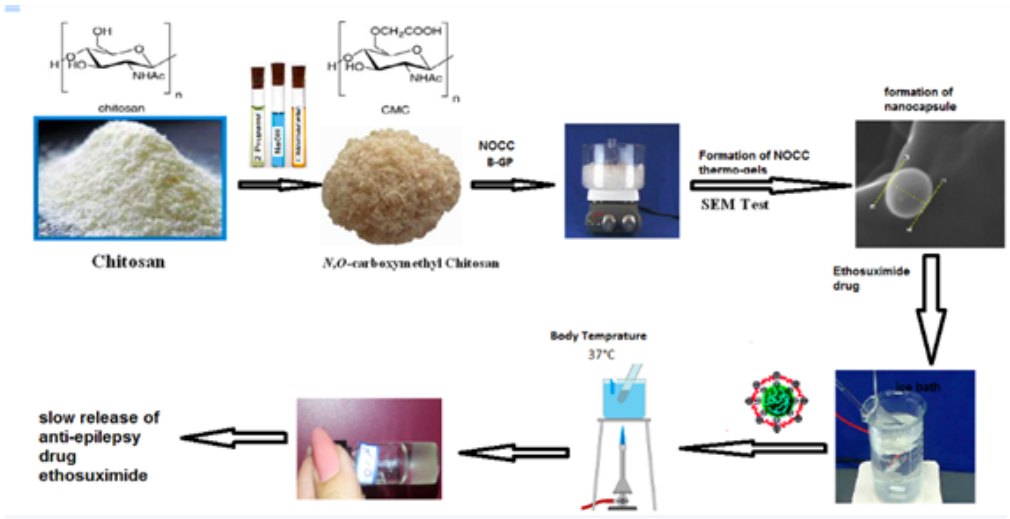


Figure 1. Schematic representation of preparation of NOCC thermogel and ethosuximide encapsulation.

SL-049

Dihydrochalcone glucosides from the subaerial parts of *Thonningia sanguinea* and their *in vitro* PTP1B inhibition

Stefan Schwaiger[1], Luca Pompermaier[1], Elke H. Heiss[2], Mostafa Alilou[1], Fabian Mayr[1,3], Mawunu Monizi[4], Thea Lautenschlaeger[5], Daniela Schuster[3,6], Hermann Stuppner[1]

[1]Institute of Pharmacy/Pharmacognosy, CMBI, University of Innsbruck, Innsbruck, Austria, [2]Department of Pharmacognosy, Faculty of Life Sciences, University of Vienna, Vienna, Austria, [3]Institute of Pharmacy/Computer-aided molecular design group, CMBI, University of Innsbruck, Innsbruck, Austria, [4]Universidade Kimpa Vita, Province of Uíge, Uíge, Angola, [5]Department of Biology, Institute of Botany, Faculty of Science, Technische Universität Dresden, Dresden, Germany, [6]Department of Pharmaceutical and Medicinal Chemistry, Institute of Pharmacy, Paracelsus Medical University Salzburg, Salzburg, Austria

Six new and four known dihydrochalcone glucoside derivatives (1-10), the phenylpropanoid coniferin (11) and the lignans (+)-pinoresinol (12) and lariciresinol (13) were isolated from the subaerial plant parts of *Thonningia sanguinea* in the course of a screening campaign for new antidiabetic lead structures. Their structures were elucidated by HRESI-MS, NMR, GC-MS and ECD data evaluation. 2'-*O*-(3-galloyl-4,6-*O*-*a*S-hexahydroxydiphenoyl-*β*-D-glucopyranosyl)-3-hydroxyphloretin (4), 2'-*O*-(4,6-*O*-*aS*-hexahydroxydiphenoyl-*β*-D-glucopyranosyl)phloretin (5), 2'-*O*-(3-*O*-galloyl-4,6-*O*-*aS*-hexahydroxydiphenoyl-*β*-D-glucopyranosyl)phloretin (6) and thonningianin B (9) showed moderate protein tyrosine phosphatase-1B (PTP1B) inhibition in an enzyme assay (IC_{50} values ranging from 19 to 25 µM), whereas thonningianin A (10) was identified as potent inhibitor (IC_{50} = 4.4 µM). Observed activity differences could be explained by molecular docking experiments. The activity of 10 could further be confirmed in HEPG2 liver carcinoma cells, where the compound was able to increase the level of phosphorylated insulin receptors in a concentration dependent manner.

SL-050

Antifungal discovery for the treatment of White Nose Syndrome (WNS) in Bats

Christine Salomon, Yudi Rusman

Center for Drug Design, University of Minnesota, Minneapolis, United States

White nose syndrome (WNS) is a disease of hibernating bats in North America caused by the fungus *Pseudogymnoascus destructans*. *P. destructans* is a psychrophilic dermatophyte that infects hibernating bats and leads to mortality as high as 95-100% for some species. There are currently no viable field methods for preventing or treating infection, and WNS is rapidly spreading through bat colonies across the United States and Canada. As part of a project to develop new treatments, we are collecting native microbes found in hibernacula to develop biological control agents for direct and sustained inhibition of *P. destructans* on hibernaculum substrates and bats. We have identified >100 bacterial and fungal isolates from subterranean habitats that inhibit the growth of *P. destructans in vitro*. The most potent strains were cultured in larger volumes, extracted and analyzed for antifungal metabolites. Some of the active compounds include norditerpene lactones from *Oidiodendron* sp., resorcylic acid lactones from *Ilyonectria radicicola* and cyclic depsipeptides from *Streptomyces albidoflavus*. To determine the potential cytotoxicity of the compounds (and producing microbes) to bats, primary fibroblast cell cultures were established from a WNS susceptible species (Northern Long Eared Bat, *Myotis septentrionalis*) and a resistant species (Gray Bat, *Myotis grisescens*). All isolated compounds were tested against both fibroblast cell lines using standard MTT cell viability assays and found to vary in their activity. Additional experiments were conducted to measure the ability of biocontrol candidate strains to grow on natural cave substrates. This information together with the antifungal potency and fibroblast cytotoxicity data were used to prioritize the most promising biocontrol strains for field studies. We will also discuss our current efforts to characterize the microbial diversity of subterranean environments in the context of this screening work and future application of biological control agents.

SL-051

The quantification of health claim-relevant tyrosol and hydroxytyrosol after direct hydrolysis improves customer understanding and mitigates market distortion

Thomas Jakschitz

Austrian Drug Screening Institute GmbH - ADSI, Innsbruck, Austria

The EFSA-approved claim related to olive oil being beneficial to cardiovascular health suffers from ambiguities that allow for a vague and possibly subjective interpretation of underlying analytical data. Not only does this lead to misconceptions among customers but might also lead to market distortions. Herein, a rapid and facile analytical technique is presented, circumventing the ambiguities by capturing the content of presumably health-benefiting compounds as the sum of tyrosol and hydroxytyrosol, cleaving such moieties of more complex constituents such as oleuropein and oleocanthal. Since direct hydrolysis of the olive oil is the method's central element, the reaction temperature, time, reagent concentration and reagent type have been optimized. Furthermore, the influence of co-solvents is investigated, possibly aiding the intermittent miscibility of the two phases during hydrolysis. The analytical and economic implications are discussed especially in connection with a conventionally employed technique.

SL-052

Anti-angiogenic activity of some iridoids isolated from medicinal plants

M P Germanò[1], V D'Angelo[1], N De Tommasi[2], M D'Ambola[2], KA Beladjila[3], AM Iannuzzi[4], A Braca[4], A Bader[5]

[1]Department of Chemical, Biological, Pharmaceutical and Environmental Sciences, University of Messina, Messina, Italy, [2]Department of Pharmacy, University of Salerno, Fisciano, Fisciano, Italy, [3]Laboratoire d'Obtention des Substances Thérapeutiques (LOST), Département de Chimie, Université des Frères Mentouri-Constantine, Constantine, Algeria, [4]Department of Pharmacy, University of Pisa, Pisa, Italy, [5]Faculty of Pharmacy, Umm Al-Qura University, Makkah, Saudi Arabia

Naturally occurring iridoids are secondary metabolites found in a large number of plant families usually as glycosides. Chemotaxonomically they are useful as markers of several genus. In general, iridoids have been regarded as defense chemicals against herbivores and pathogens, due to their antifeedant and growth inhibitory activities against insects [1]. Conversely, many iridoids are associated with a wide range of pharmacological activities such as cardio and neuroprotective, anti-inflammatory, and anticancer [2]. Nowadays angiogenesis inhibition might be a promising approach for anti-inflammatory and anticancer therapies [3]. In this context, the aim of the present study was the isolation and characterization of iridoid derivatives from species belonging to Lamiaceae, Rubiaceae, and Scrophulariaceae families, along with the assessment of their effects on angiogenesis. Two *in vivo* models were used to evaluate the new blood vessel formation: *Danio rerio* (zebrafish) embryo and the chick chorioallantoic membrane [4]. Results showed that among the iridoids tested, asperuloside, geniposidic acid, iridoid V1, and 6-α-L-(2"-caffeoyl)-rhamnopyranosyl-catalpol were able to reduce microvessel formation in both assays showing different potency (0.5-2.0 μg) as compared to retinoic acid and 2-methoxyestradiol, used as reference substances. In conclusion, our results may suggest that iridoids represent interesting anti-angiogenic molecules and a new scaffold to develop anti-angiogenic agents.

Keywords: Anti-angiogenic activity, iridoids, chick chorioallantoic membrane, zebrafish

References:

[1] Dinda B, Debnath S, Harigaya Y. Chem Pharm Bull 2007; 55: 159–222
[2] Dinda B, Debnath S, Banik R. Chem Pharm Bull 2011; 59: 803–833
[3] Folkman J Nat Med 1995; 1: 27–31
[4] Certo G, Costa R, D'Angelo V, Russo M, Albergamo A, Dugo G, Germanò MP. Nat Prod Res 2017; 31: 2850–2856

SL-053

Bioactivity of medicinal plants: is it the plant, endophyte or both?

Hajara Alfa, Randolph Arroo, Susannah Walsh, Ketan Ruparelia, Avninder Bhambra, Adrian Slater

De Montfort University, Leicester, Leicester, United Kingdom

Boswellia dalzielii (*Burseraceae*) has ethnopharmacological importance and is claimed to have anti-infection and immunomodulatory effects [1]. In the Northern part of Nigeria, a region with a tropical dry climate, an aqueous infusion of this plant is used in the treatment of infections and tumours. Thus, the effect of temperature on the said infusion was studied. Microorganisms can remain dormant for a very long duration [2], therefore, the impact of endophytic microbes present in aqueous infusion of *B. dalzielii* was also studied.
Activity-guided fractionation against *Staphylococcus aureus* and its methicillin resistant strain was used to identify two antibacterial compounds namely gallic acid and pyrogallol. The MIC for pyrogallol and gallic acid against *S. aureus* growth are 508 and 753 μM, while against MRSA growth are 254 and 2032 μM, respectively. A growth inhibition assay showed the activity of gallic acid as bacteriostatic, and pyrogallol as bacteriocidal against tested microorganisms. Interestingly, the bacteriocidal compound was found to arise by conversion of gallic acid by the endophyte *Enterobacter cloacae.* Similarly, MTT cytotoxicity assay against breast cancer cell line (MCF7) led to identification of catechol with an EC_{50} of 86μM and endophytic *Klebsiella pneumoniae* species were shown to be responsible for bioconversion of protocatechuic acid to catechol. All isolated compounds were identified using HPLC, TLC, NMR, FTIR and MS/MS.

Acknowledgements: Dr Laura Smith and Dr Ken Beresford are acknowledged for their assistance

Keywords: *Boswellia dalzielii, S. aureus,* MRSA, MCF7, endophytes, *Enterobacter cloacae, Klebsiella pneumoniae*, bioconversion, antibacterial assay, cytotoxicity

References:
[1] Dalziel JM. Royal Bot Gard Kew 1910; 1910: 133–142
[2] Young JM. N Z J Agric Res 1974; 17: 115–119.

SL-054

Exploring the potential of apple dihydrochalcones on novel cosmetical, nutritional and pharmaceutical applications (ExPoApple2)

Stefan Martens[1], Francesco Guzzonato[1], Luisa Palmieri[1], Salvatore Multari[1], Stefano Benini[2], Rosanna Caliandro[2], Hermann Stuppner[3], Fabian Mayr[3], Daniela B. Schuster[3], Stefan Schwaiger[3]

[1]Edmund Mach Foundation, San Michele All'adige (tn), Italy, [2]Free University of Bozen - Bolzano, Bozen, Italy, [3]University of Innsbruck, Innsbruck, Austria

The growing knowledge about the health attributes of apple fruits and apple derived products, and the increasing public awareness for nutritious and health promoting food, have led to an increasing demand for apple fruits and products with high nutritional and healthy qualities. The apple tree itself (*Malus* sp.) is an agriculturally and economically very important crop and the source of several foods and beverages but also of some nutritional or pharmaceutical products so far. Especially the region Trentino-Alto Adige/Südtirol but also the "Genussregion Oberland" (in Tyrol) are well known for the important production of high quality apples. Fruits but also leaves are known to accumulate various polyphenolic compounds with dihydrochalcones as characteristic bioactive metabolite of the genus *Malus*, with otherwise only limited distribution in higher plants. Although this class of phytochemicals is, compared to other prominent secondary metabolites in apple (e.g. flavonoids, anthocyanins and proanthocyanidins), less understood, there is strong evidence that they greatly contribute to the health-beneficial activities of apple itself and derived products. This fact led to the hypothesis that dihydrochalcones might have also great potential for yet not identified applications in nutrition, cosmetics and pharmacy.
The specific aims of the project are:
1)to explore the distribution and to identify dihydrochalcone derivatives in various apple genotypes and species,
2)to understand the biological and genetic pathway including functionality of involved proteins and their potential as novel-biocatalysts in microbial cell factories,
3)to identify new targets and applications of natural and modified dihydrochalcones for applications in cosmetic, nutritional and pharmacy,
4)to establish and optimize biotechnological approaches for pre-industrial production/bio-catalysis of most promising dihydrochalcone derivatives involving structural characterised and optimised proteins

SL-055

Challenges in authenticating essential oils

Prabodh Satyal, William N. Setzer

Aromatic Plant Research Center, Pleasant Grove, United States

Based on the numerous side effects of synthetically-derived medicines, many people are trying to transform their lifestyles through naturopathy. Unfortunately, due to a lack of complete instrumental techniques and strict government regulation, adulteration of natural products is rampant all over the world. It is estimated that approximately 80% of commercially-available, so-called "natural" essential oils are adulterated in some ways. Through research and the establishment of proper standards, which include detailed chemical profiles of what authentic essential oils look like chemo-metrically, and thorough analysis of essential oils by synthetic markers and biomarkers, progress is being made to overcome the instrumental limitation in essential oil analysis and identify adulterated essential oils.

SL-056

Combining supercritical fluid extraction and chromatography with molecular networks generation for plant dereplication

Florent Olivon, Nicolas Elie, Louis-Félix Nothias-Scaglia, Marc Litaudon, David Touboul

CNRS ICSN, Gif-sur-yvette, France

For the most effective analytical strategies, development and validation include optimization of some analytical parameters such as resolution, detectability, sensitivity, simplicity, cost effectiveness, flexibility, and speed. Nevertheless, other aspects concerning operator safety and environmental impacts are usually not considered at the same level. Thus, an absurd situation emerged, due to the side effects of analytical methods developed to investigate natural compounds in complex samples that generate a large amount of chemical waste and lead to strong negative environmental impacts. Taking into account current public concerns on environmental questions, analytical studies and the consequent use of toxic reagents and solvents have increased to such a level that it becomes indefensible to continue without environmentally friendly methods. One of the largest consumption of toxic solvents is occurring when performing extraction and separation of natural compounds in complex mixtures.
The use of supercritical fluid (SF), in particular SF CO_2, offers unique advantages for the efficient extraction (SFE) of enriched fraction of natural compounds and fast and robust separations (SFC) [1]. Coupled to high resolution tandem mass spectrometry (MS/MS), SFC offers an excellent platform for generating molecular networking.
We propose here to exemplify SFE and SFC-MS/MS for the structural characterization of diterpenoids from *Euphorbia* showing diverse antiviral activities [2]. Moreover, the new molecular network software "METGEM" allowing the generation of networks in few seconds on a standard PC by non-classical t-sne algorithm will be introduced.

Acknowledgements: This work has benefited from a grant managed by Agence Nationale de la Recherche (ANR-16-CE29-0002-01 CAP-SFC-MS).

Keywords: supercritical fluid chromatography, supercritical fluid extraction, tandem mass spectrometry, molecular networking, dereplication.

References:

[1] Laboureur L, Ollero M, Touboul D. Int J Mol Sci 2015 Jun 17;16(6):13868–84.
[2] Nothias LF *et al*. J Nat Prod 2017 Oct 27;80(10):2620–2629.

SL-057

Carobs, the black gold of Cyprus: science meets industry

Antonis Kakas, Christiana Neophytou, Andreas Constantinou

University of Cyprus, Nicosia, Cyprus

"BlackGold" is a multidisciplinary project that is funded by the Research Promotion Foundation of Cyprus starting in 2019. The consortium is led by the University of Cyprus and is comprised by the Ministry of Health, the Agricultural Research institute, five private companies as well as the Coop Carob federation. The major objective of the project is to re-mobilize and expand the carob industry driven by extensive interdisciplinary scientific research to fully characterize the endemic varieties of Cyprus carobs. The specific aims of the project are: (1) to characterize the genetic, morpho-physiological and compositional variability of local carob genetic resources, (2) to improve carob cultivation in Cyprus, (3) to identify the chemical/biological properties of carobs and define their functional value, (4) to identify the profile of Cypriot Carob products and characterize their authenticity, and (5) to expand the commercial value of Cypriot carob in the international markets. Based on the solid scientific outcomes of this 3-year project, we expect to develop new and innovative food and nutritional supplements that would give us a competitive advantage in the growing international market of biological and gluten-free products. These products would then form the basis for new export activities, initially from the companies that are engaged in the project and later-on from other companies that will form the same type of cooperation with the project's scientific consortium. Importantly, by exposing farmers to the new economic incentives, we expect to motivate them to re-cultivate the carobs in response to the expected increased market demand for novel carob-based products. In conclusion, the "BlackGold" project aims to revitalize the carob industry of Cyprus by fully characterizing the biochemical, genetic and agricultural properties of Cypriot carobs and by forming working links between the academic/research institutions and the private industry.

SL-058

Cannabis and Its Discontents: Regulatory Choices Impacting the Success of Medical Cannabis Programs in the U.S. and EU

Elizabeth Ashford

Medicanna, Business Development, USA

In this session, regulatory and legislative decisions in the United States will be presented as well as how legal cannabis markets are driven in 32 U.S. states, and lessons other nations and how the EU can take to improve the efficacy of emerging medical cannabis programs. Topics covered will include the impacts of: physician prescriptions verses over-the-counter medicines; the existence or absence of a recreational market; taxation and fees; Schedule I classifications and the U.S. Drug Enforcement Administration; laws governing research funding; and more.

SL-059

Analysis of pesticides and mycotoxins in legal cannabis using flow based LCMSMS system

Ariovaldo Bisi

PerkinElmer Chromatography and LCMSMS LL EMT

Over half of the U.S. has legalized the use of medical cannabis due to its therapeutic benefits for ailments such as cancer, multiple sclerosis, and ALS.1-3 and some EU countries are adopting similar rules legalizing cannabis use for medical and recreative purpose. On top of the control of cannabinoids content in recreative cannabis that must be below a regulated concentration we need to check also contaminants like pesticides and mycotoxins

Like traditional agriculture crops, pesticides are sometimes used in cannabis cultivation to protect plants from pests and improve growth yield. Chronic exposure to pesticides can pose serious health risks; therefore, pesticide analysis in cannabis is an important consumer safety topic and LCMSMS is the most suitable technique for this purpose; extracts of cannabis are oily and needs either extensive, time and money consuming cleanup or a rugged MSMS analyzer that is not prone to contamination.

In this lecture we'll discuss an analytical workflow with simple extraction steps and LCMSMS analysis of a wide panel of pesticides and mycotoxins; all the data have been validated and shows sensitivity, linearity and recovery far better than legal requirements

SL-060

Method development of chlorophyll pigments separation by Supercritical Fluid Chromatography (SFC)

Thibault Lefebvre, Arwa Talbi, Emilie Destandau, Eric Lesellier

ICOA (Institute of Organic and Analytical Chemistry) UMR7311, Orléans, France

Supercritical Fluid Chromatography (SFC) is a suited method for separation of complex mixtures. Use carbon dioxide as main fluid of the mobile phase presents a lot of advantages, ecological, economical and physicochemical. CO_2 can be mixed with numerous co-solvents called modifiers, for tuning the mobile phase polarity inducing great retention changes and potential strong improvement of separation performances.
In order to study the chlorophyll pigments in natural extracts of plants, ethanolic extracts of ivy were made and SFC method was developed. Due to the numerous parameters acting on retention and separation, related in part to the compressibility of the mobile phases used, this method development can be subtle and should be carefully achieved. In that order, the use of UV/visible coupled detector is suited for the peak tracking from one experiment to another. Graphics of retention factor (Log k), separation factor (alpha) or discrimination factor (do) should be used for the selection of the optimal conditions, and also to study the parameter effects on interaction changes between the compounds and the supercritical fluid.
Due to the high log P value of chlorophyll, C_{18} bonded phase was first selected. Systematic experiments were achieved to understand and modelize the chromatographic behavior of the compounds regarding the modifier nature, its percentage, temperature, flow rate and back pressure. Chromatographic profile of the extract is dramatically modified by the nature of the modifier, and, due to the complexity of the extract, optimal conditions obtained are somewhere unusual for supercritical chromatography. Moreover, the observed chromatographic behavior was rarely described in literature for other compounds.
Besides, these systematic studies achieved by SFC, allow a better understanding of the solubility changes of these compounds during supercritical fluid extraction (SFE). This approach should be favored to provide selected extract by SFE in the future.

SL-061

The application of hydrostatic counter-current chromatography in the purification of pharmacologically active phenolics and alkaloids

Wirginia Kukula-Koch

Medical University of Lublin, Lublin, Poland

Counter-current chromatography is a liquid-liquid separation-type distribution technique in which the separation of compounds from complex matrices takes place based on the differences in their affinity to each of two immiscible phases, on a rotating column. Low consumption of reagent grade solvents, short operation time, high selectivity and sample recovery, the possibility of direct injection of crude extracts, or the use of a wide range of solvents for the construction of a biphasic system are the greatest benefits of this technique.
The application of hydrostatic counter-current chromatographs (known as centrifugal partition chromatographs) in the isolation protocols of some selected natural products from *Curcuma longa*, *Zingiber officinale*, *Argemone mexicana*, *Aesculus hippocastanum*, and *Berberis* spp. will be discussed. Several details on the separation conditions: the applied operation modes (e.g. pH-zone refining mode, or elution-extrusion mode), the composition of biphasic solvent systems and the apparatus settings will be presented to discuss about their impact on the purification efficiency.
Also, the results of bioactivity studies of the isolates will be shown, including online acetylcholinesterase inhibition by TLC-LC-MS/MS or LC-MS/MS, anticancer and MAO-A inhibition assays.

Funding information: National Science Center, Warsaw, Poland, Grant No: 2015/17/D/NZ7/00822

SL-062

Purification of bioactive natural products by centrifugal partition chromatography (CPC)

Tsvetelina Mandova[1,2], Grégoire Audo[2], Sylvie Michel[1], Raphael Grougnet[1]

[1]Laboratoire de Pharmacognosie, Université de Paris Descartes, Sorbonne Paris Cité, Faculté de Pharmacie de Paris, UMR-CNRS 8638 COMETE, 4 avenue de l'Observatoire, 75006 Paris, France, [2]Gilson Purification SAS, Application Laboratory, 22 rue Bourseul, F-56890 Saint-Avé, France

CPC is a continued liquid-to-liquid solvent partition where the target compounds are competitively distributed between the two-phase solvents due to their different partition coefficient. Using a centrifugal force, one phase is kept stationary while the other phase is pumped through the stationary one. In this work, three case studies on the purification of natural products from plants will emphasize the great potential of the CPC technique. *Centaurium erythraea* Rafn. (Gentianaceae) is a widespread herbaceous plant, the most abundant seco-iridoid glycosides, swertiamarin, sweroside and gentiopicrin, three structurally closely-related target natural products, were separated by CPC with swertiamarin purified from 8.72 mg/g in the crude extract until 770 mg/g (98.2%S) in enriched fractions. Experimental conditions were optimized in a different elution mode and different CPC devices coupled with MS detector. Indolizidine alkaloids, one ent-kaurane diterpene and its glycoside are some of the compounds isolated from two other species – an Argentinean spiny deciduous tree from *Prosopis* spp (Fabaceae) and the above ground parts of *Achillea clypeolata* Sibth. et Sim (Asteraceae), a Balkan endemic species. Therefore, we conclude that a suitable elution system using the CPC technology coupled to MS can afford good results in natural compound purification, with the great advantage of higher yields when compared to other separation techniques.

Keywords: Centrifugal Partition Chromatography (CPC), *Centaurium*, seco-iridoids, *Achillea*, *Prosopis* spp.

References:

[1] Ingrid Werner *et al*. Z. Naturforsch 2007; 62b: 267–271.
[2] Mandova *et al*. Phytochem Lett 2017; 20: 401–405.
[3] Santhaseelan Henciya *et al*. J Food Drug Anal 2017; 25:187–196.

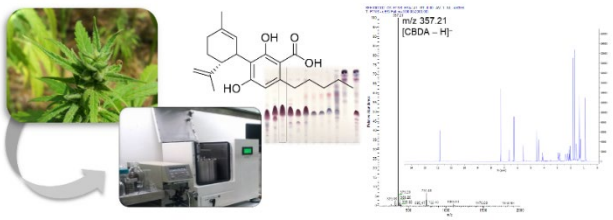
SL-063

A simple and effective approach for the isolation of acidic cannabinoids from *Cannabis sativa* L. using pH-zone-refining centrifugal partition chromatography

Johannes R. Popp[1,2], Eleftherios A. Petrakis[1], Apostolis Angelis[1], Maria Halabalaki[1], Günther K. Bonn[2], Hermann Stuppner[2], Alexios-Leandros Skaltsounis[1]

[1]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, Athens, Greece, [2]Institute of Pharmacy/Pharmacognosy, Center for Molecular Biosciences, University of Innsbruck, Innsbruck, Austria

Fibre-type *Cannabis sativa* L. (hemp) has been associated with a considerable therapeutic potential, as revealed by on-going research, largely due to the cannabinoids contained [1]. Cannabidiol (CBD) and its biogenetic precursor cannabidiolic acid (CBDA) represent the major non-psychoactive cannabinoids in hemp, possessing numerous pharmacological properties [1,2]. In this context, the development of simple and efficient protocols for their rapid isolation is increasingly required. This study reports an effective methodology for the isolation of the major cannabinoids originally present in the plant material from *Cannabis sativa* L., based on their ionisable character. The selective separation of cannabidiolic acid (CBDA), as a paradigm, from the extract obtained through supercritical fluid extraction (SFE) was performed by using centrifugal partition chromatography (CPC) in pH-zone-refining mode [3]. The fractionation was carried out using a biphasic solvent system, consisted of *n*-hexane/ethyl acetate/ethanol/water 8:2:5:5 (v/v/v/v). Trifluoroacetic acid was used as retainer in the organic stationary phase, while triethylamine was added as eluter in the aqueous mobile phase. The most promising fraction obtained by CPC was subsequently submitted to liquid-liquid extraction, in order to recover CBDA from the salt form as well as for further purification. Following this two-step procedure, more than 40% (w/w) of CBDA was totally recovered with higher purity compared to previously reported methodologies [4], i.e. greater than 95%, as revealed by HPLC-UV analysis. Taking into account the ideal scale-up of CPC, this methodology can be easily transferred to industrial scale and could be readily adaptable for the isolation of other acidic cannabinoids.



Keywords: Cannabidiolic acid (CBDA), cannabidiol (CBD), phytocannabinoids, *Cannabis sativa* L., preparative isolation

References:

[1] Izzo AA, Borrelli F, Capasso R, Di Marzo V, Mechoulam R. Trends Pharmacol Sci 2009; 30: 515–527.
[2] Morales P, Reggio PH, Jagerovic N. Front Pharmacol 2017; 8: 422.
[3] Ito Y. J Chromatogr A 2013; 1271: 71–85.

[4] Hazekamp A, Simons R, Peltenburg-Looman A, Sengers M, van Zweden R, Verpoorte R. J Liq Chromatogr Relat Technol 2004; 27: 2421–2439.

SL-064

Understanding the taste of wine: isolation and identification of new taste-active compounds in oak wood

Delphine Winstel, Pierre Waffo-Téguo, Blandine Cretin, Marine Gammacurta, Denis Dubourdieu, Axel Marchal

Unite de recherche Œnologie, EA 4577, USC 1366 INRA, ISVV, Université de Bordeaux, Villenave D'ornon, France

For several years, the chemistry of taste has aroused high interest both from academics and industrials. New taste-active compounds exhibiting mainly sweetness, but also bitterness, have been identified in various plants [1]. These molecules belong to diverse chemical families and their sensory properties are strongly affected by slight structural modifications. As a consequence, the investigation of natural taste-active products in a given matrix often appears as a major challenge for chemists. Such studies are particularly relevant in oenology since they allow a better understanding of wine taste.

Recently, taste-guided approaches have been developed in our group to isolate new molecules likely to contribute to wine sensory perception. Such methodology has been applied to various matrices and particularly to oak wood used for barrel aging. Purifications were achieved by liquid-liquid extraction, solid phase extraction, Centrifugal Partition Chromatography and Preparative High Performance Liquid Chromatography. After each fractionation step, the extracts were submitted to sensory analysis to select the most active one. Structural elucidation of purified compounds was carried out by Fourier Transform Mass Spectrometry and Nuclear Magnetic Resonance. Following these procedures, new sweet triterpenes called Quercotriterpenosides have been identified [2] and the structure-activity relationship has been investigated [3]. Furthermore, isolation of bitter lignanes has allowed the study of the influence of stereochemistry on taste properties by combining sensory analysis and Vibrational Circular Dichroism [4,5].

These results highlight the interest of inductive approach, hyphenating analytical technics and sensory analysis, to discover new taste-active compounds. These studies provide promising perspectives for a better understanding of the molecular phenomena associated to wine taste and, more generally, to taste perception.

References:

[1] Kinghorn *et al.*, 2010. Natural Products as Sweeteners and Sweetness Modifiers.
[2] Marchal *et al.* Anal Chem 2011.
[3] Marchal *et al.* Anal Chim Acta 2015.
[4] Cretin *et al.* Anal Chim Acta 2015.
[5] Sindt *et al.* J Nat Prod 2016.

SL-065

Application of CCC in the separation of active compounds from natural products

Xinyi Huang, Jianfei Liu, Yuan Gong, Kaijun Quan, Dong Pei, Duolong Di

Lanzhou Institute of Chemical Physics, Chinese Academy of Sciences, Lanzhou, China

Counter-current chromatography (CCC) is a liquid-liquid partition chromatography technique that is free to solid support matrix. By comparison with traditional chromatography technique with solid stationary phase, CCC has unique advantages of high efficiency and high recovery because it uses two immiscible solvents or solutions as stationary and mobile phase, which can eliminate the irreversible adsorption of samples on stationary phase and the solutes have access to the whole volume of the phase [1]. Consequently, this technique has been widely used for the preparative and analytical separation, isolation and purification of various products [2]. Our research group is concerned with the applications of CCC to natural product and conducted the research of the combination of CCC and other technologies. 1) High shear technique coupled with CCC was successfully used for the extraction and online isolation of seven highly polar chemical constituents from the *Brassica napus* L. The lower phase of solvent system was used as mobile phase and extract solvent. Seven compounds were obtained in a one-step extraction and separation process [3]. 2) CCC coupled with post-column on-line evaluation was developed to screen, isolate and identify the major anti-diabetic compounds present in the leaves of *Olea europaea* L. Five major constituents of the olive extracts with potential anti-diabetic activity have been successfully found and obtained by this method [4]. 3) The target guidance of the DPPH-HPLC-DAD experiment coupled with CCC was applied to screen and separate radical scavengers from the water extract of *Cynomorium songaricum* Rupr. Three compounds with potential antioxidant activities were obtained from crude sample through one-step separation [5].

These works suggest that the CCC technique combined with other techniques, which provide an effective way to separate target compounds from complex matrix of natural product.

SL-066

Large-scale isolation of high-purity peptide VVYP from Globin Peptide using MCI gel column combined with consecutive high-speed counter-current chromatography: aqueous two-phase systems

Dong Pei, Xin-Yi Huang, Duo-Long Di

Lanzhou Institute of Chemical Physics (LICP), Chinese Academy of Sciences (CAS)., Lanzhou, China

Peptides have gained increased interest over the past several decades because of their therapeutic properties. More than 7000 peptides have been identified, and these often have crucial roles in human physiology, including actions as hormones, neurotransmitters, growth factors, ion channel ligands, or anti-infectives [1-4]. Then, peptides purification has been a topic of considerable interest in the past few decades [5]. In this research, a strategy combining MCI GEL column chromatography and high-speed countercurrent chromatography was developed for the separation of high-purity peptide Val-Val-Tyr-Pro from Globin Peptide. After purification using MCI GEL CHP20/P120 column with a mixture of ethanol/water /TFA (85:15:0.1, v/v/v), a fraction of Val-Val-Tyr-Pro mixtures with a purity of 15.7% was obtained. High-speed countercurrent chromatography with a aqueous two phase systems of EtOH/ACN/1-PrOH/$(NH_4)_2SO_4$ satd•soln/H_2O (0.5:0.5:0.25:1.5:0.7, v/v) was used to separate the Val-Val-Tyr-Pro. Ammonium sulfate from HSCCC fractions was removed from target compounds by MCI GEL column using ethanol/water in stepwise elution mode. 140 mg of VVYP was successfully purified with the purities of 98.86 % from 40 g crude Globin Peptide. The amino acid sequence of the VVYP was determined by ESI-HRMS/MS. The method presents a practical strategy for the large-scale separation of pure peptide VVYP from Globin Peptide, and provides a reference method for obtaining high-purity peptide from other Polypeptide mixture.

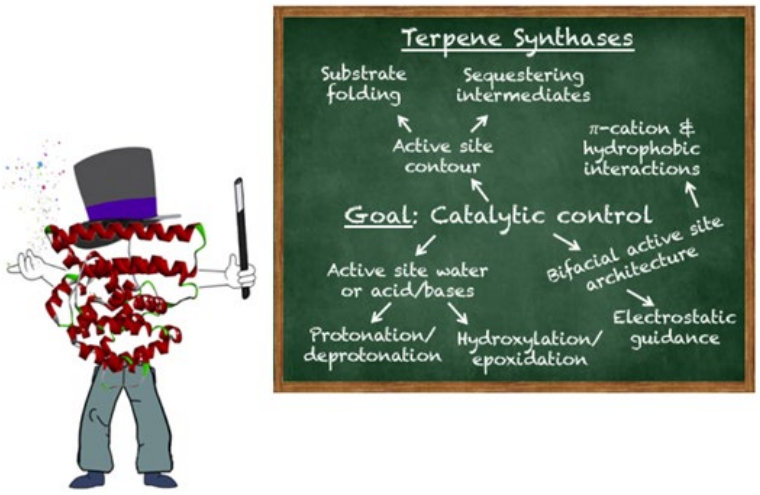
SL-067

Redefining enzyme catalysis: chemical control in the biosynthesis in terpene cyclases

Dan T Major

Bar-ilan University, Ramat Gan, Israel

Terpene cyclases are responsible for the initial cyclization cascade in the multistep synthesis of more than 80,000 known natural products. This abundance of compounds is generated using a very limited pool of substrates based on linear isoprenoids. The astounding chemodiversity obtained by terpene cyclases suggests a tremendous catalytic challenge to these often-promiscuous enzymes. In the current presentation, we will focus on representative mono-, sesqui-, and diterpene cyclases. Specifically, we will describe a novel view of enzyme catalysis based on the biosynthesis of these terpene synthases. For these enzymes, the catalytic challenge is chemical control of hyper-reactive intermediate carbocation species, rather than rate acceleration. Based on multiscale *in silico* simulations, we propose that to ensure catalytic specificity these enzymes employ their active site contour to fold the initial substrate and sequester intermediates. Chemical control is obtained through conformational selection, electrostatic steering, dispersive interactions that adapt to changing electronic requirements during catalysis, as well as carefully placed basic residues.1-4 A detailed understanding of terpene cyclase mechanisms is expected to be useful in the design of novel enzymes that can synthesize complex terpenes efficiently.



References:

[1] Weitman M, Major DT. J Am Chem Soc 2010; 132: 6349–6360
[2] Major D T, Weitmann M. J Am Chem Soc 2012; 314: 19454–19462
[3] Major DT, Freud Y, Weitman M. Curr Opi. Chem Biol 2014; 21: 25–33
[4] Dixit M, Weitman M, Gao J, Major D T. ACS Catal. 2017; 7: 812–818

SL-068

Novel pathways constructed through metabolic engineering in microbial biofactories to produce highly valuable natural products

Emmanouil Trantas[1], Maria Halabalaki[2], Leandros Skaltsounis[2], Filippos Ververidis[1]

[1]Plant Biochemistry and Biotechnology Group, Laboratory of Biological and Biotechnological Applications, Department of Agriculture, School of Agriculture, Food and Nutrition, Technological Educational Institute of Crete, P.O. 1939, GR 710 04, Heraklion, Greece, [2]Division of Pharmacognosy and Natural Product Chemistry, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, GR-15771, Athens, Greece

One of the most abundant phenolic compounds traced in olive tissues is Hydroxytyrosol (HT) and Resveratrol (Rs) in grapes and some other plant tissues. These molecules have been attributed with a pile of beneficial effects, well documented by many epidemiological studies and thus adding value to products containing it. Strong antioxidant capacity and protection from cancer are only some of their exceptional features making them ideal as potential supplements or preservatives to be employed in the nutraceutical, agrochemical, cosmeceutical, and food industry. Although Rs biosynthetic pathway is well documented in plants, the HT pathway in plants is not well apprehended yet. However, based on its structure, it can be presumed that HT might be derived from tyrosine which is then biotransformed to HT through sequential biosynthetic steps involving a hydroxylation, a decarboxylation, and a deamination reaction. In this presentation, we will present metabolic engineering strategies encompassing a dual pathway introduced in *Escherichia coli*, or *Saccharomyces cerevisiae* leading them to produce HT and Rs respectively. Phenylalanine, or Tyrosine were depicted as the precursor molecules and were fed either externally or utilized through an overproduction approach straight from glucose. Various gene combinations derived from plants or bacteria were used to form a novel, artificial biosynthetic dual pathway managing to redirect the carbon flow towards the production of HT or Rs directly from glucose. Although, various biosynthetic bottlenecks were faced, we have currently achieved Rs production at promising levels, while, equimolar concentration of HT to tyrosine as precursor when overproduced straight from glucose, reaching the level of 1.76 mM (270.8 mg/L) within 24-48h as analyzed by LC-HRMS.

SL-069

Balkan Botanic Garden of Kroussia: Protection, conservation and sustainable use of plant genetic recourses

Eleni Maloupa, Krigas Nikos

Laboratory of Conservation and Evaluation of Native and Floricultural Species, Balkan Botanic Garden of Kroussia, Institute of Plant Breeding and Genetic Resources, Hellenic Agricultural Organization Demeter, GR-57001 Thermi, Thessaloniki, Greece

Plant diversity in Greece and the Balkans is exceptionally rich and unique, presenting a higher degree of endemism in relation to surface than any other comparable area of Europe or the Mediterranean region.
Since its establishment, the BBGK has been focused on the conservation and the sustainable use of native plants of Greece and the Balkans ('Only Native Plants' Policy), leaving aside the exotic and ornamental plants. All plant displays in BBGK have been created using plant material originating in the wild that has been sustainably managed.
The Balkan Botanic Garden of Kroussia (BBGK), Greece was funded in the framework of the Inter-Regional Developmental Initiatives of the European Union (INTERREG II-External Borders). The BBGK was founded on 19 May 2001. It is member of the Botanic Gardens Conservation International (BGCI) and National Network of Greek Botanic Gardens. The research and services of BBGK are supported by the Laboratory of Protection and Exploitation of Native and Floricultural Plants located at Thermi of Thessaloniki. Aiming first to promote to urban citizens environmental awareness issues regarding native plant conservation and second in order to facilitate the maintenance of the increasing mother plantations, the BBGK has recently established the Garden of Environmental Awareness (GEA) in Thermi at sea level.
The BBGK is situated in northern Greece, about 70 km from Thessaloniki (the second biggest Greek city), near the mountain village of Pontokerasia in the Prefecture of Kilkis (41°05'N/23°06'E).
Greece has an exceptionally rich flora with more than 6,600 taxa (species and subspecies), about 50% of European Flora. Approximately 15-20% are unique, (Greek endemic taxa), presenting the highest degree of endemism for any comparable territory in Europe and the Mediterranean region. Almost 20% of the Greek flora have medicinal and aromatic properties. These plants produce a great variety of chemical compounds. As a result, the beneficial properties in the human body are countless and complex. The importance of diet for maintenance of optimal health was known from the ancient times. Aromatic and medicinal plants extensively used in the Mediterranean Diet as flavorings and seasonings, for the preservation and storage of various foods and help to maintain their organoleptic properties. Plants that are often used in Mediterranean diet are: Oregano (*Origanum vulgare* ssp. *hirtum*), Sage (*Salvia officinalis*), Thyme (*Thyumus* spp.) and Rosemary (*Rosmarinus officinalis*). But have you ever cooked with dittany (*Origanum dictamnus*), mountain tea (*Sideritis* spp.), or wild rose (*Rosa canina*). This presentation approaches scientific knowledge in a simple way, enriches the Mediterranean diet, opens new horizons of gastronomic and gives new impetus to the cultivation of aromatic and pharmaceutical plants.

SL-070

Gene activation strategies in fungi – Sex, Drugs, and Genetics?

Donovon A. Adpressa, Paige. E. Mandelare, George F. Neuhaus, Lanelle R. Connolly, Kristina M. Smith, Michael Freitag, Sandra Loesgen

Oregon State University, Corvallis, United States

Many microorganisms produce secondary metabolites that are believed to have critical roles in intra- and inter-species communication, competition for resources, or defense of a symbiotic partner. Despite the importance of these metabolites, it is estimated that only a tiny fraction of microbial secondary metabolites has been identified. By now, far more than 1,000 fungal genomes have been sequenced. The high abundance of biosynthetic gene clusters encoding for secondary metabolites compared to the small number of compounds seen produced under standard laboratory conditions give hope for future natural products discoveries. However two prominent challenges are that most fungi do not express their secondary metabolites under standard laboratory culture conditions, and there is no straightforward method for identifying novel secondary metabolites from complex extracts. Here we present the discovery of new fungal chemistry from gene activation experiments using co-cultivation, chemical perturbation, and deletion mutants. We employ multi-channel metabolomics to rapidly identify new metabolites, and leverage a panel of assays to assess biological function. We found new, cytotoxic bianthrones called allianthrones A-F from the co-culture of two developmental forms of a marine alga-derived *Aspergillus alliaceus* strain. HDAC inhibitor treatment of *Chalara* sp. resulted in the production of the new chalanilines A and B with cytotoxic and antibacterial properties. Finally, we examined metabolites produced by *Fusarium graminaerum* deficient in histone methylation by virtue of deletion of the kmt6 histone methyltransferase gene, and found overexpression of many known compounds but also new terpenes that we connected to their respective biosynthetic gene clusters.

SL-071

New insights on the structure of Kraft lignin & its fractions with tangible practical implications

Dimitris Argyropoulos

North Carolina State University, Raleigh, United States

The complexity of native softwood lignin when coupled with the complexity of the Kraft pulping process is known to lead to a rather heterogeneous material that has eluted us to date.
During this lecture a new structural constitutional scheme will be proposed for Softwood Kraft Lignin. This effort attempts to unify and rationalize our current knowledge of Kraft pulping chemistry with a series of focused NMR & Chromatographic measurements.
This effort will thus introduce the foundations for describing our systematic efforts in the following areas aimed at arriving at practical applications for an otherwise intractable raw material.
More specifically the lecture will cover our efforts in:
(i) Refining technical Kraft lignin so as to expose its potential as a source for reactive polyphenols of well-defined molecular weight polymers and oligomers. We will then demonstrate that a continuum of narrow fractions can be isolated from softwood Kraft lignin, common to a variety of such sources irrespective of the manufacturing details of the pulping process. Such consistently homogeneous lignin streams from technical lignins offer significant commercial ramifications.
(ii) Creating heat stable Kraft lignin copolymers with heat stabilities approaching 300 °C.
(ii) Creating Novel blends with polylolefins
(iii) Creating new thermoplastic lignin polymers and precursors to carbon fibers by applying propargylation derivatization chemistry followed by thermal treatments.
This approach offers a versatile novel route for the eventual chain extension & utilization of technical lignins with a significant amount of molecular control.

SL-072

Metabolite profiling of the roots of 26 different accessions of soil-grown non-sterile *Arabidopsis thaliana* involved in plant interaction with rhizosphere microorganisms

Kourosh Hooshmand[1,2], Enoch Narh Kudjordjie[1], Rumakanta Sapkota[1], Tong Shen[2], Oliver Fiehn[2], Mogens Nicolaisen[1], Inge Fomsgaard[1]

[1]Department of Agroecology - Aarhus University, Slagelse, Denmark, [2]NIH West Coast Metabolomics Center, UC Davis Genome Center, University of California, Davis, CA 95616, USA, DAVIS, United States

The tritrophic interaction between plant, beneficial microorganisms and pathogens have been understudied in the past. It is currently believed that plants are able to regulate the microbial composition inhabiting its root and rhizosphere by exuding distinct bioactive chemical compounds and possibly thereby acquire resistance to biotic stresses.
In order to investigate the molecular mechanism underlying this interaction, 26 accessions (differences in genetic adaptation) of *Arabidopsis thaliana* (wild type) were grown in field soil under controlled conditions. The root (n=6) and its attached soil was harvested, snap-frozen, lyophilized, ground and extracted in a monophasic solvent mixture. The fraction of extracted material was subjected to derivatization (methoximation+silylation) for primary metabolites profiling by employing GC-TOFMS platform. The raw data was processed by BinBase (Fiehn 2016) for metabolite annotation as well as post-processed to remove the redundant features.
The remaining fraction was subjected to solvent-solvent extraction by utilizing a biphasic solvent mixture (Matyash *et al.* 2008). The non-polar phase was analyzed by using reverse phase LC-MS/MS on a high resolution Thermo Q Exactive HF mass spectrometer for lipids profiling (lipidomics). The data for polar layer was acquired by using HILIC-TripleTOF to determine polar metabolite composition. Both lipidomics and metabolomics raw data were processed by MS-DIAL (Tsugawa *et al.* 2015) followed by post-processing with MS-FLO (DeFelice *et al.* 2017) to remove the insignificant features. Furthermore, the microbial composition was determined by the Illumina MiSeq sequencing platform.
The data from multiple platforms will be combined and different statistical approaches such as one-way ANOVA, PCA, PLS-DA, HCA and heat map will be employed to highlight the statistical significant features associated with a certain group of plant genotype as well as their significant impact on rhizosphere microbial community composition.

Keywords: Rhizosphere, metabolite, microorganism, metabolomics, lipidomics, LC-MS/MS, GC-MS/MS

References:

[1] DeFelice BC, Mehta SS, Samra S, Čajka Ts, Wancewicz B, Fahrmann JF, Fiehn O. Anal Chem 2017; 89 (6): 3250–3255
[2] Fiehn O. Curr Protoc Mol Biol 2016; 114:30.34.31-30.34.32.
[3] Matyash V, Liebisch G, Kurzchalia TV, Shevchenko A, Schwudke D. J Lipid Res 2008; 49(5): 1137–1146.
[4] Tsugawa H, Cajka T, Kind T, Ma Y, Higgins B, Ikeda K, Kanazawa M, VanderGheynst J, Fiehn O, Arita M. Nat Methods 2015; 12(6): 523.

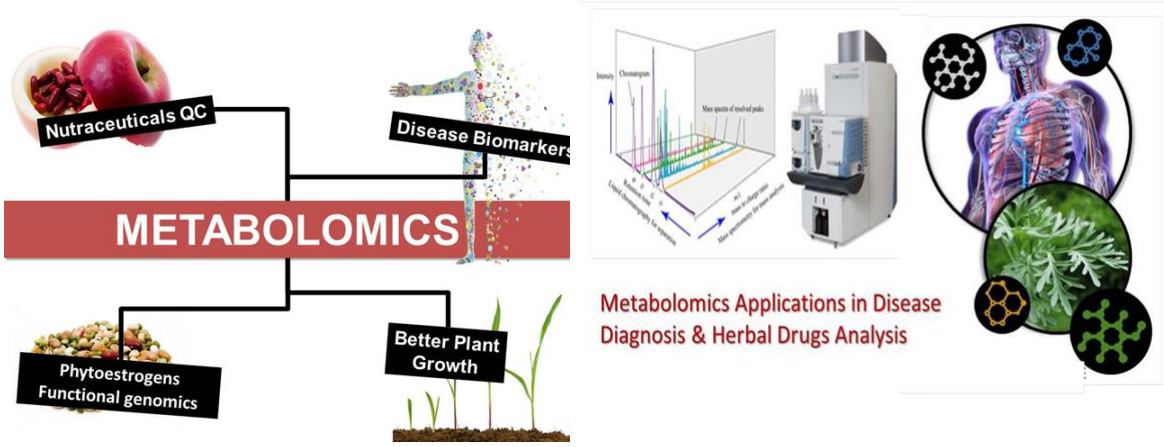
SL-073

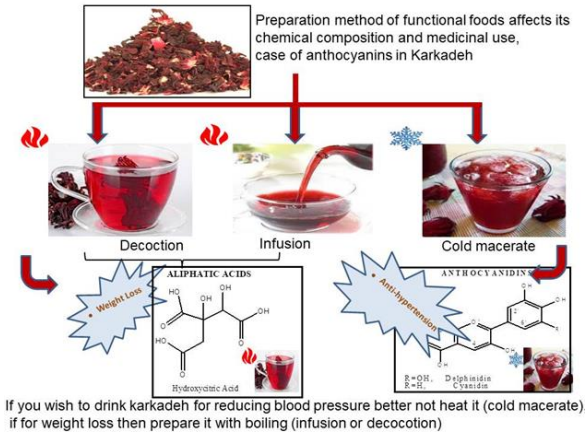
Metabolomics novel application in functional foods analysis & drug discovery

Mohamed Farag

American University in Cairo, Cairo, Egypt

The ability to sequence whole plant and human genomes has taught us that our knowledge with respect to gene function is rather limited. Functional genomics analyses include investigations at the level of gene expression (transcriptomics), protein translation (proteomics) and more recently the metabolite network (metabolomics). Metabolomics is the study of global metabolite profiles in a system (cell, tissue, or organism) under a given set of conditions. The analysis of the metabolome is particularly challenging due to the diverse chemical nature of metabolites in a cell. This presentation provides an overview of metabolomics and discusses its complementary role within system biology. It highlights how metabolome analyses are being conducted using different spectroscopic techniques NMR and MS, and how the highly complex data generated are analyzed. Specific examples will then be presented to illustrate how metabolomics can lead to valuable information relative to natural products drug discovery and herbal medicines quality control analysis.





SL-074

Feature Fishing – How to catch bioactives in a complex mixture of lanostane triterpenes

Ulrike Grienke[1], Paul A. Foster[2,3], Julia Zwirchmayr[1], Ammar Tahir[1], Judith M. Rollinger[1], Emmanuel Mikros[1,4]

[1]Department of Pharmacognosy, Faculty of Life Sciences, University of Vienna, Vienna, Austria, [2]Institute of Metabolism and Systems Research, University of Birmingham, Birmingham, United Kingdom, [3]Centre for Endocrinology, Diabetes, and Metabolism, Birmingham Health Partners, Birmingham, United Kingdom, [4]Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Athens, Panepistimiopolis Zografou, Athens, Greece

The discovery of bioactive compounds from complex natural product mixtures is a tedious task with many drawbacks, e.g. the loss of activity over fractionation processes or the re-isolation of known bioactives. In the past years, chemometric methods and statistical correlation of spectral with bioactivity data have improved the classical bio-guided isolation. However, hit discovery from an extract containing close structural analogues remains challenging.
The present study is the first report on the application of a heterocovariance (HetCA) approach [1] to a complex mixture of the same compound class providing a natural biosynthetic library of analogues. The approach is exemplified in the discovery of steroid sulfatase (STS) inhibiting lanostane triterpenes from the extract of *Fomitopsis pinicola* Karst. STS catalyses the conversion of sulfated precursors to free steroids and has evolved as an attractive drug target for hormone-dependent cancers [2].
Prior to any isolation the aim of this study was to identify structural features that contribute to STS inhibition observed for the mixture and to rule out the ones that do not contribute or are detrimental for bioactivity. To reduce the complexity of the extract, it was separated into 32 fractions. As opposed to bio-guided isolation, special attention was payed to achieve a concentration variation of the components over several fractions. Using the statistical method of covariance, ^{1}H NMR spectra were correlated with STS inhibition data and complemented with HRMS data.
The effectiveness of this approach was demonstrated by disclosing chemical features crucial for STS inhibition, thus taking advantage from the innate library produced by the polypore's biosynthetic machinery. As a proof of concept two lanostanes equipped with these imperative features were isolated and showed IC_{50}s in the low micromolar range.

References:

[1] Aligiannis N *et al*. ChemistrySelect 2016; 1: 2531–2535.
[2] Mueller JW *et al*. Endocr Rev 2015; 36: 526–563.

SL-075

Can nature's pharmacy influence the health of honey bees?

Nanna Hjort Vidkjær, Per Kryger, Inge S. Fomsgaard

Aarhus University, Faculty of Science and Technology, Department of Agroecology, Forsøgsvej 1, DK-4200 Slagelse, Denmark

Declining honey bee (*Apis mellifera*) populations are receiving increasing attention especially because honey bees are important pollinators of our food crops. The underlying cause of the decline is hypothesized to be multifactorial and honey bees face many stressors including pathogens, xenobiotics, and changes in floral resources.
Multitudes of bioactive plant secondary metabolites (PSMs) – widely utilized in human medicine – are present in the honey bees pollen and nectar diet. Some PSMs can reduce the virus loads in honey bees [1] whereas others are known inducers of their detoxification- and immune system [2,3] but the influence of PSMs on honey bee health remain largely unexplored. Detailed studies of the impact of PSMs on honey bee health are thus essential and PSM uptake and metabolization requires clarification since limited knowledge presently exists.
In a controlled feeding experiment, honey bees were exposed to different PSMs including alkaloids, cyanogenic glycosides, iridoid glycosides and diterpenoids. A global overview of the honey bees' response to individual PSMs was attained using a combination of untargeted GC-TOF-MS metabolomics focusing on effects on the honey bees' primary metabolism and RT-qPCR targeting expression of genes related to the detoxification- and immune system. Uptake of the PSMs was investigated with LC-MS. The results of this experiment establish the foundation for a larger field experiment focusing on the implication of PSMs in honey bee disease resistance and detailed studies of the honey bees metabolization of PSMs.

Keywords: Honey bees, plant secondary metabolites, uptake and metabolization, disease resistance, metabolomics

References:

[1] Palmer-Young EC *et al.* J Econ Entomol 2017; 110(5): 1959.
[2] Mao W *et al.* PNAS 2013; 110(22): 8842.
[3] Johnson RM *et al.* Plos One 2012; 7(2): e31051.

SL-076

High performance separation techniques for identification, characterization and quantification of plant secondary metabolites with health-promoting properties

Danilo Corradini, Isabella Nicoletti

CNR, Institute of Chemical Methodologies, Via Salaria Km 29.300, 00015 Monterotondo Stazione, Rome, Italy

The wide range of bioactive compounds produced in plants by the secondary metabolism comprise food ingredients having disease prevention and health-promoting properties, in addition to give specific sensorial characteristics to the aliment. The identification and quantification of these target compounds in plants and agro-food matrices is a challenging task, continuously requesting the development of more robust, efficient and sensitive instrumental analytical techniques. This communication discusses fundamental and practical aspects of both reversed phase high performance liquid chromatography (RP-HPLC) and capillary zone electrophoresis (CZE) employed for the analysis of plant secondary metabolites occurring in food and cosmetic matrices. The two analytical separation techniques might display complementary capability in separating secondary metabolites, as it is discussed for the analysis of phenolic compounds in plant extracts. The different selectivity exhibited by RP-HPLC and CZE in separating phenolic compounds has been ascribed to the concomitant presence of hydrophilic, hydrophobic and ionogenic groups displayed by most of these compounds, which is expected to influence to different extents the separation mechanisms operating in CZE and in RP-HPLC of molecules bearing multifunctional moieties. The presentation discusses the influence of various operational parameters and experimental conditions employed in CZE and in RP-HPLC on the separation performance of phenolic compounds, which are widely distributed in the plant kingdom, form an integral part of human diet, and have a remarkable position as active components in cosmetics, functional foods and food supplements.

The influence of the composition of either the electrolyte solution (BGE) or the mobile phase on the selective separation of bioactive secondary metabolites in CZE and RP-HPLC, respectively, is discussed. Appropriate selection of either the BGE in CZE or the mobile phase in RP-HPLC involves the evaluation of the equilibrium in solution that might take place between the analytes and the components of such solutions. Also discussed is the practical application of RP-HPLC to study the occurrence of secondary metabolites in selected edible plants as a function of their genetic differences and environmental grown conditions, as well as the determination of phenolic compounds in agro-food matrices during the transformation of raw ingredients into food and in the production of food supplements.

SL-077

New isomer of pancracine as a potential anticancer agent

Kateřina Breiterová[1], Anna Hošťálková[1], Lubomír Opletal[1], Darja Koutová[2], Jana Maříková[3], Lucie Cahlíková[1]

[1]ADINACO Research Group, Department of Pharmaceutical Botany, Faculty of Pharmacy in Hradec Králové, Charles University, Akademika Heyrovského 1203, 500 05 , Hradec Králové, Czech Republic, [2]Department of Medical Biochemistry, Faculty of Medicine in Hradec Králové, Charles University, Šimkova 870, 500 03 , Hradec Králové, Czech Republic, [3]Department of Organic and Bioorganic Chemistry, Faculty of Pharmacy in Hradec Králové, Charles University, Akademika Heyrovského 1203, 500 05 , Hradec Králové, Czech Republic

Amaryllidaceae alkaloids (AmA) – as the name suggests – are specific for the Amaryllidaceae family. Plants from this family are interesting source of bioactive compounds. So far, nearly 600 alkaloids of various structural types have been detected. Among the most important biological activities of AmA belong activities associated with Alzheimer's disease (AD) and the antineoplastic activity. These and other diseases of affluence are becoming increasingly widespread all over the world. From this reason the development of new potential drugs is needed. Galanthamin is already used as a reversible selective inhibitor of human erythrocytic acetylcholinesterase (HuAChE; IC_{50} HuAChE = 1.5 ± 0.2 μM) [1] in patients with AD. Many AmA have been screened for their activity to inhibit the growth of different cancer cell lines and active compounds can be also used as lead structures for a synthesis of their semisynthetic analogues. Among the most widely used templates of AmA belong lycorine and haemanthamine.

From summary alkaloidal extract made from 34 kilograms of fresh bulbs of *Narcissus* cv. PROFESSOR EINSTEIN 21 different alkaloids have been isolated and identified by MS, HRMS, 1D- and 2D-NMR techniques and X-ray. All alkaloids were tested on their activities associated with AD (AChE, BuChE, POP, GSK 3β inhibition) and their ability to inhibit the growth of several cancer cell lines. From isolated alkaloids, the newly isolated isomer of pancracine gave the best results. The most sensitive lines after 48 hours treatment with 10 μM solution of pancracine isomer were MOLT-4 (viability 1±0 %), Jurkat (17±5 %) and MCF-7 (18±2 %). Now it has been undergoing IC_{50} determinations and determining the site of interference in the cell cycle.

Acknowledgements: The study has been supported by SVV 260 292 and 260 412.

References:

[1] He M, Qu CH, Gao O, Hu X, Hong X. RSC Advanced 2015; 21: 16562–16574.

SL-078

Angiotensin converting-enzyme inhibitory activity of bioactive extracts from Philippine plants

Christine Hernandez, Rezzaira Astorga, Lyen Castro, Mavis Fabian, Jasmin Tutor

Institute of Chemistry, University of The Philippines Dilliman, Quezon City, Philippines

Hypertension is a major health concern not just of the Philippine society, but it is a persistent problem worldwide. It is the third leading cause of morbidity according to the Department of Health's statistics. Recognized as the "silent killer", it gradually destroys the body without any symptoms. It damages major organs in the body resulting to other diseases. Untreated hypertension may lead to stroke, blindness, heart attack, kidney, and heart failure. Plants continue to be an excellent source of lead compounds. Under the Discovery and Development of Health Products program in the country, 2.400 plant samples were evaluated for their potential anti-hypertensive effect. Using the angiotensin-converting enzyme or ACE inhibitory assay, several plant extracts were prioritized and the bioactive compounds were isolated and identified. The results of this study show that plant extracts can be developed as a potential anti-hypertensive health product.

Acknowledgement: This project is funded by the Department of Science and Technology through the Philippine Council for Health Research and Development

Keywords: hypertension, medicinal

SL-079

Philippine honey possesses unique natural products composition and promising antimicrobial and antioxidant properties

Jonel P. Saludes[1,2], Angelica Faith L. Suarez[1], April Dawn G. Tirador[1], Zenith M. Villorente[3], Catherine F. Bagarinao[3], Jan Vincent N. Sollesta[3], Doralyn S. Dalisay[1,2]

[1]Center for Chemical Biology and Biotechnology (C2B2), University of San Agustin, Iloilo City, Philippines, [2]Center for Natural Drug Discovery and Development (CND3), University of San Agustin, Iloilo City, Philippines, [3]Maridan Industries, Inc., Jaro, Iloilo City, Philippines

The nutritional and marketing value of honey is anchored on its antibacterial, antioxidant, anti-inflammatory, and wound-healing properties. Its composition and properties are influenced by floral source and environmental conditions, honeycomb collection and processing, and final honey extraction. Here, we present a metabolomics-based approach to correlate the natural products and biological activity of Philippine honey. Fifteen honey samples were collected and studied from various islands in the Philippine archipelago. The small molecule natural products were extracted using a nonionic polymeric resin and the crude honey extracts were analyzed by untargeted HPLC-DAD (High-Performance Liquid Chromatography with Diode-Array Detection) and LCMS-IT-TOF (Liquid Chromatography/Mass Spectrometry-Ion Trap-Time of Flight) that led to the annotation of 269 metabolites (flavonoids, phenolics, glycosides, and peptides), some of which are unique in Philippine honey. Multivariate statistical analysis using principal component analysis (PCA) scatter plot showed distinct clustering patterns of honey metabolites on the first principal component that coincided with differences based on source location, bioactivities, and metabolites. The viability of Philippine honey as antibacterial agent against skin pathogen (Methicillin resistant *Staphylococcus aureus*, MRSA) and its antioxidant activity were evaluated. Results showed that Philippine honey demonstrate moderate antibacterial activity against MRSA and strong antioxidant activity. We also analyzed the total phenolic content (TPC) and total flavonoid content (TFC) of honey extracts to correlate these to biological activity. The TPC analysis used chemometric model (partial least-squares regression) while TFC was determined using Aluminum chloride colorimetric method. Results showed that the TFC and TPC of Philippine honey significantly correlates with its biological activities. Our findings demonstrate valuable information on the secondary metabolites of Philippine honey that act as antimicrobial and antioxidant agents.

Acknowledgements: This work is funded by United States Agency for International Development (USAID) through its Science, Technology, Research and Innovation for Development (STRIDE) Program and University of San Agustin.

SL-080

Hijacking breast cancer oncogenicity by methoxylated flavonoidal compounds isolated from *Cleome droserifolia*

Rana Youness[1], Mohamed Gad[2], Amira Abdelmotaal[3]

[1]Pharmaceutical Biology Department, Faculty of Pharmacy and Biotechnology, German University in Cairo, Cairo, Egypt, [2]Biochemistry Department, Faculty of Pharmacy and Biotechnology, German University in Cairo, Cairo, Egypt, [3]Pharmacognosy Department, Faculty of Pharmacy, King Khalid University, Abha, Saudi Arabia

According to the latest reports, Breast Cancer (BC) ranks as the second most common malignancy among both sexes. Yet, it is the most common malignancy among females, thus represents a top biomedical research priority. A positive correlation has always been noticed between flavonoid rich diet and lower risk of BC which lead to the question whether flavonoids can only acts as chemopreventive agents, or can also disrupt the interplay between different genes and proteins involved in the pathogenesis of BC. Recently, epidemiological studies suggest an inverse association between a higher intake of flavonols and flavones in particular, but not other flavonoid subclasses or total flavonoids, is associated with a decreased risk of BC, especially among post-menopausal women. Our research group has recently highlighted a pivotal role of several methoxylated flavonoidal compounds isolated from *Cleome droserifolia* in halting the oncogenic profile of liver cancer cells through activation of TP53/miR-15/miR-16 tumor suppressor axis. Currently, our research group is unveiling the capacity of such methoxylated flavonoidal compounds in harnessing BC oncogenicity and unraveling the underling mechanisms downstream their potential activity. The results showed a preferential and selective activity of flavonoidal glycosides isolated from *C. droserifolia* in halting the most aggressive subtype of BC which is the triple negative breast cancer cells through active and thus emphasizing their prominent capacity to act as anti-cancer agents. Our study was extrapolated to unveil the mechanism by which those natural compounds halt the TNBC oncogenic profile and boost the innate immune system recognition through inducing the expression of the natural killer cells activating ligands such as UL-16 binding proteins.

SL-081

Anti-proliferative Guaianolides from the Aerial Parts of *Chrysophthalmum montanum* (DC.) Boiss.

Fatma Ayaz[1], Esra Emerce[2], Nezhun Gören[3], İhsan Çalış[4], Mujeeb Ur Rehman[5], Muhammad Iqbal Choudhary[5], Nurgün Küçükb\oyacı[6]

[1]Department of Pharmacognosy, Faculty of Pharmacy, Selcuk University, 42250 Konya, Turkey, [2]Department of Pharmaceutical Toxicology, Faculty of Pharmacy, Gazi University, 06330 Ankara, Turkey, [3]Department of Molecular Biology and Genetics, Faculty of Science and Arts, Yıldız Technical University, 34220 Istanbul, Turkey, [4]Department of Pharmacognosy, Faculty of Pharmacy, Near East University, Nicosia 99138, Turkish Republic of Northern Cyprus, [5]HEJ, Research Institute of Chemistry, International Center for Chemical Sciences, University of Karachi, 75270 Karachi, Pakistan, [6]Department of Pharmacognosy, Faculty of Pharmacy, Gazi University, 06330 Ankara, Turkey.

Chrysophthalmum montanum (DC.) Boiss. (Asteraceae), also named "tutça" in Turkish folk medicine, has been used traditionally to ease respiration against flu and heal part of the body as well as wound of animal [1]. In our recent studies on *C. montanum* through bioassay guided fractionation, it was demonstrated that bioactive constituents were isolated [2]. As a part of our ongoing studies on *C. montanum*, we aimed to isolate secondary metabolites from the chloroform subextract of the plant and evaluate their antiproliferative effects using MTT assay. After the multistep chromatographic purifications on the chloroform subextract, three novel guaianolides (**1**-**3**), namely 1*β*,4*β*-dihydroxy-guaia-10(14),11(13)-dien-8*α*,12-olide (**1**), 4*α*, 6*α*-dihydroxy-9*β*,10*β*-epoxy-1*β*H-guaia-11(13)-en-8*α*,12-olide (**2**) and 4*α*,9*β*-dihydroxy-6*α*-acetoxy-1*β*H-guaia-10(14),11(13)-dien-8*α*,12-olide (**3**), together with known compounds, a guaianolide, (4*α*,5*α*,8*β*,10*α*)-4,10-dihydroxy-1,11(13)-guaiadien-12,8-olide (**4**), a lignan, pinoresinol (**5**), a flavonoid, chrysosplenol C (**6**), two triterpenes, a mixture of taraxasterol acetate and Ψ-taraxasterol acetate (**7**) and a mixture of taraxasterol and Ψ-taraxasterol (**8**), were isolated. Their structures were elucidated by means of spectroscopic analysis. The isolates, except for **5** and **8**, were investigated for antiproliferative effects against Hela, MCF-7, A549, and BEAS-2B cells at a concentration range of 0.6-20 µg/ml for 48 h. Our data showed that **1**, **4** and **6** caused significant decrease in cell viability (53–64%) at a concentration of 20 µg/ml in MCF-7 cells. Exposure to **4** and **6** also caused inhibition of cell viability (approx. 64%) on Hela cells at the highest concentration. Our results suggested that **1**, **4** and **6** might be promising for developing anticancer agents for breast and cervix cancer and they have potential for further studies.

Acknowledgements: This study was supported by Gazi University Research Foundation [grant number 02/2017-03].

Keywords: Antiproliferative effect, *Chrysophthalmum montanum*, guaianolide, MTT assay.

References:

[1] Arasan S, Kaya I. J Food Nutr Res 2015; 3(5): 337–340.
[2] Ayaz F *et al*. Turk J Biochem 2016; 41 (S4).

TOPIC A: Chemistty of Natural Products

PS1-A-001
Activated and non-activated charcoal derived from durian (*Durio zibethinus murr*.) peelings as treatment to improved physico-chemical and microbiological parameters of Davao river
Venchie Badong, Maria Angelique Anne Alapag, Mark Anthony Casimina

PS1-A-002
Phytochemical screening and antioxidant activity of methanolic extract of Argan leaves (*Argania spinosa*) from north Algeria
Mahmoud Belalia, Louiza Belkacemi

PS1-A-005
Chemical analysis and metal chelating power of extracts from three medicinal plants: *Peganum Harmala* L., *Cinnamomum Zeylanicum* and *Rosmarinus Officinalis*
Souad Djellali, Rachid Saharaoui, Ibtissem Debab

PS1-A-006
Liquid chromatography combined with time-of-flight high resolution mass spectrometry (LC/Q-TOF/HRMS) for the chemical characterization of *Smilax aspera* L. leaves
Eleni Kakouri, Charalabos Kanakis, Panayiotis Trigas, Petros Tarantilis

PS1-A-007
New insular red propolis from Colombia: botanical origin, biological and chemical markers
Guillermo Salamanca Grosso, Mónica Patricia Osorio Tangarife

PS1-A-008
Mechanistically degenerate kinetic resolution in [5+2] cycloisomerization
Kai Ma

PS1-A-009
Phytochemical diversity of *Salvia* species in Hengduan Mountains
Ayumi Ohsaki, Hiroshi Kawabe, Kana Kuwada, Mariko Funasaki, Hiroshi Hirota, Haruka Tsukada, Teppei Komiyama, Hiroyuki Kagechika, Yasuko Okamoto, Motoo Tori, Yoshinori Saito, Gong Xun

PS1-A-010
Search for new compounds from *Portulaca pilosa*
Minori Nonaka, Emi Hara, Ayumi Seike, Yasuko Okamoto, Miwa Kubo, Mariko Funasaki, Yoshiyasu Fukuyama, Pilosa Keiichi Matsuzaki, Hiroshi Hirota, Ayumi Ohsaki

PS1-A-011
Oxidation of lignan structures
Patrik Runeberg

PS1-A-012
Isolation of gallic acid and flavonoids from *Terminalia brownii* leaves
Intisar Elzein, Mahgoub El Tohami, Aisha Almagboul

PS1-A-013
Reaction products formed with food constituents and norlignans dissolving from sugi barrel for sake
Fumio Kawamura

PS1-A-014
Saffron aqueous extract: phytochemistry & determination of crocetin's serum and tissue pharmacokinetics after oral and intravenous administration to C57/BL6J mice
Maria-Eleni Grafakou, Eirini Christodoulou, Helen Skaltsa, Nikolaos Kadoglou, Nikolaos Kostomitsopoulos, Georgia Valsami

PS1-A-015
Chemical variation in essential oils from the oleo-gum resin of *Boswellia carteri*
William Setzer, Anjanette DeCarlo, Stephen Johnson, Ambika Poudel, Prabodh Satyal, Loren Bangerter

PS1-A-016
Sesquiterpenoid skeleton type drimane produced by the bioluminescent mushroom *Neonothopanus gardneri*
Teresinha De Jesus Aguiar Dos S. Andrade, Maria das Dores A. Oliveira, Joaquim Soares C. Junior, Nerilson Marques Lima, Jioji N. Tabudravu, Dulce Helena S. Silva, Antonia Maria das Graças Lopes Citó

PS1-A-017
Chitosan/carboxymethyl cellulose sulfuric acid hydrogels and their nanocomposits: Preparation and application in tartrazine removal
Ali Reza Karimi

PS1-A-018
4-oxo-*β*-apo-13-carotenone from the cyanobacterium *Anabaena cylindrica* PCC 7122
Teresa Martins, Mariana Reis, Pedro Leão

PS1-A-019
Characterization of sulfhydryl conjugates with betanidin and gomphrenin quinoids by means of HPLC-DAD-ESI-MS/MS and LC-MS-IT-TOF
Agnieszka Kumorkiewicz, Sławomir Wybraniec

PS1-A-020
Secondary metabolites from *Sideritis germanicopolitana*
Hasan Kırmızıbekmez, Murat Erdoğan, Umur Erdem[1], Norbert Kúsz, Fatih Demirci, Judit Hohmann

PS1-A-021
Essential oil content of *Sideritis* spp. cultivated in Northern Greece
Andreas Douros, Helen Skaltsa, Catherine Grigoriadou, Eleni Maloupa

PS1-A-022
Significant secondary metabolites from *Crepis commutata* (Spreng.) Greuter
Ioanna Kotti, Christina Barda, Helen Skaltsa

PS1-A-023
Chemical profile of wild *Crithmum maritimum* L.
Vasiliki Zafeiropoulou, Aikaterini-Michaela Tomou, Helen Skaltsa

PS1-A-024
Essential oil content of cultivated *Crithmum maritimum* L.
Vasiliki Zafeiropoulou, Helen Skaltsa

PS1-A-025
Isolation of bioactive compounds from *Sideritis* ssp.
Ekaterina-Michaela Tomou, Helen Skaltsa, Paschalina Chatzopoulou

PS1-A-026
Kinetic and chromatographic studies on non-enzymatic oxidation of gomphrenin pigment from the fruit juice of *Basella alba* L.
Agnieszka Kumorkiewicz, Tomasz Świergosz, Sławomir Wybraniec

PS1-A-027
Preliminary study: Secondary metabolites of *Melissa officinalis* L.
Dimitrios Dimas, Ekaterina-Michaela Tomou, Helen Skaltsa

PS1-A-028
Seasonal variation of the esential oil of *Rosmarinus officinalis* L. (Lamiaceae), growing wild in the island of Cephalonia (Greece)
Aristeidis Pritsas, Gerasimia Tsasi, Theophilos Mailis, Yannis Samaras, Helen Skaltsa

PS1-A-029
Antioxidant capacity of fractions obtained from wild *Ugni molinae*, Turcz. leaves
Rodrigo Pérez, Carla Delporte

PS1-A-030
Crystalline sponge method efficiently and exhaustively reveals stereo-configurations of polycyclic compounds derived from beer's bitter acids
Yoshimasa Taniguchi, Takashi Kikuchi, Makoto Fujita

PS1-A-031
Isolation and identification of components from hybrids *Phlomis* × *commixta* Rech. f. (*P. cretica* × *P. lanata*)
Michalis Stefanakis, Diamando Lazari, Haralambos E. Katerinopoulos

PS1-A-032
Phytochemical investigation of *Achillea coarctata* Poir. (Asteraceae)
Konstantina Papakosta, Christina Barda, Helen Skaltsa

PS1-A-033
Comparative Chemical analysis of seven endemic Greek *Citrus* hybrids
Eleni Anastasopoulou, Konstantia Graikou, Anastasios Makritis, Ioanna Chinou

PS1-A-034
Phytochemical study on Greek endemic *Rindera graeca* aerial parts. Antioxidant Activity
Christos Ganos, Tomasz Mroczek, Nektarios Aligiannis, Ioanna Chinou, Konstantia Graikou

PS1-A-035
Impact of the soil substrate on essential oil composition of *Origanum vulgare*
Amalia Katsifara, Eleni Sazakli, Soterios Varnavas, Michalis Leotsinidis

PS1-A-036
Effects of different cooking techniques on vitamin levels of green leaf vegetables; A HPLC method development study and an anti-oxidative capacity study
Tuğba İduğ, Ümit Can Erim, Hilal Hızlı, Ali Şen, Barkın Berk

PS1-A-037
Selective copper(II) catalyzed amination of aryl-himachalene to secondary benzylamines derivatives in water via chloromethylation
Intissar Ait Lahcen

PS1-A-038
Chemical Constituents of *Galium asparagifolium* Boiss. et Heldr
Umit Toktas, Nazli Boke Sarikahya, Husniye Kayalar

PS1-A-039
Phytochemical investigation and antimicrobial activity of isolated sesquiterpene lactones from *Crepis incana* Sm.
Christina Barda, Ana Ciric, Marina Sokovic, Michael Tsoukalas, Helen Skaltsa

PS1-A-041
Novel anti-inflammatory steroidal compounds
Jesus Sandoval-Ramirez, Reyna Zeferino-Díaz, María A Fernández-Herrera, Nuvia Kantún-Moreno, Leticia Olivera-Castillo

PS1-A-042
Synthesis of novel 22-oxocholestane glycosides with potential anticancer activity
Jazmin Ciciolil Hilario-Martinez, Zhendong Jin, María Antonieta Fernández-Herrera, Jesús Sandoval-Ramírez

PS1-A-043
Chemical composition of the essential oil of leaves and roots of *Stevia serrata* Cav. from Guatemala
Juan Francisco Perez Sabino, Manuel Alejandro Muñoz Wug, Max Samuel Mérida Reyes, Bessie Evelyn Oliva Hernández, Edwin Adolfo Taracena Monzón, José Vicente Martínez Arévalo, Antonio Jorge Ribeiro da Silva

PS1-A-044
Response of dry yield and saponin content of some saponin containing plants to planting media and drying methods
Salah Eldeen M Mahmoud, Halla M Gamal El-Dein, S M Abou-Shleel, Marwa M Darweesh

PS1-A-045
Antiplasmodial and antitryponosomal activities of extracts, fractions and compounds from *Beilschmiedia* spp
Waleguele Christine Claire, Lenta Ndjakou Bruno, Tsamo Etienne Voltaire, Rui Krause

PS1-A-046
New phenylethanoid glycosides from *Cistanche phelypaea* and their activity as inhibitors of monoacylglycerol lipase
Khadidja Aya Beladjila, Djemaa Berrehal Berrehal, Assia Khalfallah, Nunziatina De Tommasi, Carlotta Granchi, Marinella De Leo, Giulia Bononi, Alessandra Braca, Zahia Kabouche

PS1-A-047
Phytochemical screening of *Drimia numidica* bulbs and rhizomes
Eleni Kakouri, Charalabos Kanakis, Panayiotis Trigas, Petros Tarantilis

PS1-A-048
Quantification of grape seed oils' FAMEs of varieties traditionally cultivated in the Ionian islands
Nefeli Sofia Sotiropoulou, Ioanna Oikonomou, Dimitra Daferera, Efstathia Skotti, Christos Pappas, Petros Tarantilis

PS1-A-049
Chemical constituents isolated from the leaves and the flowers of *Achillea grandifolia* Friv. (Asteraceae)
Olga Tsiftsoglou, Christos Gounaris, Christina Papitsa, Maria Nanouli, Diamanto Lazari

PS1-A-050
Azomethine ylide cycloaddition: A versatile way of semi–synthetic modification towards novel spirooxindolo analogues of piperine
Meenakshi Singh

PS1-A-051
Gomphrenins stability during tea brewing of purple flowers of *Gomphrena globosa* L.
Natalia Szmyr, Sławomir Wybraniec

PS1-A-052
Acylated gomphrenins and their stability in *Gomphrena globosa L.* extracts
Natalia Szmyr, Sławomir Wybraniec

PS1-A-053
Synthesis of a regioisomeric *N*–methyl aspidostomide D, its derivatives via Lewis acid mediated epoxide opening and their antibacterial assessment
Mulla Althafh Hussain, Althafh Hussain Mulla, Amhed Khan

PS1-A-054
Structural correction of a dimeric phthalide with progesterone–like activity
José Luis Ávila, Dr. Guillermo Delgado

PS1-A-055
Qualitative composition of volatile constituents in three wild grown species of aromatic plants commercially known as oregano
Paraskevi Yfanti, Eleni Leneti, George Patakioutas, Dimitra Douma, Marilena Lekka

PS1-A-056
Isolation, identification and investigation of the antioxidant activities of compound from *Costus afer*
Victor Fadipe

PS1-A-057
Revealing an unprecedented biosynthetic pathway in *Aspergillus oryzae*
Naoya Maeda, Dr. Yuta Tsunematsu, Dr. Kenji Watanabe

PS1-A-058
Biochemistry analysis and physicochemical properties for the quality assessment of the Greek honey bee
Dimitrios Kafetzopoulos[1], Chrissi Petraki, Georgios Kafetzopoulos

PS1-A-059
Pharmaceutical formulations of *Capsicum frutescens* extracts
Enkelejda Goci, Entela Haloci

PS1-A-060
Indole alkaloids from medicinal plants of Cameroon with cancer chemopreventive activity
Joseph Thierry Ndongo, Josephine Ngo Mbing, Muriel Cuendet, Dieudonné Emmanuel Pegnyemb, Hartmut Laatsch

PS1-A-061
Efficient removal of cationic dyes from aqueous medium using a recyclable activated carbon from Phoenix fruit pits: A study of adsorption parameters and thermodynamics
Abdullah Aldawsari

PS1-A-062
Skadar lake *Nymphaea alba* and *Nuphar luteum* leaves chemical profile
Nina Djapic

PS1-A-063
Study on the synthesis of vinpocetine
Kai Qiao, Dong Zhang[1], Y Wu X, Hong Qin, Zheng Fang, Kai Guo

PS1-A-064
Surface and aggregation properties of Nα-lysine based surfactants
Ting Shi T, Zheng Fang, Kai Guo

PS1-A-065
Diterpenes from *Zhumeria majdae* Rech.f. & Wendelbo roots as potential HSP90 interactors
Student Reza Zadali, Abbas Hadjiakhoondi, Massimiliano D'ambola, Matthias Hamburger, Nunziatina De Tommasi, Samad Nejad Ebrahimi

PS1-A-066
Antioxidant capacity *Rubus glaucus* and soil, physico-chemical, nutritional and Enzymatic relationship against three chemical inhibitors
Jairo Granados, Dolffi Rodríguez

PS1-A-067
Synthetic routes to royal jelly constituents 10-hydroxy-2-decenoic acid and 3-hydroxy-decanoic acid
Rodalia Babaiti, Adelajda Shahu, Maria Theodoropoulou, George Koutoulogenis, Maroula Kokotou, George Kokotos

PS1-A-068
Synthetic routes to 3-hydroxy saturated fatty acids
Adelajda Shahu, Maria Thedoropoulou, Asimina Bourboula, George Kokotos

PS1-A-069
Asymmetric synthesis of 7-hydroxy saturated fatty acids
Maria Theodoropoulou, Adelajda Shahu, Olga Mountanea, George Kokotos

PS1-A-070
Optimization and preparation of methylcellulose edible film combined with *Ferulago angulata* essential oil (FEO) nanocapsules for food packaging applications
Akbar Esmaeili

PS1-A-071
Antioxidant activity of polyphenolic extracts from carob pods and their effect on acrylamide formation in the asparagine/fructose Maillard reaction system
Iliana Tsenkova, Anna Marina Grigoriou, Eftychia Pinakoulaki

PS1-A-072
Effect of pure polyphenols and polyphenolic extracts from carobs to acrylamide formation in the asparagine/glucose Maillard reaction system
Charia Hadjipakkou, Eftychia Pinakoulaki

PS1-A-073
Triterpene saponins content in Swiss chard (*Beta vulgaris* L.)
Agnieszka Mroczek, Urszula Klimczak, Anna Stochmal, Mariusz Kowalczyk

PS1-A-074
Phytochemical investigation of fruits from *Paliurus spina-christi* Mill
Vaios Amountzias, Dionysios Abatis, Nektarios Aligiannis

PS1-A-075
Further constituents of the stem bark of *Acacia auriculiformis*
Augustine Ahmadu, Nikolaos Tsafantakis, Nikolas Fokialakis

PS1-A-076
Fast Centrifugal Partition Chromatography – gustatometry as a valuable tool for the identification of new natural sweet compounds from wood chips used in wine ageing
Eirini Koui, Antigoni Cheilari, Evanthia Dina, Dennis Abatis, Michalis Stefanakis, Yorgos Kotseridis, Nektarios Aligiannis

PS1-A-077
Polyphenols from *Thymus thracicus* Velen
T. Papagrigoriou, O. Tsiftsoglou, D. Lazari

PS1-A-078
Synthesis of Blue Dye from Loganin, its Application in Dyeing, Ultraviolet Protection of Proteinious Fabric and Docking studies
Sapna Patil

PS1-A-079
Exploitation of aromatic plants' by-products for the development of bioactive extracts with antioxidant and antimicrobial properties
Evanthia Ntina, Christina Bakali, Dionisis Abatis, Konstantina Stathopoulou, Eleutheria Arapi, Spyros Economou, Hristo Iliev, Ilias Smyrnioudis, Nikolaos Fokialakis, Nektarios Aligiannis

PS1-A-080
Study of the basidiomycete of *Ganoderma adspersum* (Ganodermataceae)
Raichan Chafouz, Olga Tsiftsoglou, Dimitra Hadjipavlou-Litina, Pavle Maskovic, Diamanto Lazari

PS1-A-081
Determination of biophenols in virgin olive oils by liquid chromatography supporting the health claim by European Food Safety Authority (EFSA)
Anna Stefanitsi, Vasilios Sakkas, Panagiotis Stathopoulos, Maria Halabalaki, A-L Skaltsounis

PS1-A-082
Investigation of bioactive secondary metabolites from *Alkanna* species of Greece
Evangelia Tsiokanou, Nikolaos Tsafantakis, Eirini Baira, Eleftherios Kalpoutzakis, Andreana Assimopoulou, Nektarios Aligiannis, Nikolas Fokialakis

PS1-A-083
New diterpenes from two populations of the brown alga *Dictyota linearis* from the Aegean Sea
Aikaterini Koutsaviti, Charoula Machaira, Christina Keramea, Vassilios Roussis, Efstathia Ioannou

PS1-A-084
Development of an inhalant based on African ginger (*Siphonochilus aethiopicus*) for the management and/or treatment of coughs, colds, flu and as a decongest
Leylene Kruger, Vinesh Maharaj, Thomas Klimkait

PS1-A-085
LC/MS guided isolation of protopanaxadiol ginsenosides having 5~6 sugars from *Panax vietnamensis*
Van Le, Long Vu, Khoi Nguyen[1], Hill Park, Duc Nguyen

PS1-A-086
Sesquiterpene lactones from the Greek endemic *Inula subfloccosa* Rech. f. (Asteraceae)
Dafni Ioanna Diakaki, Aikaterini Koutsaviti, Efstathia Ioannou, Olga Tzakou

PS1-A-087
Synthesis of the natural product (+) Civet and its enantiomer (–) Civet
Fátima Garrido

PS1-A-088
Qualitative analysis of Herbofix® herbal extracts and comparison with extracts prepared by infusion using HPTLC, HPLC-DAD and UPLC-MS
Ioulia K. Tseti, Efstathia Thoma, Aikaterini Argyropoulou and Alexios-Leandros Skaltsounis

PS1-A-089
Alkaloids from grains of Peganum harmala from Azerbaijan flora
E.A.Garayev, I.S.Movsumov, T.A.Nasibova, G. Herbette, E.E.Garayev

Topic C: Medicinal chemistry and synthesis

PS1-C-001
Caffeic acid-derived biopolymers of medicinal plants, synthesis of its monomer, methylated analogue and their comparative anticancer efficacy
Vakhtang Barbakadze

PS1-C-002
Pharmacomodulation of ellagic acid, a promising antiplasmodial agent, to improve its bioavailability
Gilles Degotte, Aurore Hans, Olivia Jansen, Allison Ledoux, Pauline Desdemoustier, Bernard Pirotte, Pierre Francotte, Michel Frederich

PS1-C-003
Preparation of chiral phenylethanols using various vegetables grown in Algeria
Louisa Aribi-Zouioueche, Manhel Bennamane, Samra Razi, Saoussen Zeror

PS1-C-004
Invasive weed lantadene derivatives: experimental and computational studies
Monika Chauhan, Manu Sharma, Sharad Kumar Suthar, Hong Boo Lee, Richa Dhingra, Neelima Dhingra

PS1-C-005
Studies on the total synthesis of Palhinine A
Shu Xu, Liang Li, Xiaolei Wang, Yaling Gong, Shichao Lu

PS1-C-006
Studies on the total synthesis of Mollanol A
Yixuan Zheng, Jianzhuang Miao, Mei Li, Yuyan Liang, Linna Wang, Shu Xu

PS1-C-007
Approach to C–20 epimers of vitamin D analogues
Hugo Santalla**,** Fátima Garrido

PS1-C-008
Semisynthetic approaches to dicadalenol and some analogues
José Luis Ávila, Guillermo Delgado

PS1-C-009

Discovery of new 5-arylcarboximidamidopyrazolo[3,4-c]pyridines with potent antiproliferative activity

Panagiotis Marakos, Athanassios Papastathopoulos, Nikolaos Lougiakis, Nicole Pouli, Harris Pratsinis, Dimitris Kletsas

PS1-C-010

Synthesis of novel multisubstituted pyrazolo[3,4-c]pyridines and evaluation of their antiproliferative and antiangiogenic activity

Nikolaos Lougiakis, Stavros Kampoukos, Vassili Konstantin, Nicole Pouli, Panagiotis Marakos, Heleni Loutrari

PS1-C-011

Adventures in the synthesis and biological evaluation of novel "reactive" colchicine-derived compounds

Andreas Stein, Persefoni Thomopoulou, Tim Schulte, Benoit Gigant, Aram Prokop, Hans-Günther Schmalz

PS1-C-012

Castor oil as feedstock for the production of 12-Fatty Acid esters of Hydroxy Fatty Acids (12-FAHFAs)

George Koutoulogenis, Valentini Avramidi, Evrydiki Katsikari, George Kokotos

PS1-C-013

Design, synthesis and pharmacological evaluation of novel TDP1 inhibitors

Maria Karelou[1], Keli Agama, M.D., Yves Pommier, Ioannis Kostakis

PS1-C-014

Hydroxytyrosol, a versatile natural compound as chemical probe in drug design

Aikaterini Komianou, Ioanna Kalpaktsi, Alexios-Leandros Skaltsounis, Ioannis Kostakis

PS1-C-015

Design and Synthesis of new Modified hybrids of bisindole Derivatives as Chemical tools in The study of Anticancer Activity

Amalia Dimitrios Kalampaliki, Eleftheria Athanasios Georgiou, Ioannis Konstantinos Kostakis

PS1-C-016

Design and synthesis of novel staurosporine and rutaecarpine hybrids as therapeutic leads in the treatment of cancer

Amalia Dimitrios Kalampaliki, Ioannis Konstantinos Kostakis

PS1-C-017

Spiro-oxazolidinone C-nor-D-homo steroids as potential inhibitors of hedgehog pathway

Roumana Aggeliki, Dimanthi Pliatsika, Sotiris Nikolaropoulos, Athanassios Giannis, Manolis Fousteris

PS1-C-018
Design and synthesis of stilebenoid and chalconoid analogues as potent tyrosinase modulators
Argyro Vontzalidou, S.M, Dimitrakoudi, K Tsoukalas, E Chaita, G Zoidis, C Cheimonidou, I.P Trougakos, G Lambrinidis, E Mikros, A.L Skaltsounis, N Aligiannis

PS1-C-019
Further exploration of the indirubin scaffold affords 5'-6 disubstituted analogues with potent cytotoxicity and *in vivo* efficacy targeting Src family kinases
Nicolas Gaboriaud-Kolar, Marina Kritsanida, Vassilios Myrianthopoulos, Nikolaos Lougiakis, Anastasia Papachristodoulou, Anna Tsantili, Ioannis Rerras, Laurence Wennogle, Emmanuel Mikros, Sangil Nam, Alexios-Leandros Skaltsounis

PS1-C-020
Design and synthesis of new substituted nucleosides as potential anti-HCMV agents
Nicole Pouli, Maria Gerasi, Georgios Papadakis, Nikolaos Lougiakis, Panagiotis Marakos

PS1-C-021
An improved synthetic route to Abyssomicin C
Veroniki P. Vidali, Aggelos Petroulias, Aleksander Canko, Emmanuel A. Bouzas and Elias A. Couladouros

Topic E Natural compounds from marine organisms

PS1-E-001
LC-PDA-ESI-MS^n metabolite fingerprinting and anticancer screening of an endophyte from marine algae *Dichotomaria marginata*
Teresinha De Jesus Aguiar Dos S. Andrade

PS1-E-002
Natural carotenoids from a sustainable source: a comparative study of five microalgae species
Ginevra Lombardi-Boccia, Gabriella Di Lena, Irene Casini, Massimo Lucarini

PS1-E-003
Biotechnological syntheses of maritime high-value diterpene type natural products
Markus Reinbold, Marion Ringel, Daniel Garbe, Thomas Brück

PS1-E-004
Structurally diverse secondary metabolites from a marine fungus isolated from the Atlantic Ocean
Mallique Qader, Larry Mweetwa, Manal Eshelli, Lalith Jayasinghe, Marcel Jaspars, Mostafa E. Rateb

PS1-E-005
Isolation and structure determination of two new peptides from marine cyanobacteria
Keitaro Iwasaki, Arihiro Iwasaki, Simpei Sumimoto, Kiyotake Suenaga

PS1-E-006

A marine lipopeptide, minnamide A: Synthetic studies for assignment of the C-9 methyl stereochemistry

Masayuki Kobayashi, Shinpei Sumimoto, Seiichi Sinomiya, Toshiaki Teruya, Arihiro Iwasaki, Kiyotake Suenaga

PS1-E-007

Kakeromamide A, a new cyclic pentapeptide from the marine cyanobacterium *Moorea bouillonii*

Fumiaki Nakamura, Hiroshi Maejima, Midori Kawamura, Daisuke Arai, Meng Zhao, Tao Ye, Nobuhiro Fusetani, Yoichi Nakao

PS1-E-008

Isolation and sstructure determination of four new compounds from marine cyanobacteria

Ikuma Shiota, Arihiro Iwasaki, Shimpei Sumimoto, Hiroshi Tomoda, Kiyotake Suenaga

PS1-E-009

Deep-sea marine life molecules: assessing bioactivity potential of deep-sea microorganisms

Adriana Rego, Eduarda Almeida, Mariana Girão, Inês Ribeiro, Tiago Ribeiro, Ralph Urbatzka, Maria de Fátima Carvalho, Pedro Leão

PS1-E-010

Structures and cytotoxic activities of new macrolides from the marine dinoflagellates *Amphidinium* species

Masashi Tsuda, Keiko Kumagai, Masayuki Tsuda

PS1-E-011

Genome mining for novel bioactive compounds from the marine cyanobacterium *Nodosilinea nodulosa* LEGE 06152

Kathleen Abt, Pedro Leão

PS1-E-012

Skin protective effects of marine invertebrates and symbionts from the mesophotic zone

Sofia Letsiou, Konstantinos Gardikis, Eleni Spanidi

PS1-E-013

Preparation of chemical probes from marine cyclic peptides

Rie Kamihira, Jun K. Yamashita, Yoichi Nakao

PS1-E-014

Isolation, separation and structure elucidation of natural products from marine endophytes with potential activity towards bacterial infections and cancer cell lines

Gaia Burgio, Mostafa E. Rateb

PS1-E-015

How the culture conditions affect the production of a potent anticancer metabolite by *Nostoc*

Alexandros Polyzois, Quoc Dang Thai, Elie-Bertrand Duran, Sylvie Michel

PS1-E-016

Chemical profile of indole alkaloids from the nudibranch *Tambja stegosauriformis*

Douglas Siqueira Almeida Chaves, Rosiane Silveira, Marcin Ozarowski, Piotr Kachlicki, Hoffgan Félix, Raquel Rennó Braga

PS1-E-017

Bioactive 3,7–cyclized cembranes and cembranes from the soft coral *Klyxum flaccidum*

Chiung–Yao Huang Huang, Atallah F. Ahmed, s. Wan–Ru Tseng, Yi–Ying Tsai, Tsong–Long Hwang, Jyh–Horng Sheu

PS1-E-018

Metabolites from a bioactive organic fraction of an unidentified Red Sea sponge

Atallah F. Ahmed, Chi–Jen Tai, Raha S. Orfali, Walied M. Alarif, Jyh–Horng Sheu

PS1-E-019

Extraction of chlorophylls and phycobiliproteins from Indonesian red seaweed

Hakiki Melanie**,** Ambo Tuwo, Marcos A. Neves, Mitsutoshi Nakajima

PS1-E-020

Antithrombotic and anticoagulant activity of Brazilian and Antarctic brown algae

Ana Cláudia Philippus**,** Gabriele Andressa Zatelli, Stephanie Milis Syracuse, Nathalia da Silva Moura, Bianca Regina Alberton, Rubia Karine de Souza, Aline Paternostro Martins, Paulo Antunes Horta, Pio Colepicolo Neto, Ana Carolina Rabello de Moraes, Miriam de Barcellos Falkenberg

PS1-E-021

Agaclathratosides A and B, two new monogalactosylacyl glycerols from the brown alga *Agarum clathratum* subsp. *yakishiriense*

Yukyoung Jeon, Hwan Su Yoon, Jong Hwan Kwak

PS1-E-022

A new and seven known phlorotannins from *Eisenia bicyclis* and their anti-diabetic activity in Zebrafish model

Eunbin Kim, Yukyoung Jeon, Youn Hee Nam, Tong Ho Kang, Jong Hwan Kwak

PS1-E-023

6-Bromoindole derivatives from the marine sponge *Geodia barretti*: isolation and anti-inflammatory activity

Xiaxia Di, Caroline Rouger, Ingibjorg Hardardottir, Jona Freysdottir, Tadeusz F. Molinski, Deniz Tasdemir, Sesselja Omarsdottir

PS1-E-024

AlgaeCeuticals: Development of microalgae-based natural carotenoids as cosmeceuticals and nutraceuticals

Stefan Martens, Panagiotis Madesis, Nikolaos Labrou, Leonardo Ceracino, Nico Salmaso, Luisa Palmieri, Silvia Fluch, Pablo F. Ruiz, Ángel M. San Martin, Jose L. Mullor, Natalie Vagioni, Marina Chatzikonstantinou

PS1-E-025
Chemical profiles of *Hypoxylon monticulosum* fungal strains isolated from marine red algae *Acanthophora spicifera* and *Dichotomaria marginata*
Dulce Silva, Alana Honorio, Beatriz Barcellos, Rebeca Medina, Iata Mendonca, Victor Rufino, Marcos Soares

PS1-E-026
2,5-Diketopiperazines from marine-derived bacteria isolated from marine sediments collected in the Eastern Mediterranean Sea
Maria Harizani, Panagiota Georgantea, Eleni Katsini, Vassilios Roussis, Efstathia Ioannou

Topic F: Metabolomics and phytochemical analysis

PS1-F-001
Comparative analysis of antidiabetic, anti-obesity and antioxidant activities of leaves and flowers of *Ocimum basilicum* and identification of chemical constituents in their essential oils and extracts
Dildar Ahmed, Zoy I Noor, Muhammad Tariq Qamar

PS1-F-002
Identification of antioxidant and hypoglycemic compounds in aqueous-methanol fraction of methanolic extract of *Ocimum canum* leaves
CJ Ononamadu, MS Sule, AJ Alhassan, GO Ihegboro, TA Owolarafe, FC Nwachukwu

PS1-F-003
Automated metabolomic analysis of LC-MS data
Jiri Gruz**,** Andrea Luterova, Miroslav Strnad

PS1-F-004
Metabolism of isotopically labeled benzoic and cinnamic acids in oat
Klára Supíková, Andrea Luterová, Miroslav Strnad, Jiří Grúz

PS1-F-005
Probing lipid catabolism in cyanobacteria
Sandra Figueiredo, Teresa Martins, Kathleen Abt, Pedro Leão

PS1-F-006
Capsicum-derived biomass quantum dots coupled with alizarin red S as an inner-filter-mediated illuminant nanoprobe for imaging of intracellular calcium ion dynamics
Shu-lin Zhao

PS1-F-007
Lipid and polyphenol composition in *Vaccinium* wild berries and cultivars
Ilona Vanaga, Egija Kitija Meijere, Linards Klavins, Maris Klavins, Anete Rateniece, Zane Greiza, Ugis Kletnieks

PS1-F-008
Quality control of cultivated greek thyme (*Thymus vulgaris*)
Anastasia Karioti, Anna Rita Bilia

PS1-F-009

Targeted & untargeted profiling of Muscat of Alexandria grapes

Maria Marinaki, Miss Konstantina Liva, Christina Virgiliou, Anastasia Ketsetzi, Helen Gika, Georgios Theodoridis, Dimitrios Christofilos, Andreana Assimopoulou

PS1-F-010

HPLC–DAD–MS guided investigations on the Greek cultivated medicinal plants *Origanum dictamnus* and *Thymus vulgaris* (Lamiaceae) reveal new natural products

Anastasia Karioti, Sofia Govari, Charikleia Paloukopoulou, Ilias Stefanis, Athina Soulioti

PS1-F-011

Simultaneous analysis of flavonoids and artemisinin with its analogues in *Artemisia annua* and real–time monitoring of its interaction with Bcl–2 with in–cell NMR spectroscopy

Ioannis Gerothanassis, Vassiliki Kontogianni, Alexandra Primikyri, Marianna Sakka

PS1-F-012

Metabolomic analysis of micromolecular diversity from Caatinga using LC–ESI–MS/MS

Danielle Rocha Pinho, Alan Cesar Pilon, Norberto Peporine Lopes

PS1-F-013

Dereplication by ^{13}C NMR in the presence of high boiling point solvents

Marine Canton, Stéphane Poigny, Richard Roe, Jean–Hugues Renault, Jean–Marc Nuzillard

PS1-F-014

Targeted and untargeted UHPLC-HRMS-based metabolomics of Boraginaceae roots

Nebojša Rodić, Angeliki Vlachou, Euaggelia Michailidou, Evangelos Tzimpilis, Helen Gika, George Theodoridis, Vassilios Papageorgiou, Andreana Assimopoulou

PS1-F-015

Interaction between blue light and ABA during tomato seed germination

Pavla Šimurová (Pokorná), Veronika Turečková, Miroslav Strnad, Martin Fellner

PS1-F-016

Aqueous ethanolic extracts from arid halophyte species *Arthrocnemum macrostachyum* and *Tetraena qatarensis*

Samar Al-Jitan, Saeed Ahmed Alkhoori, Michael Ochsenkühn, Shady A. Amin, Lina F. Yousef

PS1-F-017

An LC/HRMS method for the determination of naturally occurring saturated hydroxy fatty acids

Maroula Kokotou, Christiana Mantzourani, George Kokotos

PS1-F-018

Study of glucosinolate-hydrolysis products in broccoli and brussels sprouts by gas chromatography-mass spectrometry and liquid chromatography-mass spectrometry

Irene Mesimeri, Maroula G. Kokotou, Violetta Constantinou-Kokotou

PS1-F-019

Improving the definition of maca products' quality using NMR, HPTLC and HPLC

Francesca Scotti, Pinelopi Nika, Raman Suthar, Michael Heinrich

PS1-F-020

High resolution mass spectrometry studies of sulforaphane and indole-3-carbinol in broccoli

Maroula G. Kokotou, Panagiota-Kyriaki Revelou, Christos Pappas, Violetta Constantinou-Kokotou

PS1-F-021

Variability of phytohormones in Brassicaceae determined by high resolution mass spectrometry

Panagiota-Kyriaki Revelou, Maroula G. Kokotou, Violetta Constantinou-Kokotou

PS1-F-022

New strategies to strigolactone determination in complex sample matrices

Kristyna Flokova, Mahdere Z. Shimels, Beatriz Andreo Jimenez, Yanting Wang, Harro J Bouwmeester

PS1-F-023

Custom-made algorithm: a powerful tool for ^{13}C-NMR dereplication of complex mixtures. Proof of concept on a *Garcinia mangostana* fruit peel extract

Antoine Bruguiere, Joel Dietsch, Derbre Severine, Sorphon Suor-Cherer, Dimitri Breard, Pascal Richomme

PS1-F-024

Targeted isolation of *trans*-crocin 4 (TC4) from *Crocus sativus* L. employing step gradient Countercurrent Chromatography (CPC), followed by a metabolome study in mice after i.p. administration of TC4

Evangelia Karkoula, Apostolis Angelis, Ioanna Dagla, Nikolaos-Stavros Koulakiotis, Evagelos Gikas, Nikolaos Kokras, Christina Dalla, Maria Halabalaki, Alexios-Leandros Skaltsounis, Anthony Tsarbopoulos

PS1-F-025

Untargeted metabolomics study reveals Greek propolis novel anti-tyrosinase agents using ultrahigh-performance liquid chromatography-hybrid quadrupole-orbitrap mass spectrometry

Aikaterini Termentzi, Maria-Ioanna Stavropoulou, Konstantina-Georgia Stathopoulou, Kyriaki Machera, Evagelos Gikas, Nektarios Aligiannis

PS1-F-026

A comparative and integrated study for the extraction and determination of phenolic compounds in olive fruits and stones

Georgia Sarikaki, Konstantina Touvleliou, Maria Halabalaki, Sofia Mitakou, Leandros A Skaltsounis

PS1-F-027

Assessment of the effect of extraction solvent on cannabinoid yield and phytochemical profile of fibre-type Cannabis sativa L. using UPLC-PDA and HPTLC

Petros S. Tzimas, Eleftherios A. Petrakis, Apostolis Angelis, Maria Halabalaki, Leandros A. Skaltsounis

PS1-F-029
Development of a GeLC-MS/MS based strategy for the identification of novel drug or diagnostic targets against Breast and prostate cancer
Ioanna-Maria Orfanou, Theodoros Karampelas, Manousos Makridakis, George Mermelekas, Konstantinos Vougas and Constantin Tamvakopoulos

PS1-F-030
An optimized analytical methodology for the determination of olive oil biophenols, based on IOC recommended method
E. Bata, P. Stathopoulos, A. Rodi, M. Halabalaki, A-L Skaltsounis

PS1-F-031
Development and validation of an HPTLC densitometric method for the rapid quantification of bioactive lignans in sesame seeds: comparison with HPLC-PDA
Eleni V. Mikropoulou, Eleftherios A. Petrakis, Aikaterini Argyropoulou, Sofia Mitakou, Maria Halabalaki and Leandros A. Skaltsounis

PS1-F-032
Essential oil composition of *Juniperus oxycedrus* ssp. *macrocarpa* from Greece
Nikolaos Armenis, Maria Couladis

PS1-F-033
Validation and implementation of an HPLC-DAD method for the simultaneous quantification of chlorogenic acid, flavonoids and flavonolignans in *Cecropia* species
Andrés Rivera-Mondragón, Sebastiaan Bijttebier, Tania Naessens, Deborah Custers, Catherina Caballero-George, Sandra Apers, Luc Pieters, Kenn Foubert

PS1-F-034
Cannabis-related Bioanalysis Applications Using a Compact Mass Spectrometer (CMS)
Ben Nie, Jack D Henion, George Maylin

Topic G: Authenticity, traceability, safety of natural products

PS1-G-001
Gasca D as potent diabetic weight and blood glucose reduction formulation
Salihu Ismail Ibrahim, Gezawa Ibrahim, Fakhradden Yahya M, Chedi Basir, Nafisatu Kabir, A A Imam

PS1-G-002
Effective HPLC methods for the detection of *Sophora japonica* adulteration in *Ginkgo biloba* leaf extracts
Evangelia Bampali, Zarko Kulic, Stefan Germer

PS1-G-003
Hepatoprotective potentials of *Picralima nitida* against *in vivo* carbon tetrachloride-mediated hepatotoxicity
Macdonald Idu, Oghale Ovuakporie-Uvo, Gerald Eze

PS1-G-004
Correlating carob pods (*Ceratonia Siliqua* L.) aroma profile with geographical origin
Andriana Krokou, Agapios Agapiou, Marinos Stylianou, Rebecca Kokkinofta

PS1-G-005
Combinatory chromatographic and spectroscopic methods-approach for honeys botanical origin authentication based on the volatile fractions
Ewa Makowicz, Izabela Jasicka-Misiak, Dariusz Teper, Paweł Kafarski

PS1-G-006
HPTLC – fast and accurate technique for determination of botanical origin of honeys based on the fluorescent compounds
Ewa Makowicz, Izabela Jasicka-Misiak, Paweł Kafarski

PS1-G-007
HPTLC fingerprints as useful tools for profiling of honeys to determine authenticity
Natalia Stanek, Izabela Jasicka–Misiak, Paweł Kafarski

PS1-G-008
Characterization of *Abies Sibirica* L. needle polyprenols and safety of their liposome product in marathoners
Ilona Vanaga, Ilva Nakurte, Sandra Rozenstoka, Simona Steina, Dace Suna, Ugis Kletnieks, Baiba Jansone

PS1-G-010
Identification of quality marker compounds in Greek EVOOs using integrated LC-HRMS & FIA FTICR MS platforms and chemometrics
Theodora Nikou, Matthias Witt, Panagiotis Stathopoulos, Aiko Barsch, Leandros A Skaltsounis, Maria Halabalaki

PS1-G-011
Isotopic traceability (^{13}C and ^{18}O) of Greek olive oils
Karalis P., Halabalaki M., Skaltsounis L.A., Diamantopoulos G., Dotsika E.

PS1-G-012
Isotopic model for detecting original wine vinegar
Diamantopoulos G., Karalis P., Gougoura S., Palaigeorgiou E., Dotsika E.

Topic H: Separation and extraction methodologies

PS1-H-001
Phenolic content of the fruit of *Ceratonia siliqua* L. (Carob) determined by HPLC-DAD-MS analysis
S Schwaiger, Alexandru Vasincu, Christiana Neophytou, S Aigner, Hermann Stuppner, Andreas Constantinou

PS1-H-002
Innovative extraction techniques for the recovery of phenolic compounds from olive oil by-products
Sofia Chanioti, Paraskevi Siamandoura, Constantina Tzia

PS1-H-003

Inclusion of borneol, camphor and thujone monoterpens from sage infusion in native *α*- and *β*-cyclodextrin

Nefeli Sofia Sotiropoulou, Katerina Fourtaka, Elias Christoforides, Christos Pappas, Kostas Bethanis, Petros Tarantilis

PS1-H-004

HPLC-DAD-ESI-TOF-MS analysis and antiulcer effects of some extracts from *Vernonia kotschyana* Sch. Bip. ex Walp

Alexandru Vasincu, Christiana Neophytou, Luca-Simon Vlad, Anca Miron, Ioana-Mirela Vasincu, Bogdan-Gabriel Şlencu, Daniela Carmen Ababei, Oana-Dana Arcan, Delia Bulea, Monica Neamţu, Veronica Bild

PS1-H-005

Development a highly sensitive and selective GC-MS method for the determination of Dioxane in cosmetic products

Ibrahim Alsohaimi, Hazim Ali, Tarek Seaf Elnasr

PS1-H-006

Extraction method development for the creation of a bank of extracts

Thibault Lefebvre, Sandrine Zubrzycki, Manon Bunout, Chloé Catan, Eric Lesellier, Emilie Destandau

PS1-H-007

Experimental archaeology: The scent of antiquity reborn by KORRES for the National Archaeological Museum

Iordanis Samanidis, Lena Philippou

PS1-H-008

Optimization of ultrasound–assisted extraction of polyphenolic compounds from *Plantago lanceolata* using response–surface methodology

Jelena Zivkovic, Katarina Savikin, Nada Cujic, Teodora Jankovic

PS1-H-009

Simultaneous HPTLC densitometric estimation of acetyl-11-keto-*β*-boswellic acid and 11-keto-*β*-boswellic acid from *Boswellia serrata*

Meenu Mehta, Saurabh Satija

PS1-H-010

Development of novel HPTLC fingerprint method for simultaneous estimation of berberine and rutin in medicinal plants and their pharmaceutical preparations followed by its application in anti-oxidant assay

Saurabh Satija, Meenu Mehta

PS1-H-011

Isolation and identification of endophytic fungi from *Piper nigrum* L. and investigation of their chemical constituents

Pragya Paramita Pal

PS1-H-012
Oil and fatty profile on the film from the *Pistacia lentiscus,* region of Collo. east in Algeria
Nassima Boudiaf

PS1-H-013
***N*-doped magnetic hydrochar to toxic metals from water: synthesis, characterization, and adsorption studies**
Ayoub Alqadami, Zeid Alothman, Moonis Khana, Mohammad Algamdi

PS1-H-014
Extraction and purification of metabolites from natural substance or biotechnology by Centrifugal Partition Chromatography
Sébastien Chollet, Luc Marchal, Jean-Hugues Renault

PS1-H-015
Cannabis Process Separation of Cannabinoids
Mouroutis-Prionistis, Yiannis

PS1-H-016
A combination of liquid phase microextraction and spectrophotometric determination of ascorbic acid in foods
Yaroslav Bazel, Tetiana Riabukhina

PS1-H-017
Application of the liquid phase microextraction to separation and extraction of biological active compounds from plant samples
Vasil Andruch

PS1-H-018
Environmentally-friendly enzyme-assisted extraction of Naphthoquinones from *Alkanna tinctoria* roots
Elodie Bossard, Angelos Argyriadis, Anthi Karnaouri, Nikos Tsafantakis, Nektarios Aligiannis, Andreanna Assimopoulou, Evangelos Topakas, Nikolas Fokialakis

PS1-H-019
A holistic green strategy for the isolation of bioactive compounds from fennel seed oil and the corresponding by-products, with pilot scale potentials
Dimitris Michailidis, Apostolis Angelis, Alexios-Leandros Skaltsounis

PS1-H-020
Ultrasound-Assisted Extraction (UAE): induced physicalimpacts on Rosemary (*Rosmarinus officinalis* L.) and Artichoke (*Cynara scolymus* L.) leaves
Boutheina Khadhraoui, El Maâtaoui Mohammed, Isabelle Bornard, Rémi Imbert, Philippe Robinet, Anne-Sylvie Fabiano-Tixier, Njara Rakotomanomana, Farid Chemat

PS1-H-021
Oleocanthalic and oleaceinic acids: Isolation, identification and semi-synthesis of new secoiridoids compounds of extra virgin olive oil
Lemonia Antoniadi, Apostolis Angelis, Panagiotis Stathopoulos, Maria Halabalaki, Leandros A. Skaltsounis

PS1-H-022
CORNET Project SaliChem: Halophytes cultivation for sequential biomass valorisation
Marie-Caroline Jonville, Job Tchoumtchoua, Stefan Dröge, Cécile Beauve, Patrick Ballmann, Michael Müller

Topic J: Biodiversity and nutrition

PS1-J-001
HS-SPME analysis of seven truffles (*Tuber* sps) growing wild in Greece. Nutritional value & biological activity
Eleni Stavraki, Konstantia Graikou, Olga Gortzi, Ioanna Chinou

PS1-J-002
Characterization of volatiles, phenolics and antioxidant properties of different populations of *Sideritis clandestina*
Ioannis Bakratsas, Virginia Dimaki, Lygeri Papaioannou, Gregoris Iatrou, Fotini Lamari

PS1-J-003
Phenolic compounds profile and antioxidant properties of different sweet cherry (*Prunus avium* (L.) L.) varieties from Spain
Dolores Muñoz Mingarro, Antonia García, Nuria Acero, Ana Gradillas

PS1-J-004
Development of a novel functional goats' milk yoghurt enriched with *Pistacia atlantica* resin extracts and *Saccharomyces boulardii*: Stability and organoleptic effects
Elena Hadjimbei, George Botsaris, Vlasios Goulas, Eleni Alexandri, Vassilis Gekas, Ioannis Gerothanassis

PS1-J-005
Chemical characterization of seed oils obtained from different apple cultivars
Jelena Zivkovic, Mihailo Ristic, Josephine Kschonsek, Anna Westphal, Angelika Malarski, Volker Bohm

PS1-J-006
Deciphering bacterial composition in the rhizosphere of *Baphicacanthus cusia* (NeeS) Bremek
Meijuan Zeng, Yongjia Zhong, Shijie Cai, Yong Diao

PS1-J-007
Investigation of sea fennel nutritional value under the effect of iodine biofortification using different metabolomics approaches
Eirini Sarrou, Anastasios S. Siomos, Samantha Riccadona, Pavlos Tsouvaltzis, Paschalina Chatzopoulou, Stefan Martens

PS1-J-008
A holistic approach to sustainable use of Greek native medicinal/aromatic plants' by-products to produce innovative feeds for domestic animals
Katerina Grigoriadou, Ilias Giannenas, Eftyxia Agapidou, Panagiotis Bagatzounis, Nektarios Aligiannis, Efterpi Christaki, Panagiota Florou-Paneri

Topic K: Biodeversity and chemical ecology

PS1-K-001
Green propolis: how resin collected by honey bees from *Baccharis dracunculifolia* could be influenced by the gender, number of galls and chemical components of this plant?
Debora Munhoz Rodrigues, Caroline Arruda, Jairo Kenupp Bastos, Rodrigo Augusto Santinelo Pereira

PS1-K-002
Natural products diversity of *Tithonia diversifolia* (Asteraceae) and its specialist insect herbivore *Chlosyne lacinia* (Nymphalidae)
Marilia Gallon, Eduardo Silva-Junior, Juliano Amaral, Norberto Lopes, Leonardo Gobbo-Neto

PS1-K-003
Oregano spices in the European market: Taxonomic identification and quality control
Dimitrios Mertzanidis, Dimitrios Koureas, Andreana Assimopoulou, Stella Kokkini

PS1-K-004
Volatile metabolomic analysis of the heartwood of *Pinus heldreichii* H. Christ: Chemodiversity insights
Elena Zioga, Olga Tsiftsoglou, Diamanto Lazari

PS1-K-005
Evaluation of silymarin content and flavonolignans composition in native *Silybum marianum* populations from Greece
Dimitrios Arampatzis, Anestis Karkanis, Nikolaos Tsiropoulos

PS1-K-007
An ethnobotanical study and phytochemical analysis of medicinal plants in the Greek islands of North Aegean region
Evangelos Axiotis, Maria Halabalaki, Sofia Mitakou, Leandros A. Skaltsounis

PS1-A-001

Activated and non-activated charcoal derived from durian (*Durio zibethinus murr*.) peelings as treatment to improved physico-chemical and microbiological parameters of Davao river

Venchie Badong, Maria Angelique Anne Alapag, Mark Anthony Casimina

University of the Immaculate Conception, Davao, Philippines

River plays a major role in hydrologic cycle but anthropogenic activities have deeply caused deterioration on its physico-chemical and microbiological quality.
It is the goal of this study is to look for a low-cost effective treatment to remove or decrease the contaminants from river water. The researchers utilized activated and non-activated charcoal derived from waste Durian (*Durio zibethinus* murr.) peelings for treatment to improve the physico-chemical and microbiological quality samples collected from Davao River under the following test parameters: pH, color, turbidity, nitrates, sulfates, total coliform and fecal coliform.
Results showed that activated charcoal have shown potential to reduced the levels of contaminants from the collected river water samples 5 to 25 minutes post treatment. The adsorption capacity of the naturally produced activated charcoal was due to the smaller pore size as detected from Scanning Electron Microscope analysis.
Statistical analysis further proved that there was no significant difference in the adsorption capacity of activated and non-activated charcoal durian peelings. The overall results showed significant improvement on the test parameters posttreatment with non-activated and activated charcoals.
Based from the findings of the study, both non-activated and activated charcoals have the capacity to improve the physico-chemical of river water. However, activated charcoal can significantly reduce the amount of bacteria present in the surface water.

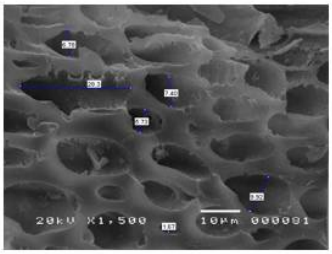


Figure 1: SEM Analysis of Durian Non-Activated Charcoal

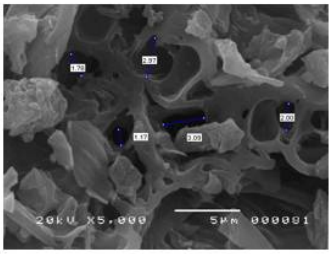


Figure 2: SEM Analysis of Durian Activated Charcoal

Table 1

Physico-chemical and Bacteriological Characteristics of the River Water Sample Before and After Treatment with Non-Activated Charcoal

Test Parameters		Initial	Post treatment, minutes		
			5	15	25
Physical	pH	7.91	8.75	8.5	8.49
	Color, PCU	20	400	400	400
	Turbidity, NTU	740	22	18	18
Chemical	Nitrates, ppm	14.61	13.2	13.49	9.09
	Sulfates, ppm	106	77.85	83.92	84.33
Bacteriological	Total Coliform, MPN	160,000	-	-	160,000
	Fecal Coliform, MPN	160,000	-	-	160,000

Table 2

Physico-chemical and Bacteriological Characteristics of the River Water Sample Before and After Treatment with Activated Charcoal

Test Parameters		Initial	Post treatment, minutes		
			5	15	25
Physical	pH	7.91	10.15	10.14	10.1
	Color, PCU	20	10	5	5
	Turbidity, NTU	740	13	17	21
Chemical	Nitrates, ppm	14.61	5.52	2.87	1.47
	Sulfates, ppm	106.00	5.59	5.11	5.00
Bacteriological	Total Coliform, MPN	160,000	-	-	24000
	Fecal Coliform, MPN	160,000	-	-	513

PS1-A-002

Phytochemical screening and antioxidant activity of methanolic extract of Argan leaves (*Argania spinosa*) from north Algeria

Mahmoud Belalia[1], Louiza Belkacemi[2]

[1]Laboratoire SEA2M, Université Abdelhamid Ibn Badis, 27000 Mostaganem, Algérie, Mostaganem, Algeria, [2]Laboratoire de technologie alimentaire et nutrition, Université Abdelhamid Ibn Badis, 27000 Mostaganem, Algérie, Mostaganem, Algeria

The argan tree, geographically distributed in southwestern Algerian Sahara (north of Tindouf), was recently observed in the Algerian west coast (Mostaganem). This study aimed to assess the phytochemical composition and the antioxidant effect of the methanolic extract of argan leaves (*Argania spinosa*) growing on the west coast of Mostaganem. The methanolic extract was obtained according to the Soxhlet method. A phytochemical screening and an evaluation of the antioxidant activity (DPPH, TBARS) were carried out. Qualitative phytochemical tests show the presence of polyphénols (flavonoids and tannins), triterpenes and reducing sugars. The polyphénols, flavonoid and tannins assay revealed that the methanolic extract contained 168.5 ± 0.002 mg Eq gallic acid / g extract, 47.86 ± 2.37 mg Eq quercetin / g extract and 97.33 mg Eq catechin/ g extract, respectively. Antioxidant activity, evaluated by the DPPH free radical reduction method, reveals an IC_{50} of up to 0.838 mg / ml. The extract at the concentrations tested, seems to have a non-negligible ($p<0.05$) effect on the lipid oxidation of the meat kept at 4 °C. In fact, the TBA values recorded after five days of storage reached 1.99 mg equivalent MDA / kg for the untreated meat against 0.369 and 0.243 mg equivalent MDA / kg in the meat treated with the concentrations 0.1 and 0.2 mg / ml, respectively.
The methanolic extract of the Argan tree leaves growing in the west coast of Algeria has an interesting antioxidant potential to be exploited

Keywords: *Argania spinose*, methanolic extract, antioxidant activity, north Algeria, polyphenols

PS1-A-005

Chemical analysis and metal chelating power of extracts from three medicinal plants: *Peganum Harmala* L., *Cinnamomum Zeylanicum* and *Rosmarinus Officinalis*

Souad Djellali[1,2], Rachid Saharaoui[3], Ibtissem Debab[1]

[1]Department of Matter Sciences, Faculty of Science and Technology, Bordj Bouarreridj University, 34000, Bordj Bouarreridj, Algeria, Bordj Bouarreridj, Algeria, [2]Laboratory of Physico-Chemistry of High Polymers, Faculty of Technology, Ferhat Abbas University, 19000, Setif., Setif, Algeria, [3]Department of Biology and Plant Ecology, Ferhat Abbas University, 19000, Setif, Algeria, Setif, Algeria

This work had focused on the chemical analysis and the study of the metal chelation capacities of three medicinal plants widely used in Algeria: *Rosmarinus officinalis*, *Peganum harmala* and *Cinnamomum zeylanicum*. Plants were collected during the flowering period. Flowers and leaves were cleaned of impurities, dried in shade at room temperature then grounded to powder before being subjected to adequate extraction. Polyphenols and alkaloids were the two major classes of secondary metabolites extracted. In the chemical analysis, the contents of total polyphenols, flavonoids, condensed tannins, percentage of ash and metals content were evaluated. The capacities of polyphenols and alkaloids to form complexes with iron and copper ions were determined. The results obtained showed that these plants contain variable contents of polyphenols, flavonoids and condensed tannins. For total polyphenols, the highest level was obtained for *Cinnamomum zeylanicum* while the lowest was obtained for *Peganum harmala*. For alkaloids, the analysis showed the presence of various compounds such as morphine, and codeine. In addition, it was found that the ashes of the plants contain low amounts of toxic metals such as Hg and Pb which makes these plants safe to use. Concerning the chelating capacity of the secondary metabolites studied, the results showed that both classes have high capacities to bind copper and iron. However, alkaloids have shown a greater efficiency to form chelates with the copper ions than polyphenols. The presence of many secondary metabolites (polyphenols, tannins, flavonoids and alkaloids) that have well-known therapeutic properties may justify the multiple therapeutic indications for which these plants are used in traditional therapy.

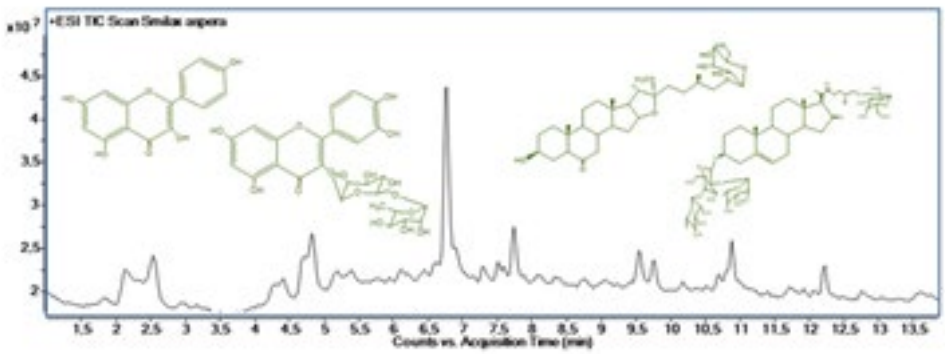
PS1-A-006

Liquid chromatography combined with time-of-flight high resolution mass spectrometry (LC/Q-TOF/HRMS) for the chemical characterization of *Smilax aspera* L. leaves

Eleni Kakouri, Charalabos Kanakis, Panayiotis Trigas, Petros Tarantilis

Agricultural University of Athens, Greece

Medicinal plants are known for their potential therapeutic activity due to their content of bioactive compounds. Among them, *Smilax aspera* L. (Smilacaceae) is known for its tonic, diuretic, antidiabetic and antioxidant properties which are attributed to the underground organs of the plant [1]. In addition, its berries have been demonstrated to possess strong antioxidant capacity. However, leaves of the plant remain an unexplored source of potentially useful bioactive compounds, that need to be elucidated. The aim of the current research is to evaluate the chemical profile of *S. aspera* leaves with the use of liquid chromatography combined with a time-of flight high resolution mass spectrometry (LC/Q-TOF/HRMS). The dried leaves of the plant were extracted using methanol (70% v/v) with the assistance of ultrasonic bath water. The organic solvent was evaporated in a rotary evaporator and the received hydro-extract was freeze dried. The HPLC analysis was performed using a solvent system of water LC/MS and acetonitrile LC/MS both with 0.1% formic acid. Chromatograms were recorded at 280,360,440 nm. The Q-TOF mass spectrometer was operated with a dual ESI source in the positive ionization mode. Results of our study indicated that the leaves of the plant are a rich source of flavonoids and steroid saponins presented as furostanol, isospirostane and spirostane glycosides. These compounds have significant biological activity and could be used in food and/or pharmaceutical industry [2].



Total ion chromatogram of the extract of Smilax aspera leaves

Keywords: *Smilax aspera*, LC/Q-TOF/HRMS analysis, flavonoids, steroidal saponins

References:

[1] Tian LW, Zhang Z, Long HL *et al*. Nat Prod Bioprospect 2017; 7: 283.
[2] Güçlü-Üstündağ O, Mazza G. Crit Rev Food Sci Nutr 2007; 47(3): 231–58.

PS1-A-007

New insular red propolis from Colombia: botanical origin, biological and chemical markers

Guillermo Salamanca Grosso[1], Mónica Patricia Osorio Tangarife[2]

[1]Universidad Del Tolima, Ibagué, Colombia, [2]Mellitopalinological and Physicochemical Properties of Foods Research Group. Universidad del Tolima, Ibagué, Colombia

Propolis is a complex natural product based on exudates, pollen wax and resinous material mixture collected by bees from various plant sources, normally it is a dark yellow, brownish or green resinous material. A broad spectrum of biological properties has been recognized. As a result of this wide range of biological activities, propolis is one of these products which have attracted the researchers' attention. Raw propolis contains such diverse chemical classes of compounds as flavonoids, phenylpropanoids, terpenes, stilbenes, lignans, coumarins, and their prenylated derivatives, showing a pattern consistent with around 300 previously reported compounds. The chemical characteristics of propolis are linked to the diversity of geographical location, plant sources and bee species. The interest in the composition and biological activity of green propolis, gave way to the study of red propolis, which to date has only been reported in Cuba, Venezuela, Brazil, Mexico, China and Nigeria. In Colombia, the studies related to the botanical origin, composition and biological activity of propolis have been gaining interest on the part of some researchers. For effect-directed analysis (EDA), HPTLC-Finger print, electrospray ionization mass spectrometry (ESI(-)-MS), spectroscopy (UV-V, FT-IR), total flavonoids and phenols. antioxidant activity assays, and the antimicrobial and cytotoxic and genotoxic bioassays was developed in characterization of ethanoic extract of Colombian red propolis (EECRP). Visual exam of HPTLC multiple chemicals. Characteristic ESI(-)-MS spectra show ions *m/z* 121, 147, 165, 213, 239 low relative frequency, 255, 271, 283, 371, 373, 501 and 502 abundant, 315, 339, 371, 373, 401 medium, that are representative of chrysin, kaempferol, quercetin, naringenin, pinocembrin, formononetin, biochanin A and daidzein. Diffusional test for microbiological sensibility on *Staphylococcus aureus* (ATCC 29213), and *Listeria monocytogenes* (ATCC 4677)], suggesting that the EECRP has components with antimicrobial activity. This is the first scientific report in relation to the Colombian red propolis.

PS1-A-008

Mechanistically degenerate kinetic resolution in [5+2] cycloisomerization

Kai Ma

University of Oxford, Oxford, United Kingdom

Kinetic resolution is a highly attractive strategy for the synthesis of enantioenriched compounds from racemic starting materials, whereby one enantiomer of a substrate reacts in preference, or in a different manner, to the other. In the field of asymmetric transition metal-catalyzed cycloisomerization, where acyclic starting materials are converted to relatively complex cyclic (chiral) products under the influence of a chiral metal catalyst, kinetic resolutions are scarce – and to date unknown for the [5+2] cycloisomerization of vinylcyclopropanes, a venerable method for the synthesis of seven-membered rings. Here we show how a rhodium(I)-phosphoramidite catalyst is able to affect the first examples of kinetic resolution in [5+2] cycloisomerization. Depending on the substrate, either a classical resolution (where one enantiomer remains unreacted), or a diastereodivergent resolution (in which each enantiomer gives a different product diastereomer) are achieved, both with exceptional selectivity. This difference in the mode of resolution is shown to arise from a divergence in the mechanistic pathway followed, as supported by detailed theoretical studies; such mechanistic degeneracy is, to our knowledge, unprecedented in the wider arena of kinetic resolution. The chemistry operates across a range of vinylcyclopropane substrates, and includes the first example of kinetic resolution by cycloisomerization of a chiral silane, as well as an application to natural product synthesis through resolution of an allenyl vinylcyclopropane.

PS1-A-009

Phytochemical diversity of *Salvia* species in Hengduan Mountains

Ayumi Ohsaki[1], Hiroshi Kawabe[1], Kana Kuwada[1], Mariko Funasaki[1], Hiroshi Hirota[1], Haruka Tsukada[2], Teppei Komiyama[2], Hiroyuki Kagechika[2], Yasuko Okamoto[3], Motoo Tori[3], Yoshinori Saito[4], Gong Xun[5]

[1]College of Humanities and Sciences, Nihon University, Setagaya-ku, Japan, [2]Institute of Biomaterials and Bioengineering, Tokyo Medical and Dental University, Surugadai, Japan, [3]Faculty of Pharmaceutical Sciences, Tokushima Bunri University, Tokushima 770-8514, Japan, [4]Graduate School of Biomedical Sciences, Nagasaki University, Nagasaki 852–8521, Japan, [5]Kunming Institute of Botany, Chinese Academy of Science, Kumming 650204, China

Salvia is one of the largest genera of Lamiaceae, distributed all over the world. Some species, such as *S. miltiorrhiza*, *S. przewalskii*, and *S. yunnanensis*, have been used as traditional Chinese medicines known as 'Tanjin' for the treatment of angina pectoris, inflammation, coronary artery diseases, chronic renal failure etc. Many constituents have been isolated from the genus Salvia, especially, diterpenoids with abietane and clerodane skeletons [1]. Some of the diterpenoids from *Salvia* species have shown various activities, e.g., antioxidant, anti-inflammatory and antifeedant activities. According to Flora of China, 126 species of *Salvia* are found in China, for which there are still many taxonomical problems, especially in Yunnan and Sichuan Provinces. Thus, during the course of our study of the diversity of chemical constituents of six *Salvia* species (*S. przewalskii, S. przeawlskii var. przewarskii, S. yunnanensis, S. plattii, S. grandifolia* and *S. maximowicziana*) by a comprehensive analysis, we isolated more than fifty compounds including new compounds with unusual skeletons arisen a consequence of the rearrangement of their carbon framework [2-4]. We will discuss the chemotaxomy of *Salvia* species from a standpoint of the structural diversity of the isolated compounds.

Acknowledgment: This work was supported in part by JSPS (Grant-in-Aid for Scientific Research No. 25303010 and 24510293) and the Japan – China Scientific Cooperation Program from JSPS and NSFC.

References:

[1] Wu Y-B, Ni Z-Y, Shi Q-W, Dong M, Kiyota H, Gu Y-C, Bong B. Chem Rev, 2012; 112: 5967–6026.
[2] Ohsaki A, Kawamata S, Ozawa M, Kishida A, Gong X, Kuroda C. Tetrahedron Lett, 2011; 1375–1377.
[3] Tsukada H, Kawabe H, Ohtaka A, Saito Y, Okamoto Y, Tori M, Kagechika H, Hirota H, Gong X, Kuroda C, Ohsaki. A Nat Prod Commun, 2016; 11: 159–161.
[4] Kawabe H, Suzuki R, Hirota H, Matsuzaki K, Gong X, Ohsaki. A Nat Prod Commun, 2017; 12: 1177–1179.

PS1-A-010

Search for new compounds from *Portulaca pilosa*

Minori Nonaka[1], Emi Hara[1], Ayumi Seike[1], Yasuko Okamoto[2], Miwa Kubo[2], Mariko Funasaki[1], Yoshiyasu Fukuyama[2], Pilosa Keiichi Matsuzaki[3], Hiroshi Hirota[1], Ayumi Ohsaki[1]

[1]College Of Humanities and Sciences, Nihon University, Setagaya-ku, Japan, [2]Faculty of Pharmaceutical Sciences, Tokushima Bunri University, Yamashiro-cho, Japan, [3]School of Pharmacy, Nihon University, 7-7-1, Narashinodai, Funabashi, Japan

The genus *Portulaca,* which includes about 100 species, is mainly distributed throughout the South America. However, few reports are available on their chemical constituents except *Portulaca oleracea*, which is known as herbal medicine. We previously reported on the skeletal transition in diterpenoid constituents and phylogenetic relationship among three *Portulaca* plants, *P. grandiflora, P. cv. jewel* and *P. pilosa*. The skeletal types of the major diterpenoid constituents of *P. cv. jewel, P. pilosa* and *P. grandiflora* are clerodane ([6.6]-fused ring), printziane ([7.6]-fused ring) and portulane ([7.5]-fused ring), respectively. In addition*, P. cv. jewel* and *P. pilosa* produce printziane and clerodane diterpenes in minor amounts, and *P. grandiflora* yields all types of diterpenoids. Thus, the biosynthetic pathway was proposed follows: clerodane → printziane → portulane. Further the phylogenetic sequence in three plants was discussed. [1, 2]

We studied the minor constituents of *Portulaca pilosa* which could be evidence for inference of the biosynthesis pathway. As the results, we isolated 16 new printziane diterpenoids and 4 new clerodane diterpenoids together with 2 known printziane diterpenoids. These compounds were elucidated on the basis of spectroscopic analyses of NMR and HRESI including 2D NMR (^{1}H-^{1}H COSY, HSQC, HMBC and NOESY).

References:

[1] Ohsaki A, Shibata K, Tokoroyama T, Kubota T. J Chem. Soc Chem Commun 1987; 151–153.
[2] Ohsaki A, Shibata K, Kubota T, Tokoroyama T. Biochem Syst Ecol 1999; 27: 289–296.

PS1-A-011

Oxidation of lignan structures

Patrik Runeberg

Åbo Akademi University, Turku, Finland

Lignans are biologically active natural compounds found in various plants. Studies have shown that lignans have various interesting properties. For example they are strong antioxidants, they exhibit anticancer properties and lower the risk of coronary heart disease [1,2].
The lignan hydroxymatairesinol can readily be extracted from wood knots of Norway spruce (*Picea abies*) and further modified to a wide range of lignan structures. [3-5].
Our study has found that various TEMPO-catalyzed methods selectively oxidize primary hydroxyls of lignans to their corresponding aldehydes, without further oxidation to carboxylic acids.
The hypervalent iodine reagent known as [Bis(trifluoroacetoxy)iodo]benzene, or PIFA, works as a strong oxidation agent. Oxidation of the lignan matairesinol by PIFA lead to oxidative ring closure. Depending on if DCM or TFA was used as solvent in the reaction, the ring closing step was selective towards formation of either six or eight membered rings. By increasing the amount of PIFA, aromatization of the formed ring was obtained.
The faith of lignans during UV-irradiation was also investigated using a wide range of analytical techniques, such as NMR, HPSEC, LCMS and GCMS. The study showed that UVC irradiation of olefinic lignans lead first to a *cis*- and *trans*-isomerization of the double bond. If the irradiation continued, further reactions were observed. With the major reactions being oligomerization and intramolecular reactions to monomeric products, also cleavage of double bond to aldehydes and carboxylic acids was observed to a smaller extent. For non-olefinic lignans, UV-irradiation primarily lead to oligomerization.

Acknowledgements: Academy of Finland, The Swedish Cultural Foundation in Finland, and Raision Tutkimussäätiö are acknowledged for financial support

References:

[1] J Agric Food Chem, 2003; 51: 7600–7606.
[2] Org Biomol Chem, 2005; 3: 3336–3347.
[3] Phytochem Rev, 2004; 2: 331–340.
[4] J Org Chem, 2002; 67: 7544–7546
[5] J Chem- Soc, Perkin Trans., 2002; 1: 1906–1910.

PS1-A-012

Isolation of gallic acid and flavonoids from *Terminalia brownii* leaves

Intisar Elzein[1], Mahgoub El Tohami[2], Aisha Almagboul[3]

[1]Division of Chemistry, Faculty of Applied and industrial Science, University of Bahri, Khartoum bahri, Sudan, [2]Division of Pharmacognosy, faculty of Pharmacy, Omdurman Islamic University, omdurman, Sudan, [3]Division of Microbiology, Medicinal and Aromatic Plants research Institute, khartoum, sudan

The present work is concerned with the chemical structure of the active components in leaves of *Terminalia brownii*; that may be responsible of some of its reported biological effects including antibacterial and antifungal properties.

The extracts from dried *Terminalia brownii* leaves by miscellaneous organic solvents were investigated for antimicrobial effects against four types of bacteria: *Bacillus subtilis*, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa* and two fungi *Aspergillus niger* and *Candida albicans.* The most active constituents were isolated and their chemical structures were identified by spectroscopic methods of analysis NMR and LC-MS, which indicate the presence of gallic acid and flavonoids.

Acknowledgement: The author would like to thank Prof. Dr. R.Verpoorte/ Division of Pharmacognosy/ Section of Metabolomics/ Institute of Biology/ Faculty of Science and Mathematics/ Leiden University/ the Netherlands for hosting and all facilities present to perform this work.

Keywords: Antimicrobial effects, active constituents, chemical structures, *Terminalia brownii*.

References:

[1] Abdel Mageed MAM. 1996; M.Sc. Thesis, Pharm., University of Khartoum.
[2] Collins GH, Lynes PM and Grange JM 1995; Microbiology Methods, 7th ed, Butterwort-Heinemann Ltd, Britain, 175–190.
[3] Eldahshan OA, Ayoub NA, Singab A-NB and Al-Azizi MM 2008; Rec Nat Prod, 2(3), 83–93.
[4] El Ghazali GEB., El Tohami MS, El Egami AAB, Abdalla WS and Mohammed M.G. 1997; Medicinal Plants of the Sudan, part IV, Medicinal Plants of Northern Kordofan.
[5] El Ghazali GEB and Abdalla WS 2000; Bibliography of Sudanese Medicinal Plants, National Centre of Research, Medicinal and Aromatic Plants Research Institute, Sudan, Khartoum.

PS1-A-013

Reaction products formed with food constituents and norlignans dissolving from sugi barrel for sake

Fumio Kawamura

Department of Forest Resources Chemistry, Forestry and Forest Products Research Institute, Tsukuba, Japan

The aim of this study was to estimate the influence of the co-existence of sugi (*Cryptomeria japonica*) heartwood and sake (rice wine) constituents in wooden barrel sake (Japanese name, "Taruzake"). Two norlignans (C_6-C_3-C_2-C_6 compounds), sequirin-C (SQ-C) and agatharesinol (AR) were isolated from the heartwood of sugi (ca. 70-year-old). Each norlignan (SQ-C, AR) was paired with eight typical sake constituents (amino acids, organic acid, and furfurals), and dissolved in 15% ethanol to prepare the model solution of wooden barrel sake. These model solution was aged (25 °C, dark, static), then analyzed by HPLC. The solution of norlignan only was used as control. Solutions of [AR + sake constituent] showed less reactivity, because of no difference with control. In contrast, model solution of SQ-C and amino acid showed remarkable difference from control; it was confirmed that peculiar products were formed by co-existence of both constituents. Especially, co-existence of L-alanine (L-Ala) expressed significant influence; seven major products (P1-P7) were confirmed. Aging experiment (12 d) of SQ-C and L-Ala was scale upped, then five major products (P1-P5) were isolated or separated by preparative HPLC. The product, which was formed at largest amount (P5), was very unstable; it easily transformed to other products. P5 was presumed to be an intermediate for the formation of a diversity of products. Isolated P1-P4 were instrumental analyzed using NMR. Characteristics of P1, P3 and P4 were as follows: disappearance of catechol nucleus, saturation of side chain, linkage of L-Ala on side chain. Characteristics of P2 were as follows: side chain formed a ring, highly oxygenated structure. The results in this study demonstrated high reactivity of SQ-C and L-Ala. L-Ala and other amino acids are the constituents, which have great influence on flavor of sake. Therefore, influence of constituents dissolved from the sugi barrel on the flavor of sake was suggested.

PS1-A-014

Saffron aqueous extract: phytochemistry & determination of crocetin's serum and tissue pharmacokinetics after oral and intravenous administration to C57/BL6J mice

Maria-Eleni Grafakou[2], Eirini Christodoulou[1], Helen Skaltsa[2], Nikolaos Kadoglou[3], Nikolaos Kostomitsopoulos[4], Georgia Valsami[1]

[1]Laboratory of Biopharmaceutics and Pharmacokinetics, Department of Pharmacy, School of Health Sciences, National & Kapodistrian University of Athens, Panepistimiopolis, Zografou, Athens, Hellenic Republic, [2]Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, School of Health Sciences, National & Kapodistrian University of Athens, Panepistimiopolis, Zografou, Athens, Hellenic Republic, [3]Oxford University, Oxford, United Kingdom , [4]Center of Clinical Experimental Surgery and Translational Research, Biomedical Research Foundation, Academy of Athens, Athens, Hellenic Republic

Saffron, the dried stigmata deriving from the flower of *Crocus sativus* L. (Iridaceae), is known since antiquity as a medicinal spice and a dye pigment. Several studies have been conducted in order to unveil saffron's extracts potential pharmaceutical applications [1]; however, its serum and tissue pharmacokinetics have not yet been evaluated.

The chemical investigation of the saffron standardized aqueous extract (SFE), purified on RP18-HPLC (linear gradient elution from 20% to 100% ACN in H_2O), revealed the presence of all-trans crocin, picrocrocin and its aglycon, as well as free sugars. The structures of the isolated compounds were elucidated by high-field NMR spectroscopy [2]. SFE was found to be stable in room temperature for almost 15 months and was quantified for the main secondary metabolite crocin (approximately 27%) using an HPLC-PDA method.

An HPLC-PDA method has also been applied for the analysis of serum and tissue samples after I.V. and oral administration to C57/Bl6J mice in order to measure crocetin levels and apply pharmacokinetics analysis. Crocetin is crocin's primer metabolite, which derives from hydrolysis right after saffron administration.

An one-compartment distribution model has adequately described the pharmacokinetics of crocetin and its metabolite after single dose (I.V. and oral) administration of SFE (60 mg/kg of body weight) to C57Bl/6J male mice. A first order kinetic constant described the rate of crocetin's I.V. biotransformation to its conjugated form, as well as crocetin's oral absorption. Relative bioavailability was calculated at 1.17 for total crocetin. Tissue PKs were described by non-compartmental analysis and revealed extensive crocetin distribution to liver and kidneys.

Keywords: *Crocus*, saffron, extract, NMR, pharmacokinetics, crocin, crocetin

References:

[1] Razak SIA, Hamzah MSA, Yee FC, Kadir MRA, Nayan NHM. J Herbs Spices Med Plants 2017; 23(2): 98–116.

[2] Sobolev AP, Carradori S, Capitani D, Vista S, Trella A, Marini F, Mannina L. Foods 2014; 3(3): 403–419.

PS1-A-015

Chemical variation in essential oils from the oleo-gum resin of *Boswellia carteri*

William Setzer[1], Anjanette DeCarlo[2], Stephen Johnson[2], Ambika Poudel[2], Prabodh Satyal[2], Loren Bangerter[3]

[1]University Of Alabama In Huntsville, Huntsville, United States, [2]Aromatic Plant Research Center, Winston-Salem, United States, [3]doTERRA International, Pleasant Grove, United States

Frankincense, the oleo-gum resin of *Boswellia* species, has been an important element of traditional medicine for thousands of years. Frankincense is still used for oral hygiene, to treat wounds, and for its calming effects. Different *Boswellia* species show different chemical profiles, and *B. carteri*, in particular, has shown wide variation in essential oil composition. In order to provide insight into the chemical variability in authentic *B. carteri* oleoresin samples, a hierarchical cluster analysis of 42 chemical compositions of *B. carteri* oleo-gum resin essential oils has revealed at least three different chemotypes, (I) an *α*-pinene-rich chemotype, (II) an *α*-thujene-rich chemotype, and (III) a methoxydecane-rich chemotype.

PS1-A-016

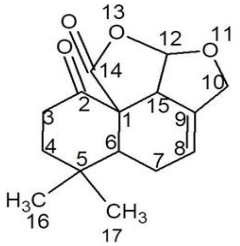
Sesquiterpenoid skeleton type drimane produced by the bioluminescent mushroom *Neonothopanus gardneri*

Teresinha De Jesus Aguiar Dos S. Andrade[1], Maria das Dores A. Oliveira[2], Joaquim Soares C. Junior[3], Nerilson Marques Lima[4], Jioji N. Tabudravu[5], Dulce Helena S. Silva[6], Antonia Maria das Graças Lopes Citó[2]

[1]Nucleus of Applied Research to Sciences-NIAC, Federal Institute of Education, Science and Technology of Maranhão-IFMA, Timon, Brazil, [2]Federal University of Piauí, Department of Chemistry, Campus Ministro Petrônio Portela, Teresina, Brazil, [3]Federal Institute of Piauí, Department of Chemistry, Campus Central, Teresina, Brazil, [4]Department of Chemistry - ICE, Federal University of Juiz de Fora, University Campus,, Juiz de Fora, Brazil, [5]School of Forensic and Applied Sciences, Faculty of Science and Technology, University of Central Lancashire, Lancashire, UK, [6]Nucleus of Bioassays, Biosynthesis and Ecophysiology of Natural Products (NuBBE), Department of Organic Chemistry, Institute of Chemistry, São Paulo State University (UNESP), Araraquara, Brazil

A sesquiterpenoid skeleton type drimane was isolated from the luminescent mushroom *Neonothopanus gardneri*, which is widespread in Cocais Forest, a transitional biome between the Amazon Forest and Caatinga. The structure of this compound was subsequently elucidated using spectroscopic and spectrometric data. The acetonitrile fraction (2.52 g) was fractionated on C18 flash column eluted with H_2O:CH_3OH gradient which yielded subfractions (Ng1-Ng8). Subfraction Ng3 (165 mg) was chromatographed by RP-HPLC-UV and afforded a sesquiterpene skeletal type drimane (12.0 mg), obtained as a crystalline solid. Its UV-vis spectrum showed λmax 256 nm, whereas IR data showed bands for carbonyl group (1777 cm^{-1}) and sp^3 carbons (2924.4 cm^{-1}). The ^{1}H NMR and HSQC spectrum showed signals for two methyl at δH 0.87 (δC 25.08), δH 1.42 (δC 24.00), olefinic at δH 6.11 (δC 118.82), a methine hydrogen at δH 6.06 (δC 104.39). The 13C NMR and DEPT-135 spectra associated with HMBC and HSQC evidenced signals for two carbonyls at δC 174.29 (C-14) and δC 2011.84 (C-2), correlations of H-12, H-8 with C-9 and C-10. In the HMBC, δH 1.59 (m) of H-4 correlated with C-3, C-16 and H-3 with C-1, C-14 and δH carbons 0.87 (s) of H-16 with the C-1, C-6 and C-15 carbons and correlations δH 3.59 (d) of H-6 with C-8 and C-9 and δH 1.66/1.84 (m) of H-7 with C-2, C-15. The molecular formula was established as $C_{15}H_{19}O_4$ by HRESIMS spectrum with *m/z* 263.1779 $[M+H]^+$. To the best of our knowledge, this is the first report of the chemical constituents of *N. gardneri* and the first report of the isolation of tetracyclo [7,5,1,0^{16},0^{12},15]-5,5-dimethyl-11-oxa-2-oxo-pentadec-8-en-14(13)-lactone (fig.1) from *N. gardneri*.

Figure1: Tetracyclo [7,5,1,$0^{1,6}0^{12}$,15] -5,5-dimethyl-11-oxa-2-oxo-pentadec-8-en-14 (13)-lactone



Keywords: bioluminescent mushroom, secondary metabolites, lactone.

References:

[1] Capelari M et al. Mycologia 2011;103: 1433–1440.
[2] Kirk PM et al. Ainsworth & Bisby's Dictionary of the Fungi. Ed.10, CABI Europe–UK, 2008: 771.

PS1-A-017

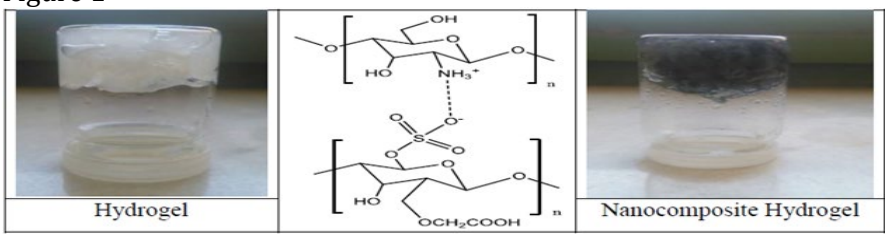
Chitosan/carboxymethyl cellulose sulfuric acid hydrogels and their nanocomposits: Preparation and application in tartrazine removal

Ali Reza Karimi

Department of Chemistry, Faculty of science, Arak University, Arak 38156-8-8349, Iran.

Non-covalent interactions such as hydrogen bonds and electrostatic between amino and hydroxyl groups of chitosan with different cross-linkers leads to formation of physically cross-linked chitosan hydrogels. The cross-linking of chitosan not only improve their resistance to acidic environments, but also increase their absorption capacity.
Herein, carboxymethyl cellulose sulfuric acid (CMC-SO_3H) was prepared through reaction of chlorosulfonic acid with CMC. Then, preparation of physically cross-linked hydrogels was carried out by simple mixing of chitosan and CMC-SO_3H in acetic acid (0.1%, v/v) solution. The mixture was stirred vigorously for approximately 2 min at room temperature. The various ratios of chitosan: CMC-SO_3H mixtures were used for synthesis of hydrogels. In continuation, their hydrogel nanocomposites were prepared by crosslinking of chitosan and CMC-SO_3H (with ratio of 1:1, w/w) in the presence of 0.25% and 0.5% (w/w) multi-walled carbon nanotubes (MWCNTs).
The adsorption of tartrazine, by prepared hydrogels was investigated using batch equilibration method at different pHs (pH: 4, 7). It was found that chitosan : CMC-SO_3H hydrogel with ratio of 1:1 was the appropriate adsorbent for tartrazine. Therefore, the hydrogel with ratio of 1:1 was selected. The effect of pH was studied for the adsorption of tartrazine at different pHs by chitosan: CMC-SO_3H hydrogel (1:1). It was observed that the best result was obtained at pH 7. Introducing MWCNT-COOH in the structure of chitosan : CMC-SO_3H hydrogels has an effect on the adsorption of tartrazine. It was seen that the maximum and best adsorption capacity was obtained for hydrogel nanocomposites (0.25%). The maximum percentage of tartrazine adsorption for nanocomposite hydrogel (0.25%) was 94%.

Figure 1



References:

[1] Dragan ES. Chem Eng J 2014; 243: 572.
[2] Sahnoun S, Boutahala M. Int J Biol Macromol 2018; 114: 1345.

PS1-A-018

4-oxo-*β*-apo-13-carotenone from the cyanobacterium *Anabaena cylindrica* PCC 7122

Teresa Martins, Mariana Reis, Pedro Leão

Interdisciplinary Centre of Marine and Environmental Research (CIIMAR/CIMAR), Matosinhos, Portugal

Carotenoids are isoprenoid compounds with light-harvesting functions in photosynthesis, widely distributed among plants, algae and bacteria. Carotenoid cleavage dioxygenases (CCDs) transform carotenoids into the smaller apocarotenoids. Present in either carotenoid- and non-carotenoid-synthesizing organisms, these enzymes are responsible for the production of a large diversity of apocarotenoids with several signalling functions regarding development, environmental interactions and hormonal regulation. In addition, apocarotenoids aromatic and flavour properties are particularly interesting for food and cosmetic applications.
In cyanobacteria, apocarotenoids have been associated with a photoprotective role in thylakoid membranes and a large number of CCDs have been characterized in this phylum. However, despite the large number of apocarotenoids already characterized, their diversity is expected to be even higher.
In this work [1], we describe the isolation and structural elucidation of the methyl ketone apocarotenoid 4-oxo-*β*-apo-13-carotenone from the cultured freshwater cyanobacterium *Anabaena cylindrica* PCC 7122, corresponding to the first report of this compound from natural sources. Because this apocarotenoid has been shown to be generated ex vivo under oxidative conditions, we provide metabolomics and bioinformatics evidences for its cyanobacterial biogenesis.

PS1-A-019

Characterization of sulfhydryl conjugates with betanidin and gomphrenin quinoids by means of HPLC-DAD-ESI-MS/MS and LC-MS-IT-TOF

Agnieszka Kumorkiewicz, Sławomir Wybraniec

Cracow University of Technology, Cracow, Poland

Betacyanins, water-soluble plant pigments which are a subgroup of betalains, belonging to most families of the Caryophyllales, are extensively used in industry as food colorants. However, they display not only prominent coloring attributes but also a wide variety of biological properties, namely antioxidant, chemopreventive and cytotoxic. For that reason, there has been a growing interest in research on mechanism of betalains action in living organisms and their potential applications as therapeutic agents capable of stopping tumor growth in search of anticancer preparations.

The aim of this study was to investigate the possibility of different sulfhydryl-based scavengers to form conjugates with betalains. The detection of the adducts was performed by high performance liquid chromatography coupled to tandem mass spectrometry (HPLC-DAD-ESI-MS/MS). In addition, determination of molecular formulae as well as research on the fragmentation paths of the conjugates by ion-trap high-resolution mass spectrometry (LCMS-IT-TOF) were performed. According to our research, the use of automated measurement system with the utilization of a trapping agents such a glutathione, cysteine, *N*-acetylcysteine and DL-dithiothreitol enables the detection of short-lived oxidation intermediates of betanidin and gomphrenin pigments (quinones, quinone methides and aminochromes). In the case of glutathione conjugates, the characteristic protonated molecular ions of glutathionyl-betanidin $[M+H]^+$ at *m/z* 694 as well as glutathionyl-gomprhrenin $[M+H]^+$ at *m/z* 856 were detected. Furthermore, their fragmentation under collision-induced dissociation (CID) results in a characteristic neutral loss corresponding to the pyroglutamic acid (129 Da) moiety. When deprotonated GSH conjugates are formed, the fragment ions of *m/z* 273 corresponding to deprotonated *γ*-glutamyl-dehydroalanyl-glycine are formed. Further extensive studies also compared stability of the conjugates for different trapping agents.

Acknowledgements: This research was financed by Polish National Science Centre for years 2015-2018 (Project No. UMO-2014/13/B/ST4/04854).

PS1-A-020

Secondary metabolites from *Sideritis germanicopolitana*

Hasan Kırmızıbekmez[1], Murat Erdoğan[1], Umur Erdem[1], Norbert Kúsz[2], Fatih Demirci[3], Judit Hohmann[2]

[1]Department of Pharmacognosy, Faculty of Pharmacy, Yeditepe University, TR-34755, Kayışdağı, İstanbul, Turkey, [2]Department of Pharmacognosy, Faculty of Pharmacy, University of Szeged, 6720, Szeged, Hungary, [3]Department of Pharmacognosy, Faculty of Pharmacy, Anadolu University, 26470, Eskişehir, Turkey

The genus *Sideritis* (Lamiaceae) contains annual and perennial species which are mainly distributed in the Mediterranean basin [1]. Turkey, with 46 *Sideritis* species and high endemism rate, is considered to be one of the gene centers of this genus [2]. The aerial parts of *Sideritis* species are used to prepare infusions and decoctions for the treatment of gastritis, gastric ulcer, inflammations of mucous membrane as well as against common cold and flu in the traditional medicines of Mediterranean countries [2,3]. The phytochemical composition of the genus is mainly represented by flavonoids, phenylethanoid glycosides, iridoid glycosides, diterpenes and volatile principles [1,2]. *S. germanicopolitana* Bornm., an endemic species to Turkey, is a perennial herb which is distributed in Inner and Northern Anatolia [4]. In the continuation of our studies on the isolation of bioactive secondary metabolites from *Sideritis* species, we describe in the present work the isolation and structure elucidation of the secondary metabolites from the aerial parts of *S. germanicopolitana*. Chromatographic separation of the MeOH extract led to the isolation of three iridoid glycosides, 5-*O*-allosyloxy-aucubin, melittoside and ajugol, five phenylethanoid glycosides, decaffeoylverbascoside, verbascoside, martynoside, leucoseptoside A and lamalboside, four flavonoids, xanthomicrol, isoscutellarein 7-*O*-[6'''-*O*-acetyl-*β*-allopyranosyl-(1-2)]-*β*-glucopyranoside, 4'-*O*-methylisoscutellarein 7-*O*-[6'''-*O*-acetyl-*β*-allopyranosyl-(1-2)]-*β*-glucopyranoside, 3'-hydroxy-4'-*O*-methylisoscutellarein 7-*O*-[6'''-*O*-acetyl-*β*-allopyranosyl-(1-2)]-*β*-glucopyranoside, two lignan glycosides dehydrodiconiferylalcohol 4-*O*-*β*-glucopyranoside and pinoresinol 4'-*O*-*β*-glucopyranoside. The structures of the isolates were elucidated by NMR and HR-MS analyses. All secondary metabolites within this work are reported for the first time from *S. germanicopolitana*, while the isolated lignans are new for the genus *Sideritis*.

References:

[1] Fraga BM. Phytochemistry 2012; 76: 7–24.
[2] González-Burgos E, Carretero ME, Gómez-Serranillos MP. J Ethnopharmacol 2011; 135: 209–225.
[3] Sağır ZO, Çarıkçı S, Kılıç T, Gören A.C. Int J Food Prop 2017; 20: 2994–3005.
[4] Huber-Morath A. Sideritis L. In: Davis PH. editor. Flora of Turkey and East Aegean Islands, University Press Edinburgh, 1982, Vol. 7, 178–199.

PS1-A-021

Essential oil content of *Sideritis* spp. cultivated in Northern Greece

Andreas Douros[1], Helen Skaltsa[1], Catherine Grigoriadou[2], Eleni Maloupa[2]

[1]National & Kapodistrian University of Athens, School of Pharmacy, Department of Pharmacognosy & Chemistry of Natural Products, Athens, Greece, [2]Laboratory for the Conservation and Evaluation of Native and Floricultural Species-Balkan Botanic Garden of Kroussia. Agricultural Research Center of Northern Greece, Central Macedonia, Greece

For centuries, *Sideritis* spp. are widely used in the traditional medicine due to their antioxidant, anti-inflammatory and analgesic properties [1, 2]. *Sideritis* L. is one of the largest genera within the Lamiaceae family. *Sideritis* herb is referred to *S. scardica*, *S. raeseri* and *S. clandestina* or a mixture of them and it is approved by EMA as traditional herbal medicinal product against cough and mild gastrointestinal discomfort. Numerous studies revealed well-defined pharmacological activities of both essential oils and plant extracts. Their main constituents are terpenes and phenolic derivatives. Driven by our interest on this genus, we investigated the essential oils of *S. raiseri*, *S. perfoliata*, *S. scardica*, cultivated at the Laboratory of Conservation and Evaluation of the Native and Floricultural Species. EOs were obtained by hydrodistillation in a modified Clevenger-type apparatus and their chemical analyses were performed by GC-MS. The identification of the components was based on comparison of their mass spectra with those of Wiley and NBS Libraries and those described by Adams [3], as well as on comparison of their retention indices and with literature values. The main constituent of *S. scardica* was *β*-caryophylenne (30.2%), of *S. raiseri β*-pinene (20.61%) and of *S. perfoliata α*-pinene (39.4%). According to our analyses, the chemical content of *Sideritis* spp. reacted positively to cultivation, based on the literature data.

Keywords: *Sideritis* spp., essential oil

References:

[1] Charami MT, Lazari D, Karioti A, Skaltsa H, Hadjipavlou-Litina D and Souleles C. Phytother Res 2008; 22: 450–454.
[2] Aligiannis N, Kalpoutzakis E, Chinou IB, Mitakou S, Gikas E, Tsarbopoulos A. J Agric Food Chem 2001; 49: 811–815.
[3] Adams R. Identification of Essential oil components by Gas Chromatography/Quadrupole Mass Spectroscopy, 2007.

PS1-A-022

Significant secondary metabolites from *Crepis commutata* (Spreng.) Greuter

Ioanna Kotti, Christina Barda, Helen Skaltsa

Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, School of Health Sciences, National & Kapodistrian University of Athens, Athens, Greece

C. commutata (Spreng.) Greuter. (Asteraceae; common name glikossirides) is an edible plant mainly used in traditional diet of Crete, fresh or cooked in salads [1]. It is an annual plant, native in W. Asia and Europe (Bulgaria, Cyprus, Greece) [2]. *C. commutata* was collected in Anabyssos (Attiki) and the aerial parts have been exctracted with a mixture of solvents containing cyclohexane: ether: methanol 1:1:1, according to the Bohlmann isolation method [3]. The non-polar extract was prefractionated by VLC on silica gel, using cyclohexane-EtOAc-Me_2CO mixtures of increasing polarity as eluents to give several fractions. Further purification on silica gel CC and on RP18-HPLC (MeOH-H_2O 3:2) yielded four sesquiterpene lactones: integrifolin (1), 8-epi-grosheimin (2), 8-epiisolipidiol (3), its 3-*O*- acetyl ester (4), two flavonoids: luteolin (5), luteolin-7-*O*-*β*-D-glucuronide (6), and two phenolic acids: *p*-anisic acid (7), caffeic acid (8). It is noteworthy that the present study is the first phytochemical analysis of *C. commutata*. The main compound was 8-epiisolipidiol, while luteolin consists a significant chemosystematic marker for the genus *Crepis* L. Compounds (1), (2), (6) and (8) have been previously isolated from the genus *Crepis* L. while compounds (4) and (7) are reported here for the first time in *Crepis* sp. The study is still in progress.

Acknowledgement: The authors wish to thank Associate Prof. Theophanis Constantinidis (Department of Ecology & Systematics, Faculty of Biology, NKUA) for the collection and identification of the plant material.

Keywords: *C. commutata*, 8-epiisolipidiol

References:

[1] Vardavas CI, Majchrzak D, Wagner KH, Elmadfa I, Kafatos A. Food Chem 2006; 99: 822–834.
[2] Babcock EB. The genus Crepis 1–2. 1947. Publications in Botany 21: i–ix, 1–197, 22: i–x, 199–1030.
[3] Bohlmann F, Zdero C, King RM, Robinson H. Phytochemistry 1984; 23: 1979–88.

PS1-A-023

Chemical profile of wild *Crithmum maritimum* L.

Vasiliki Zafeiropoulou, Aikaterini-Michaela Tomou, Helen Skaltsa

Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, School of Health Sciences, National & Kapodistrian University of Athens, Athens, Greece

Crithmum maritimum L. (Apiaceae), the only species of the genus, commonly known as sea fennel or rock samphire, is a facultative halophyte growing on maritime cliffs and sometimes in sand along the Mediterranean and Black sea coasts [1]. It is edible aromatic herb with powerful scent. Its organs (roots, leaves and fruits) are rich in several bioactive substances that could be used as aromatic, medicinal, antimicrobial and insecticide products and therefore it shows substantial economical and medicinal potentials [2]. Its healing properties are already known since antiquity. It is mentioned in Corpus Hippocraticum and later on in Plinius' Historia Naturalis for soothe vesical pains and in cases of amenorrhea [3]. The plant material was selected from maritime cliffs in a small coastal town, Parga (W. Greece) and the aerial parts have been extracted with cyclohexane, dichloromethane, methanol, methanol: water 5:1, successively. The phytochemical analyses, performed by means of analytical techniques and NMR spectroscopy, allowed us to isolate and identify nine compounds belonging to different phytochemical groups, namely falcarindiol, *O*-geranylvanillin, chlorogenic, caffeic, 3, 4-*O*-dicaffeoylquinic acids, rutin, quercetin-3-*O*-robinobioside, hyperoside and isoquercitrin. Furthermore, we investigated the chemical profiles of *C. maritimum* decoction and infusion. The NMR analyses revealed that both of them contain free sugars, chlorogenic and dicaffeoylquinic acids, while the decoction was more abundant concerning the mentioned phenolic acids.

Keywords: *C. maritimum*, phenolic acids, flavonoids, falcarindiol

References:

[1] Coiffard L. Rev Hist Pharm 1991; 38: 313–317.
[2] Meot-Duros L, Magné C. Plant Physiol Biochem 2009; 47: 37–41.
[3] Coiffard L, Piron-Frenet M, Amicel L. Int J Cosmet Sci 1993; 15: 15–21.

PS1-A-024

Essential oil content of cultivated *Crithmum maritimum* L.

Vasiliki Zafeiropoulou, Helen Skaltsa

Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, School of Health Sciences, National & Kapodistrian University of Athens, Athens, Greece

Crithmum maritimum L. (Apiaceae), known as Sea fennel, is a perennial plant growing wild mainly on maritime rocks along the Mediterranean countries, Pacific and Atlantic coasts [1]. Its leaves are used in traditional medicine as a tonic, an antiscorbutic, carminative, vermifuge and diuretic [2]. In the present study, the essential oils (EOs) of the aerial parts obtained from cultivated populations of two consecutive years (2016; 2017; Larissa-Central Greece) were analyzed by GC-MS on a Hewlett-Packard 7820A-5977B MSD system, using a fused silica HP-5 MS capillary column. The identification of the components was based on comparison of their mass spectra with those of Wiley and NIST Research Library and those described by Adams [3], as well as on comparison of their retention indices and with literature values. The main compounds of the EO derived from the cultivar of 2016 were sabinene (17.6%), *γ*-terpinene (17.5%), *p*-cymene (16.7%), *β*-phellandrene (15.5%), thymol methyl ether (9.3%), terpinen-4-ol (4.8%), and dillapiole (2.0%) while in the sample of 2017 were *β*-phellandrene (30.9%), *γ*-terpinene (19.6%), sabinene (15.8%), thymol methyl ether (7.6%), terpinen-4-ol (4.8%), and dillapiole (0.2%). Finally, a comparison was made between the two samples, as well as with the literature data. None of the previous investigated Sea fennel EOs from Greece revealed the presence of dillapiole [4,5].

Keywords: *C. maritimum*, cultivated populations, EOs, sabinene, *γ*-terpinene, dillapiole.

References:

[1] Cunsolo F, Ruberto G, Amico V, Piatelli M. J Nat Prod 1993; 56: 1598–1600.
[2] Senatore F, Napolitano F, Ozcan M. Flav Fragr J 2000; 15: 186–189.
[3] Adams R., 2007. Identification of Essential Oil Components by Gas Chromatography/Mass Spectroscopy, 4th edition. Carol Stream, Allured Publishing Co., Illinois.
[4] Katsouri E, Demetzos C, Perdetzoglou D, Loukis A. J Essent Oil Res 2001; 13(5): 303–308.
[5] Tsoukatou M, Tsitsimpikou C, Vagias C, Roussis V. Z. Naturforsch. 2001; 56c: 211–215.

PS1-A-025

Isolation of bioactive compounds from *Sideritis* ssp.

Ekaterina-Michaela Tomou[1], Helen Skaltsa[1], Paschalina Chatzopoulou[2]

[1]Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, School of Health Sciences, National & Kapodistrian University of Athens, Hellenic Republic, Panepistimiopolis/ Zografou/Athens, Greece, [2]Hellenic Agricultural Organization DEMETER, Institute of Breeding and Plant Genetic Resources, IBPGR, Department of Medicinal and Aromatic Plants, Thermi/Thessaloniki, Greece

The genus *Sideritis* L. is remarkably rich in bioactive secondary metabolites. Many studies have revealed its important pharmacological activities such as anti-inflammatory, antioxidant, antimicrobial and gastroprotective activities [1,2]. The decoction of the herba *Sideritis* ssp. (Greek mountain tea) is well known for its traditional uses against mild gastrointestinal discomfort and common cold and it is used widely in Europe and the Mediterranean region [1,2]. Approximately 17 endemic species grow in Greece [1,3]. In continuation of our research on the Lamiaceae family, aerial parts of some *Sideritis* spp. have been studied. Among them, aerial parts of cultivated population of *S. euboea* Heldr., a narrow endemic threatened species, were extracted. The plant material was offered by the Institute of Breeding and Plant Genetic Resources. A voucher specimen was deposited in the herbarium of Aromatic and Medicinal Plant Department; under the code 19-17.

The air-dried aerial parts were successively extracted with cyclohexane, dichloromethane and methanol. The methanol extract was concentrated and the residue was re-dissolved in boiling water. The water-soluble fraction was filtered and extracted with EtOAc and *n*-BuOH, successively.

So far, one phenolic acid, four flavones, four phenylethanoids have been isolated from the EtOAc and *n*-BuOH residues, while three diterpenes have been yielded by the dichloromethane extract. Their structural elucidations were elucidated by high-field NMR spectroscopy.

Keywords: *Sideritis*, anticancer activity, bioactive secondary metabolites

References:

[1] HMPC/39453/2015—European Union Herbal Monograph on *Sideritis scardica* Griseb; *Sideritis clandestina* (Bory & Chaub.) Hayek; *Sideritis raeseri* Boiss. & Heldr; *Sideritis syriaca* L., herba-final.

[2] Charami MT, Lazari D, Karioti A, Skaltsa H, Hadjipavlou-Litina D, Souleles C. Phytother Res 2008; 22: 450–454.

[3] Strid A, Tan K. Mountain Flora of Greece. Edinburgh University Press: Edinburgh, 1991: 84–91.

PS1-A-026

Kinetic and chromatographic studies on non-enzymatic oxidation of gomphrenin pigment from the fruit juice of *Basella alba* L.

Agnieszka Kumorkiewicz, Tomasz Świergosz, Sławomir Wybraniec

Cracow University of Technology, Cracow, Poland

The mechanism of betacyanins oxidation is of significant interest because of recent emergence of these pigments as highly active natural compounds with antioxidative properties and potential benefits to human health. Recent studies indicate a particularly beneficial effect of betacyanins as antioxidants in different types of pathologies associated with oxidative stress. For this reason, the studies are focused on betacyanins which can act as the so-called "scavengers" of free radicals that prevent free-radical damage induced by active oxygen or oxidation of biological molecules, membranes and tissues.
Although recent enzymatic studies have shed some light on oxidation pathways of 5-*O*-glucosylated betacyanins, full reaction mechanism data are still lacking. Therefore, we continued our research on 6-*O*-glucosylated betanidin (gomphrenin) which was oxidized by ABTS cation radicals at different physicochemical conditions. The non-enzymatic oxidation kinetics of gomphrenin was monitored by spectrophotometry in a microplate reader at pH 3, 5 and 7. The first inspection of the consecutive visible spectra registered during the course of oxidation reveals a fast initial decrease of each of the main absorption bands of the pigment. After this point, further changes are much slower or negligible. The λmax main absorption band of gomphrenin is shifted from the starting value of 538 to 520–525 nm within the first few minutes of the experiment, but is not further changed for the remaining 2 h. Furthermore, the non-enzymatic oxidation of gomphrenin results in a formation of additional absorption bands at λmax 445 nm suggesting a formation of dehydrogenated derivatives of gomphrenin. Chromatographic separation of the reaction products with spectrophotometric and mass spectrometric detection (LC-DAD-MS/MS) was also performed. The presence of two prominent oxidation products, 2-decarboxy-2,3-dehydrogomphrenin (*m/z* 505) and 2,17-bidecarboxy-2,3-dehydrogomphrenin (*m/z* 461) was detected.

Acknowledgements: This research was financed by Polish National Science Centre for years 2015-2018 (Project No. UMO-2014/13/B/ST4/04854).

PS1-A-027

Preliminary study: Secondary metabolites of *Melissa officinalis* L.

Dimitrios Dimas, Ekaterina-Michaela Tomou, Helen Skaltsa

Section of Pharmacognosy& Chemistry of Natural Products, Department of Pharmacy, School of Health Sciences, National &Kapodistrian University of Athens Hellenic Republic, Panepistimiopolis, Zografou, Athens, Greece

Lemon balm (*Melissa officinalis* L.) is a well-known medicinal plant widely used in traditional medicine over the world for the treatment of several ailments. According to the monograph of EMA, it is a traditional herbal medicine for the symptomatic treatment of mild gastrointestinal disorders, as well as for the relief of mild symptoms of mental stress [1]. Several studies have reported its various pharmacological activities such as antioxidant and ant-inflammatory activities [2].
In continuation to our research on plants belonging to the Lamiaceae family, we report herein our first results on the investigation of *M. officinalis*. The air-dried aerial parts from a wild population were successively extracted by dichloromethane and methanol. The dichloromethane extract was fractionated by VLC and CC on silica gel revealing, so far, the presence of two triterpenoids, namely ursolic acid and pomolic acid. Ursolic acid is a common metabolite of plants belonging to the Lamiaceae family and the main compound of triterpenoids in *M. officinalis* [3,4]. Our study confirms the presence of pomolic acid in *M. officinalis* reported only once in 1967 [5]. Their structural elucidations were elucidated by high-field NMR spectroscopy.

Keywords: *Melissa officinalis*, Lamiaceae, ursolic acid, pomolic acid

References:

[1] EMA/HMPC/196745/2012-Community herbal monograph on *Melissa officinalis* L., folium
[2] Miraj S, Rafieian-Kopaei M, Kiani S. JEBCAM 2017; 22(3): 385–396.
[3] Awad R, Muhammad A, Durst T, Trudeau VL, Arnason JT. Rhytother Res 2009; 23: 1075–1081.
[4] Janicsák G, Veres K, Kakasy AZ, Mathe I. Biochem Syst Ecol 2006; 34: 392–396.
[5] Brieskorn HC & Wunderer H. Chem Ber 1967; 100: 1252–1265.

PS1-A-028

Seasonal variation of the esential oil of *Rosmarinus officinalis* L. (Lamiaceae), growing wild in the island of Cephalonia (Greece)

Aristeidis Pritsas[1], Gerasimia Tsasi[1], Theophilos Mailis[1], Yannis Samaras[2], Helen Skaltsa[1]

[1]Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, School of Health Sciences, National & Kapodistrian University of Athens, Hellenic Republic, Panepistimiopolis, Zografou, Athens, Greece, [2]Department of Food Technology, Technological Educational Institute (T.E.I.) of Ionian Islands, Argostoli, Cephalonia, Greece

Seasonal variations in the composition of the essential oils obtained from the same rosemary plant growing wild in the island of Cephalonia (Greece) were determined by GC/MS analyses on a Hewlett-Packard 7820A-5977B MSD system, using a fused silica HP-5 MS capillary column. Retention indices for all compounds were determined according to the Van den Dool approach [1], using *n*-alkanes as standards. The identification of the components was based on comparison of their mass spectra with those of Wiley and NIST Research Library and those described by Adams [2]. In each case, oxygenated monoterpenes and monoterpene hydrocarbons were the dominating groups (31.2%-67.2% and 19.6-43.2%, respectively). In all samples, the predominant constituents are *α*-pinene (6.3-28.0%), 1,8-cineole (11.4-24.4.0%), *trans*-caryophyllene (4.7-33.5%) and borneol (7.0-18.5%); followed by camphene (1.3-6.2%), *β*-pinene (0.6-5.4%), linalool (1.2-5.3%), camphor (tr-12.9%), *α*-terpineol (1.8-9.2%), verbenone (2.0-5.6%) and bornyl acetate (0.5-7.4%). The essential oils revealed mainly quantitative differences during the vegetative cycle. The highest amounts of *α*-pinene and 1,8-cineole were found in the sample with young and incompletely developed leaves collected in March. In contrast, the amounts of both compounds decreased, while *trans*-caryophyllene reached its maximum concentration (33.5%) on the sample collected in July (old shoots). Borneol was persistent ca. 10% in all months, except for the sample collected in August (18.5%). This study confirms the suggestion that the chemical composition of the essential oils sometimes critically depends on the time of collection [3,4].

Keywords: *Rosmarinus officinalis*, Essential oil, Seasonal variations, *α*-pinene, 1,8-cineole, *trans*-caryophyllene, borneol

References:

[1] Adams R. Identification of Essential Oil Components by Gas Chromatography/Mass Spectroscopy. 4th edition. Carol Stream. Allured Publishing Co. Illinois 2007.
[2] Van den Dool H, Kratz P D. J Chromatogr 1963; 11: 463–471.
[3] Lakušić D *et al*. Chem Biodivers 2012; 9(7): 1286–1302.
[4] Lakušić D *et al*. Nat Prod Comm 2013; 1: 131–134.

PS1-A-029

Antioxidant capacity of fractions obtained from wild *Ugni molinae*, Turcz. leaves

Rodrigo Pérez[1,2], Carla Delporte[1]

[1]Natural Products Laboratory, Chemical and Pharmaceutical Sciences Faculty, University of Chile, Santiago, Chile, [2]Laboratory of Proteostasis Control and Biomedicine, Faculty of Medicine, University of Chile, Santiago, Chile

Several studies suggest that high consumption of polyphenolic compounds (PC) can reduce the risk of chronic diseases, improving human health [1,2,3]. Although PC are not essential for plant development, they are found in high concentrations in several species, and until date 8000 different structures have been reported [2]. A great source of PC are berries, and Chile is one of the top producers of the southern hemisphere. Murtilla (*Ugni molinae*, Myrtaceae.) is a chilean shrub whose leaves have been traditionally used by indigenous people for the treatment of urinary tract pains, as astringents, stimulants [4], and for their anti-inflammatory and analgesic properties [5]. The aim of this study was to compare the antioxidant capacity of fractions obtained from a global methanolic extract (GME) of wild murtilla leaves. Fractions were obtained using a Silica60 chromatographic-column and increasing polarity solvents. Eleven fractions were obtained and qualitatively characterized through TLC. Five fractions (F5, F7, F9, F10, and F11) were selected for further analysis due to higher yields. The total phenolic content (TPC) and antioxidant activity of the selected fractions (SF) were analyzed through the Folin-ciocalteu and DPPH• assay. Significant differences ($p<0,05$) were obtained for the TPC between the SF, the highest value being for F7 (254,6 ± 14,1 mg EAG/ g dried fraction). The highest antioxidant capacity was also for F7 (IC_{50} = 4,2 ± 1,7), but no differences ($p<0,05$) were observed between the SF.

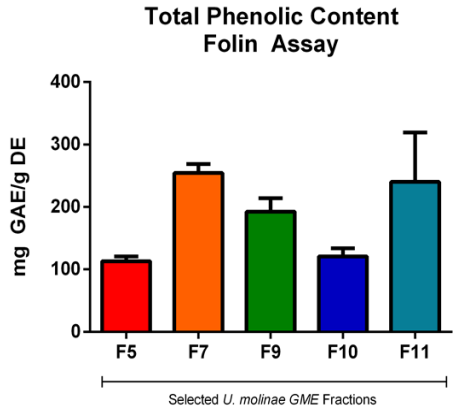


Fig. 1. Total phenolic content of the selected fractions from the GME of wild *U. molinae* leaves

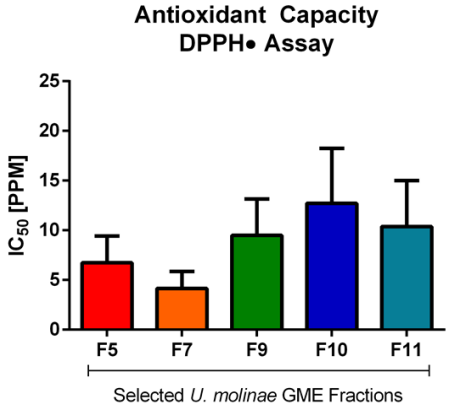


Fig. 2. Antioxidant capacity of the selected fractions from the GME of wild *U. molinae* leaves

Acknowledgements: CONICYT n°21150769 FONDECYT n°1130155

Keywords: Polyphenols, Antioxidants

References:

[1] Costa C *et al*. Food Chem Toxicol 2017; 110: 286–299
[2] Del Rio D *et al*. Antioxid Redox Signal 2013; 18: 1819–1891.
[3] Mcdougall G J. Proc Nutr Soc 2016; 76: 163–171.
[4] Rubilar M *et al*. J Agric Food Chem 2006; 54: 59–64.
[5] Delporte C *et al*. J. Ethnopharmacol 2007; 112: 162–165.

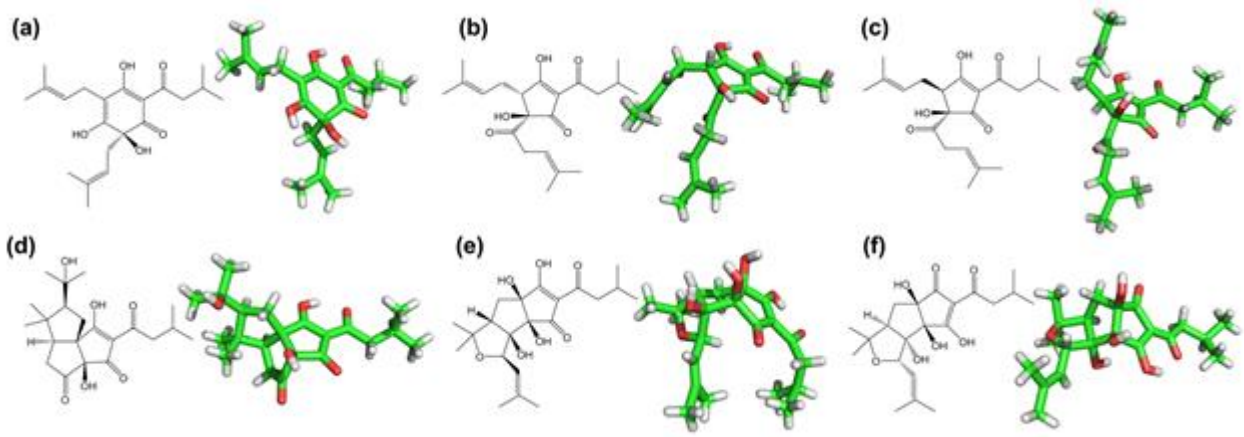
PS1-A-030

Crystalline sponge method efficiently and exhaustively reveals stereo-configurations of polycyclic compounds derived from beer's bitter acids

Yoshimasa Taniguchi[1,2,3], Takashi Kikuchi[4], Makoto Fujita[2,3]

[1] Kirin Company Limited, Yokohama, Japan, [2]Department of Applied Chemistry, School of Engineering, The University of Tokyo, Tokyo, Japan, [3]JST-ACCEL, Kawaguchi, Japan, [4]Rigaku Corporation, Akishima, Japan

The bitter acids derived from hops (*Humulus lupulus* L.) and their derivatives affect quality of beer as well as show multi effects on physiological function as potentially beneficial agents. Information on molecular characteristics of these compounds is particularly important to develop science-based brewing technology and also to explore their bioactivities. However, their structural analyses, especially stereo-configuration analyses, are very arduous because they have various scaffolds and many continuous quaternary carbon atoms and chiral centers. Here, we showed that the crystalline sponge (CS) method1 could easily and efficiently reveal the relative and absolute configurations of bitter acids and their derivatives. The absolute configurations of (−)-humulone and (−)-*trans*- and (+)-*cis*-isohumulones, the representative bitter acids, were confirmed without the need to prepare single crystals of their derivatives containing heavy atoms or reference chiral centers (Figure 1). The absolute configurations of (+)-tricyclooxyisohumulones, a series of bitter acid oxides, were also determined and 6 novel compounds were identified. The relative configurations of *rac*-scorpiohumulinols and *rac*-dicyclohumulionols, another series of bitter acid oxides, were confirmed for the first time by the CS method. Furthermore, we succeeded in elucidating the absolute configurations of a series of polycyclic compounds, including several novel ones, which are generated from (−)-*trans*-isohumulone during storage of beer. As a result, the CS method is highly practical to analyze stereo-configurations of various bitter compounds derived from hops.



Acknowledgements: This research was carried out as a part of the JST-ACCEL project in which M.F. is a principal investigator.

Keywords: Crystalline sponge, Configurational analysis, Bitter acids, Hop, *Humulus lupulus* L., Beer

References:

[1] Nature 2013; 495: 461–466.

PS1-A-031

Isolation and identification of components from hybrids *Phlomis* × *commixta* Rech. f. (*P. cretica* × *P. lanata*)

Michalis Stefanakis[1], Diamando Lazari[2], Haralambos E. Katerinopoulos[1]

[1]University of Crete - Department of Chemistry, Heraklion, Greece, [2]Aristotle University of Thessaloniki - School of Pharmacy, Thessaloniki, Greece

The present work describes the component analysis of the hybrid *P.* × *commixta*, a plant that belongs to the genus *Phlomis*. The plant material was divided into flowers, leaves and roots. The secondary metabolites of the plant were recovered via extractions, isolated by various chromatographic techniques such as V.L.C., C.C., HPLC, and identified by spectroscopic methods such as UV / Vis, NMR, GC-MS and LC-MS.

The compounds isolated from the flowers were a) the flavonoids: naringenin, eriodyctiol, luteolin, apigenin-7-*O*-*β*-D-glucoside, eriodyctiol-7-*O*-*β*-D-glucoside, and diosmentin-7-*O*-*β*-D-glucoside; b) the phenylethanoic glycosides: martinoside, verbascoside, and forsythoside B; c) the chromone: 5,7-dihydroxychromone; d) the phenolic acids: caffeic acid, *p*-hydroxybenzoic acid, chlorogenic acid, chlorogenic acid methyl ester, gallic acid, and *p*-coumaric acid and e) an aliphatic hydrocarbon: Docos-1-ene.

The leaf sample contained a) flavonoids: quercetin, apigenin, hesperetin, luteolin-7-*O*-*β*-D-glucoside, and quercetin-3-*O*-*β*-D-glucoside; b) the phenylethanoid glycosides: echinacoside, verbascoside and forsythoside B; c) a phenolic acid: vanillic acid.

The root sample contained a) metabolites of the phenylethanoid glycoside class: allysonoside, verbascoside, and forsythoside B; b) the steroids: brassicasterol and stigmasterol and c) the tyrosol esters: tyrosol lignoserate and tyrosol methyl ether palmitate, the latter one being isolated as a natural product for the first time.

Although *Phlomis* species thrive almost all over the world, the present work is the first report on the ingredients of the *P.* × *commixta* hybrid. The pharmaceutical profile of the components isolated from the hybrid is in agreement with the folklore medicine applications reported in the literature, and is indicative of the potential use of the plant in contemporary medicine.

Keywords: *P.* × *commixta*, *Phlomis*, secondary metabolites, flavonoids, phenylethanoid glycosides, phenolic derivatives, tyrosol esters, steroids

References:

[1] Georgescu L, Stefanakis M K, Kokkini S, Katerinopoulos H E, Pirintsos S A. Phytochemistry 2016; 122: 91.

PS1-A-032

Phytochemical investigation of *Achillea coarctata* Poir. (Asteraceae)

Konstantina Papakosta, Christina Barda, Helen Skaltsa

Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, School of Health Sciences, National & Kapodistrian University of Athens, Athens, Greece

The genus *Achillea* L. is comprised of over 100 widely distributed, medicinal herbs, known since antiquity [1]. The name Achillea refers to Greek hero Achilles, who used it to treat the soldiers' wounds during the literary Trojan War. *Achillea* spp. have been traditionally used for many ailments, such as wound healing, against bleeding, headaches, inflammations, pains, spasmodic diseases, flatulence and dyspepsia. *A. millefolium* L. herb and flos are quoted by EMA as traditional herbal medicinal products [2]. *A. coarctata* Poir., a perennial herb found from sub-montane to montane level in Greece [1], was collected at Lake Polifitou (Kozani, Greece), where it is locally used to treat gastrointestinal disorders. Its aerial parts have been extracted according to the Bohlmann isolation method [3]. The extract was fractionated by VLC, CC and RP18-HPLC yielded five sesquiterpene lactones: arteludovicinolide A (1), rupicolin A (2), rupicolin B (3), artecalin (4) and 3-acetylridentin B (5), a nor-isoprenoid: 3S,5R-loliolide (6), three flavonoids: luteolin (7), centaureidin (8), casticin (9) and a phenolic acid: 3,5-di-*O*-caffeoylquinic acid (10). Compounds 1-4 and 7-9 are common metabolites of *Achillea* L., while (5) and (6) have been reported for the first time in this genus. In addition, due to the cytotoxic activity of *A. coarctata* Poir., the initial extract, as well as the isolated metabolites are subjected to biological tests, still in progress

Acknowledgements: The authors wish to thank Mrs Elli Lampretsa (pharmacist, Kozani) for the collection of the plant material and Associate Prof. Theophanis Constantinidis (Department of Ecology & Systematics, Faculty of Biology, NKUA) for the identification of the plant material

Keywords: *Achillea coarctata* Poir., sesquiterpene lactones.

References:

[1] Strid A, Tan K. Mountain Flora of Greece. Edinburgh University Press: Edinburgh 1991; 431–450.
[2] Saeidnia S *et al*. DARU 2011; 19: 173–186.
[3] Bohlmann F *et al*. Phytochemistry 1984; 23: 1979–88.

PS1-A-033

Comparative Chemical analysis of seven endemic Greek *Citrus* hybrids

Eleni Anastasopoulou[1], Konstantia Graikou[1], Anastasios Makritis[2], Ioanna Chinou[1]

[1]Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, National & Kapodistrian University of Athens, Greece, [2]National Organization for Medicines (EOF), Ministry of Health and Welfare, Athens, Greece

In this study it is reported the volatiles chemical analysis of the cold pressed peel oils of:
•grapefruit: *Citrus paradisi* cv Star Ruby × *Citrus aurantium* (1)
•four sweet oranges: *Citrus sinensis* cv Newhall × *citrumelo* (2), *C. sinensis* cv Newhall × *trifoliata* (3), *C. sinensis* cv Valencia Porou × *citrumelo* (4), *C. sinensis* cv Valencia Porou × *trifoliata* (5)
•pomelo: *C. grandis* cv Cuban shaddock × *trifoliata* (6) and
•mandarin*: Citrus reticulata* cv Minneola tangelo × *trifoliata* (7)
All volatiles have been analyzed through GC-MS and forty-five constituents were identified. Terpenes appeared as the most abundant secondary metabolites (94-99%) in all studied *Citrus* hybrids among which limonene monoterpenes revealed as the most characteristic (80-95%). Especially in the oil of hybrid 1 the sesquiterpene nootkatone has been detected (3.76%), which is very valuable due to its pharmacological activities and its high commercial value. From the peels of *Citrus* hybrids 4 and 7, six polymethoxyflavones (PMF) have been isolated and structurally determined by NMR spectroscopy as: 3',4',5,6,7-pentamethoxyflavone (sinensetin), 3',4',5,6,7,8-hexamethoxyflavone (nobiletin), 3',4',3,5,6,7-hexamethoxyflavone, 4',5,6,7-tetramethoxyflavone, 3,5,6,7,8,3',4'-heptamethoxyflavone (3-methoxy-nobiletin) and 4',5,6,7,8-pentamethoxyflavone (tangeretin).
PMFs (approx. 20) exist almost exclusively in the *Citrus* genus and particularly in the peels of sweet oranges and mandarin, while their type and concentration may serve taxonomic purposes in botanical and agricultural sciences. Moreover PMFs, due to their hydrophobic nature of methoxy groups, may have higher permeability through the small intestine and are readily absorbed in to the blood circulation system of the human body exhibiting a broad spectrum of biological activities.

Acknowledgements: The author would like to thank the Special Account for Research Grants and the National and Kapodistrian University of Athens for funding their participation in this meeting.

PS1-A-034

Phytochemical study on Greek endemic *Rindera graeca* aerial parts. Antioxidant Activity

Christos Ganos[1], Tomasz Mroczek[2], Nektarios Aligiannis[1], Ioanna Chinou[1], Konstantia Graikou[1]

[1]Section of Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, National & Kapodistrian University of Athens, Greece, [2]Department of Pharmacognosy with Medicinal Plant Laboratory Unit, Medical University, Lublin, Poland

In the framework of our phytochemical studies, on plants of Boraginaceae family [1,2], the Greek endemic species *Rindera graeca* Boiss & Heldr. (tribe Cynoglosseae) is studied for the first time.

The methanolic extract of its aerial parts was subjected to qualitative LC/MS analysis. Through this analysis 12 phenolic metabolites were identified among which 7 phenolic acids: chlorogenic, caffeic, rosmarinic, lithospermic B and salvianolic A acid, together with rabdosiin and rabdosiin disodium salt and 5 flavonoids: kaempferol-3-robinoside-7-rhamnoside (robinin), kaempferol-3-*O*-rutinoside (nicotiflorin), kaempferol-3-*O*-glucoside (astragalin), quercetin-3-*O*-rutinoside (rutin) and quercetin-3-*O*-rutinoside-7-rhamnoside. The same extract was submitted to various chromatographic separations to afford: rutin, quercetin-3-*O*-rutinoside-7-rhamnoside, rosmarinic acid and disodium rabdosiin, which is a new natural product. The structures of all the isolated metabolites were identified by means of 1D ^{1}H-/^{13}C-NMR and 2D NMR spectroscopy.

Additionally, Rindera's methanolic extract was screened for its content of pyrrolizidine alkaloids (PAs) through Mattocks-Molyneux visualization reagent and SPE clean out method as proposed by BfR, with positive results, while LC-MS analysis confirmed the presence of PA rinderine and its *N*-oxide.

Moreover, the total phenolic and flavonoid content were estimated by the Folin-Ciocalteu method (TPC 66.5 mg GAE/g, TFC mg QUE/g 9.7) and the free radical scavenging activity was determined by DPPH and ABTS assays.

Acknowledgements: The author would like to thank the Special Account for Research Grants and the National and Kapodistrian University of Athens for funding their participation in this meeting

Keywords: *Rindera graeca*, LC/MS analysis, phenolics, PAs, antioxidant activity

References:

[1] Marini G, Graikou K, Zengin G, Karikas G, Gupta M, Chinou I. Ind Crops and Prod 2018; 120: 84–89.

[2] Damianakos H, Jeziorek M, Sykłowska Baranek K, Pietrosiuk A, Buchwald W, Chinou I. Phytochem Lett 2016; 15: 234–237.

PS1-A-035

Impact of the soil substrate on essential oil composition of *Origanum vulgare*

Amalia Katsifara[1], Eleni Sazakli[1], Soterios Varnavas[2], Michalis Leotsinidis[1]

[1]Lab of Public Health, Medical School, University of Patras, Patras, Greece, [2]Department of Geology, University of Patras, Patras, Greece

Aromatic and medicinal plants are known to be beneficial for human health since ancient times. Essential oils of these plants provide antimicrobial, antifungal and antioxidant properties. *Origanum* genus has been widely applied in medicine and health care. The aim of this study was to investigate the influence of different soil substrates on the growing process, the biogeochemical characteristics and eventually the composition of essential oil of the plant species *Origanum vulgare* L. ssp. *hirtum* (LINK) IETSWAART.

The study was carried out in the area of the Oblos Mountain, where the species under investigation are indigenous. Soil samples were subjected to chemical analyses to determine the bioavailable fraction of the elements Fe, Ca, Mg, K, Na, Mn and Zn. Representative plant samples were taken from different plant clusters, in two sampling periods, during spring. Plants were fully matured in the second sampling. Essential oil was extracted from each sample and analyzed by GC/MS.

By applying PCA on bioavailable metals in soil, three factors were extracted. Magnesium, Mn and K were loaded on the first factor, Ca on the second and Na on the third. Iron was almost equally loaded on the first two factors. It was observed that two out of 5 plants were grown on soils with main bioavailable metals those that loaded on the first factor, while the rest 3 plants were grown on soils with Ca as the main bioavailable element. Thymol was identified as the dominant compound (mean value 44.6%) in the first group of plants, while in the second, Carvacrol dominated (mean value 59.8%). Essential oil composition changed between the two samplings. Higher number and increased concentrations were observed in fully matured plants.

It is concluded that the essential oil composition of *Origanum* plants depends on degree of maturity and soil substrate.

PS1-A-036

Effects of different cooking techniques on vitamin levels of green leaf vegetables; A HPLC method development study and an anti-oxidative capacity study

Tuğba İduğ[1], Ümit Can Erim[2], Hilal Hızlı[3], Ali Şen[4], Barkın Berk[5]

[1]School of Pharmacy, Department of Pharmacognosy, Medipol University, İstanbul/Beykoz, Turkey, [2]School of Pharmacy, Department of Analytical Chemistry, Medipol University, İstanbul/Beykoz, Turkey, [3]Faculty of Health Science, Department of Nutrition and Dietetics, Medipol University, İstanbul/Beykoz, Turkey, [4]Faculty of Pharmacy, Department of Pharmacognosy, Marmara University, İstanbul/Kadıköy, Turkey, [5]School of Pharmacy, Department of Pharmaceutical Chemistry, Medipol University, İstanbul/Beykoz, Turkey

Qualitative and quantitative vitamin content analysis of complex matrixes with HPLC methods are very popular these days as applicable to various fields of pharma and food industries [1].
In this study, we developed validated HPLC methods for determination of Vitamin B1, Vitamin B6 and ascorbic acid contents of four different vegetable samples (broccoli, spinach, brussel sprouts, and leeks) that were prepared either as raw or cooked with steaming and boiling techniques.
In this context first samples were chopped into small pieces and different cooking techniques were applied for an average of five minutes. After grinding the vegetables either with or used after decantation of cooking water, all of the samples were macerated with methanol.
Finally validated HPLC methods for different experimental conditions were developed and compared for the qualitative and quantitative vitamin analysis.
Further DPPH analysis of these samples were studied for their anti-oxidative capacity.

References:

[1] Nollet L M L. Food Analysis by HPLC Second Edition Revised and Expanded. USA: CRC Press 2000.

PS1-A-037

Selective copper(II) catalyzed amination of aryl-himachalene to secondary benzylamines derivatives in water via chloromethylation

Intissar Ait Lahcen

Equipe De Chimie De Coordination Et Catalyse, Département De Chimie, Université Cadi Ayyad, faculté Des Sciences Semlalia, Marrakech, Morocco

The benzylamines moiety is a significant building block since they are frequently encountered motifs in several molecules used in different fields. They play a prominent role in numerous pharmaceutically compounds such as: Imatinib, a standard anti-cancer drug for the treatment of chronic myelogenous leukaemia and gastrointestinal tumours, Rivastigmine, a cholinergic agent for treating dementia due to Parkinson's disease;
Nonetheless, several general strategies can be envisaged for the preparation of the benzylamine moiety, such as reductive amination of aldehydes, hydroamination of alkenes, reduction of nitrile, alcohols amination, proton shift of triazenes, nucleophilic substitution of halides by amines and Suzuki–Miyaura coupling with trifluoroborates.
A simple and efficient synthesis of aryl-himachalene benzylamines derivatives catalyzed by copper in water via chloromethylation was investigated under mild conditions. Various aromatic amines were used to provide the corresponding secondary benzylamines in good yields. The reactivity of amines bearing electron-withdrawing and electron-donating groups has been studied. The reaction offers a first practical synthetic method for the preparation of a range of sesquitepenes benzylamines derivatives.

PS1-A-038

Chemical Constituents of *Galium asparagifolium* Boiss. et Heldr

Umit Toktas[1], Nazli Boke Sarikahya[2], Husniye Kayalar[1]

[1]Department of Pharmacognosy, Faculty of Pharmacy, Ege University, Bornova, Izmir, Turkey, [2]Department of Chemistry, Faculty of Science, Ege University, Bornova, Izmir, Turkey

The one of the medicinal plant genus *Galium* L. (Rubiaceae) is represented in Turkey by 101 species gathered in 10 sections. *Galium* species are traditionally used to coagulate milk because of an enzyme in their chemical composition. For this reason, this plant is known as "yoghurt herb".

Galium L. contains various secondary metabolites including mostly iridoids and their glycosides, rarely anthraquinones and flavonoids. It is known that according to the literature findings these natural compounds exhibit wide range of biological activities.

In this study, it was aimed to obtain pure natural compounds from *Galium aspragifolium*, to make structure determination of these compounds and to carry out biological activity studies. The *n*-butanol extract was examined by various chemical and chromatographic methods (MPLC, VLC, CC, TLC) for pure glycosides. The structures of these compounds were determined by extensive NMR techniques (^{1}H, ^{13}C, COSY, HSQC, HMBC) and chemical methods (alkaline hydrolysis, acidic hydrolysis, silylation). Up to now, 7 compounds have been isolated and identified. Our phytochemical studies on *Galium aspragifolium* are ongoing.

PS1-A-039

Phytochemical investigation and antimicrobial activity of isolated sesquiterpene lactones from *Crepis incana* Sm.

Christina Barda[1], Ana Ciric[2], Marina Sokovic[2], Michael Tsoukalas[1], Helen Skaltsa[1]

[1]National and Kapodistrian University of Athens, Athens, Greece, [2]Department of Plant Physiology, Mycological Laboratory, Institute for Biological Research, University of Belgrade, Belgrade, Serbia

Crepis incana Sm. (Asteraceae) is an endemic perennial herb growing wild in Greece. The genus *Crepis* L. belongs to the Cichorieae tribe, where most taxa are edible [1]. The present preliminary study is focused on the chemical investigation and provides full Spectroscopic Data (^{1}H-NMR ^{13}C-NMR) for crepiside D not available, so far [2]. Furthermore, the isolated sesquiterpene lactones (SLs) from this plant were tested for antimicrobial potential activity [3]. The fresh plant material was dried and extracted at room temperature with cyclohexane: Et_2O:MeOH (1:1:1) and MeOH:H_2O (5:1), successively. Two costus type guagianolides, grosheimmin and crepiside E and two germacranolides, taraxinic acid and its 1'-*O*-*β*-*D*-glucopyranoside were isolated by RP18-HPLC from the non polar extract. Moreover three flavonoids, luteolin, luteolin-3-*O*-*β*-D-glucopyranoside and quercetin-7-*O*-*β*-D-glucopyranoside were isolated by column chromatography from the polar extract. The structures of the isolated compounds were elucidated by high-field NMR spectroscopy (^{1}H-NMR, ^{1}H-^{1}H COSY, NOESY, HSQC and HMBC). The antimicrobial activity of this five SLs were investigated against (Gram-positive bacteria): *Staphylococcus aureus*, *Bacillus cereus*; (Gram-negative bacteria) *Pseudomonas aeruginosa*, *Escherichia coli* and *Enterococcus faecalis*; (fungi) *Aspergillus fumigatus A. versicolor*, *A. niger*, *Trichoderma viride*, *Penicillium ochrochloron* and *P. verrucosum* var. *cyclopium*. Their activities were in range of MIC 0.015–0.09 mg mL^{-1} and MFC 0.30–0.12 mg mL^{-1}.

Acknowledgements: The authors wish to thank Associate Prof. Th. Constantinidis (Department of Ecology & Systematics, Faculty of Biology, NKUA) for the identification of the plant material.

Keywords: *Crepis incana*, Crepiside D, sesquiterpene lactones, antimicrobial activity

References:

[1] Kavvadas D. Botanical Dictionary. Crepis vol 4. Athens 1956: pp. 2137–2140.
[2] Miyase T, Ueno A, Noro T, Kuroyanagi M, Fukushima S. Chem Pharm Bull 1985; 33:4451–4456.
[3] Barda C, Ciric A, Sokovic M, Tsoukalas M, Skaltsa H. Biochem Sys Ecol 2018; 80: 59–62.

PS1-A-041

Novel anti-inflammatory steroidal compounds

Jesus Sandoval-Ramirez[1], Reyna Zeferino-Díaz[1], María A Fernández-Herrera[2], Nuvia Kantún-Moreno[2], Leticia Olivera-Castillo[2]

[1]Benemérita Universidad Autónoma de Puebla, Puebla, Mexico, [2]CINVESTAV-Mérida, Merida, Mexico

Inflammation, a natural response for the protection of organisms against any damage or infection, involves cellular events and chemical signals. The most prescribed anti-inflammatory compounds are corticosteroids and analogs, despite of many undesirable consequences. In this sense, the search of new drugs has been undertaken. A new family of 22-oxocholestane hydroxyimino derivatives has been synthesized and evaluated by means of an acute mouse ear edema induced by 12-*O*-tetradecanoylphorbol 13-acetate. Herein the obtaining of 3 new oximes, and their anti-inflammatory activity is related. According to these results, the best active compounds were selected to be analyzed vis a vis the gene markers, expressed in inflammation processes: Tumor Necrosis Factor alpha (TNF-α), Interleukin 6 (IL-6), Cyclooxygenase 2 (COX-2) and Macrophage Inhibition Factor (MIF).
The steroidal oximes were synthesized from diosgenin through selective fission of spirostan E and F rings and further oxidation: the steroidal 26*E* and 26*Z* oximes (25*R*)-26-hydroximino-22-oxocholest-5-en-3β,16β-diyl diacetate, 1 and 2, the C-25 oximes of 27-nor-22,25-dioxocholest-5-en-3β,16β-diyl diacetate. The biological evaluation was carried out under a murine model *(Mus musculus)*, which consisted in the topical application of the proto-inflammatory TPA, at the mouse right ear. This procedure increases the relative expression of the genes COX -2, TNF-α, IL-6 and MIF, involved in the inflammatory process. The new steroidal oximes 1, 2, and 3 were tested to reverse the inflammation, comparing their activity with dexamethasone (DXA). Oximes 1 and 3 showed higher anti-inflammatory activity than DXA. Oximes 1 and 3 inhibit the expression of the TNF-α, COX-2, Il-6 and MIF genes. MIF is associated with several cutaneous pathologies. Steroidal oximes inhibit the expression of MIF, so these substances have a high dermatological potential.

PS1-A-042

Synthesis of novel 22-oxocholestane glycosides with potential anticancer activity

Jazmin Ciciolil Hilario-Martinez[1], Zhendong Jin[2], María Antonieta Fernández-Herrera[3], Jesús Sandoval-Ramírez[1]

[1]Benemérita Universidad Autónoma de Puebla, Puebla, Mexico, [2]The University of Iowa, Iowa City, USA, [3]CINVESTAV-Mérida, Mérida, Mexico

Despite a great variety of new compounds have been developed for cancer therapies, all clinically used drugs exhibit undesirable side effects, ought to the lack of selectivity. For this reason, the search for novel molecules possessing selective anticancer activity continues.
The natural saponin OSW-1 has exhibited extremely potent cytotoxic activity against diverse malignant tumor cell lines, becoming an attractive synthetic target looking for selective antiproliferative steroidal candidates. It has been proposed that the C-22 carbonyl group was crucial for the activity. Our research groups have been interested in structure-activity studies of 22-oxocholestane compounds.
On the other hand, various naturally steroidal glycosides bearing a sugar chain attached at position C-3 have exhibited antiproliferative activity, reveling the importance of sugar moiety at this position.
We now report new 22-oxocholestane glycosides containing diverse sugars at C-3. For the synthesis of a chacotriosyl moiety, D-pentaacetylglucose was transformed into a thioglycoside, followed by deprotection of acetylated positions and a selective C-3 and C-6 benzoyl-protection, then, a glycosylation with α-rhamosyl imidate was performed to furnish a thiotrisaccharide. The latter was selectively deprotected at the anomeric position and subsequently converted into an imidate to be used in the glycosylation of diosgenin. In a similar way, glucose, rhamnose, lactose, and maltose diosgenyl glycosides were synthesized. Finally, the diosgenyl saponins were transformed into 22-oxocholestane derivatives through a spirostanic E and F ring opening by means of $BF_3.OEt_2/Ac_2O$ complex followed by an aqueous work-up. The prominent stability of the glyosidic bond was observed despite of the acidic conditions at the ring opening. All compounds were fully characterized by spectroscopic techniques and at this moment *in vitro* antiproliferative tests in tumor cells are being carried out.

PS1-A-043

Chemical composition of the essential oil of leaves and roots of *Stevia serrata* Cav. from Guatemala

Juan Francisco Perez Sabino[1], Manuel Alejandro Muñoz Wug[1], Max Samuel Mérida Reyes[1], Bessie Evelyn Oliva Hernández[1], Edwin Adolfo Taracena Monzón[1], José Vicente Martínez Arévalo[2], Antonio Jorge Ribeiro da Silva[3]

[1]University Of San Carlos of Guatemala, Faculty of Chemistry and Pharmacy, Guatemala, Guatemala, [2]University Of San Carlos of Guatemala, Faculty of Agronomy, Guatemala, Guatemala, [3]Federal University of Rio de Janeiro, Rio de Janeiro, Brazil

Stevia serrata Cav. (Asteraceae) is a perennial herb found usually over 1500 m in the western highlands of Guatemala [1]. The intense blue essential oil of the *S. serrata* leaves contains chamazulene, *β*-caryophyllene and germacrene D as major components and showed an important antinociceptive activity and potential to develop anti-inflammatory products [2]. In the present study, the essential oil of leaves and roots of *S. serrata* collected in September 2017 in a locality of western Guatemala (Sipacapa, department of San Marcos) was analyzed, with the purpose of comparing the composition of the oil of both parts of the plant because the authors had references from the local population that infusions of the plant were used to treat stomach diseases. The essential oils of dried leaves and roots of *S. serrata* were obtained separately by means of hydrodistillation with a Clevenger apparatus, obtaining a yield of 0.2% for leaves and 0.2% for roots. The oils were analyzed by GC-FID and GC/MS, and showed a high content of sesquiterpenes, being germacrene D (37.6%), chamazulene (31.5%) and *β*-caryophyllene (8.0%) being the major components of the oil from leaves and *α*-longipinene (23.5%), 1*R*,3*Z*,9*s*-4,11,11-trimethyl-8-methyllenebicyclo[7.2.0]undec-3-ene (22.2%) and santolina triene (12.6%), the major components of the oil from roots. The essential oil of the leaves of *S. serrata* from Sipacapa analyzed in this study had a lower content of chamazulene than the oil from Sololá analyzed by Simas *et al*. [2]. This is the first report on the composition of the essential oil of roots of S. serrata, and the results should be useful as baseline for the management of the species. Further studies regarding antimicrobial activity are suggested to evaluate possible uses of the oil as potential raw material for therapeutic products.

Acknowledgments: SENACYT, Guatemala (FINDECYT/FODECYT 17-2017).

Keywords: Chamazulene, *α*-longipinene, essential oil, *Stevia serrata*

PS1-A-044

Response of dry yield and saponin content of some saponin containing plants to planting media and drying methods

Salah Eldeen M Mahmoud[1], Halla M Gamal El-Dein[3], S M Abou-Shleel[2], Marwa M Darweesh[3]

[1]Horticulture Dep., Fac. of Agric., Al-Azhar Univ., Nasr City, Cairo, Egypt, [2]Environment and Bio-Agricultural Dep., Fac. of Agric., Al-Azhar Univ., Cairo, Egypt, [3]Biological and Environmental Sciences Dep., Fac. of Home Economy, Al-Azhar Univ., Tanta, Egypt

This study was carried out at the experimental farm of the Environmental and Bio-Agricultural Department, Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo, Egypt and the farm and laboratories of the Biological and Environmental Science Department, Faculty of Home Economics, Al-Azhar University, Tanta, Egypt. The experiment was divided into two parts: -
I- Three different types of soil (Clay, Sand, Mixed 1:1 Clay: Sand) were used for growing of both verbascum and fennugreek plants. Three groups of Pots of 25 cm size were prepared with the above mentinal soils in rate of 20 pots for each treatment. The seeds were sown on (October, 1st) for both the two plants.
II- Different drying methods were used for verbascum leaves and flowers as well as fenugreek herb and seeds. The samples were dried in shade, sun, and ovens at temperature of 55, 65 and 75 for two days (until the dry weight was fixed).
Using growing media of Clay soil produced the higher values of growth, flowering and yield of both *Verbascum thapsus* L. and *Trigonella foenum-graecum* L. plants. Verbascum plants which were cultivated in sand soil improved saponin contents, while using mixture soil improved fenugreek saponin contents at both of the experimental seasons. Furthermore, drying plant material of verbascum and fenugreek in shade recorded the highest value of dry matter and saponin content, while drying in oven at 75 °C recorded the lowest value in all tested samples that collected from plants grown in the different types of soils.

PS1-A-045

Antiplasmodial and antitryponosomal activities of extracts, fractions and compounds from *Beilschmiedia* spp

Waleguele Christine Claire[1,2], Lenta Ndjakou Bruno[1], Tsamo Etienne Voltaire[1], Rui Krause[2]

[1]University Of Yaounde I, Yaounde, Cameroon, [2]Rhodes University, Grahamstown, South Africa

Malaria and trypanosomiasis are major cause of mortality and morbidity in many tropical countries in the world. The control and treatment of these infections continue to be complicated by development of parasite resistance strains. The situation is becoming worrisome as there is still no effective vaccine for malaria and trypanosomiasis. Therefore, there is an urgent need to search for new and more efficient drug for management of these diseases. Medicinal plants have always been used for decade for treatment of malaria in endemic areas. Indeed, plants of the Lauraceae family are used in Cameroonian traditional medicine to treat parasitic diseases like malaria and trypanosomiasis. Therefore, this study was designed to evaluate the antiplasmodial and antitrypanosomal potentials of extracts, fractions and compounds from *Beilschmiedia* spp from Cameroon. Methanol extracts of leaves, root, barks and stem of each plant species were prepared by maceration. Extracts were therefore fractionated by column chromatography using solvent with increase polarity from hexane, hexane-ethyl acetate, and ethyl acetate-methanol to methanol. 50 fractions pooled according to their TLC profile were collected and evaluated for their *in vitro* efficacy against the chloroquine sensitive strain of *Plasmodium falciparum* using syber green assay and pentamidine sensitive strain of *Trypanosoma brucei*. From results obtained, around 66.66% of extracts were inactive against Pf3D7 and 15,22% against *Trypanosoma brucei*, while 33.33% of gave from 50 to 100% to malaria and 74,78 to trypanosomiasis. Seven extracts were the most potent with IC_{50} ranged from 19-77.25µg/ml to malaria and eleven to trypanosomiasis with IC_{50} ranged from 1-26.25µg/ml. The results from this study suggest that extracts from *Beilschmiedia* spp are potential sources of antiparasitics diseases like malaria and trypanosomiasis compounds.

Keywords: *Beilschmiedia* spp, malaria, trypanosomiasis, *Plasmodium falciparum*, antiplasmodial, antitrypamosomal.

PS1-A-046

New phenylethanoid glycosides from *Cistanche phelypaea* and their activity as inhibitors of monoacylglycerol lipase

Khadidja Aya Beladjila[1], Djemaa Berrehal Berrehal[1], Assia Khalfallah[1], Nunziatina De Tommasi[3], Carlotta Granchi[2], Marinella De Leo[2,4], Giulia Bononi[2], Alessandra Braca[2,4], Zahia Kabouche[1]

[1]Département de Chimie, Université des Frères Mentouri-Constantine, Laboratoire d'Obtention des Substances Thérapeutiques (LOST), Campus Chaabet-Ersas, 25000 Constantine, Algerie, [2]Dipartimento di Farmacia, Università di Pisa, via Bonanno 33, 56126 Pisa, Italy, [3]Dipartimento di Farmacia, Università di Salerno, via Ponte Don Melillo, 84084 Fisciano, Italy, [4]Centro Interdipartimentale di Ricerca "Nutraceutica e Alimentazione per la Salute", Università di Pisa, via del Borghetto 80 56124 Pisa, Italy

Cistanche genus, belonging to Orobanchaceae family, comprises about 20 species, distributed in arid and semi-arid regions of Africa [1]. Several *Cistanche* species are used in the traditional Chinese medicine as tonic for treatment of different diseases [2]. In the course of our studies, we carried out both chemical study of *Cistanche phelypaea* (L.) *Coutinho* from Algeria, and the inhibitory activity of isolated compounds on human lactate dehydrogenase (LDH) and monoacylglycerol lipase (MAGL), enzymes involved in the peculiar glycolytic or lipidic metabolism of cancer cells. The phytochemical study of *C. phelypaea* aerial parts, afforded the isolation of four new phenylethanoid glycosides (1-4), 1-*β*-*p*-hydroxyphenyl-ethyl-2-*O*-acetyl-3,6-di-*α*-L-rhamnopyranosyl-*β*-D-glucopyranoside (1), 1-*β*-*p*-hydroxyphenyl-ethyl-3,6-*O*-di-*α*-L-rhamnopyranosyl-*β*-D-glucopyranoside (2), 1-*β*-*p*-hydroxyphenyl-ethyl-2-*O*-acetyl-3,6-di-*α*-L-rhamnopyranosyl-4-*p*-coumaroyl-*β*-D-glucopyranoside (3), and 1-*β*-*p*-hydroxyphenyl-ethyl-3,6-di-*α*-L-rhamnopyranosyl-4-*p*-coumaroyl-*β*-D-glucopyranoside (4), together with three known compounds, named brandioside, pinoresinol 4-*O*-*β*-D-glucopyranoside, and apigenin 7-*O*-*β*-glucuronopyranoside, were isolated from the n-butanol extract, after submission to column chromatography over Sephadex LH-20, followed by RP-HPLC. The structures of all compounds were determined by spectroscopic analysis, including NMR and HRESI-MS experiments.

All the compounds showed negligible activity on LDH, whereas some of them displayed a certain inhibition activity on MAGL. In particular, compound 1 was the most active on MAGL showing an IC_{50}= 88.0 μM, and modeling studies rationalized the supposed binding mode of compound 1 in the MAGL active site.

Acknowledgements: The authors are grateful to Algerian Ministry of Higher Education and Research (MESR) for the PNE grant to Miss Khadidja Aya Beladjila.

Keywords: *Cistanche phelypaea*, Orobanchaceae, phenylethanoid glycosides, monoacylglycerol lipase, lactate dehydrogenase.

References:

[1] Fahmy GM, El-Tantawi H, Abd El-Ghani MM. J Arid Environ 1996; 34: 263–276.

[2] Wang LI, Ding H, Yu HS, Han LF, Lai QH, Zhang LJ, Song XB. Chin Herb Med 2015; 7: 135–142.

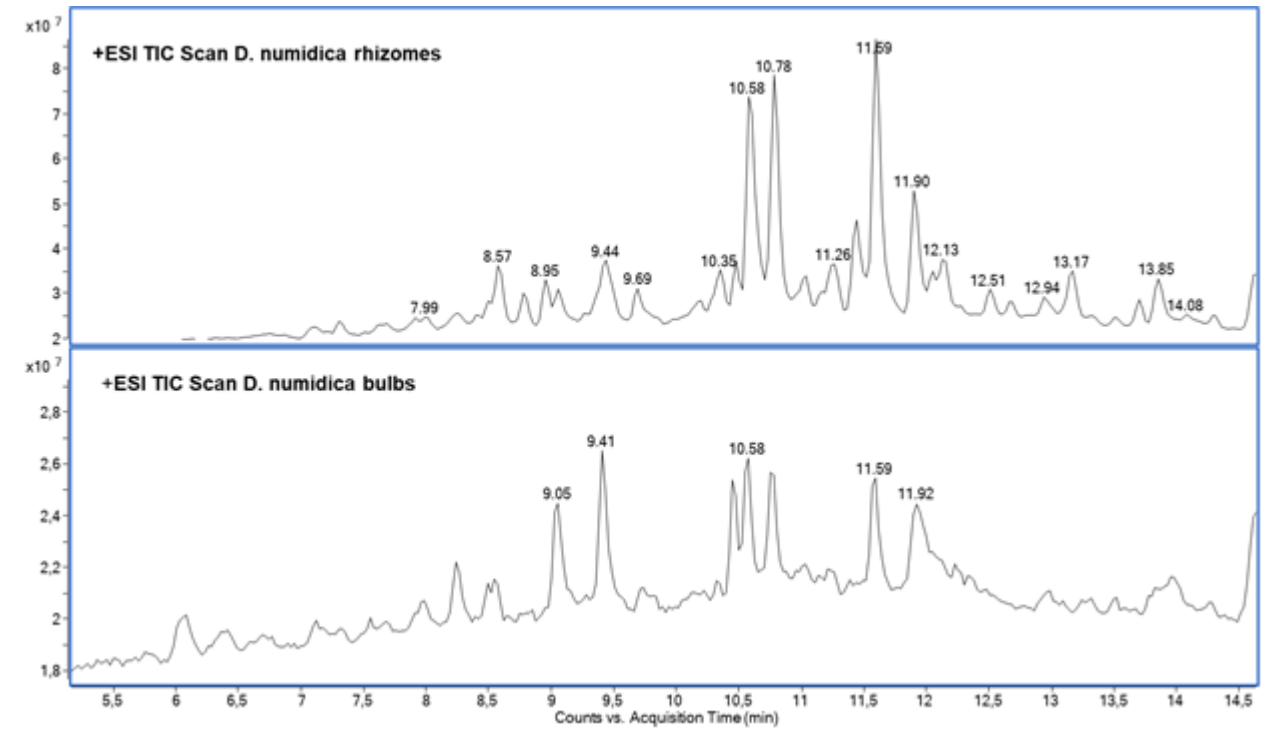
PS1-A-047

Phytochemical screening of *Drimia numidica* bulbs and rhizomes

Eleni Kakouri, Charalabos Kanakis, Panayiotis Trigas, Petros Tarantilis

Agricultural University of Athens, Athens, Greece

Drimia numidica is a bulbus plant known for its cardiotonic properties owed to its main constituents, the bufadienolides, compounds that belong to cardiac glycosides. Bufadienolides can be isolated either from animals of the genus Bufo or from plants. Natural sources of bufadienolides are the plants that belong to Crassulaceae, Hyacinthaceae, Iridaceae, Melianthaceae, Ranunculaceae and Santalaceae families. In particular, they are found in abundance at the Crassulaceae and Hyacinthaceae families (1). Bufadienolides inhibit Na-K adenosine triphosphatase (ATPase) activity in the myocardial cell membrane and consequently exert a positive inotropic action. They can also decrease heart rate, an activity described as negative chronotropic action (2).
Many studies performed on different species of the plant, focus on the chemical analysis of the bulb, which is considered the main source of bufadienolides. In this study we focused on the chemical analysis of bulbs and rhizomes of *D. numidica* (Jord. & Four) J.C, Manning & Goldblatt, with liquid chromatography combined with a time-of flight high resolution mass spectrometry (LC/Q-TOF/HRMS). The HPLC analysis was performed using a solvent system of water LC/MS and acetonitrile LC/MS both with 0.1% formic acid. Chromatograms were recorded at 280,360,440 nm. The Q-TOF mass spectrometer was operated with a dual ESI source in the positive ionization mode. Results of the chemical analysis revealed variations regarding the chemical composition of the underground part. The presence of bufadienolides at the extract of rhizomes was certainly more intense respect to that of bulbs.



PS1-A-048

Quantification of grape seed oils' FAMEs of varieties traditionally cultivated in the Ionian islands

Nefeli Sofia Sotiropoulou[1], Ioanna Oikonomou[1], Dimitra Daferera[1], Efstathia Skotti[2], Christos Pappas[1], Petros Tarantilis[1]

[1]Laboratory of Chemistry, Department of Food Science and Nutrition, Agricultural University of Athens, Athens, Greece, [2]Technological Educational Institute of the Ionian Islands, Department of Food Technology, Argostoli, Greece

Grape seeds are comprising part of the pomace produced by the wineries and are important agroindustrial waste [1]. Grape seeds are involved only in the red winemaking. Otherwise, they are immediately discarded, mixed with the skins and the rest of the pulp. Though seeds are a valuable source of bioactive constituents, essential to the human body. Oily compounds are some of them, like linoleic fatty acid. In this study, the fatty acids methyl esters (FAMEs) originated from different grape varieties traditionally cultivated in the Ionian Islands were analyzed. The oil was extracted by Soxhlet apparatus and *n*-hexane used as a solvent. The FAMEs acquired were analyzed by Gas Chromatography-Mass Spectrometry (GC-MS), after the oil's transesterification with methanol. Decanoic acid methyl ester was served as an internal standard for the quantification process. FAME analysis is indicating that the oils are rich in unsaturated fatty acids, dominated by linoleic fatty acid, ranging from 53.68 to 60.95 % and followed by oleic fatty acid, ranging from 17.55 to 25.44 %. FAMEs expressed both in the oil's concentration % w/w and in the fresh weight's concentration w/w. Fatty acid ratios obtained from GC-MS analysis seem to differentiate each variety tested, according to other researchers as well [2]. Wineries waste reclamation can contribute as a source of important health agents, that can be easily quantified and the reduction of the agro-industrial waste accomplished is in favor of the environment.

Keywords: grape seed oil, FAME, GC-MS, quantification, internal standard

References:

[1] Food and Agricultural Organization (29 July 2018), FAOSTAT, Retrieved from URL: http://www.fao.org/faostat/en/

[2] Lachman J, Hejtmánková A, Táborský J, Kotíkova Z, Pivec V, Stralkova R, Vollmannova A, Bojnanska T, Dědina M. J. LWT-Food Sci. Technol. 2015; 63: 620–625.

PS1-A-049

Chemical constituents isolated from the leaves and the flowers of *Achillea grandifolia* Friv. (Asteraceae)

Olga Tsiftsoglou, Christos Gounaris, Christina Papitsa, Maria Nanouli, Diamanto Lazari

Aristotle University of Thessaloniki, Thessaloniki, Greece

A. grandifolia represent one of about 130 species of the genus *Achillea* L., which belongs to family Asteraceae (Compositae), the largest family of vascular plants distributed throughout the world, and is widespread in Europe, North America and temperate areas of Asia [1]. Many *Achillea* species have been used in alternative treatments, especially for wound healing, abdominal pain, against diarrhea and flatulence, and for their diuretic and emmenagogue action [2]. In present study, leaves and flowers of *Achillea grandifolia* Friv. (Asteraceae) were examined for their content in nonvolatile secondary metabolites. So far, eleven secondary metabolites have been isolated using chromatographic methods: six sesquiterpene lactones, two guaianolides: rupicolin A (1) and rupicolin B (2), three eudesmanolides: artecalin (3), ridentin B (4) and ligustolide A (5) and one sesquiterpene methyl ester: (1*S*,2*S*,4*αR*,5*R*,8*R*,8*αS*)-decahydro-1,5,8-trihydroxy-4*α*,8-dimethyl–methylene-2-naphthaleneacetic acid methylester (6), and five flavonoids: luteolin (7), luteolin-7-*O*-glucoside (8), apigenin (9), apigenin-7-*O*-glucoside (10) and eupatolitin (11). They were identified using spectroscopic methods UV/Vis and NMR (^{1}H, ^{13}C, COSY, ROESY, NOESY, HMQC, HMBC).

Acknowledgements: We acknowledge support of this work by the project "Upgrading the Plant Capital (PlantUp)" (MIS 5002803) which is implemented under the Action "Reinforcement of the Research and Innovation Infrastructure", funded by the Operational Programme "Competitiveness, Entrepreneurship and Innovation" (NSRF 2014-2020) and co-financed by Greece and the European Union (European Regional Development Fund).

Keywords: *A. grandifolia*, Asteraceae, sesquiterpene lactones, flavonoids

References:

[1] Nemeth, E. Israel J. Plant Sci 2010; 58: 279–289
[2] Saeidnia S, Gohari A R, Mokhber-Dezfuli N, Kiuchi F. DARU 2011; 19 (3): 173–186.

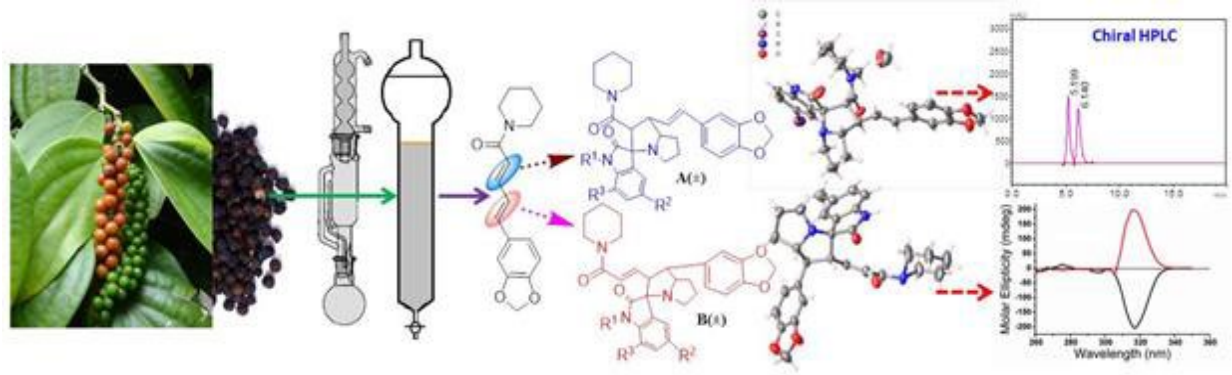
PS1-A-050

Azomethine ylide cycloaddition: A versatile way of semi–synthetic modification towards novel spirooxindolo analogues of piperine

Meenakshi Singh

National Institute of Pharmaceutical Education and Research– Kolkata, (NIPER–K) India, Kolkata, India

Spirooxindoles are notable synthetic targets with its unique three-dimensional structures and conformational restriction imparted by the spiro carbon. Moreover, this framework is often considered to be a privileged motif with respect to their broad biological activities and applications in pharmaceutical lead discovery. This thesis entitled "Azomethine ylide cycloaddition: A versatile way of semi–synthetic modification towards novel spirooxindolo analogues of piperine" depicts the synthesis of a series of twentytwo novel spirooxindolo pyrrolizidine adducts of alkaloid piperine via an azomethine ylide cycloaddition reaction using secondary amino acid proline and isatins as the reagents. The main rationale behind using natural product piperine as a core structure for this semisynthetic modification is due to its superabundance of biological activities and medicinal importance. The structural modification by azomethine ylide cycloaddition resulted in two regioisomeric products in racemic nature with good yields. The products were characterized by extensive 1D/2D NMR analysis, IR spectroscopy and single–crystal X–ray crystallographic studies. The enantiomers of two racemic regioisomers formed in a reaction were separated by HPLC on a Chiralcel OD–H column and were indeed confirmed by the CD spectra of the separated enantiomers.



PS1-A-051

Gomphrenins stability during tea brewing of purple flowers of *Gomphrena globosa* L.

Natalia Szmyr, Sławomir Wybraniec

Department of Analytical Chemistry, Cracow University of Technology, Cracow, Poland

Dried flowers of *Gomphrena globosa* L. (Amaranthaceae) are commonly used for preparation of tea infusions in China. Nowadays, the number of people suffering from civilization diseases, such as diabetes, hypertension, as well as cancer, is increasing. Therefore, natural products comprising of high biological activity compounds attract much attention. These products, implemented in daily diet can effectively prevent or retard generation of such health problems. The violet *Gomphrena globos*a L. inflorescences are a precious source of many antioxidants involving betalains. These compounds are vacuolar plant pigments occurring in plants of Caryophyllales order. Betacyanins acylated by hydroxycinnamic acid derivatives are present in gomphrena flowers in relatively high levels. The main pigments are trivially denominated as gomphrenins [1]. Unfortunately, these natural colorants have relatively low stability and degrade in certain conditions.

The current study aims at investigation of the extraction of betacyanins occuring in *Gomphrena globosa* L. flowers in course of tea brewing with and without citric acid addition and preliminary determination of betacyanin degradation products. Previously isolated single diastereomers of gomphrenin were used for additional heating experiments for preliminary determination of degradation products. In each tea brewing experiment, one flower was extracted at 90 °C in 3 ml of solvent (pure water or 1% aqueous citric acid). The samples were collected within 60 min of heating for analysis by high–performance liquid chromatography with diode array and mass spectrometric detection (LC–DAD–ESI–MS).

Acknowledgements: This research was financed by National Science Centre, Poland, for years 2018–2021 (Project No. UMO–2017/27/B/NZ9/02831).

Keywords: gomphrenins, tea brewing, *Gomphrena globosa* L., betacyanins, stability, LC–DAD–ESI–MS.

References:

[1] Kugler F, Stintzing FC, Carle R. Anal Bioanal Chem 2007; 387: 637–648.

PS1-A-052

Acylated gomphrenins and their stability in *Gomphrena globosa L.* extracts

Natalia Szmyr, Sławomir Wybraniec

Department of Analytical Chemistry, Cracow University of Technology, Warszawska 24, 31–155, Poland

Gomphrenins belong to betacyanins which are a subgroup of betalains. Betacyanins are water–soluble, vacuolar plant pigments belonging to families of Caryophyllales plant order [1]. Due to their numerous pro–health properties, they can potentially find applications in the pharmaceutical industry and as food colorants [2]. Purple *Gomphrena globosa* L. blossoms are a precious source of betacyanins acylated by hydroxycinnamic acid derivatives. The most known betacyanin source used on a commercial scale is red beet root (*Beta vulgaris* L.). However, it contains relatively poor betalain profile and undesirable flavor, therefore, alternative sources of betalains are searched for. However, as most of natural products, betacyanins are less stable than synthetic dyes, therefore, studies need to be carried out on their stability as well as on determination of their degradation products.
In this study, the degradation products of acylated gomphrenins extracted from purple *Gomphrena globosa* L. flowers were formed in the course of extract heating at 90 °C in aqueous and ethanolic solutions at selected pH values. The samples of the solutions were collected for analysis within the 60 min process of reaction. An impact of applied solvents on gomphrenins degradation rate was investigated. High–performance liquid chromatography coupled to electrospray ionization mass spectrometry (LC–DAD–ESI–MS) was applied in order to tentatively identify the resulting product mixtures of decarboxylated and dehydrogenated gomphrenin derivatives.

Acknowledgements: This research was financed by National Science Centre, Poland, for years 2018–2021 (Project No. UMO–2017/27/B/NZ9/02831)

Keywords: acylated gomphrenins, *Gomphrena globosa L.*, betacyanins, stability

References:

[1] Strack D, Vogt T, Schliemann W. Phytochemistry 2003; 62:247–269
[2] Esatbeyoglu T, Wagner AE, Motafakkerazad R, Nakajima Y, Matsugo S, Rimbach R. Food Chem Toxicol 2014; 73: 119–126

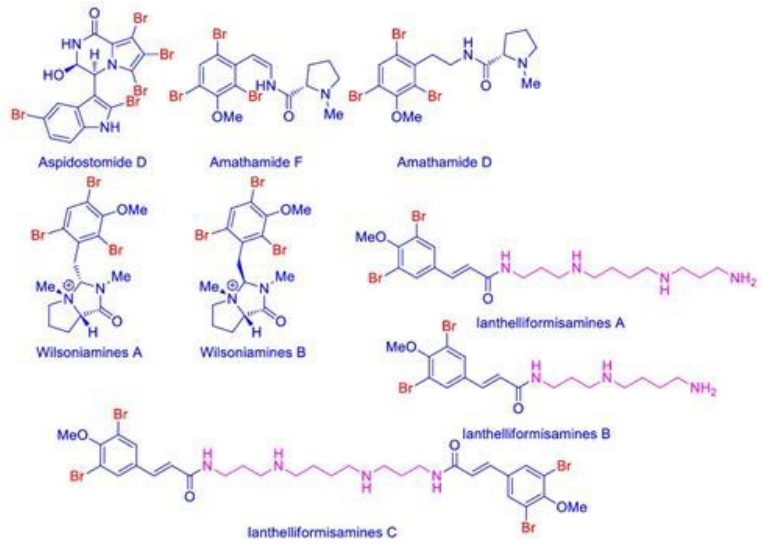
PS1-A-053

Synthesis of a regioisomeric *N*–methyl aspidostomide D, its derivatives via Lewis acid mediated epoxide opening and their antibacterial assessment

Mulla Althafh Hussain[1],Althafh Hussain Mulla[2], Amhed Khan[2]

[1]ACADEMIC institution, Department of Chemistry, Indian Institute of Technology, Hyderabad, India, [2]Department of Chemistry, Indian Institute of Technology Hyderabad, Kandi, Sangareddy, 502285, Telangana, India.

The marine atmosphere has established to be a very rich source of tremendously potent compounds that have demonstrated significant biological activities such as antitumor, anti–inflammatory, antibacterial, analgesia, immunomodulation, allergy, and antiviral assays [1]. Because of these dominant activities we became interested in synthesis of marine natural products, and reported the syntheses marine natural products wilsoniamines A and B [2] amathamide D and F,2 ianthelliformisamines A and their analogues [3]. In this presentation, we will disclose our present efforts towards the synthesis of aspidostomide D, [4] which led to the synthesis of regioisomeric *N*–methylated aspidostomide D. Through epoxide opening strategy.



References:

[1] David J, Newman Gordan M, Cragg. J Nat prod 2004; 67: 216.
[2] (a) Khan FA, Ahmad S. Tetrahedron Lett. 2013; 54: 2996. (b) Khan FA, Ahmad SJ. Org Chem 2012; 77: 2389. (c) Ahmad S, Choudhury S, Khan FA. Tetrahedron 2015; 71:4192.
[3] Khan FA, Ahmad S, Kodipelli N, Shivange G, Anindya R. Org Biomol Chem2014; 12: 3865.
[4] a) b) Patino CLP, Muniain C, Knott ME, Puricelli L, Palermo JA. J Nat prod 2014; 77: 1170.

PS1-A-054

Structural correction of a dimeric phthalide with progesterone–like activity

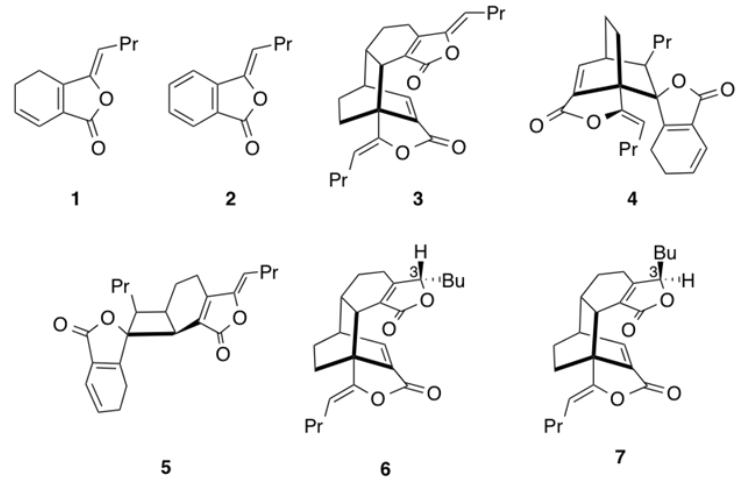
José Luis Ávila, Dr. Guillermo Delgado

Instituto de Química, Universidad Nacional Autónoma de México, Ciudad Universitaria, Coyoacán 04510, Mexico

Ligusticum porteri (Apiaceae) is a plant used in Northern Mexico for the treatment of stomachache, colds and in rituals [1]. Phthalides, such as (*Z*)–ligustilide (1), (*Z*)–butylidenephthalide (2), diligustilide (3), tokinolide B (4) and riligustilide (5), have been isolated from this species [2,3]. On the other hand, *Ligusticum chuanxiong* is used in Traditional Chinese Medicine for menstrual disorders. Compounds 1–6 were isolated from it, identifying 6 as a progestin almost as potent as progesterone [4].

Previous studies on the reactivity of 3 led to 6, when it was submitted to catalytic hydrogenation [5], but spectroscopic constants reported for 6 isolated as natural product or prepared semisynthetically are different [4,5]. It led us to hypothesize that the structure of the natural product was the epimer 7.

In order to prove it, compound 3 was hydrogenated, confirming the structure 6 through X–ray crystallography. After epimerization with DBU, compound 7 was obtained and its NMR constants were consistent with those reported for L. chuanxiong metabolite. Consequently, the structure for the compound with remarkable progestagenic activity is 7 [4].



Acknowledgements: The authors acknowledge Conacyt (grant no. 294731), DGAPA UNAM (PAPIIT IG200318), and Programa de Maestría y Doctorado en Ciencias Químicas (UNAM) for financial support.

Keywords: Phthalides, progestins, *Ligusticum porteri*

References:

[1] Bye R A, Linares E. J Ethnobiol 1986; 6: 289–306.
[2] León A, Del–Ángel M, Ávila JL, Delgado G. Prog Chem Org Nat Prod 2017; 104: 127–246.
[3] León A, Chávez MI, Delgado G. Magn Reson Chem 2011; 49 (8): 469–476.
[4] Lim LS, Shen P, Gong YH, Yong EL. Phytochemistry 20061; 67 (7): 728–734.
[5] Delgado G, Reza–Garduño RG, Toscano RA, Bye R, Linares E. Heterocycles 1988; 27 (6): 1305–1312.

PS1-A-055

Qualitative composition of volatile constituents in three wild grown species of aromatic plants commercially known as oregano

Paraskevi Yfanti[1,2], Eleni Leneti[2], George Patakioutas[2], Dimitra Douma[2], Marilena Lekka[1]

[1]Department of Chemistry, University of Ioannina, University campus, P.C. 45110, Ioannina, Greece, [2]Department of Agricultural Technology, Technological Education Institute of Epirus, Kostakii, P.C. 47100, Arta, Greece

Aromatic plants commercially known as "Oregano", have an important economic value [1]. The aim of the present study was to investigate the essential oil profile of three plant species with "Oregano" odor, wild grown in the municipality Ziros, Epirus, Greece. The selected species *Origanum vulgare* ssp. *hirtum*, *Coridothymus capitatus* and *Satureja horvatii* spp. *macrophylla* (Lamiaceae) were collected at the flowering stage. The essential oil obtained by hydro–distillation using a Clevenger type apparatus, was chromatographically analyzed by a Gas Chromatography–Mass Spectrometry (GC–MS) system equipped with a capillary column BPX–5 (30m x 0.25mm x 0.25μm). Carrier gas was Helium (He) at a flow rate of 0.7 mL min^{-1} and split ratio 1:30. Oven temperature program was 60 to 110 °C at 3 °C min^{-1}, 110 °C isothermal for 10 min, 110 to 150 °C at 3 °C min^{-1}, 150 to 280 °C at 30 °C min^{-1}, 280 °C isothermal for 5 min. The MS operating parameters were: ionization potential 70 eV, ion source temperature 200 °C, acquisition rate 5 spectra s^{-1} and mass range from 50 to 550 *m/z*.
All three species are characterized by a high content of essential oil (*O. vulgare* ssp. *hirtum* 4,8 %, *C. capitatus* 2,9 %, *S. horvatii* spp. *macrophylla* 1,5% dw). Twentynine components were indentified in *O. vulgare* ssp *hirtum*, twentyfour *C. capitatus* and forty in *S. horvatii* ssp. *macrophylla* essential oil. The oxygenated monoterpenes comprised a large portion, among which carvacrol (66,94 %, 74,78 %, 43,97 respectively) was the major one. The results suggest that the populations of the three aromatic plants tested are characterized by high content of essential oil which belongs to carvacrol chemotype.

Acknowledgements: to the OPENSCREEN–GR network

Keywords: essential oil, GC–MS

References:
[1] Kokkini S, Vokou D. Flavor Fragrance J 1989; 4: 1–7.

PS1-A-056

Isolation, identification and investigation of the antioxidant activities of compound from *Costus afer*

Victor Fadipe

Federal Ministry of Science and Technology, Abuja, Abuja, Nigeria, [2]Raw Material Research and Development Council, Abuja, Nigeria, Abuja, Nigeria, [3]International Centre for Chemical and Biological Science, University of Karachi, Pakistan, Karachi, Pakistan

Costus afer has been used in foods and drugs in the treatment of various diseases in Nigeria and some West African Countries. The objective of the research is to Isolate and identify bioactive compounds from *Costus afer* and also investigate their antioxidant activities.
The aerial part of *C. afer* was dried and powdered (1.0 kg ±2.0 g), and then extracted with hexane methanol. Solvent (3 × 1 L) was used for each extraction for a week at room temperature. The crude methanol extract was dried and re–dissolved in methanol and then filtered and subsequently dried. The methanol extract subjected to chromatography to isolate the active compounds. 10 fractions were obtained after pooling with TLC. The fractions were subjected to antioxidant activity using DPPH and Superoxide anion radical scavenger. The fraction with the highest value of DPPH was further subjected to chromatograph on recycling HPLC. A single compound was isolated and the structures of isolated compound were elucidated by 1 D, 2 D NMR and LC–TOF/MS. The obtained spectroscopic data was compared with the values from the literatures and the compound was identified as Harmine (1). Harmine (1) has the most antioxidant effects on all assays and this has to do with the chemical structure of the compound bearing the acidic protons. Harmine is an ubitiqous medicinal compound, a beta–carboline alkaloid, that is widely distributed in the plants, marine creatures, insects, mammalians. Harmine was originally first isolated from seeds of *Peganum harmal*. The information above validate the use of the plant as foods and drugs. The isolation of harmine from *C. afer*, indicate that the plant may serves as alternative source for the compound. Research collaboration involving biotechnologist, botanist and chemist may as well provide opportunity to come up with species of the plant that are rich in the compound.

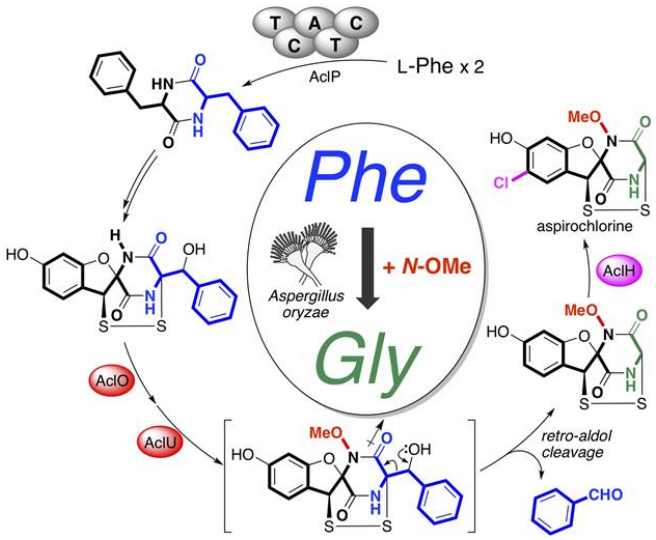
PS1-A-057

Revealing an unprecedented biosynthetic pathway in *Aspergillus oryzae*

Naoya Maeda, Dr. Yuta Tsunematsu, Dr. Kenji Watanabe

University of Shizuoka, Shizuoka, Japan

Aspirochlorine is an unusual antifungal cyclopeptide produced by *Aspergillus oryzae*, an important mold used for food fermentation. Not like other epidiketodithiopiperazine(ETP) class natural products, this small molecule bears an untypical bicyclo[3.2.2] ring system with a spiro–carbon. Furthermore, whereas its structure suggested that a non–ribosomal peptide synthetase assembles the cyclopeptide from phenylalanine and glycine building blocks, labeling studies indicated that one Phe moiety is transformed into Gly after peptide formation. By means of genetic engineering, heterologous expression, biotransformations, and *in vitro* assays we dissected and reconstituted four crucial steps in aspirochlorine biosynthesis that involve two cytochrome P450 monooxygenases, (AclL and AclO), a methyltransferase (AclU), and a halogenase (AclH). We found that the installation of the *N*–methoxylation of the peptide bond sets the stage for a retro–aldol reaction that leads to the Phe–to–Gly conversion. We demonstrated that a seemingly small change may lead to a substantial modification of the carbon framework in a peptide. As this amino acid conversion is a prerequisite of antifungal activity of the natural product, this scenario represents an unusual case of natural lead optimization by peptide editing, which markedly differs from classical amino acid exchanges involving codon changes or altered adenylation domains.
We are currently looking for an enzyme, which is responsible for the formation of the spiro ring in aspirochlorine pathway. We identified a candidate gene aclX, due to a loss of the production of spiro ring formed metabolites in aclX–deficient mutant of *A. oryzae*. Current efforts to isolate and to characterize a precursor of the spiro ring formed metabolite as a substrate of enzymatic reaction of AclX will also be presented.



PS1-A-058

Biochemistry analysis and physicochemical properties for the quality assessment of the Greek honey bee

Dimitrios Kafetzopoulos[1,2], Chrissi Petraki[2], Georgios Kafetzopoulos[3]

[1]University of the Aegean, Myrina, Lemnos, Greece, [2]Hellenic Open University, Patra, Greece, [3]University of Thessaly, Magnisia, Volos, Greece

The present study was carried out to determine the physicochemical properties and biochemical activities of the Greek honey in order to identify possible deviations or adulterations and the level of conformance with quality parameters set by the law. For this reason, 119 honey samples were randomly collected from different markets in Greece, coming from three different botanical origins (thyme, pine and orange blossom). The quality criteria of the product with regard to its composition, macroscopic and microscopic appearance are analyzed. Each criterion whether considered alone or in combination with others provides information on the beekeeping practices followed and on its plant origin. Composition analyses of water content, sugar concentration, Hydroxymethylfurfural (HMF) concentration, High Fructose Corn Syrup (HFCS) and pollen analysis, were determined. Moreover, PH, Electrical Conductivity (EC) and activity of the enzyme diastase were measured using the method appropriate for each case. In addition, biochemical analyses for total phenolic content and total flavonoid content were determined in 7 samples of honey from each botanical origin in order to characterize Greek honeys. The analysis and evaluation of the laboratory results indicate that from the 119 samples tested, 10 of them (8.4 %) were found to be non-compliant in relation to EC, HMF, moisture content and sugar composition while 3.1% was non-compliant regarding the botanical origin. From the 21 samples, thyme honeys contain the highest amount of total phenolic substances while orange honeys have the highest flavonoid content. The results of this study allow us to assess the level of adulterations and the quality of Greek honeys so that those involved in the beekeeping industry can be informed about ways to prevent the occurrence of variations. The importance of controlling this traditional product will help not only to safeguard the interests of consumers and producers, but also to protect its quality.

PS1-A-059

Pharmaceutical formulations of *Capsicum frutescens* extracts

Enkelejda Goci[1], Entela Haloci[2]

[1]Aldent University, Tirane, Albania, [2]University of Medicine, Tirana, Tirane, Albania

Capsicum species are native plants of America, where they have been cultivated for thousands of years by Native Americans of tropical America and are now cultivated worldwide. Capsaicin is the main capsaicinoid of chili peppers, followed by dihydrocapsaicin. Local capsaicin creams are used for therapeutic purposes in the treatment of peripheral pain in pathologies such as rheumatoid arthritis, diabetic neuropathy, osteoarthritis, muscular pain, herpes, etc.
The purpose of this study is the pharmaceutical formulation of topical preparations 0.025% of alcoholic extracts of chili pepper on different bases and evaluation of their physico-chemical characteristics.
Materials and Methods: Medical herbal drugs were undergone to pharmacopoeial requirements on humidity and ash content. The alcoholic extract obtained from the maceration was included on hydrophilic bases of macromolecular polymers and lipophilic bases.
The obtained preparations underwent an evaluation of organoleptic control, rheological control, pH determination and capsaicin identification.
Results and Conclusions: The herbal drug fulfils the pharmacopoeia requirements according to the allowed percentage of moisture and total ash content. The hydrophilic preparations presented good characteristics of homogeneity, high spreadability capacity and pH values (6.7 – 6.87) within the limits allowed for dermatological preparations in comparison with lipophilic ointment.

PS1-A-060

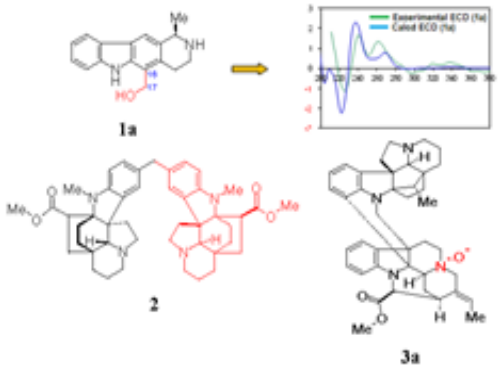
Indole alkaloids from medicinal plants of Cameroon with cancer chemopreventive activity

Joseph Thierry Ndongo[1], Josephine Ngo Mbing[2], Muriel Cuendet[3], Dieudonné Emmanuel Pegnyemb[2], Hartmut Laatsch[4]

[1]Department of Chemistry, Higher Teacher Training College, University of Yaoundé 1, P.O. Box 47, Yaoundé, Cameroon, Yaoundé, Cameroon, [2]Department of Organic Chemistry, Faculty of Science, University of Yaoundé 1, P.O. Box 812, Yaoundé, Cameroon, Yaoundé, Cameroon, [3]School of Pharmaceutical Sciences, University of Geneva, University of Lausanne, Rue Michel-Servet 1, CH-1211 Genève 4, Switzerland, Geneva, Switzerland, [4]Institute of Organic and Biomolecular Chemistry, University of Göttingen, Tammannstrasse 2, D-37077 Göttingen, Germany, Göttingen, Germany

My research aim is to identify indole alkaloids from medicinal plants of Cameroon, using modern techniques of isolation of natural products, including Molecular Networking as a Dereplication Strategy and methods of 1D and 2D NMR, and mass spectrometry. The isolated compounds will be tested for cytotoxicity; active hits will be submitted to pharmacological studies for the development of anticancer drugs.
In our present study, janetinine (1a), pleiokomenine A (2), and huncaniterine B (3a), and 13 other alkaloids were isolated from the stem bark of *Pleiocarpa pycnantha*. Janetinine (1a) is a carbazole alkaloid; in pleiokomenine A (2), two aspidofractinine-type alkaloids are bridged by a methylene unit, and huncaniterine B (3a) is a pleiocarpamine–aspidofractinine-type dimer. The structures and relative configurations of these compounds were elucidated on the basis of the mentioned methods above. The absolute configuration of these compounds was determined by comparison of experimental and calculated ECD spectra. The cancer chemopreventive activity of the isolated compounds was evaluated through quinone reductase (QR) induction and NF-κB inhibition activity, two well-established targets in cancer chemoprevention
Compounds 1, 2, 3b, 4, 6, 9, and 12 displayed cancer chemopreventive properties through either quinone reductase induction (CD = 30.7, 30.2, 29.9, 43.5, and 36.7 μM for 1, 4, 6, 9, and 12, respectively) and/or NF-κB inhibition with IC50 values of 13.1, 8.4, 9.4, and 8.8 μM for 2, 3b, 6, and 12, respectively.

Table of Contents/Graphical Abstract



PS1-A-061

Efficient removal of cationic dyes from aqueous medium using a recyclable activated carbon from Phoenix fruit pits: A study of adsorption parameters and thermodynamics

Abdullah Aldawsari

Prince Sattam Bin Abdulaziz University, Riyadh, Saudi Arabia

Because of unique surface chemistry and textural characteristics activated carbon (AC) has always been a center of attraction for environmental and energy scientists. Phase (solid/liquid and solid/gas) separation and catalysis are among the major environmental applications of AC. In recent years, ligno-cellulosic biomass (LCB) has emerged as a cost effective, eco-friendly, and renewable precursor material for AC, surpassing fossil, non-renewable precursors viz. coal and petroleum pitch. Besides, LCB produces highly pure non-graphitizable char with appropriate hardness and bulk density.

Date pits, an agricultural waste have been utilized to prepare date pits char. The developed char was activated using alkaline metal hydroxides NaOH. The date pit activated carbon (DPAC) was utilized in the removal of methylene blue (MB) from aqueous medium. The DPAC activation conditions viz. activation temperature and impregnation ratio were optimized to achieve maximum MB and to obtain highest yield of DPAC. The sample (DPAC) with activation temperature – 800 °C and NaOH: date pit char impregnation ratio (wt. %: wt. %) – 3:1 showed maximum MB removal from water and highest (56.77%) yield of DPAC. The maximum monolayer adsorption for MB at 323K was 146.3 mg/g. There was no significant effect of pH on MB adsorption on DPAC. Hence, it could be righteous to call the developed DPAC as a green and sustainable adsorbent for effective removal of MB and heavy metals from aqueous phase.

PS1-A-062

Skadar lake *Nymphaea alba* and *Nuphar luteum* leaves chemical profile

Nina Djapic

University of Novi Sad, Technical Faculty "Mihajlo Pupin", 23000 Zrenjanin, Serbia

Supercritical CO_2 extraction and hydrodistillation was used for the extraction of chemical compounds from *Nymphaea alba* and *Nuphar luteum* leaves. The extracts were analyzed by GC-MS. The most dominant compounds in *N. alba* supercritical CO_2 extract were: hexahydrofarnesyl acetone, phytol and squalene. The *N. alba* essential oil compounds present in high percentage were: hexahydrofarnesyl acetone, neophytadiene and hexadecanal. *N. luteum* supercritical CO_2 extract had high quantity of: hexahydrofarnesyl acetone, phytol and γ-sitosterol, while its essential oil mostly consisted of: hexahydrofarnesyl acetone, phytol and squalene. Obtained results permitted an observation on two species chemical profile.

Keywords: *Nymphaea alba*, *Nuphar luteum*, leaves, GC-MS

PS1-A-063

Study on the synthesis of vinpocetine

Kai Qiao[1], Dong Zhang[1], Y Wu X[1], Hong Qin[1], Zheng Fang[1], Kai Guo[1,2]

[1]College of Biotechnology and Pharmaceutical Engineering, Nanjing Tech University, Nanjing 211816, P. R. China, Nanjing, China, [2]State Key Laboratory of Materials-Oriented Chemical Engineering, Nanjing Tech University, Nanjing 210009, P. R. China, Nanjing, China

Cardiovascular disease (CVD) as a common disease, is a serious threat to human health, which could be divided into hemorrhagic and ischemic cerebrovascular diseases according to the pathological changes. Vinpocetine, as a vasodilator, could maintain or restore the physiological dilation of cerebral vascular, increase the normal cerebral blood flow in the ischemic area, improve the utilization of oxygen in the brain and the metabolism of anoxic brain tissue. Nevertheless, traditional approaches for the synthesis of vinpocetine suffer from tedious operations, lower yield, higher costs and other shortcomings. We use the tryptamine as the starting material, which is firstly under the addition of the home-made α-ethyl-pentanolid, and then reacts with $POCl_3$, the four rings Wenkert's enamine, as a key intermediate, is obtained by the Bischler-Napieralski cyclization reaction. Then, the enamine is sequentially in the addition of hydrazone synthesized by ethyl bromopyruvate and 2,4- dinitrophenylhydrazine, reduced by $NaBH_3CN$ selectively, cyclized in the presence of $TiCl_3$ and resolved by L-(+)-tartaric acid to obtain the five rings product vinpocetine with a total yield of 21.2%.

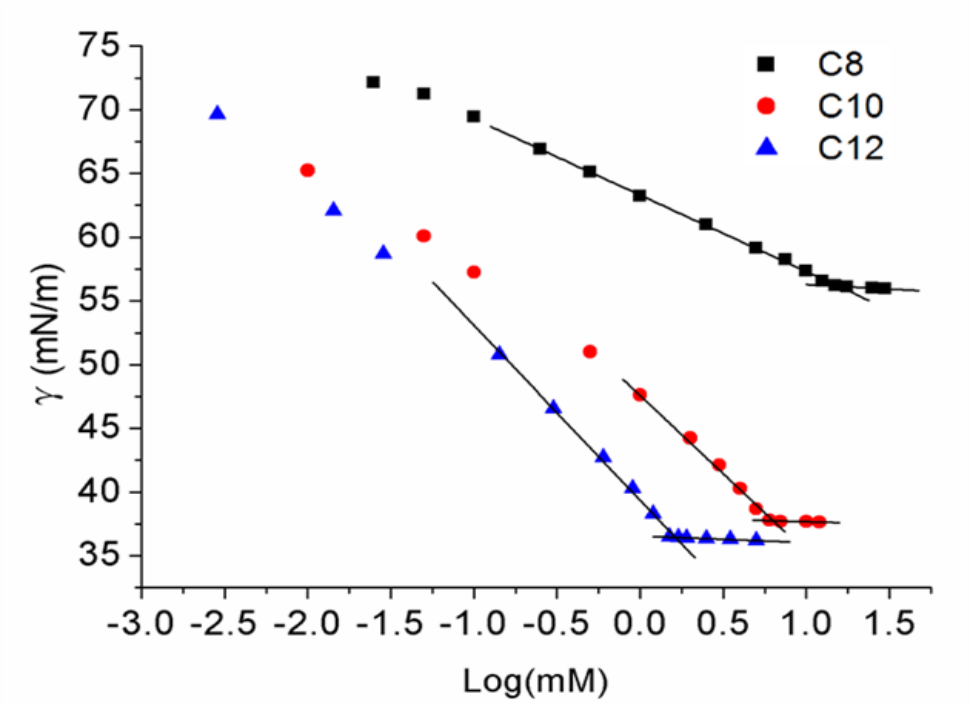
PS1-A-064

Surface and aggregation properties of Nα-lysine based surfactants

Ting Shi T[1], Zheng Fang[1], Kai Guo[1,2]

[1]College of Biotechnology and Pharmaceutical Engineering, Nanjing Tech University, Nanjing, China, [2]State Key Laboratory of Materials-Oriented Chemical Engineering, Nanjing Tech University, Nanjing, China

Surfactants synthesized from amino acid possess biocompatibility, biodegradability and multifarious aggregation properties. In this paper, several Nα-lysine based surfactants with 8-18 even chains have been synthesized, with particular focus on the behavior of sodium Nα-octanamide lysine (C_8), sodium Nα-capramide lysine (C_{10}) and sodium Nα-lauramide lysine (C_{12}). Surface activities of C_8-C_{12} have been investigated by surface tension. The aggregation behavior of the three amphiphiles at medium to high concentrations in aqueous medium have been studied by transmission electron microscopy (TEM), optical polarizing, scanning electron microscopy (SEM), and small angle x-ray scattering (SAXS). The three Nα-lysine based surfactants can all self-assemble into vesicles. Upon increasing the concentration, different crystalline phases for C_8-C_{12} and only liquid-crystalline phases for C_{12} at higher weight ratio have been detected. The self-assembled and crystal behaviors of the compounds are discussed.



PS1-A-065

Diterpenes from *Zhumeria majdae* Rech.f. & Wendelbo roots as potential HSP90 interactors

Student Reza Zadali[1], Abbas Hadjiakhoondi[1], Massimiliano D'ambola[2], Matthias Hamburger[3], Nunziatina De Tommasi[2], Samad Nejad Ebrahimi[4]

[1]Department of Pharmacognosy, Tehran University of Medical sciences (TUMS), Tehran, Iran, [2]Università di Salerrno, Dipartimento di Farmacia, Fisciano, Italy, [3]Division of Pharmaceutical Biology, University of Basel, Basel, Switzerland, [4]Department of Phytochemistry, Shahid Beheshti University, Teheran, Iran

Zhumeria majdae (Lamiaceae) is the unique member of *Zhumeria* genus endemic in Iran [1], locally used to treat gastrointestinal disease and dysmenorrhea[1]. Several studies have been reported on anti-protozoal, anti-convulsant, anti-nociceptive and anti-inflammatory activity of *Z. majdae* extracts [2]. Previous phytochemical investigation described the presence of diterpenes such as 12,16- dideoxy aegyptinone B and 12-deoxy-salvipisone, along with manool [3].

In the frame of a project oriented to study diterpenes as Heat Shock Protein 90 (HSP90) inhibitors, the phytochemical investigation of *Z. majdae* was carried out.

Chromatographic separations of *Z. majdae* roots *n*-hexane extract led to isolation of five pure compounds of which one never reported in literature before. The structures of isolates were elucidated by NMR and HRMS spectroscopy.

A surface plasmon resonance (SPR) assay was used to screen diterpenes towards HSP90. HSP90, is involved in the turnover, trafficking, and folding of several onco-proteins; thus, there is an interest in the development of molecules targeting Hsp90 [4].

SPR sensorgrams exhibited an interesting interaction of *Z. majdae* diterpenes with HSP90, showing thermodynamic constant (KD) values in nanomolar range, particularly diterpene Lanugon Q was the most interesting binder with a KD of 2.98±1.9 nM (radicicol was used as positive control 0.22±0.17nM). Meanwhile, the cytotoxic activity on rat myoblast (L6) cell line was determined. Lanugon Q was the most active compound (IC_{50} =0.66±0.08µM).

Keywords: diterpenes, SPR, HSP90

References:

[1] Jalili A, Jamzad Z. Red Data Book of Iran Res. Inst. of Forest and Rangelands, Tehran 1999.
[2] Moein MR, Pawar RS, Khan SI, Tekwani BL, Khan IA. Phytother Res 2008; 22: 283–285.
[3] Rustaiyan A, Samadizadeh M, Habibi Z, Jakupovic J. Phytochemistry 1995; 39: 163–165.
[4] Piaz FD, Vassallo A, Temraz A, Cotugno R, Belisario M, Bifulco G, Chini M, Pisano C, De Tommasi N, Braca A. J Med Chem 2013; 56: 1583–1595.

PS1-A-066

Antioxidant capacity *Rubus glaucus* and soil, physico-chemical, nutritional and Enzymatic relationship against three chemical inhibitors

Jairo Granados[1], Dolffi Rodríguez[2]

[1]Chemistry department, University of Cundinamarca, Fusagasugá, Colombia, [2]UNAD, School of basic sciences, technologies and engineering, Bogotá, Colombia

In Colombia, *Rubus glaucus* de Castilla, presents a result of high degree of perishability and little firmness, leading to a short shelf life; thus obtaining large postharvest losses [1]. The purpose of this study was to characterize the physicochemical, nutritional, enzymatic [2] behavior and antioxidant capacity of fruits of blackberry (*Rubus glaucus*), before and after applying three chemical ripenning inhibitors: ascorbic acid; Calcium chloride and sodium chloride [3]. In addition, we studied the correlation of these variables with the edaphic component of each crop in the respective sampled farms; It applied a design completely random sampling of soil, water and foliage in 8 places of Cundinamarca and Boyaca, respectively. Each farm was performed characterization of the geographical and climatic indicators; 3 fruit farms were sampled at random, in order to evaluate physicochemical, enzymatic and antioxidant capacity indicators; for this, they applied a design randomized block , with 2x3x3 factorial arrangement, blocking was performed with 3 farms and the factors evaluated were: f1: Maturation State; f2: Inhibitory; f3: concentration; design produced 18 treatments with 3 replicates per treatment, 2 per replicate experimental units for a total of 432 fruits intended for laboratory analysis. The values obtained were subjected to statistical analysis using SAS package (2012), using statisticians of central tendency, statisticians dispersion, simple and two-way ANOVA, multiple comparison tests Duncan mean and regression analysis and correlation between variables studied. The results showed a significant impact ($P<0.05$) and enzymatic soil physico-chemical parameters such as: % organic carbon, total nitrogen, cation exchange capacity effectively, catalase, protease and urease, on antioxidant properties and activities of peroxidase, catalase, polyphenol in fruits and pectinmethylesterase *Rubus glaucus*. Moreover, a positive statistical effect ($P <0.05$) of chemical inhibitors as ascorbic acid and sodium chloride, on physicochemical and enzymatic indicators studied the fruits was found.

PS1-A-067

Synthetic routes to royal jelly constituents 10-hydroxy-2-decenoic acid and 3-hydroxy-decanoic acid

Rodalia Babaiti, Adelajda Shahu, Maria Theodoropoulou, George Koutoulogenis, Maroula Kokotou, George Kokotos

National and Kapodistrian University of Athens, Athens, Greece

Royal jelly is a secretion from the hypopharyngeal glands of worker bees (*Apis mellifera L.*). Unlike fatty acids of most animal and plant materials, which consist mainly of triglyceride fatty acids each one having 14–20 carbon atoms, royal jelly fatty acids are medium-chained (8–10 carbon atoms) free fatty acids, terminally and/or internally hydroxylated, with terminal mono- or dicarboxylic acid functions, either saturated or monounsaturated at the 2-position [1]. The predominant fatty acid in royal jelly is trans-10-hydroxy-2-decenoic acid (10-HDA) and the amount of this unique fatty acid in pure royal jelly varies depending on the origin of the jelly and characteristics of the bee. 3-Hydroxy-decanoic acid (3-HDA) is another rare fatty acid found in royal jelly. Here we present convenient synthetic routes to these two fatty acids. The first step for the synthesis of 10-HDA was the monoprotection of 1,8-octanediol by the tetrahydrofuranyl group, which was achieved applying a photochemical protocol involving the *in situ* formation of 2-chlorotetrahydrofuran. The terminal hydroxyl moiety was oxidized to the corresponding aldehyde, which then reacted with the appropriate phosphorus ylide to give the desired unsaturated ester. After saponification and removal of the tetrahydrofuranyl group, 10-HDA was obtained. For the synthesis of 3-hydroxy-decanoic acid, octanal was converted into the corresponding 4-hydroxy terminal alkene by treatment with allylzinc bromide. After ozonolysis and treatment with dimethylsulfide, the aldehyde obtained was oxidized to 3-HDA by Pinnick reaction. An LC/HRMS method, employing an AB SCIEX TripleTOF® 4600 system coupled with a micro-LC Eksigent, for the rapid simultaneous determination of 10-HDA and 3-HDA was developed.

Acknowledgements: The support from the "Special Account for Research Grants" of the National and Kapodistrian University of Athens is gratefully acknowledged.

Keywords: Royal jelly, fatty acids, synthesis

References:

[1] Cornara L, Biagi M, Xiao J, Burlando B. Front Pharmacol 2017; 8: 412

PS1-A-068

Synthetic routes to 3-hydroxy saturated fatty acids

Adelajda Shahu, Maria Thedoropoulou, Asimina Bourboula, George Kokotos

National and Kapodistrian University of Athens, Athens, Greece

Saturated hydroxy fatty acids, although relatively common, have been scarcely studied as potential signaling molecules. Recently, the immunostimulatory G protein-coupled receptor GPR84 has been shown to be activated by medium-chain free fatty acids, while 2- or 3-hydroxy fatty acids have been proven comparatively more potent agonists than the nonhydroxylated fatty acids [1]. Using chemical isotope labeling-assisted LC–MC, various hydroxy fatty acids have been detected in plant tissues (for example, rice and *Arabidopsis thaliana*) [2], while 3-hydroxy fatty acids have been found in milk and cheese. The aim of the present study was to develop a general method for the synthesis of 3-hydroxy fatty acids in both racemic and chiral forms. For the synthesis of racemic 3-hydroxy fatty acids, a variety of long chain aldehydes were treated with allylzinc bromide to produce the corresponding 4-hydroxy terminal alkenes. Cleavage of the terminal double bond by ozonolysis, followed by treatment with dimethylsulfide and subsequent Pinnick oxidation, afforded the target compounds. The synthesis of chiral 3-hydroxy fatty acids was based on the organocatalytic conversion of aldehydes into chiral terminal epoxides. Organocatalytic α-chlorination of aldehydes, followed by reduction with NaBH4 and treatment with KOH, afforded the terminal epoxides in a one-pot procedure. Treatment with vinyl magnesium bromide led to chiral 4-hydroxy terminal alkenes, which were then converted to the target compounds by the ozonolysis-oxidation protocol described above. Various organocatalysts (either proline-based or MacMillan's imidazolinones) were studied for the key-organocatalytic step leading to varying enantiopurities of the epoxides and the corresponding long chain 3-hydroxy fatty acids.

Acknowledgements: The support from the "Special Account for Research Grants" of the National and Kapodistrian University of Athens is gratefully acknowledged.

Keywords: Hydroxy fatty acids, synthesis

References:

[1] Suzuki M, Takaishi S, Nagasaki M, Onozawa Y, Iino I, Maeda H, Komai T, Oda T.J Biol Chem 2013; 288: 10684–10691

[2] Shu J, Qiu Z, Lv S, Zhang K, Tang D. Anal Chem 2018; 90: 2425–242

PS1-A-069

Asymmetric synthesis of 7-hydroxy saturated fatty acids

Maria Theodoropoulou, Adelajda Shahu, Olga Mountanea, George Kokotos

National and Kapodistrian University of Athens, Athens, Greece

The discovery of a novel class of lipids [1], which were named Fatty Acid esters of Hydroxy Fatty Acids (FAHFAs), has recently attracted the interest on naturally occurring Hydroxy Fatty Acids (HFAs) and derivatives. FAHFAs contain two chains connected through an ester bond. Each family of FAHFAs consists of multiple regioisomers in which the hydroxyl participating in the ester bond is at different positions (5-, 7- etc). Recently, the detection of both HFAs and FAHFAs in plant tissues (rice and *Arabidopsis thaliana*) by chemical isotope labeling-assisted LC–MS was reported [2]. It has been reported that FAHFAs are involved in glucose metabolism and immune responses. The aim of our work was to develop an efficient methodology permitting the synthesis of various saturated 7-hydroxy fatty acids. The key-step of our approach was the conversion of an aldehyde into a chiral terminal epoxide under organocatalytic conditions. Monoprotected 1,8-octanediol was oxidized to aldehyde and subjected to α-chlorination in the presence of either the natural amino acid proline or urea-type proline derivatives as catalysts. Subsequently, reduction by $NaBH_4$, followed by treatment with KOH led to the formation of the terminal epoxide in a one-pot procedure. Treatment of the epoxide with an alkyl magnesium bromide provided a carbon chain, bearing the hydroxyl group at position-7. Acetylation of the hydroxyl, followed by removal of the terminal O-protecting group led to 7-acetoxy fatty acid and after deprotection to 7-HFA. Thus, a variety of 7-HFAs, which may be used for the synthesis of FAHFAs or for biological evaluation, were synthesized.

Acknowledgements: The support from the "Special Account for Research Grants" of the National and Kapodistrian University of Athens is gratefully acknowledged.

Keywords: Hydroxy fatty acids, synthesis

References:

[1] Suzuki M, Takaishi S, Nagasaki M, Onozawa Y, Iino I, Maeda H, Komai T, Oda T. J Biol Chem 2013; 288: 10684–10691

[2] Shu J, Qiu Z, Lv S, Zhang K, Tang D. Anal Chem 2018; 90: 2425–2429

PS1-A-070

Optimization and preparation of methylcellulose edible film combined with *Ferulago angulata* essential oil (FEO) nanocapsules for food packaging applications

Akbar Esmaeili

Department of Chemical Engineering, North Tehran Branch, Islamic Azad University, Po Box 19585/936, Tehran, Iran., Tehran, Iran

The films based on methylcellulose with biodegradable and antioxidant activity incorporated with nano capsule suspension containing *F. angulata* essential oil were developed. Oil extraction and identification of *F. angulata* essential oil compounds was done. Nano capsule suspension containing *F. angulata* essential oil was prepared by ultrasonic bath. The films were prepared by a casting method in three different ratios. The mechanical properties, colour, light transmission, antioxidant activity and release rate characteristics of the films were studied. The addition of nano capsule suspension to methylcellulose films decreased the thickness, tensile strength but increased the percentage elongation at break (%E) and lightness. High antioxidant activity and a prolonged release of *F. angulata* essential oil were also reported. Five factors design of Response Surface Methodology were used to optimize the thickness, holding time and anti-oxidant effect of edible film based on methylcellulose incorporated with nano capsule suspension containing *F. angulata* essential oil. Design of experiments was carried out by the software: Minitab 17 (Sigma package). Optimization of thickness, 2, 2-diphenyl-1-picrylhydrazyl radical scavenging and holding time would yield the best mixture proportions of methylcellulose and nano capsule suspension 30%, 30% and 70%; oil<0.05, 0.07 and 0.05 mg; at 20, 24 and 24 min; 50°, 40°, 50° C were obtained respectively.

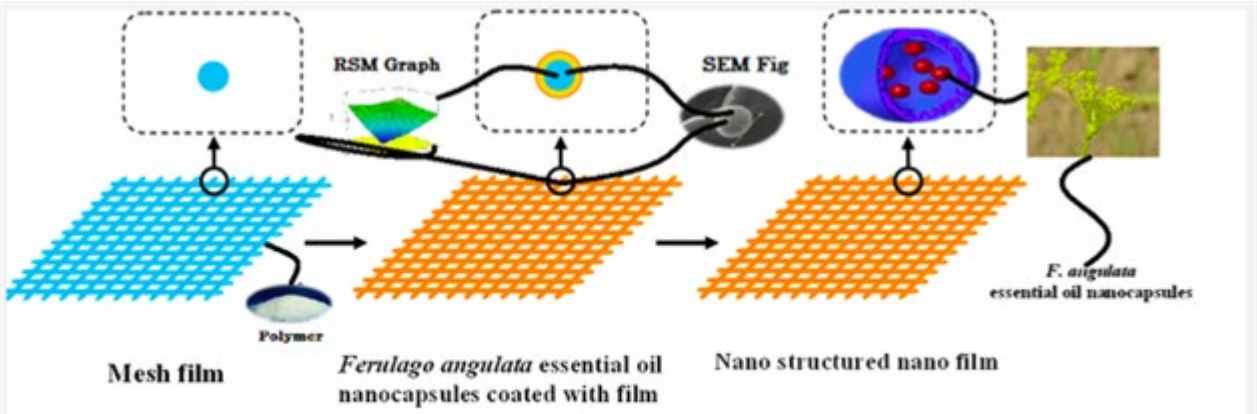


Figure1. The films based on methylcellulose with biodegradable and antioxidant activity incorporated with nano capsule suspension containing FEO were developed.

PS1-A-071

Antioxidant activity of polyphenolic extracts from carob pods and their effect on acrylamide formation in the asparagine/fructose Maillard reaction system

Iliana Tsenkova, Anna Marina Grigoriou, Eftychia Pinakoulaki

University of Cyprus, Nicosia, Cyprus

Carob is the fruit of an evergreen tree (*Ceratonia siliqua* L.) cultivated in the Mediterranean area and has been recognized as a valuable source of bioactive compounds including polyphenols in recent years. The concentration of polyphenols in carob depends strongly on genetic, environmental and farming factors, while the method used for the extraction of polyphenols significantly affects their concentration and profile [1]. In this work polyphenolic extracts were obtained from Cypriot carobs and the antioxidant activity and total polyphenolic content of these extracts were determined. The effect of the carob extracts on the formation of acrylamide in the asparagine/fructose Maillard reaction system was examined, as recent studies have indicated that plant extracts may mitigate acrylamide formation. Acrylamide is a probable human carcinogen that is formed mainly by the Maillard reaction mainly between L-asparagine and reducing sugars, such as fructose and glucose, in a range of fried and oven-cooked foods [2]. Acrylamide formed upon the reaction of the asparagine and fructose at different heating times and temperatures was quantified by high performance liquid chromatography (HPLC) with UV detection. The effect of carob polyphenolic extracts on acrylamide formation in the asparagine/fructose system was compared to that of pure polyphenolic compounds, which were present in significant quantities in the carob extracts, as determined by HPLC.

Acknowledgments: This work was supported within the initiative "Carob: the Black Gold of Cyprus" of the University of Cyprus.

Keywords: Polyphenols, maillard reaction, acrylamide, carob, HPLC

References:

[1] Loullis A, Pinakoulaki E. Eur Food Res Technol 2018; 244: 959–977.
[2] Rydberg P, Eriksson S *et al*. J Agric Food Chem 2003; 51, 7012–7018.

PS1-A-072

Effect of pure polyphenols and polyphenolic extracts from carobs to acrylamide formation in the asparagine/glucose Maillard reaction system

Charia Hadjipakkou, Eftychia Pinakoulaki

University of Cyprus, Nicosia, Cyprus

Acrylamide has been found in various foods, which is formed during the Maillard reaction. Many studies have been conducted to mitigate or inhibit acrylamide formation by adding exogenous additives such as polyphenols [1]. In this work, the formation of acrylamide versus temperature and heating time was studied by employing a model system for the Maillard reaction and the effect of three different pure polyphenolic compounds on acrylamide formation was examined. Extraction of water-solubilized polyphenols from carob kibbles was performed by two methods. Furthermore, the effect of the carob polyphenolic extracts on acrylamide formation was studied for the first time in the asparagine/glucose system. Reversed phase high-performance liquid chromatography (RP-HPLC) with UV detection was used to detect and quantify acrylamide. Time and temperature affect the formation of acrylamide, since higher temperatures, from 160 to 200 °C, produce higher amounts of acrylamide. The effect of gallic acid on the formation of acrylamide at pH 7 was not significant, while at pH 5.8 an increase in acrylamide levels was observed, up to 64 ± 1% when high concentration of gallic acid was used. On the other hand, addition of quercetin and catechin, led to reduced acrylamide levels, reaching up to 24 ± 1% and 33 ± 2% acrylamide reduction, respectively. Effective extraction of polyphenols from carob kibbles was then achieved using water as the extraction solvent. The obtained extracts displayed high total polyphenolic content (up to 62 mg GAE/g of extract) and high antioxidant activity. Finally, with the addition of extracts to the asparagine/glucose model, reduction of acrylamide (up tp 18 %) was observed for short heating periods, although for extended heating periods increase of acrylamide levels was detected.
Keywords: Maillard reaction, polyphenols, acrylamide, carob

References:

[1] Rannou *et al*. Food Res Int 2016; 90: 154–176.

PS1-A-073

Triterpene saponins content in Swiss chard (*Beta vulgaris* L.)

Agnieszka Mroczek[1], Urszula Klimczak[1], Anna Stochmal[2], Mariusz Kowalczyk[2]

[1]Department of Plant Biochemistry, Faculty of Biology, University of Warsaw, Miecznikowa 1, 02-960 Warsaw, Poland, Poland, [2]Department of Biochemistry, Institute of Soil Science and Plant Cultivation, State Research Institute, Czartoryskich 8, 24-100 Puławy, Poland

The family Amaranthaceae is a widespread and cosmopolitan family that can be found from the tropics to cool temperate regions. The phytochemical profile of Amaranthaceae plants comprises essential oils, betalains, phenolics and triterpene saponins. Different pattern of triterpene saponin occurrence was characterized in almost 30 species belonging to Amaranthaceae which can be considered as promising and highly available sources of these biologically active compounds. Recently, triterpene saponins were described in red beet (*Beta vulgaris* ssp. *esculenta* L.). So far nothing was known about the occurrence of saponins in other plant from *Beta* genus - Swiss chard (*B. vulgaris* ssp. *cicla* L.). This vegetable is valued not only for the taste of its leaves, but also because of its health-promoting properties. Swiss chard was used in folk medicine in the treatment of diabetes and kidney diseases. In the contemporary studies extracts from the plant leaves showed anti-cancer, anti-inflammatory and antioxidant properties.

Due to the wide spectrum of activities saponins could be at last partially responsible for biological activities of this plant. Thus, the aim of the present study was qualitative and quantitative analysis of saponins in leaves of five cultivar varieties of Swiss chard: Lukullus, Rhubarb, Vulkano, Silber and Perpetual Spinach. Ultra-high performance liquid chromatography coupled with tandem mass spectrometry (UHPLC-MS/MS) was used as the analytical method. Results obtained indicated that leaves of Swiss chard contained 18 saponins - derivatives of oleanolic acid, hederagenin, phytolaccagenic acid, akebonic acid and serjanic acid. Additionally, it was stated that analyzed varieties differ in the total concentration of saponins. Moreover, differences in the relative content of individual saponins among varieties were observed.

Acknowledgements: The study was supported by the Ministry of Science and Higher Education through the Faculty of Biology, University of Warsaw intramural grant DSM nr. 501-D114-86-0117600-24

Keywords: Triterpene saponins, Swiss chard, Amaranthaceae, UHPLC-MS/MS

PS1-A-074

Phytochemical investigation of fruits from *Paliurus spina-christi* Mill

Vaios Amountzias, Dionysios Abatis, Nektarios Aligiannis

Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, University of Athens, Panepistimiopolis, Zografou, 15771, Athens, Greece, Greece

Paliurus spina-christi Mill. (Rhamnaceae family) is a much-branched, deciduous, thorny shrub up to four meters tall with green fruits which become brown during maturation. The shrub is found in dry slopes in Mediterranean, Southwest and Central Asia and North America. It is used as an ornamental and for building hedges, so it is cultivated in some areas like Fiji Islands. It is also known as *P. aculeatus*, *P. australis* and *Rhamnus paliurus*, while its common name is 'Christ's thorn' because it is thought that the spiny branches were used to make the crown of thornes which had been placed on Christ's head before his crucifixion. *P. ramosissimus* Lour., *P. orientalis* Franch. and *P. hemsleyanus* Rehder are well known species from Eastern Asia, while *P. microcarpus* E. Willm., is distributed to North Greece [1]. Traditionally, *P. spina-christi* is used for its diuretic and antihypercholesterolemic properties, as well as palliative agent of gastrointestinal pain [2-5]. Thus, it is important to investigate the phytochemical profile of this shrub. In this project, powdered mature fruits were subjected to exhaustive extraction by maceration technique with Cyclohexane, Dichloromethane, Methanol and Water as solvents. In order to remove fats and sugars, the dried methanolic extract subjected to liquid-liquid extraction with EtOAc and Water. The EtOAc fraction was further evaluated using Normal Phase - Vacuum Liquid Chromatography (NP-VLC), while Preparative Thin Layer Chromatography (Prep-TLC) and Normal Phase - High Pressure Liquid Chromatography (NP-HPLC) were used for the isolation of secondary metabolites. Structure elucidation was performed using Nuclear Magnetic Resonance (NMR) and Mass Spectroscopy (MS) and revealed the presence of secondary metabolites belonging to phenolics, triterpenoids, sterols and cyclopeptide alkaloids.

PS1-A-075

Further constituents of the stem bark of *Acacia auriculiformis*

Augustine Ahmadu[1], Nikolaos Tsafantakis[2], Nikolas Fokialakis[2]

[1]Kaduna State University, Kaduna, Nigeria, [2] National Kapodistrian University of Athens, Athens, Greece

Acacia auriculiformis also referred to as ear leaf Acacia is an important medicinal plant. The plant is a widely distributed member of the family Fabaceae. An infusion of the bark of this plant is used to treat inflammation among the aborigines of Australia (Gijasahnkar, 2010). The antimutagenic and chemoprotective activities of extracts of the stem bark of *Acacia auriculiformis* as well as the ability of the ethylacetate and acetone fractions of the stem bark to scavenge radical have been reported (Kauer et ,2002; Singh *et al*,2007). A new triterpenoid trisaccharide and three new triterpenoids have been isolated from this plant (Mahato *et al*,1989; Saraswal and Mahato,1997) and the antimicrobial activity of the isolated saponins was reported by (Mandal *et al*, 2005). As part of our investigation of the genus *Acacia* for protein kinase inhibitory agents we report herein the isolation from the dichloromethane extract of the stem bark of *Acacia auriculiformis*, a new Ferulic acid ester, dodecyl-4-hydroxy-3-methoxy-*trans*-cinnamate named auriculate (I) alongside with *α*-spinasterol (II) and Lupenol (III). The structure of compound I was elucidated using NMR and HR-ESI-MS, while compounds II and III were deduced based on the comparison of their spectral data with literature. Compound I is being reported for the first time in the genus *Acacia*.

Keywords: *Acacia auriculiformis*, dodecanyl ferulate

References

[1] Gijasahnkar V. J Med Plant Res 2010; 5: 482–486.
[2] Kauer K, Aroara S, Hawthone ME, Kauer S, Kumar S, Mehta R. Chem Toxicol 2002; 25(1): 639–648.
[3] Mahato SB, Pal BC, Price KK. Phytochemistry 1989; 28(1): 207–210.
[4] Mandal P, Sinhababbu SP, Mandal NC. Fitoterapia 2005; 76: 462–465.
[5] Saraswat G, Mahato SB. Phytochemistry 1997; 44(1): 137–140.
[6] Singh R, Singh S, Kumar S, Arova S. Food Chem Toxicol 2007; 45: 216–223

PS1-A-076

Fast Centrifugal Partition Chromatography – gustatometry as a valuable tool for the identification of new natural sweet compounds from wood chips used in wine ageing

Eirini Koui[1], Antigoni Cheilari[1], Evanthia Dina[1], Dennis Abatis[1], Michalis Stefanakis[2], Yorgos Kotseridis[2], Nektarios Aligiannis[1]

[1]Deptartment of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [2]Department of Food Science and Technology, Agricultural University of Athens, Athens, Greece

Ancient Greeks and Romans were aware of artificial means of accelerating natural ageing of wine since centuries. However, not all wines have the potential to be aged and for those that can, ageing is a complex chemical reaction that has been partially described.
Beside methodology, grape variety, and environmental conditions, the barrels (type of wood, level of toasting, etc.) play a crucial role in wine ageing [1]. In addition, taste-guided fractionation and purification (CPC-gustatometry) is a novel and promising approach for the isolation and identification of new compounds exhibiting a sweet taste and has been implemented successfully [2]. For this purpose, four different wood species were selected in order to study their composition in correlation to the flavors and aromas given to the wine: the popular in wine industry French (*Quercus petraea*) and American oaks (*Quercus alba*), the Slavonian oak (*Quercus robur*) and Acacia (*Robinia pseudoacacia*).
All samples, provided in the form of chips and medium toasted, were extracted successively (EtOAc, MeOH, H_2O and EtOH/H_2O 1:1) with accelerated solvent extraction (ASE Dionex-300) and their phytochemical profile was compared by HPTLC (CAMAG). The hydroalcoholic extracts of the Slavonian oak and *Acacia* were selected for further analysis and were fractionated by FCPC with a biphasic step-gradient (hept/EtOAc/MeOH/H_2O) and a three-phase (hept/MTBE/ACN/H_2O) solvent system, respectively. All FCPC fractions were characterized for their flavor and taste by Descriptive Sensory Analysis that was conducted using 8-10 panelists. The most interesting ones, at present with sweet taste, were further subjected into prep-TLC and sephadex resulting in the isolation of the major compounds of the fractions. Characterization and structure elucidation of the derived 15 secondary metabolites was performed by GC-MS and 1&2D NMR experiments and revealed the presence of phenolic compounds and flavonoids.

References:

[1] Trends Food Sci Technol 2006; 17(8): 438–447
[2] Anal Chem 2011; 24: 9629–9637.

PS1-A-077

Polyphenols from *Thymus thracicus* Velen

T. Papagrigoriou, O. Tsiftsoglou, D. Lazari

Laboratory of Pharmacognosy, School of Pharmacy, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece

The flora of Greece is quite diverse, boasting a wide variety of species, many of which are endemic to the country. *Thymus thracicus* Velen (Lamiaceae) is sub-endemic to the central part of the Balkan peninsula, distributed in northern Greece, as well as parts of Bulgaria, former Yugoslavia, and southwestern Turkey. It grows in rocky slopes and grasslands, at altitudes of 1200-2000m. In the present study, we report the isolation of nine phenolic derivatives from the aerial parts of this species, namely a phenolic acid derivative: oresbiusin A (1), four depsides: rosmarinic acid (RA) (2), methyl rosmarinate (MR) (3), 9”-methyl lithospermate (4), and dimethyl lithospermate (5), three phenylethanoid glucosides: calceolarioside A (6), 3-methoxy-calceolarioside A (7), and 4-methoxy-calceolarioside A (8), and one flavone glycoside: luteolin-7-*O*-*β*-D-glucuronopyranoside (9). The pure compounds were isolated using chromatographic methods (VLC, CC, TLC, HPLC) and the structures were elucidated by spectroscopic methods UV-Vis, 1D and 2D NMR. With the exception of RA, MR and luteolin-7-*O*-*β*-D-glucuronopyranoside, all the other compounds have been isolated for the first time, to our knowledge, from a *Thymus* species.

PS1-A-078

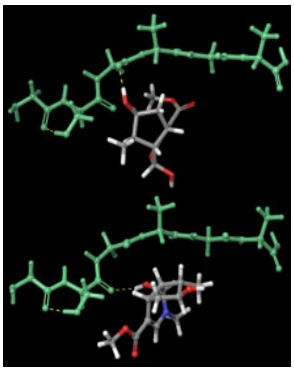
Synthesis of Blue Dye from Loganin, its Application in Dyeing, Ultraviolet Protection of Proteinious Fabric and Docking studies

Ms Sapna Patil[1]

[1]*Institute Of Chemical Technology, Mumbai, India*

The aim of the present work is mainly to study the blue colored dye, synthesized from an iridoid glycoside loganin from Strychnos nux-vomica and explore its application in textile industry. Another aim is to carry out Computer-Aided Drug Design (CADD) studies to determine the blue dye interrraction with silk monomeric units. Isolated white loganin was converted to blue dye by one step chemo enzymatic reaction, wherein loganin was transformed to aglycone, loganetin which was further allowed to react with amino acids generating a blue colorant. The colorant was further studied for its application to dye proteinous fabric like wool and silk. Color strength, fastness properties and Ultraviolet protection factor were evaluated. Mordanting studies were carried out using three different mordants.

Blue dye produced is polar, water soluble, insoluble in inorganic solvents and showed λmax at 590 nm. The dye showed variation in absorbance in different solvents. The single bath dyeing of fabric was carried out. The fabric was also subjected to pre-mordanting, meta-mordanting and post-mordanting studies. The colour strength (K/S) of wool fabric was found to be higher in case of meta-mordanting method using alum as a mordant as compared to pre and post mordanting techniques. Treated fabrics showed adequate wash, light and rubbing properties with and without mordanting. The treated silk and wool fabric showed good to very good ultraviolet protection property with mordanted fabric. The loganin, its aglycone and blue dye structures were subjected to molecular docking using Glide and SiteMap software.



PS1-A-079

Exploitation of aromatic plants' by-products for the development of bioactive extracts with antioxidant and antimicrobial properties

Evanthia Ntina[1], Christina Bakali[1], Dionisis Abatis[1], Konstantina Stathopoulou[1], Eleutheria Arapi[1], Spyros Economou[1], Hristo Iliev[2], Ilias Smyrnioudis[3], Nikolaos Fokialakis[1], Nektarios Aligiannis[1]

[1]National & Kapodistrian University of Athens, Department of Pharmacy (Pharmacognosy & Natural products chemistry), Athens, Greece, [2]Galen-N., Sofia, Bulgaria, [3]Chios Mastic Growers Association, Chios, Greece

EXANDAS project involves the exploitation of aromatic plants' by-products for the development of novel cosmeceuticals and food supplements. Thirty-two byproducts from the industrial exploitation of nineteen economically important aromatic/medicinal plants such as mastic gum, rose, mountain tea, lavender, geranium etc. were selected, and extraction protocols were developed. Depending on raw material, different environmentally friendly techniques (ASE, SFE, MAE) were used and the optimal extraction conditions were determined. In order to isolate, purify, and structurally characterize the active constituents, a variety of chromatographic techniques were used such as Centrifugal Partition Chromatography (CPC), Size Exclusion Chromatography etc. A range of bioassays was incorporated for the evaluation of antioxidant, anti-inflammatory, antimicrobial and anti-aging properties activity of the derived extracts and products.

In this work we present the cases of two very promising by-products: 1) the "aqueous waste" remaining during the hydrodistillation for the rose oil production which is a rich source of valuable phenolic and flavonoids compounds [1] and 2) "kolophony", the residue obtained after removing the essential oil from the mastic gum, which still contains the bioactive triterpenes of mastiha [2]. In the case of the rose hydrodistillation waste water, several samples of different distillation apparatuses and production dates were treated with macroporous adsorption resin XAD-4 and the optimal parameters were determined. The extracts obtained were evaluated for their Phenolic and Flavonoid Content while antioxidant evaluation was performed resulting to the identification of the most promising extract. Concerning kolophony, several trials using Supercritical Fluid Extraction technique were performed to determine the optimal conditions in order to obtain an extract enriched in neutral triterpenes and triterpenic acids.

Based on the foregoing, it is easily understood that the aforementioned by-products could be considered as a readily available source of valuable products with potential to be destined in the market of cosmetics, nutraceuticals and phytotherapeuticals.

PS1-A-080

Study of the basidiomycete of *Ganoderma adspersum* (Ganodermataceae)

Raichan Chafouz[1], Olga Tsiftsoglou[1], Dimitra Hadjipavlou-Litina[2], Pavle Maskovic[3], Diamanto Lazari[1]

[1]Aristotle University of Thessaloniki, Faculty of Health Sciences, School of Pharmacy, Laboratory of Pharmacognosy, Thessaloniki, Greece, [2]Aristotle University of Thessaloniki, Faculty of Health Sciences, School of Pharmacy, Laboratory of Pharmaceutical Chemistry, Thessaloniki, Greece, [3]University of Kragujevac, Department of Chemical and Chemical Engineering, Kragujevac, Serbia

Ganoderma is a genus of wood degrading mushrooms with medicinal importance [1]. Most of the species of *Ganoderma* spp. have been studied extensively for their secondary metabolites, biological activities and ecological value [2]. In this study, biological activities of the extracts of *Ganoderma adspersum*, growing wild on *Morus alba* tree in the region of Western Thrace, have been evaluated, as well as the petroleum ether, dichloromethanolic and methanolic extracts were studied further for their secondary metabolites. So far, five substances have been isolated by chromatographic (C.C., HPLC) and spectroscopic methods (NMR), which were classified in the following categories: a) Unsaturated fatty acids: *cis*-Oleic acid (1), b) Sterols: Ergosta-7,22-dien-3-one (2) and Ergosta-7,22-dien-3-ol (3), and c) Triterpenoids – Lanostanes: Applanoxidic acid G (4) and Applanoxidic acid A (5). Finally, the biological activities of the extracts were estimated for their antioxidant, antimicrobial and cytotoxic potent.

Keywords: *G. adspersum*, Ganodermataceae, triterpenoids, fatty acids

References:

[1] Baby S, Johnson AJ, Govindan B. Phytochemistry 2015; 114: 66–101.
[2] Xia Q, Zhang H, Sun X, Zhao H, Wu L, Zhu D, Yang G, Shao Y, Zhang X, Mao X, Zhang L, She G. Molecules 2014; 19: 17478–17535.

PS1-A-081

Determination of biophenols in virgin olive oils by liquid chromatography supporting the health claim by European Food Safety Authority (EFSA)

Anna Stefanitsi[1], Vasilios Sakkas[1], Panagiotis Stathopoulos[2], Maria Halabalaki[2], A-L Skaltsounis[2]

[1]University of Ioannina, Ioannina, Greece, [2]National and Kapodistrian University of Athens, Athens, Greece

Virgin olive oil (VOO) contains several bioactive ingredients, which contribute to the resistance of olive oil to oxidative rancidity. Among them, polyphenols or biophenols are of great sensory and biological importance. The major phenolic compounds in olive oil include hydroxytyrosol, tyrosol, oleacein, oleocanthal as well as oleuropein and ligstroside aglycons which are structurally related. The composition and percentage of these phenolic compounds in VOO depend on several factors including the variety of olives, their geographical origin and the type of oil extraction.

The European Food Safety Authority (EFSA) has evaluated the research studies linking the consumption of olive oil with the benefits on human health and has concluded that "olive oil polyphenols protect blood lipids from oxidative stress". This finding makes olive oil as a food of high nutritional value gaining a “health claim” that may be indicated on the label of olive oil as long as it contains 250 mg tyrosol/kg of olive oil and the daily consumption is 20 g of olive oil (Reg. EU 432/2012).

In the present study, the total phenolic compounds of 16 virgin olive oil samples VOO was determined using HPLC-UV/DAD after sample preparation with liquid-liquid extraction (LLE) and solid-phase extraction (SPE). The LLE method had better recovery values compared to the SPE for the total content of the phenolic compounds in the olive oil samples and the opposite results for the content of tysosol and hydroxytyrosol. The trueness of the method was tested in the interlaboratory exercise of the International Olive Council (COI) with satisfactory results. The concentration of total phenols in most of the analyzed samples was above 250 mg/kg, indicating the beneficial impact of these particular VOOs on human’s health and permitting the use of health claim.

Keywords: EFSA, health cleam, HPLC-UV/DAD, biophenols

PS1-A-082

Investigation of bioactive secondary metabolites from *Alkanna* species of Greece

Evangelia Tsiokanou[1], Nikolaos Tsafantakis[1], Eirini Baira[1], Eleftherios Kalpoutzakis[1], Andreana Assimopoulou[2], Nektarios Aligiannis[1], Nikolas Fokialakis[1]

[1]Department of Pharmacognosy and Natural Product Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis, Athens, Greece, [2]Organic Chemistry Laboratory, Faculty of Engineering, School of Chemical Engineering, Aristotle University of Thessaloniki, 54124 Thessaloniki, Greece

Studies have already shown that interlinking microbiomes with medicinal plants play a key role on plant growth processing. The plants-associated microorganisms may modulate the biosynthesis of secondary metabolites and stimulate their production. In the frame of the EU H2020 "MICROMETABOLITE" project, innovative technologies are developed by integrating microorganisms, including endophytic bacteria and fungi but also arbuscular mycorrhyzal fungi (AMF), on plant materials of Boraginaceae family.
As part of our study, flowering aerial parts of eight different populations of *Alkanna tinctoria* were collected so far at the same period from different locations close to Athens and treated similarly. On the other hand, roots and aerial parts of three other species of the genus *Alkanna* (*A. graeca*, *A. hellenica* and *A. sfikasiana*) were collected from Southern Greece, while a commercial sample of *Alkanna tinctoria* was used as a reference material.
The current work aims to assess the optimal growth environment and conditions in accordance with the metabolite production of *A. tinctoria* aerial parts from the different collections. Furthermore, the four species selected from different geographical regions of Greece were investigated and compared regarding to their metabolite variations with the commercial sample of *A. tinctoria*. Some extracts proved to be particularly rich in secondary metabolites. The extraction process was performed sequentially with solvents with increasing polarity to get a broad metabolic profile. The samples, both for roots and aerial parts, were analyzed with methods based on HPTLC, HPLC-PDA-ELSD but also UPLC-HRMS. A dereplication process combining the Dictionary of Natural Products, Molecular Networks, *in silico* MS/MS dereplication using customized libraries, were developed and it will lead to a targeted isolation and identification of promising and new compounds.

Acknowledgments: This project has received funding from the European Union's Horizon 2020 research and innovation program under the ITN - Marie Skłodowska-Curie grant agreement No 721635.

PS1-A-083

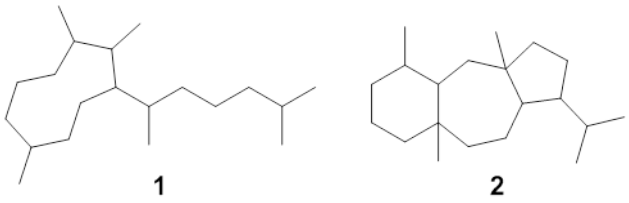
New diterpenes from two populations of the brown alga *Dictyota linearis* from the Aegean Sea

Aikaterini Koutsaviti, Charoula Machaira, Christina Keramea, Vassilios Roussis, Efstathia Ioannou

Section of Pharmacognosy and Chemistry of Natural Products, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771 Athens, Greece, Greece

Brown algae of the genus *Dictyota* (Dictyotaceae) have been the subject of thorough chemical investigations over the last 50 years that have afforded approximately 300 new natural products. Most of the isolated metabolites are sesquiterpenes and diterpenes deriving from normal biosynthetic pathways and featuring a wide range of carbon skeletons. These compounds frequently exhibit antifeedant, ichthyotoxic, algicidal, antifouling, antibacterial, antiviral, and/or cytotoxic activities.

In the course of our ongoing research on bioactive secondary metabolites from marine macroalgae occurring along the Greek coastlines, we investigated the chemistry of two populations of *Dictyota linearis* collected from Gerolimenas bay in Peloponnese and Agios Sostis at the island of Tinos. The algal tissues were extracted with mixtures of CH_2Cl_2/MeOH and the organic residues were submitted to a series of chromatographic separations, leading to the isolation of a number of diterpenes, among which several featuring the xenicane (1) and the dolastane (2) skeleton are new natural products. The structure elucidation of the isolated compounds was based on thorough analysis of their 1D and 2D NMR and MS data.



1 2

Acknowledgements: This work was partially supported by the research project "Centre for the study and sustainable exploitation of Marine Biological Resources" (CMBR, MIS 5002670) in the framework of the National Roadmap for Research Infrastructures.

Keywords: *Dictyota linearis*, diterpenes, isolation, structure elucidation

PS1-A-084

Development of an inhalant based on African ginger (*Siphonochilus aethiopicus*) for the management and/or treatment of coughs, colds, flu and as a decongest

Leylene Kruger, Vinesh Maharaj, Thomas Klimkait

University of Pretoria, Pretoria, South Africa

Siphonochilus aethiopicus (Schweinf.) B.L. Burtt. (Zingiberaceae), better known as African ginger [5], has been used by traditional healers and is one of the most important medicinal plants in South Africa [5,8]. Previous research has revealed that they have healing properties and can be used to treat colds, flu, coughs, influenza, sinus problems, asthma, etc. [2,3,5,6]. Traditional healers make use of different preparation techniques such as cold and hot infusions of the rhizomes and roots, steaming of the rhizomes and inhalation of the vapour [1,2]. Previous research has found that the plants contain 1,8-cineole (eucalyptol) and furanoterpenoid siphonochilone which has been used for anti-inflammatory treatment of asthma and allergic conditions [4,7,10,11]. Eucalyptol is a terpenoid oxide that is present in the essential oil of many plants and used in the pharmaceutical industry in drug formulation for its decongestant- and inflammatory effects, treatment of bronchitis, etc. [9].

A lot of research has been done on the plant, but little on the identification and bioassaying of the volatile compounds. Extractions and steam distillation has been done on both dried- and fresh rhizomes to identify, stabilize and purify the major compounds [7]. These extracts, essential oils and pure compounds have been stabilized with different antioxidants to prevent polymerization of the naturally occurring furanoterpenoid [12]. After these compounds have been stabilized, they have been screened for activity against the human influenza virus. The aim of the research is to produce a stable essential oil or pure compound from the rhizomes of African ginger and scientifically evaluate its traditional use so that it can be formulated as an inhalant for commercial purposes.

PS1-A-085

LC/MS guided isolation of protopanaxadiol ginsenosides having 5~6 sugars from *Panax vietnamensis*

Van Le[1], Long Vu[3], Khoi Nguyen[1], Hill Park[2], Duc Nguyen[3]

[1]University of Medicine and Pharmacy at Ho Chi Minh city, Ho Chi Minh city, Vietnam, [2]College of Pharmacy and Research Institute of Pharmaceutical Sciences, Seoul National University, Seoul, Korea, [3]Faculty of Pharmacy –Ton Duc Thang University, Ho Chi Minh city, Vietnam

Background: Seventy-three dammarane-type triterpene saponins have been reported from the different parts of *Panax vietnamensis* (Vietnamese ginseng, VG). Our recent study detected several unknown protopanaxadiol (PPD) ginsenosides have 5~6 sugars by Q-TOF-MS in different underground parts of VG.
Methods: LC/MS guided isolation together with prep-HPLC were used to isolate these targeted ginsenosides. Their structures were elucidated by Q-TOF-MS, ^{1}H-NMR, and ^{13}C-NMR.
Results and conclusion: Total six PPD ginsenosides were isolated from VG. LC/MS together with prep-HPLC are simple and efficiency tools for identifiacation and isolation of saponins from VG. Among them, a novel ginsenoside with 6 sugar molecules together with three ginsenosides with 5-6 sugar molecules were first isolated from VG.

Keywords: Panax vietnamensis, Vietnamese ginseng, ginsenoside

PS1-A-086

Sesquiterpene lactones from the Greek endemic *Inula subfloccosa* Rech. f. (Asteraceae)

Dafni Ioanna Diakaki, Aikaterini Koutsaviti, Efstathia Ioannou, Olga Tzakou

Section of Pharmacognosy and Chemistry of Natural Products, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771 Athens, Greece

Species of the genus *Inula* L. (Asteraceae), which comprises approx. 90 species including about 19 which are native to Europe, are perennial herbs, rarely shrubs, sometimes with an unpleasant smell. Several *Inula* species are used as traditional herbal medicines to treat a broad spectrum of disorders, mainly respiratory, digestive, inflammatory, dermatological, as well as microbial infections. Sesquiterpenes, mostly sesquiterpene lactones including eudesmane, guaiane, pseudoguaiane and germacrane derivatives are characteristic components of *Inula* species, many of which have exhibited a wide range of biological activities, particularly anti-tumor and anti-inflammatory.

In the context of our ongoing research towards the isolation of bioactive secondary metabolites from the Greek flora, the chemical profile of *Inula subfloccosa* Rech. f., an endemic plant of Greece that is restricted to cipolin and marbles, was investigated. The air-dried aerial parts of *I. subfloccosa*, collected during the flowering period, were exhaustively extracted with CH_2Cl_2/MeOH at room temperature. The resulting crude extract was fractionated with a series of chromatographic separations to afford 9 sesquiterpene lactones. Detailed analyses of the NMR and MS data of the isolated metabolites led to the identification of four eudesmanolides, four guaianolides, and one pseudoguaianolide, among which the pseudoguainolide derivative is a new natural product.

Acknowledgements: The authors thank the Special Account for Research Grants of the National and Kapodistrian University of Athens for funding to attend the conference.

Keywords: *Inula subfloccosa*, sesquiterpene lactones, isolation, structure elucidation

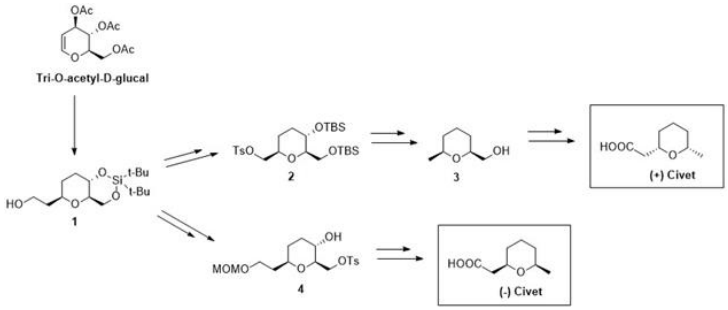
PS1-A-087

Synthesis of the natural product (+) Civet and its enantiomer (–) Civet

Fátima Garrido

Universidade De Vigo, Vigo, Spain

The research of new synthetic methodologies for the formation of carbocycles and heterocycles is one of the most active fields in organic synthesis. These structures are a source of important biological properties, either by themselves or as part of other more complex structures.
Many natural products have tetrahydropyrans in their structure, such as civet. Civet was isolated for the first time in 1978 by Maurer [1] from the perianal glandular pheromone secretion of African civet cat (*Viverra civetta*). This acid is part of a musk with great fixing power, used in perfumery.
We describe the synthesis of (+) Civet and its enantiomer (–) Civet using commercially available t*ri*–*O*–acetyl–*D*–Glucal as starting material. Using a Claisen transposition mediated synthesis of 1 as a key step, (+) Civet and (–) Civet was early obtained through sequence of known reactions [2] with this common intermediate.



Acknowledgements: This work was supported financially by the Xunta de Galicia (ED431C2017/70). The work of the NMR and MS of the research support services of the University of Vigo (CACTI)

Keywords: natural products, tetrahydropyrans, civet.

References:
[1] Maurer B, Grieder A, Thommen W. Helv Chim Acta 1979; 62: 1096.
[2] Isela G, Alioune F, Fátima G, Mohamed G, Generosa G, et al. Organic & Medicinal Chem IJ 2017; 4(4).

PS1-A-088

Qualitative analysis of Herbofix® herbal extracts and comparison with extracts prepared by infusion using HPTLC, HPLC-DAD and UPLC-MS

Ioulia K. Tseti[1], Efstathia Thoma[2], Aikaterini Argyropoulou[3] and Alexios-Leandros Skaltsounis[3]

[1]Uni-Pharma SA, 14th klm National Road 1, Kifissia, GR 145-64, Athens, Greece, [2]PharmaGnose SA, 57th km National Road Athinon-Lamia, GR 320 11 Inofyta, [3]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis, 15771 Athens, Greece

Herbofix® is a new line of herbal infusions in the form of capsules, creating an instant beverage with irresistible flavor and beneficial effects. All the benefits of the plants and their excellent perfumes are gathered in a capsule compatible with Nespresso® machines. The herbs are carefully selected, and harvested from the Mediterranean basin and elsewhere. The recognized properties of the botanicals provide an effective solution to everyday problems. In the current study, the qualitative profile of four different Herbofix® products was investigated using analytical techniques (HPTLC, HPLC-DAD and UPLC-MS). Furthermore, the commercial products were compared with extracts produced with the traditional way (infusion) in order to detect similarities and differences between them. According to the results of the HPTLC analysis, all extracts are rich in bioactive compounds while differences can be detected depending on the type of the extraction (Figure 1).

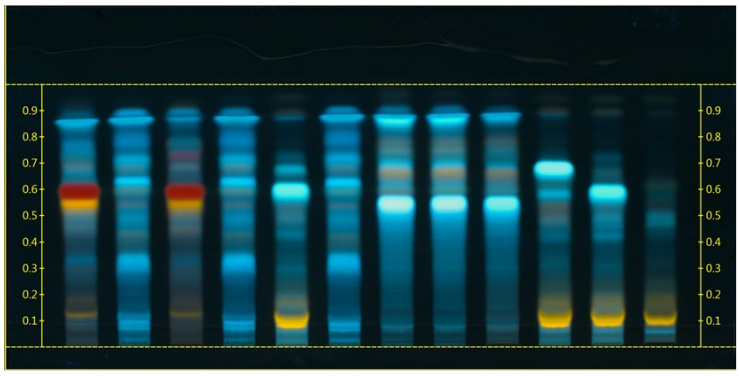


Figure 1: Results of HPTLC of all samples detection at 366 nm after PEG reagent spraying, system of development EtOAc/MeOH/H2O/FA, 50:10:7:1, v/v

PS1-A-089

Alkaloids from grains of *Peganum harmala* from Azerbaijan flora

E.A.Garayev[1], I.S.Movsumov[1], T.A.Nasibova[1], G. Herbette[2], E.E.Garayev[3]
[1]*Azerbaijan Medical University, A Bakikhanov, 21, AZ1000 Baku, Azerbaijan*
[2]*Aix Marseille Univ, CNRS, Centrale Marseille, FSCM, Spectropole, Service 511, 13397 Marseille, France.*

[3]*Neuro-sys SAS, R&D Department, 410 CD 60 Parc de l'Oratoire de Bouc, 13120 Gardanne, France*
Peganum harmala L. *(Nitrariaceae)* is a perennial herb that grows like a weed in the semi-arid steppes of Eastern Europe, Central Asia and Azerbaijan.

The plant contains alkaloids of indole (harmaline, harmine, *etc.*) and quinazoline (*dl*-peganine, peganol, *etc.*) families. The highest concentration of alkaloids is noted in grains.
In folk medicine, infusions and decoctions of *P. harmala* have long been used as a remedy with a calming, analgesic, anti-inflammatory, antiseptic, diaphoretic and diuretic effect. Infusion and decoction of herbs are used for colds, malaria, neurasthenia, nervous and epileptic seizures, for rinsing in the inflammatory processes of the mouth and throat. Baths of *P. harmala* herbs are used in the treatment of rheumatism and various skin diseases.
For the alkaloids sum extraction, dried and grinded grains of *P. harmala* are extracted 3 times by 95% ethanol. Liquid extracts are combined, evaporated on the water bath until 10 mL, mixed with 5% HCl and the obtained solution is filtered. The solution is alkalized by 25% ammonia solution and extracted by liquid/liquid extraction with chloroform 4 times. Fractions are combined, concentrated and analyzed by TLC. On TLC plates, 4 orange spots with R_f 0.69, 0.31, 0.2 and 0.1 (stationary phase "Sorbfil" (Russia), mobile phase chloroform/methanol, 9/1, v/v; revelator Dragendorff solution) are observed.
For the purification, preparative TLC plates were used (stationary phase "Machery - Nagel" (Germany), mobile phase chloroform/methanol, 8.5/1.5, v/v). Lines corresponding to each compound were scraped, extracted by ethanol and filtered.
Based on UV, IR and NMRspectroscopy data, these compounds were identified as harmaline, harmine and tetrahydronorharmine.

Keywords: *Peganum harmala*, harmaline.

References:
1. Al-Shamma A., Drake S., Flynn D.L., Mitscher L.A., Park Y.H., Rao G.S.R., Simpson A., Swayze J.K., Veysoglu T., Wu S.T.S. Antimicrobial agents from higher plants. Antimicrobial agents from *Peganum harmala* seeds // J. Nat. Prod., 1981, V. 446, p. 745-747.
2. Asgarpanah J., Ramezanloo F. Chemistry, pharmacology and medicinal properties of *P.harmala* L // African Journal of Pharmacy and Pharmacology, 2012, V. 6, I. 22, p. 1573-1580.
3. Aslam N., Wani A., Nawchoo I., Aslam M.B., Distribution and Medicinal importance of *P.harmala* // International Journal of Advanced Research, 2014, V. 2, I. 2, p. 751-755.
4. Ayoub M.T., RashanL.J. Isoharmine, a β-carboline alkaloid from *P.harmala* seeds // Phytochemistry, 1991, V. 30, I. 3, p. 1046-1047.
5. Aziz H.G.A., Kader S.M.A., El-Sayed M.M., E.A. El-Malt, E.S. Shaker. Novel β–carboline alkaloid from *P.harmala* as antibacterial agant / Tenth Radiation Physics & Protection Conference.Egypt: 2010, 10 p.
6. Benbott A., Boubendir A., Bahri L., Yahia A. Study of the chemical components of *P.harmala* and evaluation of acute toxicity of alkaloids extracted in the Wistar albino mice // Journal of Materials and Environmental Science, 2013, V.4, I.4, p. 558-565.

7. Bukhari N., Jeon C.W., Choi J.H., Khan M. Phytochemical studies of the alkaloids from *P.harmala* // Applied Chemistry, 2008, V. 12, I. 1, p. 101-104.
8.Fadhil S., Reza M.H., Rouhollah G., Rizi V., Reza M. Spectrophotometric Determination of Total Alkaloids in *P.harmala* L. Using Bromocresol Green // Research Journal of Phytochemistry, 2007, V. 1, I. 2, p. 79-82.
9. Javzan S., Selenge D., Amartuvshin N., Nedelcheva D., Christov V., Philipov S. Alkaloids from Mongolian species of *Peganum multisectum* (Maxim) Bobrov // Mongolian Journal of Chemistry, 2015, V. 16, I. 42, p. 48-53.

PS1-C-001

Caffeic acid-derived biopolymers of medicinal plants, synthesis of its monomer, methylated analogue and their comparative anticancer efficacy

Vakhtang Barbakadze

Tbilisi State Medical University Kutateladze Institute of Pharmacochemistry, Tbilisi, Georgia

According to data of ^{13}C, ^{1}H NMR, 2D heteronuclear $^{1}H/^{13}C$ HSQC, 1D NOE and 2D DOSY experiments the main chemical constituent of high molecular (>1000 kDa) water-soluble preparations from different species of two genera *Symphytum* and *Anchusa* (Boraginaceae family) was found to be poly[oxy-1-carboxy-2-(3,4-dihydroxyphenyl)ethylene] or poly[3-(3,4-dihydroxyphenyl)glyceric acid] (PDPGA). The polyoxyethylene chain is the backbone of this polymer molecule and 3,4-dihydroxyphenyl and carboxyl groups are regular substituents at two carbon atoms in the chain. The repeating unit of this regular polymer is 3-(3,4-dihydroxyphenyl)glyceric acid residue. This compound is a first representative of a new class of natural polyethers. Then the racemic monomer via Sharpless asymmetric dihydroxylation of *trans*-caffeic acid derivative using a potassium osmiate catalyst and methylated derivative of PDPGA were synthesized and compared their pharmacological properties. PDPGA is endowed with intriguing pharmacological activities as anticomplementary, antioxidant, anti-inflammatory, burn and wound healing and anticancer properties. PDPGA and its synthetic monomer exerted anticancer activity *in vitro* and *in vivo* against androgen-dependent and androgen-independent human prostate cancer (PCA) cells via targeting androgen receptor, cell cycle arrest and apoptosis without any toxicity, together with a strong decrease in prostate specific antigen level in plasma. However anticancer efficacy of PDPGA against human PCA cells is more compared to its synthetic monomer. Methylated synthetic analogue of PDPGA did not show any activity against PCA. Overall, this study identifies PDPGA as a potent agent against PCA without any toxicity, and supports its clinical application.

Keywords: poly[oxy-1-carboxy-2-(3,4-dihydroxyphenyl)ethylene], poly[3-(3,4 dihydroxyphenyl)glyceric acid]

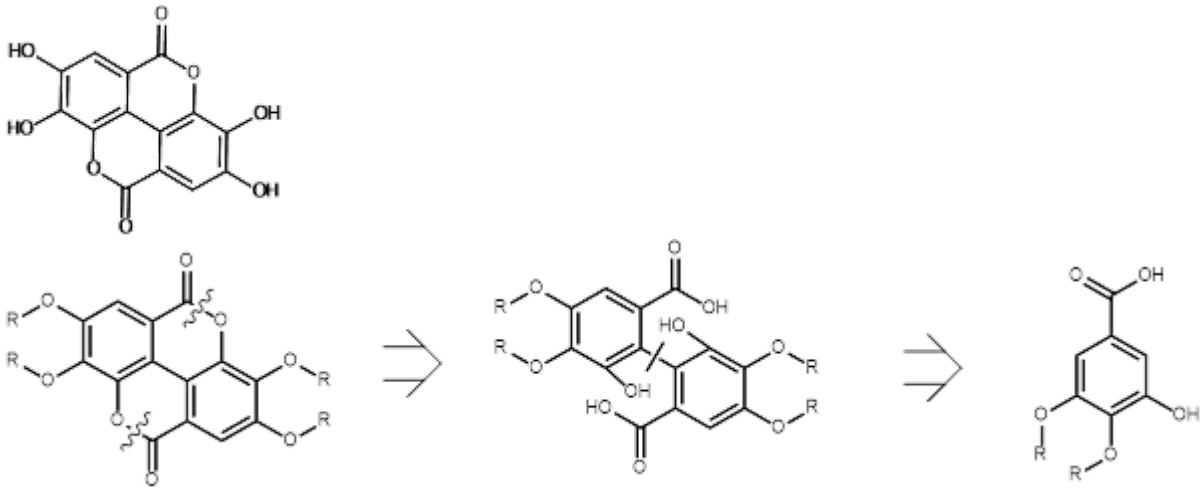
PS1-C-002

Pharmacomodulation of ellagic acid, a promising antiplasmodial agent, to improve its bioavailability

Gilles Degotte[1,2], Aurore Hans[2], Olivia Jansen[1], Allison Ledoux[1], Pauline Desdemoustier[1], Bernard Pirotte[2], Pierre Francotte[2], Michel Frederich[1]

[1]Laboratoire de Pharmacognosie, Centre Interdisciplinaire de Recherches sur le Médicament (CIRM), ULiège, Quartier Hôpital - B36 Tower 4, Avenue Hippocrate 15, Belgium, [2]Laboratoire de Chimie Pharmaceutique, Centre Interdisciplinaire de Recherches sur le Médicament (CIRM), ULiège, Quartier Hôpital - B36 Tower 4, Avenue Hippocrate 15, Belgium

216 million cases and more than 400 000 deaths per year represent two impressive figures concerning malaria, which is also known to take the life of a child every two minutes [1]. So, this parasitic disease remains a major world public health problem. As with antibiotics, the emergence of resistance of parasites (and mosquitoes) is one of the main cause of concern, particularly knowing that artemisinin is affected by such phenomenon [2]. Thus, the design of new antiplasmodial derivatives is imperative in the fight against *Plasmodium falciparum*.
One of the most widespread family of natural compounds, polyphenolic shikimic derivatives, is well known to have multiple pharmacological activities such as antioxidant [3], antitumor [4], antimicrobial [5] and antiplasmodial [6]. Among these molecules, ellagic acid (Fig.1) is one of the most promising antimalarial compounds with proven efficacy *in vitro* (105-330 nM) and *in vivo* after intraperitoneal injection. Even at >5 g/kg, no toxic effect could be demonstrated [8]. This compound seems thus attractive. However, it suffers from a poor oral bioavailability, partly explained by a reduced water solubility [9]. Thus, we work on the pharmacomodulation (Fig.2) of this scaffold to synthetize crowded analogues with enhanced solubility and bioavailability. The obtained compounds will be tested on a 3D7 strain of *Plasmodium falciparum*.



Acknowledgements: Thanks to FNRS and Fonds Léon Fredericq for financial support.

Keywords: ellagic acid, *Plasmodium falciparum*, malaria, pharmacomodulation

References:

[1] WHO World Malaria Report 2016 & 2017.
[2] Lubell *et al*. Malar J 2014; 13: 452.

[3] Wright *et al*. J Am Chem Soc 2001; 6: 1173–1183.
[4] Okuda, Food Factors for Cancer Prevention (1997).
[5] Bisignano *et al*. J Pharm Pharmacol 1999; 51: 971–974.
[6] Köhler *et al*. Zeitschrift fur Naturforsch. - Sect. C J Biosci 2002; 57: 277–281.
[7] Soh *et al*. Antimicrob Agents Chemother 2009; 53: 1100–1106.
[8] Bala *et al*. J Pharm Biomed Anal 2006; 40: 206–210.

PS1-C-003

Preparation of chiral phenylethanols using various vegetables grown in Algeria

Louisa Aribi-Zouioueche, Manhel Bennamane, Samra Razi, Saoussen Zeror

Ecocompatible Asymmetric Catalysis Laboratory (LCAE). Badji Mokhtar Annaba-University. B.P 12, 23000, ANNABA, ALGERIA. Université Badji Mokhtar Annaba Algeria, Annaba, Algeria

In a green context, several reports have described the possibility of using microorganisms or plant as the biocatalyst.[1] Compared with chemical way, biocatalysis attracted more attention due to the large biotechnological potential of enzymatic reaction; the important characteristics of these biocatalysts are their low cost, efficiency and high chemo- and regio-selectivities.[2] We have developed an enzymatic approach for the preparation of optically active alcohols using medlar (*Mespilus germanica* L) [3] or *Citrus reticulate* fruit grown in Annaba (North East Algeria) in water [4].
Ginger root, strawberry tree and mandarin growing in Algeria were evaluated for their ability to stereoselective reduction of prochiral ketones. The reactivity and the enantioselectivity are strongly dependent on the biocatalyst used, and the structure of ketone. High enantioselectivities were observed for some substrates (70–99% ee). Using two different batches of *Citrus reticulata* from two regions of our country Annaba and Skikda, the corresponding optically active alcohols were obtained with high enantioselectivity and Skikda's variety was the best biocatalyst. The results reveal that these plants species can be promising biocatalysts for the production of key intermediates. [5]

Acknowledgements: The authors acknowledge the Algerian Ministry of education and scientific research (FNR 2000 and PNR).

Keywords: Bioreduction, chiral alcohols, *Citrus reticulata*, *Arbutus unedo* L, *Zingiber officinale*.

References:

[1] Ni Y, Xy J H. Biotechnol. Adv. 2012; 30: 1279-1288. b) Birolli W G, Ferreira I M, Alvarenga N, De Santos D A, De Matos I L, Comasseto J V, Porto A L M. Biotechnol. Adv. 2015; 33: 481–510
[2] Zeror S, Collin J, Fiaud J-C, Aribi-Zouioueche L. Tetrahedron: Asymmetry 2010; 21:1211–1245
[3] Bennamane M, Zeror S, Aribi-Zouioueche L, Biocatal. Biotransfor 2014; 32: 327–332
[4] Bennamane M, Zeror S, Aribi-Zouioueche L. Chirality 2015; 27: 205–210
[5] Bennamane M, Razi S, Zeror S, Aribi-Zouioueche L. Biocatal. Agric. Biotechnol 2018; 14: 52–56

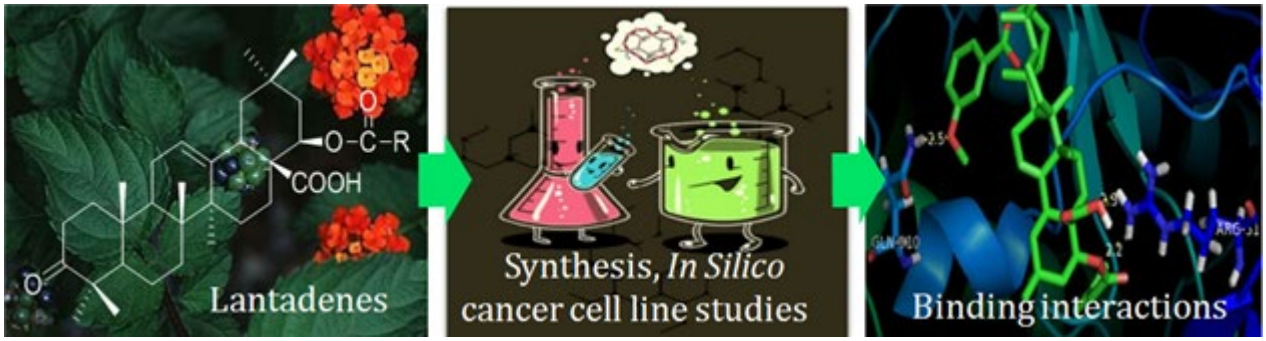
PS1-C-004

Invasive weed lantadene derivatives: experimental and computational studies

Monika Chauhan[1], Manu Sharma[2], Sharad Kumar Suthar[3], Hong Boo Lee[4], Richa Dhingra[5], Neelima Dhingra[6]

[1]University Institute of Pharmaceutical Sciences, Panjab University, Chandigarh, India, Chandigarh, India, [2]Jay Pee University of Information Technology, Solan, India, [3]Jay Pee University of Information Technology, Solan, India, [4]Cancer Research Initiative Foundation, Drug Discovery Laboratory, Taman Perindustrian UEP, Malaysia, [5]University Institute of Pharmaceutical Sciences, Panjab University, Chandigarh, India, Chandigarh, India, [6]University Institute of Pharmaceutical Sciences, Panjab University, Chandigarh, India, Chandigarh, India

Natural products and their molecular frameworks have a long tradition as valuable starting points for medicinal chemistry and drug discovery. Herbal remedies were the first medicines used by humans due to the many pharmacologically active secondary metabolites produced by plants. Some of the metabolites like Lantadenes from the weed *Lantana camara* have been known to inhibit cell division and showed anti-antitumor potential. Column chromatography was used to separate and to purify two Lantadenes the leaves of *Lantana Camara* using a two-phase solvent system composed of hexane: ethyl acetate (4:1). Further, derivatization was out at C-3 position to get different substituted (aliphatic /aromatic) analogues of lantadenes, followed by their spectral characterization and evaluation for cytotoxicity and inhibitory potential against TNF-α induced activation of NF-κB in lung cancer cell line A549. The lead molecule meta substituted benzoyloxy analogue inhibited kinase activity of IKK in a single-digit micromolar concentration. At the same time, lead molecule showed promising cytotoxicity against A549 lung cancer cells with IC_{50} of 0.98l μm. Furthermore, we have highlighted the potential of innovative computational tools in processing structurally complex natural products to predict their affinity for macromolecular targets and attempt to forecast the pre-ADME properties of lead molecule. Encouraging results indicates the Lantadene's potential to be developed as anticancer agents.



PS1-C-005

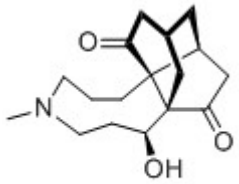
Studies on the total synthesis of Palhinine A

Shu Xu, Liang Li, Xiaolei Wang, Yaling Gong, Shichao Lu

State Key Laboratory of Bioactive Substance and Function of Natural Medicines, Beijing Key Laboratory of Active Substances Discovery and Drugability Evaluation, Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China

Palhinine A, a C16*N*–type *Lycopodium* alkaloid, was isolated from the whole plant of *Palhinhae cernua* L. (Lycopodiaceae) in 2010 [1] Structurally, it contains a unique 5/6/6/9 tetracyclic core and a nine–membered azonane ring. The low content in the natural sources largely limited its bioactivity evaluations. Chemical synthesis provides a chance to its further biological investigations. Thus far, several groups have reported their synthetic strategies [2]. However, only one racemic total synthesis has been achieved [3].

Herein, we reveal our recent result towards the total synthesis of Palhinine A. Comparing with the reported Diels–Alder or related domino Michael reaction strategies, we utilized stereoselective radical reactions to construct the caged tetracyclic core structure. Our results provide a promising pathway for the enantioselective synthesis of Palhinine A and facilitate the systematic synthetic and biological investigations of the members of Palhinine family natural products.



Palhinine A

References:

[1] Zhao FW, Sun QY, Yang FM, Hu GW, Luo JF, Tang GH, Wang YH, Long CL. Org Lett 2010;12: 3922.
[2] Zhao C, Zheng H, Jing P, Fang B, Xie X, She X. Org Lett 2012;14: 2293.
[3] Zhang GB, Wang FX, Du JY, Qu H, Ma XY, Wei MX, Wang CT, Li Q, Fan C A. Org Lett 2012; 14: 3696.
[4] Gaugele D, Maier ME. Synlett 2013; 24: 955.
[5] Sizemore N, Rychnovsky SD. Org Lett 2014;16: 688.
[6] Duan S, Long D, Zhao C, Zhao G, Yuan Z, Xie X, Fang J, She X. Org Chem Front 2016; 3: 1137.
[7] Wang FX, Du JY, Wang HB, Zhang PL, Zhang GB, Yu KY, Zhang XZ, An XT, Cao YX, Fan CA. J. Am Chem Soc 2017;139: 4282.

PS1-C-006

Studies on the total synthesis of Mollanol A

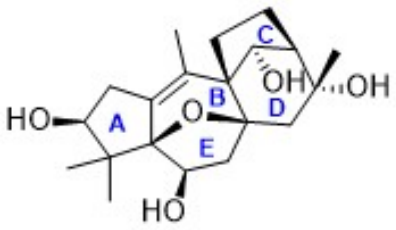
Yixuan Zheng, Jianzhuang Miao, Mei Li, Yuyan Liang, Linna Wang, Shu Xu

State Key Laboratory of Bioactive Substance and Function of Natural Medicines, Beijing Key Laboratory of Active Substances Discovery and Drugability Evaluation, Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China

Mollanol A is a diterpenoid isolated from the fruits of *Rhododenon molle* from Guang xi, China in 2014. It was found to exhibit transcriptional activation effects on the xbp1 upstream promoter in IEC–6, 293T, and RAW264.7 cells, which implied its potential application in anti–ulcerogenic colitis treatment [1]. Further examination of its biological activity could not be extensively carried out due to the low content in the plant.

Structurally, Mollanol A has a new grayananoid carbon–skeleton with an unprecedented C–nor–D–homo grayanane ring system. Due to the formation of a new oxygen–bridge between C–5 and C–8, Mollanol A has a fifth ring (E ring) than common grayanane diterpenes. Mollanol A contains 8 stereogenic centers, 5 of which are contiguous on the fused CD ring segments. The grayananoid is a big diterpene family having various kinds of bioactivities [2]. However, because of their complex structure, few examples were reported for their chemical synthesis [3]. For Mollanol A, no synthetic work has been disclosed.

Herein, we reveal our recent result towards the total synthesis of Mollanol A. On the basis of retrosynthetic analysis, our convergent strategy first achieved the highly stereoselective construction of the fused CD ring through an asymmetric Michael addition reaction and an intramolecular aldol cyclization. Another key reaction was also realized with a diastereoselective coupling addition to link the CD ring and A ring segments.



Mollanol A

References:

[1] Li Y, Liu YB, Liu YL, Wang C, Wu LQ, Li L, Ma Sb G, Qu J, Yu SS. Org Lett 2014; 16: 4320–4323.
[2] Li Y, Liu YB, Yu SS. Phytochem Rev 2013;12: 305–325.
[3] Kan T, Hosokawa S, Nara S, Oikawa M, Ito S, Matsuda F, Shirahama H. J Org Chem 1994; 59: 5532–5534.

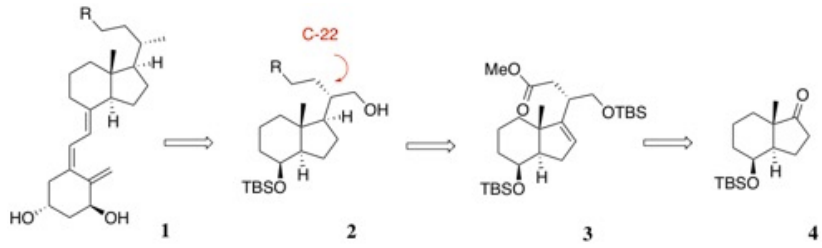
PS1-C-007

Approach to C–20 epimers of vitamin D analogues

Hugo Santalla, Fátima Garrido

Universidade de Vigo, Vigo, Spain

The 1α,25–(OH)2–vitamin D_3 has antiproliferative properties and could be used as a therapeutic agent. However, the necessary therapeutic doses involve hypercalcemia risk [1]. Research focuses on the rational design of new calcitriol analogues with higher selectivity, the so–called second–generation analogues, with side chain modification.
Second generation analogues, some of which are marketed as drugs can provide interesting improvements regarding the pharmacological and pharmacokinetic properties of the original analogue, by simple epimerization of C–20.
This work is focused on the design of a common pathway for obtaining epimers at the C–20 position of vitamin D second generation analogues. From the route initially designed for the obtention of Gemini [2] type analogues, the desired epimer's structure can be reached by the reduction of theC–22 hydroxyl group of compound 2, as shown on the scheme [3].



Acknowledgements: This work was supported financially by the Xunta de Galicia (CN 2012/184). The work of the NMR and MS divisions of the research support services of the University of Vigo (CACTI) is also gratefully acknowledged. A. M. thanks the University of Vigo for a fellowship

Keywords: Analogue, antitumour activity, Gémini, vitamin D.

References:

[1] Jones G, Strugnell SA, DeLuca HF. Physiol Rev 1998; 78: 1193.
[2] Pazos G. Tesis Doctoral. Universidad de Vigo 2011;115–163.
[3] Santalla H, Martínez A, Fátima G, Gómez G, Fall Y. Org Chem Front 2017; 4: 1999–2001.

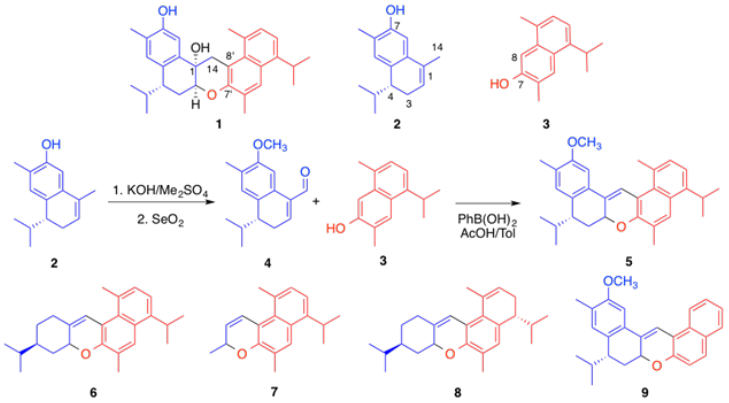
PS1-C-008

Semisynthetic approaches to dicadalenol and some analogues

José Luis Ávila, Guillermo Delgado

Instituto de Química, Universidad Nacional Autónoma de México, Ciudad Universitaria, Coyoacán 04510, Mexico

Dicadalenol (1) is a compound isolated from *Heterotheca inuloides*, a plant used in Mexico for treating inflammatory ailments. It exhibited greater anti–inflammatory activity than that of indomethacin in murine ear edema model [1]. Biogenetically, it is a dimer of cadinanes, and there are some examples of dimeric sesquiterpenoids [2]. We envisaged the possibility of preparing 1 from 3,4–dihydro–7–hydroxycadalene (2) and 7–hydroxycadalene (3), which are major metabolites from *H. inuloides* [3,4].
After experimentation of several synthetic approaches, the connectivity needed for 1 was achieved through acid–catalyzed 1,4–addition of 3 to *α,β*–unsaturated aldehyde 4 (obtained by allylic oxidation of 2), followed by cyclization and dehydration.
Compounds 6–9 were also prepared by reacting cadinanes 2 and 3 or naphthol with the appropriate *α,β*–unsaturated carbonyl compound.
The experimental results showed the intrinsic reactivity of 7–hydroxy–substituted cadinanes, and give an insight to a possible biogenetic route for 1.



Acknowledgements: The authors acknowledge Conacyt (grant no. 294731), DGAPA UNAM (PAPIIT IG200318), and Programa de Maestría y Doctorado en Ciencias Químicas (UNAM) for financial support.

Keywords: Dicadalenol, cadinanes, antiinflammatory activity

References:

[1] Delgado G, Olivares M del S.; Chávez MI, Ramírez–Apan T, Linares E, Bye R, Espinosa–García FJ. J Nat Prod 2001; 64: 861–864.
[2] Zhan ZJ, Ying YM, Ma LF, Shan WG. Nat Prod Rep 2011; 28 (3): 594.
[3] Rodríguez–Chávez JL, Egas V, Linares E, Bye R, Hernández T, Espinosa–García FJ, Delgado G. J Ethnopharmacol 2017; 195: 39–63.
[4] Egas, V.; Toscano, R. A.; Linares, E.; Bye, R.; Espinosa–García, F. J.; Delgado, G. J Nat prod 2015; 78 (11): 2634–2641.

PS1-C-009

Discovery of new 5-arylcarboximidamidopyrazolo[3,4-c]pyridines with potent antiproliferative activity

Panagiotis Marakos[1], Athanassios Papastathopoulos[1], Nikolaos Lougiakis[1], Nicole Pouli[1], Harris Pratsinis[2], Dimitris Kletsas[2]

[1]National and Kapodistrian University of Athens, Athens, Greece, [2]NCSR ''Demokritos,'' Athens, Greece, Athens, Greece

Cancer is the second cause of death globally and only in the United States about 600000 cancer deaths are estimated for 2017.1 Thus, the search for new compounds that inhibit uncontrolled cell proliferation remains an active area of research, due to the continuous need for the development of novel chemotherapeutic agents. Among the anticancer agents currently being in clinical trials or in clinical use, purine isosters are of great importance, since a number of new drugs possess such an isosteric scaffold, mainly acting as kinase inhibitors, which mimick ATP in its binding pocket.2

As part of an on-going project concerning the synthesis of purine isosters with potential antiproliferative and kinase inhibitory activity, we have identified a 5-anilinocarboximidamide-substituted pyrazolo[3,4-c]pyridine, possessing IC_{50} values in the nm range against a panel of cancer cell lines. Using this promising lead compound, we have synthesized a number of novel 5-arylcarboximidamido-substitued derivatives, which bear suitable substituents at positions 1, 2, 3 and 7 as well.

The compounds were synthesized using 2-amino-4-methylpyridine, which was first converted to 5-chloro-1*H*-pyrazolo[3,4-c]pyridine. This intermediate was suitably substituted and upon a Suzuki-type coupling provided the corresponding 3-phenyl analogues. Selected amines were then introduced at position 7 of the scaffold, followed by the catalytic insertion of a 5-nitrile moiety and its nucleophilic attack by anilines, thus resulting in the target derivatives.

In total, 42 derivatives have been synthesized and evaluated for their cytotoxic activity against MDA-MB-231, HT-1080 and PC-3 cancer cell lines. Among them, 25 compounds proved to be active, showing IC_{50} values within the range of 0.64-12.8 μM. It is noticeable that the substitution pattern of the most active analogues is in direct agreement with their potency. The investigation of the exact mechanism of action of this novel class of compounds is currently in progress.

PS1-C-010

Synthesis of novel multisubstituted pyrazolo[3,4-c]pyridines and evaluation of their antiproliferative and antiangiogenic activity

Nikolaos Lougiakis, Stavros Kampoukos, Vassili Konstantin, Nicole Pouli, Panagiotis Marakos, Heleni Loutrari

National and Kapodistrian University of Athens, Athens, Greece

Angiogenesis is a necessary procedure for the continuous growth of solid tumors and even if its prevention is a proven strategy for the treatment of dancer, it is still considered as an important unmet need [1]. The present work is a follow-up of an ongoing project of our research group, aiming in the discovery of new antiangiogenic agents. We have previously identified a series of derivatives with very promising activity [2] and this fact prompted us to attempt the amelioration of the profile of the most interesting analogue. We have thus planned the rational modification of selected parts of this lead compound, through the use of suitable substituents, a strategy that will potentially allow us to investigate in depth the molecular mechanism of action of this class of compounds and design more effective agents.
The new compounds are 1,3,5,7-tetrasubstituted derivatives of pyrazolo[3,4-*c*]pyridine and their synthesis was effected using 2-amino-4-picoline as starting material. This picoline was initially converted to the important intermediate 5-chloropyrazolo[3,4-*c*]pyridine, following a well-established procedure. Suitable manipulation of this intermediate allowed the appropriate insertion of the selected substituents in the central heterocyclic scaffold.
Totally, 22 novel derivatives were synthesized and fully characterized. The target pyrazolopyridines were initially screened for their ability to inhibit the proliferation of endothelial cells, at two representative concentrations (0.2 and 2 μM). Seven derivatives showed interesting activity and the IC_{50} value of the most promising one was determined (3.2±0.9μM) and proved to be lower than the corresponding IC_{50} value of the lead compound (7.2±2.4μM). The biological evaluation of these new compounds is currently in progress.

Keywords: pyrazolopyridine, synthesis, purine isosteres, angiogenesis

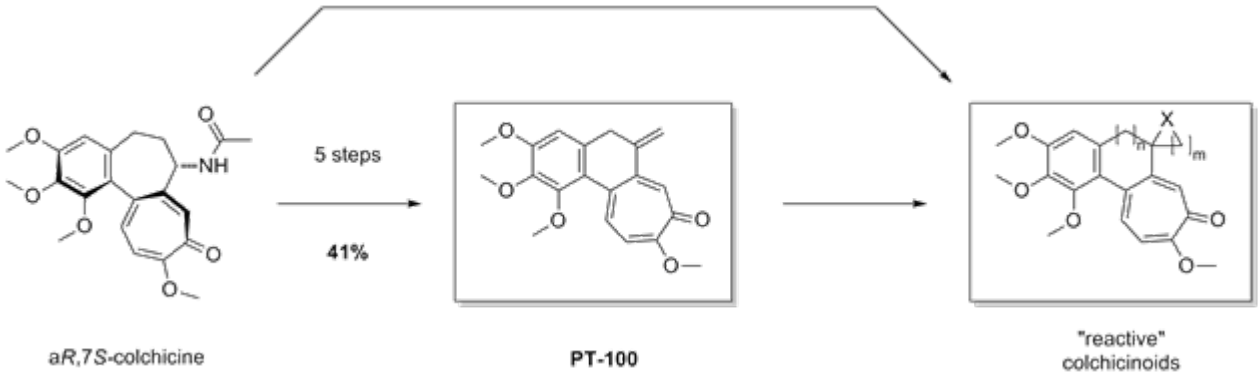
PS1-C-011

Adventures in the synthesis and biological evaluation of novel "reactive" colchicine-derived compounds

Andreas Stein[1], Persefoni Thomopoulou[1], Tim Schulte[1], Benoit Gigant[3], Aram Prokop[2], Hans-Günther Schmalz[1]

[1]Department of Chemistry, University of Cologne, Cologne, Germany, [2]Department of Paediatric Oncology, Children's Hospital Cologne, Cologne, Germany, [3]Institute for Integrative Biology of the Cell (I2BC), CEA, CNRS, Univ. Paris-Sud, Université Paris-Saclay, Paris, France

Colchicine, the main active alkaloid from *colchicum autumnale*, is known as a potent tubulin-binding agent and therefore a highly interesting lead structure for the development of new anti-cancer chemotherapeutics. In continuation of our research in the colchicine field, we describe the synthesis of novel colchicinoids with potentially reactive structural motifs, and their surprising biological activity. The investigation is centered on a compound called "PT-100" (cp. Figure 1), a recently discovered *exo*-methylene colchicinoid with a contracted B-ring. We disclose the synthesis, interesting chemical behaviour and impressive biological performance of PT-100 as a highly active anti-tumoral compound as well as the investigation of the synthesis of a variety of conceptually similar colchicinoids, and some surprising transformations in the course of this project.



PS1-C-012

Castor oil as feedstock for the production of 12-Fatty Acid esters of Hydroxy Fatty Acids (12-FAHFAs)

George Koutoulogenis, Valentini Avramidi, Evrydiki Katsikari, George Kokotos

National and Kapodistrian University of Athens, Athens, Greece

Vegetable oils are renewable resources and their fatty acids may serve as starting materials for the synthesis of bio-based products. Castor oil is an attractive feedstock for the preparation of industrially important products, because it contains ricinoleic acid (12-R-hydroxyoleic acid) as predominant fatty acid. Recently, a novel class of endogenous lipids presenting anti-diabetic and anti-inflammatory activity has been identified [1]. These lipids were named Fatty Acid esters of Hydroxy Fatty Acids (FAHFAs) and contain two long aliphatic chains connected through an ester bond. Several branched FAHFAs families have been identified and each family consists of multiple regioisomers in which the hydroxyl participating in the ester bond is at different positions (5-, 7- etc). Here, we describe the synthesis of various 12-FAHFAs using castor oil as starting material. Methanolysis of castor oil followed by saponification provided ricinoleic acid and subsequently, 12-(R)-Hydroxy Stearic Acid, after hydrogenation. 12-Palmitic Acid ester of Ricinoleic Acid (12-PARA) was synthesized after protecting the carboxyl group of ricinoleic acid with phenacyl bromide, coupling with palmitic acid, and finally deprotection with Mg/AcOH. Following a similar synthetic route using oleic acid, 12-Oleic Acid ester of Ricinoleic Acid (12-OARA) was synthesized. To obtain the potential bioactive 12-Palmitic Acid ester of Hydroxy Stearic Acid (12-PAHSA), hydrogenation of 12-PARA took place. Finally, the synthesis of 12-Ricinoleic Acid ester of Ricinoleic Acid (12-RARA) was carried out. This synthetic route required two appropriately protected derivatives of ricinoleic acid; a 12-*O*-tetrahydrofuranyl derivative and a phenacyl ester. Coupling of these derivatives and subsequent deprotection led to 12-RARA. Thus, various 12-FAHFAs suitable for biological evaluation were synthesized.

Acknowledgements: The research presented was carried out within the framework of a Stavros Niarchos Foundation grant to the National and Kapodistrian University of Athens

Keywords: Castor oil, 12-FAHFAs, ricinoleic acid

References:

[1] Yore MM, Syed I, Moraes-Vieira PM, Zhang T, Herman MA, Homan EA, Patel RT, Lee J, Chen S, Peroni OD, Dhaneshwar AS, Hammarstedt A, Smith U, McGraw TE, Saghatelian A, Kahn BB. Cell 2014; 159: 318–332

PS1-C-013

Design, synthesis and pharmacological evaluation of novel TDP1 inhibitors

Maria Karelou[1], Keli Agama[2], M.D., Yves Pommier[2], Ioannis Kostakis[1]

[1]Division of Pharmaceutical Chemistry, Department of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [2]Developmental Therapeutics Branch and Laboratory of Molecular Pharmacology, Center for Cancer Research, National Cancer Institute, NIH, Bethesda, USA

Cancer is one of the deadliest diseases, responsible for about 13% of all deaths worldwide. It is caused by genetic abnormalities related to DNA of the affected cells; therefore, targeting the DNA repair pathways could improve the efficacy of DNA-damaging anticancer drugs, such as clinically significant Topoisomerase-I and Topoisomerase-II inhibitors. Given that Tyrosyl-DNA phosphodiesterase 1 repairs stalled topoisomerase - DNA complexes, the inhibition of this DNA repair enzyme could have deadly effects in cancer cells. Here, we describe the design, synthesis and pharmacological evaluation of novel tricyclic compounds as TDP1 inhibitors. The new compounds bear the acridine or aza-acridine core, possessing one or two basic side chains. The early results suggest, in accordance with *In Silico* calculations, that the second basic side chain is essential for the activity against TDP1 while the first side chain slightly enhances the activity. Additionally, the presence of the 7-OCH_3 is crucial for the activity of the molecules.

PS1-C-014

Hydroxytyrosol, a versatile natural compound as chemical probe in drug design

Aikaterini Komianou[1], Ioanna Kalpaktsi[1], Alexios-Leandros Skaltsounis[2], Ioannis Kostakis[1]

[1]Division of Pharmaceutical Chemistry, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis-Zografou, Athens, Greece, [2]Division of Pharmacognosy and Natural Product Chemistry, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis-Zografou, Athens, Greece

Polyphenols are a wide family of compounds found in plant-based foods such as cranberries, grapes, olives, walnuts and have many diverse biological activities. One of the most important components of the polyphenol family is hydroxytyrosol (HT), a simple *o*-diphenol found in leaves and fruits of olive tree, virgin olive oil, as well as its wastewaters.

The high interest concerning this compound focuses on its attribution in many aspects of human health. There are clear epidemiological and biochemical data indicating that this compound is endowed with significant antithrombotic, antiatherogenic, anti-inflammatory, anticancer and antimicrobial activities. There are also evidences that associate HT with neuroprotective properties due to the interference with *β*-amyloid peptide and Tau protein aggregation, showing preventive effect in the Alzheimer's disease or other brain damages. Additionally, according to recent studies, HT protects against the degeneration of macula by reducing the oxidative stress in the epithelial cells of the retina, occurring by the accumulation of acrolein. Moreover, this polyphenol seems to interact in the formation of the bones and the preservation of the bone mineral density, resulting in the positive effects against osteoporosis and bone loss.

According to the above-mentioned data, it was considered interesting to synthesize a series of HT analogs in order to study their potential antifungal and antimicrobial activity as well as their protective activity against macular degeneration and osteoporosis. Additionally, selected compounds were evaluated *in vitro*, by means of their interaction with Ab(1-28) with electrospray ionization mass spectrometry and circular dichroism.

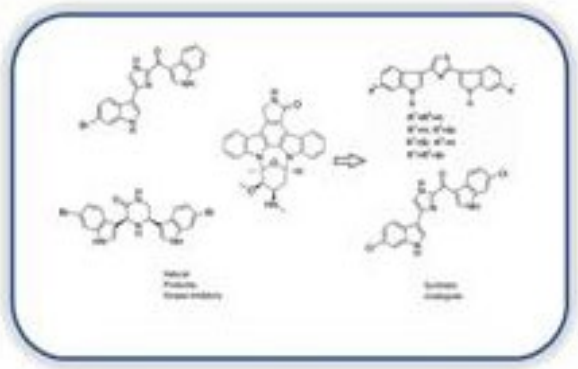
PS1-C-015

Design and Synthesis of new Modified hybrids of bisindole Derivatives as Chemical tools in the study of Anticancer Activity

Msc. Amalia Dimitrios Kalampaliki[1], Eleftheria Athanasios Georgiou[1], Dr Ioannis Konstantinos Kostakis[1]

[1]*Division of Pharmaceutical Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, 157 71, Zografou, Greece*

These days there is an indispensable need to synthesize new drugs with intended anticancer activity. Two high value heterocyclic cores that possess such characteristics and properties are indole and quinazoline rings, which have been used as anticancer agents and are part of some of the most well-known drugs. Important compounds- antibiotics with broad spectrum antitumor activity, secluded from plants or synthesized, are some bisindole alkaloids (i.e. vinblastine) and some pyrrolocarbazoles (i.e. Staurosporine, topsentin). These compounds exceed their activity by inhibiting proliferation, invasion and/ or metastasis of a variety of tumor cell lines. The mechanisms that are involved in these compounds' anticancer properties, are based on their ability to inhibit protein kinases and other distinct pathways of antitumor and/or antimicrobial activities. The main goal of this project is to design and synthesize novel hybrids of bisindole derivatives which will include in their main structure the important indole and quinazolinone moieties, which will improve and moderate the wide spectrum activities of the natural compounds. These new synthesized compounds are biologically tested in order to evaluate the effect of them on topoisomerases' and/or protein kinases' inhibition, as well as on the tumor cell growth inhibition.



Keywords: cancer; bis indole alkaloids; pyrrolocarbazoles; analogues; topoisomerases; kinases;

References:

1. A. Andreani, R. Morigi, M. Rambaldi, L. Varoli, L. Landi, C. Prata, M.V. Berridge, C. Grasso, H.-H. Fiebig , G. Kelter, A.M. Burger, M.W. Kunkel, S. Burnelli, M. Granaiola, A. Leoni, A. Locatelli, Antitumor Activity of Bis-Indole Derivatives, J Med Chem. 2008 51, 4563–4570.
2. J.-K. Son, H.W. Chang, Y. Jahng, Progress in Studies on Rutaecarpine. II.—Synthesis and Structure-Biological Activity Relationships, Molecules 2015, 20, 10800-10821.

PS1-C-016

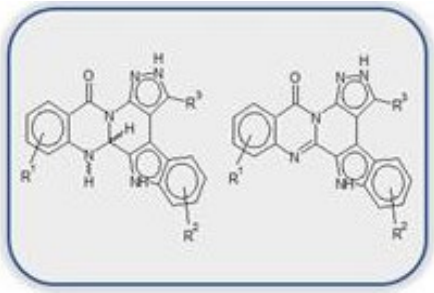
Design and synthesis of novel staurosporine and rutaecarpine hybrids as therapeutic leads in the treatment of cancer

Amalia Dimitrios Kalampaliki, Ioannis Konstantinos Kostakis

Division of Pharmaceutical Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, 157 71, Zografou, Greece

Cancer is one of the major problems these days. In 2016, more than 1.7 million people died because of cancerous diseases. This made indispensable the synthesis of new drugs with intended anticancer activity. Very important compounds, secluded from plants with such characteristics and properties are Staurosporine and Rebeccamycin, as well as Rutaecarpine and Evodiamine and their analogues. Staurosporine and Rebeccamycin are indolocarbazole antibiotics with broad spectrum antitumor activity. Rebeccamycin and its analogues stabilize the DNA-topoisomerase cleavage complex, whilst Staurosporine is one of the strongest known protein kinases inhibitors. In addition, indoloquinazoline compounds, Rutaecarpine and Evodiamine possess anti-cancer activities both *in vitro* and *in vivo* by inhibiting proliferation, invasion and metastasis. In this project, using these scaffolds as lead compounds, the design and synthesis of several hybrids considered to be very interesting, due to the multiple signaling pathways affected by the above mentioned compounds. The aim of this research is to synthesize derivatives that with small changes in their main scaffold will turn from multitargeted to selective antitumor agents. The new compounds are under pharmacological evaluation against a variety of molecular targets and cancer cell lines. Additionally, *in silico* optimization is conducted in order to escalate the potency of the new compounds. Finally, all these experiments will contribute to exclude safer structure activity relationships and to a further synthesis of new compounds which will be evaluated against the determined molecular targets.

Keywords: cancer; Staurosporine; Rutaecarpine; hybrids; topoisomerases; kinases;



References:

[1] Son J-K, Chang HW, Jahng Y. Molecules 2015; 20: 10800–10821.
[2] Sherer C, Snape TJ, Eur. J Med Chem 2015; 95: 552–560.

PS1-C-017

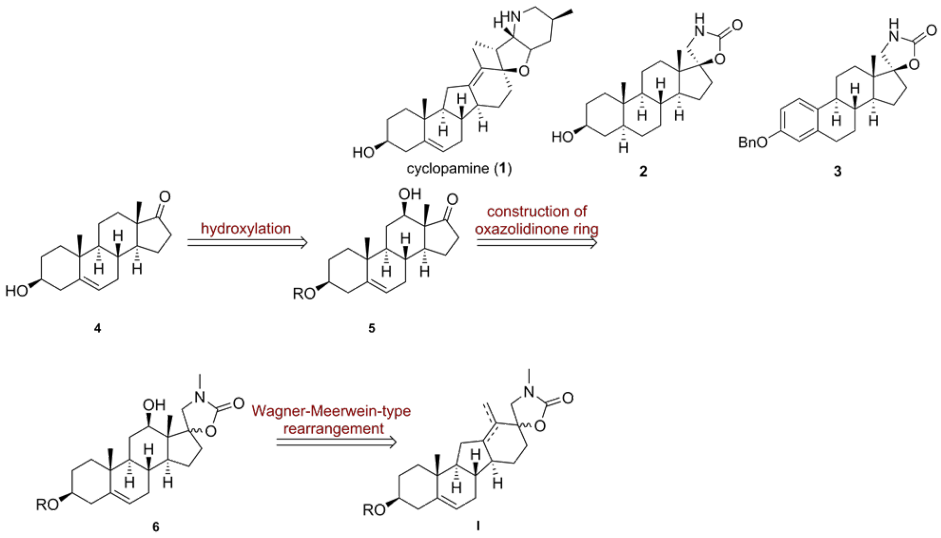
Spiro-oxazolidinone C-nor-D-homo steroids as potential inhibitors of hedgehog pathway

Roumana Aggeliki[1], Dimanthi Pliatsika[2], Sotiris Nikolaropoulos[1], Athanassios Giannis[2], Manolis Fousteris[1]

[1]Laboratory of Medicinal Chemistry, Department of Pharmacy, University of Patras, Patras GR-26500, Greece, [2]Institute of Organic Chemistry, Faculty of Chemistry and Mineralogy, University of Leipzig, Leipzig 04103, Germany

Hedgehog (Hh) signaling pathway regulates embryonic development, but its activation plays a crucial role in several solid and hematological malignancies. Cyclopamine (1), a hexacyclic isosteroidal alkaloid isolated from *Veratrum californicum*, was identified as an antagonist of the seven-pass transmembrane protein Smoothened (Smo) (IC_{50} = 5 μM), which a key component of the Hh pathway. Although, cyclopamine presents interesting antitumor activity, it suffers from poor aqueous solubility and low metabolic stability [1].FIGURE Prompted by these data, we have focused on the discovery of new structurally simplified cyclopamine-like analogues.[2] Thus, we envisaged the replacement of cyclopamine E-F ring system with a resembling, easily constructed structural motif, an intriguing modification which may result in analogues with potential Hh signaling inhibitory activity and improved chemical and metabolic stability. Based on previous findings which indicated the antiproliferative activity of spiro-oxazolidinone derivatives 2 and 3 against tumor cell lines, [3] we designed new derivatives I (Scheme), which incorporate the standard C-nor-D-homo framework and a C17 spiro-oxazolidinone ring. Starting from commercial available dehydroxyepiandrosterone 4, the synthesis of the target molecules has been accomplished via copper-mediated stereoselective 12*β*-hydroxylation, followed by E-oxazolidinone ring construction and Wagner–Meerwein-type rearrangement of the steroidal skeleton. The C17 stereochemistry was assigned by 1D- and 2D-NMR experiments. Studies regarding the Hh signaling inhibitory activity of the target molecules are in progress.

Scheme. Synthetic approach towards the new C17 spiro-oxazolidinone C-nor-D-homo steroids I.



PS1-C-018

Design and synthesis of stilebenoid and chalconoid analogues as potent tyrosinase modulators

Argyro Vontzalidou[1], S.M, Dimitrakoudi[1], K Tsoukalas[1], E Chaita[1], G Zoidis[2], C Cheimonidou[3], I.P Trougakos[3], G Lambrinidis[2], E Mikros[2], A.L Skaltsounis[1], N Aligiannis[1]

[1]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [2]Department of Pharmaceutical Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [3]Department of Cell Biology and Biophysics, Faculty of Biology, National and Kapodistrian University of Athens, Athens, Greece

Tyrosinase is a multifunctional copper-containing enzyme, playing a key role in melanin biosynthesis catalyzing the oxidation of phenol to *o*-quinone in the early stage of melanisation in animals. Moreover, the presence of the oxidation products of L-tyrosine has been linked to the demise of neurons in Parkinson's disease. Consequently, tyrosinase inhibitors can be useful for the treatment of some dermatological disorders associated with melanin hyperpigmentation whereas the study of tyrosinase activity is clearly important for the development of therapeutic agents concerning neurodegenerative disorders. Our aim was to develop lead compounds by investigating the structural requirements for optimal tyrosinase inhibition using molecular simulation studies. Previous *in silico* and *in vitro* evaluation displayed that dihydrostilbene analogues exhibited strong anti-tyrosinase activity [1]. In order to get a better insight to the stereoelectronic features for optimal activity, a number of diarylpropanoic and diarylpropenic acids, chalcones and diarylpropanes were designed and synthesized. In total, 38 compounds were synthesized, fully characterized by spectroscopic methods and were *in vitro* evaluated for their inhibitory activity against tyrosinase using L-DOPA as a substrate. According to the results diarylpropane analogues emerged as potent tyrosinase inhibitors, whereas diarylpropanoic acids seemed to enhance the enzymatic activity. 4 of the tested compounds were further evaluated in melanoma cell lines, B16F1 and B16F10, for their ability to moderate tyrosinase activity and affect melanin production. The obtained results were in accordance with the previous *in vitro* evaluation. *In silico* evaluation of their binding affinity with the tyrosinase receptor was also conducted. Considering all the above, the aforementioned compounds could be proved promising therapeutic agents for the treatment of several dermatological and/or neurodegenerative disorders [2].

References:

[1] Bioorg Med Chem Lett 2012; 22: 5523–5526.
[2] Food Biosci 2014; 6: 17–23.

PS1-C-019

Further exploration of the indirubin scaffold affords 5'-6 disubstituted analogues with potent cytotoxicity and *in vivo* efficacy targeting Src family kinases

Nicolas Gaboriaud-Kolar[1], Marina Kritsanida[1,2], Vassilios Myrianthopoulos[3], Nikolaos Lougiakis[3], Anastasia Papachristodoulou[3], Anna Tsantili[3], Ioannis Rerras[3], Laurence Wennogle[4], Emmanuel Mikros[3], Sangil Nam[*,5], Alexios-Leandros Skaltsounis[*,1]

[1]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, University of Athens, Panepistimiopolis Zografou, GR-15771, Athens, Greece, [2]UMR COMETE 8638 CNRS - Université Paris Descartes, Sorbonne Paris Cité, Faculté de Pharmacie, 4 Avenue de l'Observatoire, 75270 Paris cedex 06, France, [3]Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Athens, Panepistimiopolis Zografou, GR-15771, Athens, Greece, [4]Intra-Cellular Therapies, Inc., Alexandria Center for Life Science, 430 East 29th Street, Suite 900, New York, N.Y. USA 10016, [5]Molecular Medicine; Beckman Research Institute; City of Hope Comprehensive Cancer Center; Duarte, CA USA

Indirubins are naturally occurring bis-indoles with potent kinase-inhibitory properties. The pronounced synthetic versatility of the indirubin core has attracted considerable attention, giving rise to a series of successful synthetic efforts aiming at the development of highly potent and selective kinase inhibitors. In a continuation of our systematic studies on interrogating the interplay between structure and biological activity of indirubins, we present here the synthesis of a novel series of 6BIO derivatives carrying substitutions at positions 5' and 6'. We report their *in vitro* evaluation against kinase-related pathological cell lines, we present a profiling study of the most active derivative against a selected panel of disease-relevant kinases and finally, we provide preliminary data supporting its efficacy *in vivo*. The preparation of the novel cell-active analogues that target members of the Src family and GSK3β with high affinity but are inactive toward JAK or Akt provides an original specificity pattern that might prove of great value in a therapeutic perspective.

PS1-C-020

Design and synthesis of new substituted nucleosides as potential anti-HCMV agents

Nicole Pouli, Maria Gerasi, Georgios Papadakis, Nikolaos Lougiakis, Panagiotis Marakos

Division of Pharmaceutical Chemistry, Department of Pharmacy, School of Health Sciences, National and Kapodistrian University of Athens, Panepistimiopolis-Zografou, Athens 15771, Greece

Human cytomegalovirus (HCMV) is a widespread opportunistic pathogen that belongs to the family of DNA herpesviruses (HHV-5). Cytomegalovirus infections are associated with severe morbidity and mortality in high risk patients because of immune system disabilities. There are currently available only three systemic drugs for treatment or prophylaxis of HCMV diseases, all targeted at the viral DNA polymerase, namely Ganciclovir and its prodrug Valganciclovir, Foscarnet and Cidofovir. However, their clinical effectiveness is limited due to severe side effects and the development of resistance. Accordingly, there is a need for the discovery of new anti-HCMV agents that are less toxic, more effective, orally bioavailable and endowed with novel mechanism of action. Within this context, research efforts have led to the development of a number of polyhalogenated benzimidazole nucleosides, exemplified by 2,5,6-trichloro-1-(*β*-d-ribofuranosyl)benzimidazole (TCRB), that strongly inhibit viral replication targeting a DNA maturation and processing step, which does not occur in normal eukaryotic cells. The structurally related Maribavir proved even more potent against HCMV and has entered clinical trials. In an effort to contribute in the structure-activity relationship studies of these series and explore the spatial limitation of the target enzymes we have designed a number of new derivatives which can be considered as Maribavir isosters. Thus, we report in this presentation the preparation of the target nucleosides, which are 2-aminosubstituted analogues of the 1- and 3-position isomers of imidazo[4,5-b]pyridine.

Acknowledgments. This work has been funded by the Special Research Account (ELKE) of the National and Kapodistrian University of Athens.

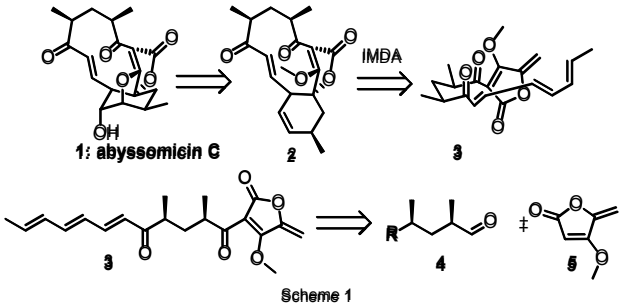
PS1-C-021

An improved synthetic route to Abyssomicin C

Veroniki P. Vidali[1], Aggelos Petroulias[1], Aleksander Canko[1,2], Emmanuel A. Bouzas[1,2] and Elias A. Couladouros[1,2]

[1]Chemistry Laboratories, Agricultural University of Athens, Iera Odos 75, Athens 118 55, Greece, [2]Laboratory of Natural Products Synthesis and Bioorganic Chemistry, Institute of Nanonscience & Nanotechnology, NCSR "Demokritos", Ag. Paraskevi Athens, Greece.

Microbial resistance, one of the greatest threats of human health worldwide, has led to the necessity for the development of new antibiotics. Abyssomicin C, **1** (Scheme 1), isolated from *Verrucosispora* strain, collected from sediment at a depth of 289 m in the Japanese sea [1,2] is a serious candidate for entering the list of new improved antibiotics, as it exhibits strong inhibitory activity against Gram-positive multiresistant bacteria MRSA (Methicillin-resistant *Staphylococcus aureus*) and VRSA (vancomycin-resistant *S. aureus*) (MIC 4 and 13 μg/mL, respectively). Its fascinating structure, characterized by a complicated polycyclic polyketide skeleton, combined with its interesting biological activity, have intrigued many research groups to develop efficient routes towards its total synthesis. [2] However, to date no scalable synthetic routes have been developed for its preparation [1,2] Synthetic strategies developed so far for abyssomicin C, **1**, are classified as biomimetic (based on its biosynthesis) and non-biomimetic. Biomimetic synthetic strategies are based on an intramolecular Diels-Alder reaction (IMDA) of key intermediate **3**, for the construction of the main carbon skeleton of abyssomicin C, **2**. Proper transformations of the latter lead to an efficient completion of the synthesis (Scheme 1). Moreover, the developed methods for the preparation of key intermediate **3**, were based on coupling reactions of an aldehyde of the general form **4** with tetronate **5**. However, all previous syntheses resulted in moderate to low and non repeatable yields of coupling reactions of aldehydes **4** with tetronate **5**, limiting the applicability of the methods for a scalable synthesis. To this end, improvement of synthetic routes is necessary. Herein, the development of a reliable synthetic route to key intermediate **3** is presented. The method is based on the synthesis of a modified aldehyde, properly substituted, which is coupled with tetronate **5**, in high and repeatable yield. Synthetic steps to this aldehyde in racemic or optically pure form and transformations performed to the completion of the synthesis of key interedriate **3**, as well as perspectives of this work are also described.



Scheme 1

References:

[1] Couladouros EA, Bouzas EA, Magos AD. Tetrahedron Lett 2006; 62: 5272.
[2] Sadaka C, Ellsworth E, Hanse PR. Molecules 2018; 23: 1371.

PS1-E-001

LC-PDA-ESI-MS^n metabolite fingerprinting and anticancer screening of an endophyte from marine algae *Dichotomaria marginata*

Teresinha De Jesus Aguiar Dos S. Andrade

Ifma, Timon, Brazil

Metabolites from endophytic fungi of marine origin occupy the center of attention in the discovery of and exploration for medicines because they possess probable therapeutic potentials [1]. Attempting to obtain more potent drugs, many studies with natural products and their analogues have been conducted, showing antitumor properties revealed new possibilities for therapeutic anticancer agents [2,3]. In our study, we used LC-PDA-ESI-MS^n to establish the metabolic fingerprinting of endophyte *Penicillium citrinum* from marine alga *D. marginata* (D. m) and anticancer screening. Algae was collected in Fortaleza beach in Ubatuba city-SP, Brazil. The endophyte was isolated, classified and the extraction of secondary metabolites as described previously [4]. The dried extract was stored in a freezer at −20◦C until LC-PDA-ESI-MS^n analyses. The fingerprinting of fungal extract was achieved on a C_{18} column using a mobile phase methanol/water with gradient elution 5-100 % in 25 min, detection at 254 nm. Detection of compounds was achieved using a Thermo Instruments HPLC system coupled a LTQ Orbitrap XL Hybrid Fourier Transform (Thermo Scientific Instruments). The use of hyphenated techniques such as LC-PDA-ESI-MS^n, combined with search in the database Antimarin, allowed quick access to structural information leading to the identification of sixteen compounds (Figure 1, Table 1). Structures of the compounds are shown in Figure 2. The results presented are relevant and promising and are an important reference for the expansion of knowledge on the chemical composition of the species studied.

Figure 1: Analytical chromatogram in LC-PDA, UV spectra (254 nm) and MS-ESI mass (+) of some compounds of the acetonitrile extract of *P citrinum*

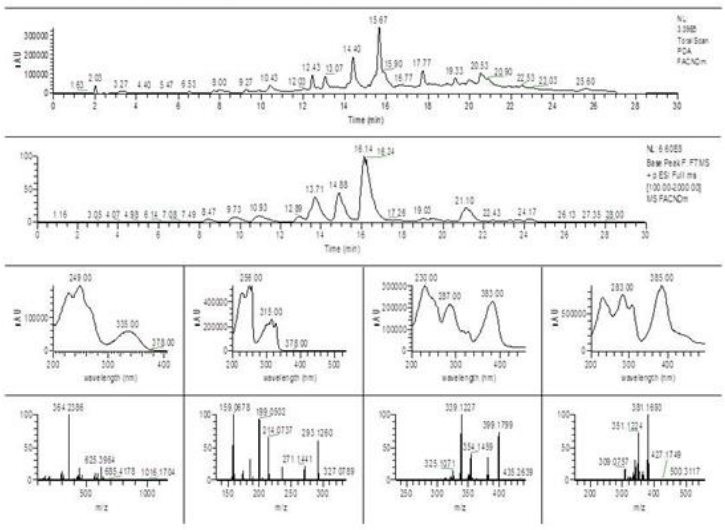


Figure 2: Chemical structures of the compounds identified from extract *ACN P. citrinum*

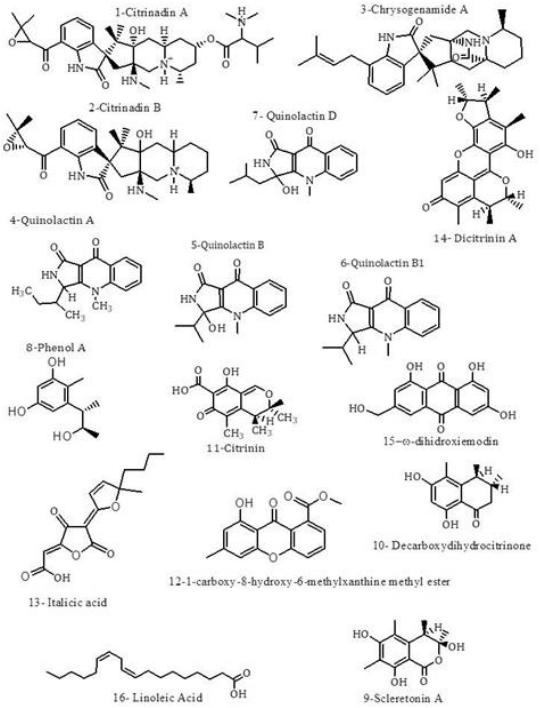


Table 1: Retention times and mass spectral data of the chromatographic peaks

No.	t_R/ (min)	$[M+H]^+$ (m/z)	MS/MS $[M+H]^+$	UV λ_{max} (nm)	DBE	Compound	Ref.
1	8.0	625.3964	594 (96), 576 (60) 449 (100), 431 (66)	229, 249,333	12	Citrinadin A	[14]
2	9.27	482.3047	437 (100), 417 (22), 202 (18)	229, 249,334	11	Citrinadin B	[14]
3	10.43	448.2971	421 (23), 391 (100) 377(34),	229, 249,335	11	Chrysogenamide A	[15]
4	12.03	271.1439	214 (76), 186 (46), 159 (39)	247, 256, 303, 314, 327	9	Quinolactin A	[15]
5	12.43	273.1234	230 (43), 214 (27),186 (100)	247, 256, 303, 314, 327	9	Quinolactin B	[15]
6	12.57	257.1284	214 (83)	247, 256, 303, 314, 327	9	Quinolactin B1	[15]
7	12.70	287.1387	230 (48), 214 (57),186 (100)	247, 256, 303, 314, 327	9	Quinolactin D	[18]
8	13.07	219.0992	135 (69), 107 (87)	228, 281	4	Phenol A	[18], [19]
9	13.68	253.1051	235 (16), 220 (36), 209 (100), 191 (23)	272, 312	6	Scleretonin A	[20], [21]
10	14.20	223.0949	193 (100), 187 (36),177 (40)	272, 312	6	Decarboxydihydrocitrinone	[20], [21]
11	14.40	251.0917	233 (49), 215 (47), 205 (100),191 (57)	318	7	Citrinin	[18]
12	15.30	285.0752	399(96), 348 (96), 318 (96)	230, 283, 304, 385	11	1-carboxy-8-hydroxy-6-methylxanthine methyl ester	[22]
13	15.67	293.1021	293 (96), 275 (96), 247 (96)	370	8	Italicic acid	[28]
14	15.77	381.1701	351 (82), 318 (27), 309 (51)	231, 283, 307, 330	12	Dicitrinin A	[27]
15	17.77	287.0752	nd	247, 266, 288, 438	11	ω-dihidroxiemodin	[23]
16	19.33	281.2476	249 (97), 219 (100), 173 (71)	269, 280	3	Linoleic acid	[26]

Keywords: *Penicillium citrinum*; MS data; identification

References

[1] Deshmukh S. K., *et al*. Front Microbiol, 2018; v. 8, 1-24.
[2] Leite M. L., Cunha N. B., *et al*. Pharmacol and Ther, 2018; v. 183, 160-176.
[3] Kumar M. S., Adki K. Biomed Pharmacother, 2018; v. 105, 233-245.
[4] Gubiani J.R., Zeraik M.L., *et al*. J Nat Prod, 2014; v. 77, 668-672.

PS1-E-002

Natural carotenoids from a sustainable source: a comparative study of five microalgae species

Ginevra Lombardi-Boccia, Gabriella Di Lena, Irene Casini, Massimo Lucarini

Crea-AN, Rome, Italy

Microalgae are among the richest and most varied producers of carotenoids, with also the advantages of a high sustainability and productivity compared to terrestrial sources [1]. With the increasing consumers' interest for natural products and healthy food and with consolidation of functional food and nutraceuticals' market, the demand for natural, health-protecting bioactive molecules has grown in the recent years.
We here present the results of a study on the carotenoid profile of biomass from five microalgae species, *Porphyridium cruentum, Isochrysis galbana, Phaeodactylum tricornutum, Tetraselmis suecica* and *Nannochloropsis gaditana* produced in industrial plant in outdoor photobioreactors. Pigments were solvent-extracted and separated by HPLC-photodiode-array. Results showed a wide differences in carotenoid profiles, among species and a prevalence of xanthophylls over carotenes. Among the carotenoids detected the most relevant ones were zeaxanthin (94.2 mg 100 g^{-1} dw) from the red alga *P. cruentum*, fucoxanthin from the golden-brown flagellate *I. galbana* (1346 mg 100 g^{-1} dw) and the diatom *P. tricornutum* (776.8 mg 100 g-1 dw, respectively), lutein (85.4 mg 100 g^{-1} dw) from the green alga *T. suecica*, violaxanthin from *T. suecica* (81.8 mg 100 g^{-1} dw) and the *Eustigmatophyta N. gaditana* (336.72 mg 100 g^{-1} dw). Beta-carotene was the only carotenoid common to all species, with *N. gaditana* showing the highest levels (100.1 mg 100 g^{-1} algal dw).
The microalgae studied, currently finding their main application in aquaculture, compare favourably to conventional sources of carotenoids; they may therefore be regarded as valuable and sustainable sources of natural carotenoids for the functional food and nutraceutic market.

Acknowledgment: Authors acknowledge Archimede Ricerche srl, part of APG Group, for providing microalgae biomass.

Keywords: carotenoids, xanthophylls, microalgae, natural pigments, food ingredients

References:

[1] Draaisma,R.B, Wijffels R.H, Slegers P.M, Brentner L.B, Roy A, Barbosa M.J. 2013; Curr Opin Biotechnol, 24, 169–177.

PS1-E-003

Biotechnological syntheses of maritime high-value diterpene type natural products

Markus Reinbold, Marion Ringel, Daniel Garbe, Thomas Brück

Werner Siemens-Chair of Synthetic Biotechnology, Technical University of Munich, Lichtenbergstr. 4, 85748 Garching, Germany

The marine diterpene glucoside family of pseudopterosins (PS), conventionally extracted from soft coral *Antillogorgia elisabethae* is commercially used as an anti-irritant additive in high-end cosmetics. Additionally, this compound class is in clinically trials for its wound healing capacity of diabetes' lesions. However, harvesting the necessary soft coral biomass causes severe damages to the coral reefs, whereby the pseudopterosin content of the biomass can vary substantially between nearby locations.
The Canadian/German cooperation project OMCBP set-out to establish a sustainable, biotechnological production route, to protect the coral reefs and to provide a sufficient PS supply. In a first attempt, the recently discovered hydropyrene synthase from *Streptomyces clavuligerus* was used to generate the bicyclic PS scaffold, termed iso-elisabethatriene A [1]. Unfortunately, the desired iso-elisabethatriene A, a structural PS analogue accounted for only 9% (w/w) of the four main terpene products generated by this enzyme. Guided by a homology model derived from another diterpene synthase, selected amino acids were mutated and analysed for their impact on the product distribution of said hydropyrene synthase. Interestingly, one mutant could be identified that showed a significant change in general product distribution, increasing the desired iso-elisabethatriene A yield by a factor of 3. The development of a selective purification strategy provided sufficient purified substances to afford NMR analysis and structural identification. Currently, the purified compounds are undergoing further *in-vitro* modification approaches, like hydroxylation.

PS1-E-004

Structurally diverse secondary metabolites from a marine fungus isolated from the Atlantic Ocean

Mallique Qader[1,2], Larry Mweetwa[3], Manal Eshelli[1,4], Lalith Jayasinghe[2], Marcel Jaspars[3], Mostafa E. Rateb[1]

[1]School of Science and Sport, The University of the West of Scotland, Paisley, United Kingdom, [2]National Institute of Fundamental Studies, Kandy, Sri Lanka, [3]Marine Biodiscovery Centre, University of Aberdeen, Aberdeen, United Kingdom, 4Faculty of Agriculture, University of Tripoli, Tripoli, Libya

Marine environment can harbour millions of macro- and micro-organisms. Recently, deep-sea habitats gained more attention as they produce a plethora of novel secondary metabolites. The bioaccumulation of metabolites in microorganisms favors their survivability in harsh conditions that differ from other habitats. Hence, marine microorganisms proved to be prolific sources of novel bioactive compounds that are of considerable interest for new drug leads. As a continuation of isolation of secondary metabolites from marine fungi, here we report the isolation of new adenosine derivative together with known brefeldin A, 3-indoleacetic acid, and triazine-3,5-dione derivative from a marine fungus. The fungus was isolated from the Atlantic Ocean in 2015 and fermented under static conditions. The crude extract was further fractionated sequentially using different solvents followed by semi-preparative HPLC isolation. The structures of the pure compounds were assigned using extensive analysis of 1D and 2D NMR, HRESIMS, and literature data. Further, one strain many compounds (OSMAC) approach was employed to study the chemical profile of the fungus fermented in shaking conditions. The dereplication of the HRESIMS data of the total crude extract identified cyclic dipeptides, indole derivatives, and few new hits apart from the above microbial metabolites. The biological screening of the isolates and isolation of metabolites from the OSMAC technique are underway.

Acknowledgements: The support provided by the EU Erasmus Mundus- gLINK project (552099-EM-1-2014-1-UK-ERA) is gratefully acknowledged.

Keywords: marine fungus; adenosine, brefeldin A, triazine-3,5-dione

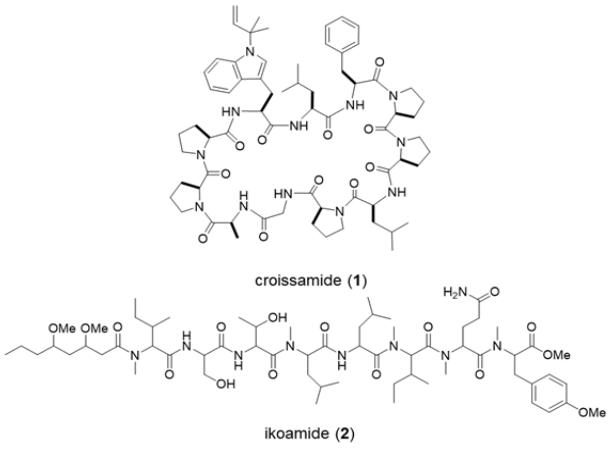
PS1-E-005

Isolation and structure determination of two new peptides from marine cyanobacteria

Keitaro Iwasaki, Arihiro Iwasaki, Simpei Sumimoto, Kiyotake Suenaga

Department of Chemistry, Faculty of Science and Technology, Keio University, Hiyoshi, Kohokuku, Yokohama, Japan

To discover novel natural products possessing remarkable structures and biological activities, we have investigated secondary metabolites of marine cyanobacteria collected in Okinawa. As a result, we isolated two new peptides, croissamide (1) and ikoamide (2) (Figure 1.).
Croissamide (1) is a cyclic peptide that consists of 11 amino acid residues including five prolines and an *N*-prenylated tryptophan. The marine cyanobacterium *Symploca* sp. (1600 g, wet weight) was collected at Minna Island, Okinawa, and extracted with methanol. The extract was filtered, concentrated, and partitioned between EtOAc and H_2O. The EtOAc-soluble material was further partitioned between 90% aqueous MeOH and hexane. The material obtained from the aqueous MeOH portion was subjected to fractionation with reversed-phase column chromatography (ODS silica gel, MeOH-H_2O) and repeated reversed-phase HPLC to give croissamide (1) (10.4 mg). Its gross structure was elucidated by spectroscopic analyses. As for the stereochemistry, all the amino acids were determined to be L-form based on chiral HPLC analysis of acid hydrolysate of 1. This compound did not exhibit cytotoxicity against HeLa cells.
Ikoamide (2) is a linear lipopeptide that consists of a unique fatty acid moiety and 8 amino acids including five *N*-methylated amino acids. The unidentified marine cyanobacterial samples (800 g, wet weight) were collected at Kuro Island, Okinawa, and extracted with methanol. The extract was filtered, concentrated, and partitioned between EtOAc and H_2O. The EtOAc-soluble material was further partitioned between 90% aqueous MeOH and hexane. The material obtained from the aqueous MeOH portion was subjected to fractionation with reversed-phase column chromatography (ODS silica gel, MeOH-H_2O) and repeated reversed-phase HPLC to give ikoamide (1) (7.8 mg). Its gross structure was elucidated by spectroscopic analyses, and the determination of its absolute stereochemistry is ongoing. This compound exhibits cytotoxicity against HeLa cells with an IC_{50} value of 1.7 μM.



PS1-E-006

A marine lipopeptide, minnamide A: Synthetic studies for assignment of the C-9 methyl stereochemistry

Masayuki Kobayashi[1], Shinpei Sumimoto[1], Seiichi Sinomiya[1], Toshiaki Teruya[2], Arihiro Iwasaki[1], Kiyotake Suenaga[1]

[1]Faculty Of Science and Technology, Keio University, Yokohama, Japan, [2]Faculty of Education, University of the Ryukyus, Nishihara-cho, Japan

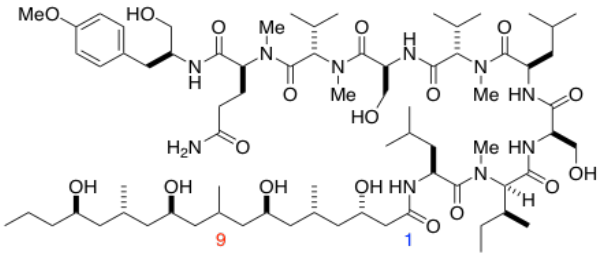
Minnamide A (1) was isolated from the marine cyanobacterium *Okeania hirsuta*, which was collected in Minna-island, Japan [1]. Minnamide A exhibits growth-inhibitory activity against HeLa cells (IC_{50}: 0.17 μM). The absolute stereochemistry of C-9 methyl group of fatty acid moiety of minnamide A had not determined yet. So, to assign the C-9 stereochemistry, we performed synthetic studies of possible two diastereomers of fragment 2, which was prepared by degradation from the natural compound, and compared their ^{1}H NMR spectra.

In synthetic studies, we prepared the bromide 3, which was derived from the known homoallylic alcohol 4 [2] and the Weinreb amides 5a and 5b prepared from the known unsaturated lactone 6 [3]. Coupling reaction between bromide 3 and the Weinreb amide 5a gave the ketone 7a. Conversion of protecting group of the ketone 7a, and then, oxidation and condensation with leucine methyl ester provided one of the target product 2a. Another target product 2b was also achieved by the same way as described above.

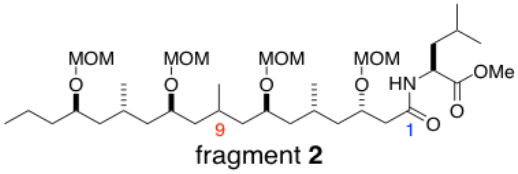
Next, we compared the ^{1}H NMR spectra of synthetic 2a and 2b, with that of fragment 2 from 1, and found that the spectrum of 2b matched that of fragment 2. As a result, we determined the absolute stereochemistry of the C-9 of minnamide A as 9-(*S*).

References:

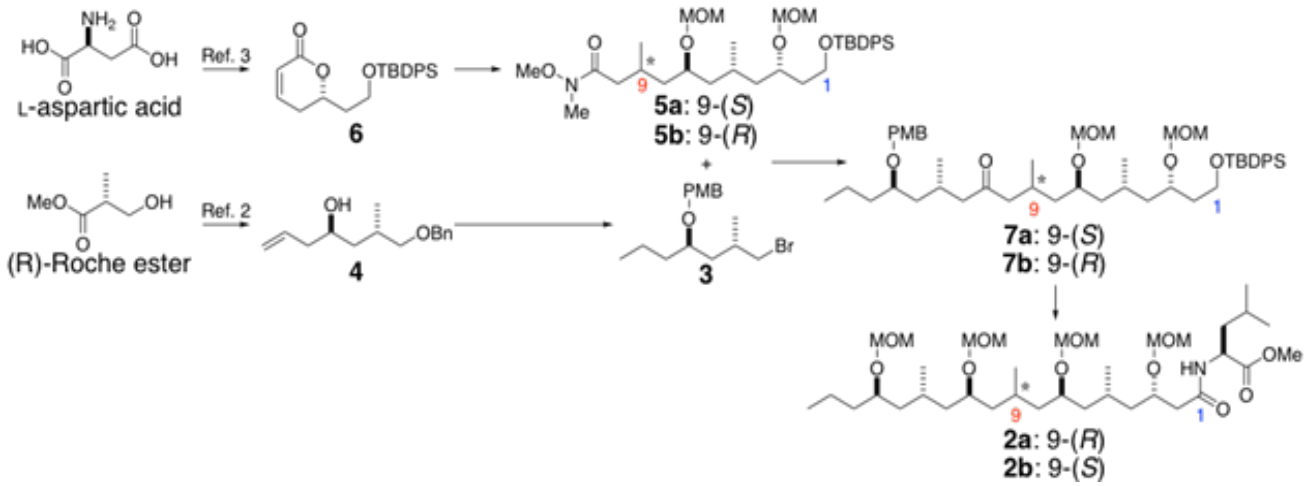
[1] Sumimoto S et al. The 97th CSJ Annual Meeting 2017: 2C7–40.
[2] Grove CI, Fettinger JC, Shaw JT. Synthesis 2012; 44: 362.
[3] Ghosh AK, Wang Y. Tetrahedron Lett 2000; 41: 2319.



Minnamide A (**1**)



fragment **2**


L-aspartic acid
Ref. 3
6
5a: 9-(S)
5b: 9-(R)
(R)-Roche ester
Ref. 2
4
3
7a: 9-(S)
7b: 9-(R)
2a: 9-(R)
2b: 9-(S)

PS1-E-007

Kakeromamide A, a new cyclic pentapeptide from the marine cyanobacterium *Moorea bouillonii*

Fumiaki Nakamura[1], Hiroshi Maejima[1], Midori Kawamura[1], Daisuke Arai[2], Meng Zhao[3], Tao Ye[3], Nobuhiro Fusetani[2,4], Yoichi Nakao[1,2]

[1]Department of Chemistry and Biochemistry, Graduate School of Advanced Science and Engineering, Waseda University, 3-4-1 Okubo, Shinjuku-ku, Tokyo 169-8555, Japan, [2]Research Institute for Science and Engineering, Waseda University, 3-4-1 Okubo, Shinjuku-ku, Tokyo 169-8555, Japan, [3]State Key Laboratory of Chemical Oncogenomics, Peking University Shenzhen Graduate School, Xili, Nanshan District, Shenzhen, 518055, China, [4]Fisheries and Oceans Hakodate, 3-1-1 Minato-cho, Hakodate, 041-8611, Japan

For more than about 40 years, many researchers have tried to discover new compounds from marine source, and as a result, more than 27,000 new metabolites have been reported to date [1]. Marine cyanobacteria are known to be a rich source of peptides. In this study, a new cyclic pentapeptide named kakeromamide A [2] was isolated from the marine cyanobacterium *Moorea bouillonii* collected at Kakeroma Island in Japan (Fig 1). Its structure containing unique amino acid residues of a thiazole ring moiety and a *β*-amino acid was elucidated by the MS and each NMR spectral analysis and the modified Marfey's method. We found a unique biological activity in kakeromamide A to induce differentiation of neural stem cells (NSCs) into astrocytes at 10 μM in the *in vitro* neural differentiation model using mouse ES cells (Fig 2).

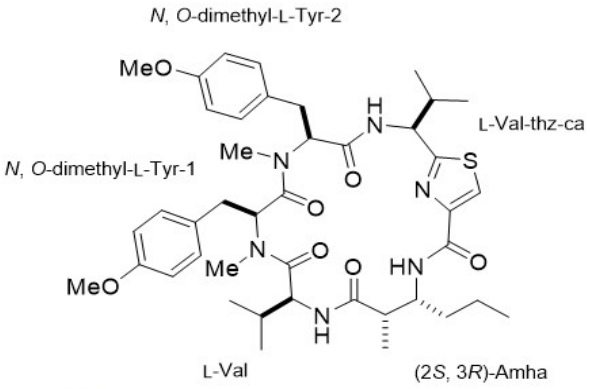


Fig. 1: Structure of kakeromamide A.

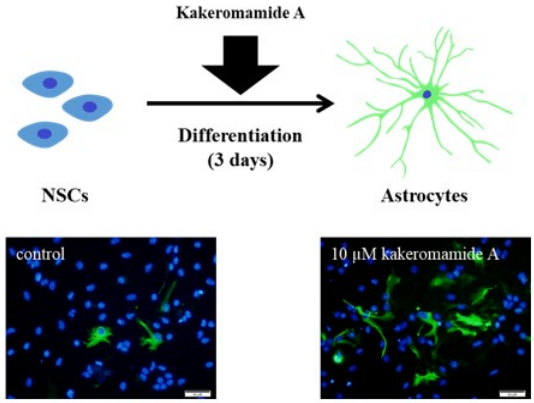


Fig. 2:Astrocyte differentiation of kakeromamide A.

PS1-E-008

Isolation and sstructure determination of four new compounds from marine cyanobacteria

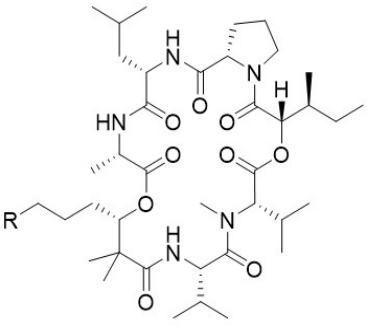
Ikuma Shiota[1], Arihiro Iwasaki[1], Shimpei Sumimoto[1], Hiroshi Tomoda[2], Kiyotake Suenaga[1]

[1]Keio University, Yokohama, Japan, [2]Kitasato University, Minato-Ku, Japan

To discover novel natural compounds bearing remarkable structure and biological activities, we have investigated secondary metabolites of marine cyanobacteria collected in Okinawa prefecture, Japan. Consequently, we isolated four compounds, kohamamides A-C (1-3) and caldorin (4) (Figure 1.).

Kohamamides A-C (1-3), cyclic depsipeptides that belong to the kulolide superfamily, were isolated from an *Okeania* sp. marine cyanobacterium. Their planar structure of 1-3 were elucidated by the NMR analysis and the fragmentation study by ESI-ion trap MS. Sterochemistries of 1-3 were unveiled by degradation reactions and chemical interconversions. Kohamamide B (2) exhibited moderate cytotoxicity against HL60 cells. Compounds 1-3 are the first members of kulolide superfamily found in the East Asian marine environment.

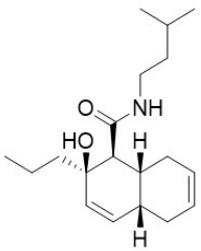
Caldorin (4), a polyletide with a *cis*-fused decalin ring scaffold, was isolated from a *Caldora penicillata* marine cyanobacterium. The gross structure and relative configuration of 4 were determined by spectroscopic techniques. To determine absolute stereochemistry of 4, we tried hydrolysis of amide bond (followed by PGME method) but failed in any reaction conditions. Then, we measured a CD spectrum of 4 but could not obtain any useful data. We also clarified that 4 is a weak sterol *O*-acyltransferase (SOAT) inhibitor and a moderate osteoblast differentiation inhibitor. Sterol *O*-acyltransferase catalyzes the esterification of cholesterol, and the accumulation of excessive cholesteryl ester causes hypercholesterolemia and related diseases. Osteoblasts are bone-forming cells. The overgeneration of osteoblasts cause certain diseases such as fibrodysplasia ossificans progressive. On the other hand, 4 did not exhibit cytotoxicity against either HeLa or HL60 cells at up to 50 μM.



Kohamamide A (**1**) : R = 

Kohamamide B (**2**) : R = 

Kohamamide C (**3**) : R = 



Caldorin (**4**)

PS1-E-009

Deep-sea marine life molecules: assessing bioactivity potential of deep-sea microorganisms

Adriana Rego[1], Eduarda Almeida[1,2], Mariana Girão[1], Inês Ribeiro[1], Tiago Ribeiro[1], Ralph Urbatzka[1], Maria de Fátima Carvalho[1], Pedro Leão[1]

[1]Interdisciplinary Centre of Marine and Environmental Research (CIIMAR/CIMAR), Matosinhos, Portugal, [2]Faculty of Sciences, University of Porto, Portugal

Deep-sea organisms have developed adaptational strategies for their unique environments, whose can be linked not only to unexplored biodiversity but also to the production of novel chemical entities. Also, the deep-sea is perhaps the last frontier in our planet for the exploration of natural products.
With the main goal of assessing the biotechnological potential of deep-sea organisms (Bacteria and Fungi), several strains from deep-sea samples are been isolated and cultivated in small-scale for screening tests.
Until now we have a total of 130 strains already identified, isolated from samples collected in Madeira and Azores, Portugal. Bacterial strains are distributed by 5 different phyla (Alpha, Beta and Gammaproteobacteria, Firmicutes and Actinobacteria) including members of *Streptomyces* genus, known producers of bioactive molecules. Seven fungi strains from Ascomycota phylum were also isolated.
Organic extracts are being tested for their bioactivity potential through cytotoxic and antimicrobial assays. An overview of the whole experimental approach and of the most promising activities will be presented.

PS1-E-010

Structures and cytotoxic activities of new macrolides from the marine dinoflagellates *Amphidinium* species

Masashi Tsuda, Keiko Kumagai, Masayuki Tsuda

Kochi University, Nankoku, Japan

Marine dinoflagellates of the genus *Amphidinium* have been recognized as rich sources of biologically active polyketides possessing unique chemical structures. We have isolated a series of cytotoxic macrolides, iriomoteolides, and polyketides, amphirionins, from the marine dinoflagellates of the genus *Amphidinium*. During our investigation on bioactive substances from the *Amphidinium* dinoflagellates, we have isolated several new cytotoxic macrolides, respectively, from the marine dinoflagellates *Amphidinium* species. In this symposium, we mainly describe the structural elucidation and biological activity of iriomoteolide-8a.

PS1-E-011

Genome mining for novel bioactive compounds from the marine cyanobacterium *Nodosilinea nodulosa* LEGE 06152

Kathleen Abt, Pedro Leão

Interdisciplinary Centre of Marine and Environmental Research (CIIMAR/CIMAR), Matosinhos, Portugal

Cyanobacteria are a rich source of secondary metabolites with biological activity which can be utilized for various biotechnological and pharmacological applications. Advances in next generation sequencing were important to obtain a picture of natural product diversity that is closer to reality. By mining whole genomes for biosynthetic gene clusters (BGCs), researchers have gathered a large pool of genomic information associated with natural products. However, and in particular for cyanobacteria, most of these BGCs are not associated with any known compound (orphan BGCs). While a large number of genome sequences is accessible in public databases, to date, a single genome of the cyanobacterial genus *Nodosilinea* is available. This genus is one of the most prevalent genera maintained in our in-house culture collection LEGE CC. The strain *Nodosilinea nodulosa* LEGE 06152 has shown potential anticancer activity, which motivated us to obtain its genome sequence. We used established genome-mining strategies to screen the genomic data for biosynthetic genes responsible for the production of natural products. Preliminary data obtained from the secondary metabolite analysis platform antiSMASH indicated the presence of terpene, arylpolyene, bacteriocin, NRPS and type I PKS clusters. Additionally, we used PRISM and Geneious for genome annotation and analysis. In this communication, we provide a thorough analysis of the BGCs found in LEGE 06152, also comparing its secondary metabolite profile with the existing data for PCC 7104.

PS1-E-012

Skin protective effects of marine invertebrates and symbionts from the mesophotic zone

Sofia Letsiou, Konstantinos Gardikis, Eleni Spanidi

Laboratory of biochemistry, Scientific Affairs Department, APIVITA S.A., Industrial Park of Markopoulo Mesogaias, 19003, Markopoulos Attikis, Athens, Greece

Nowadays, there is a huge interest on natural products obtained from marine organisms that can promote the state of health and well-being for humans. Mesophotic coral ecosystems (MCEs) appear to support a diverse biological community. The organisms living in this ecosystem need to adapt themselves to a suite of environmental conditions such as high pressure, lower sun radiation and lower temperature compare to the ones prevailing in the shallow water. Through evolution, this environmental stress has led to the development of unique structures, metabolic pathways, reproductive system, sensory and defense mechanism. Extracts from marine invertebrates and symbionts from the mesophotic zone consider as primary source of bioactive compounds that could be used as functional ingredients. The aim of the present study is to evaluate *in vitro*, the effects of these extracts on primary normal human dermal fibroblasts (NHDF), so as to investigate the potential applications in cosmeceuticals focusing on potential anti-ageing activity and angiogenesis regulation activity.
In order to gain an insight into the molecular mechanisms of extracts bioactivity, we studied the transcript accumulation for an array of genes involved in anti-ageing and angiogenesis processes. NHDF cells were purchased from Lonza CloneticsTM. NHDF incubated with extracts for 48 hours at three different concentrations. At first step, cytotoxicity was assessed by using MTT assay. In addition, anti-ageing activity and angiogenesis regulation activity of extracts on NHDF was confirmed by the regulation of several related transcripts (SIRT1, MMP9, IL8 and VEGF) involved in the relative pathways.
Current findings indicate that marine invertebrates and symbionts extracts from mesophotic zone possess strong anti-aging properties and provide new insights into the beneficial role of marine bioactive compounds in cosmetic formulations for skin protection.

Acknowledgements: This project was funded by a HORIZON 2020 project TASCMAR, Grant agreement no.: 634674.

PS1-E-013

Preparation of chemical probes from marine cyclic peptides

Rie Kamihira[1], Jun K. Yamashita[2], Yoichi Nakao[1]

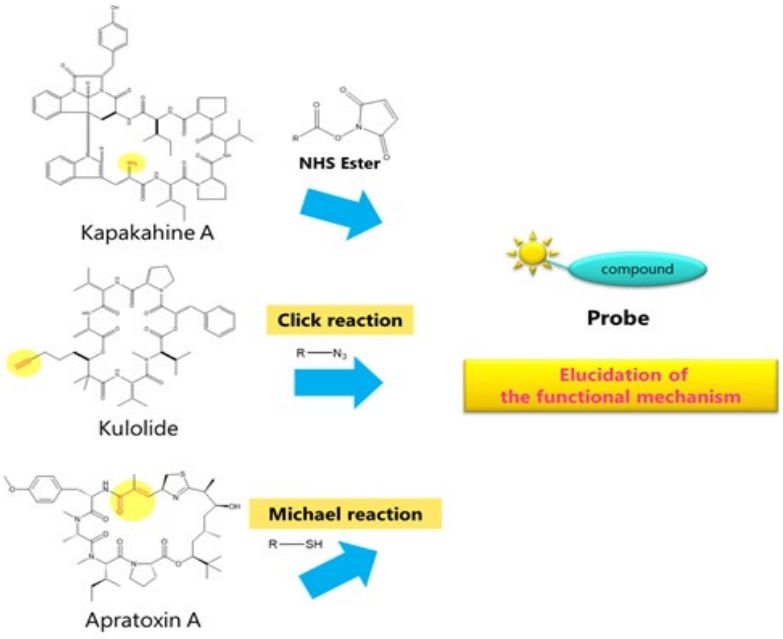
[1]Deprtment of Chemistry and Biochemistry, Graduate School of Advanced Science and Engineering, Waseda University, 3-4-1 Okubo, Shinjuku-ku, 169-8555, Tokyo, Japan, [2]Department of Cell Growth and Differentiation, Center for iPS Cell Research and Application (CiRA), Kyoto University, Kyoto, 606-8507, Japan

A large number of bioactive cyclic peptides have been isolated from marine organisms, but it is still difficult to disclose their modes of actions. To study the modes of actions, preparation of the chemical probes based on the natural products is the important process. Fluorescent probes are powerful tools to identify the localizing sites of the bioactive compounds in the cells. Also, affinity beads linked to the chemical probes are essential for targeting the binding proteins. Aurillide, a cytotoxic cyclic depsipeptide isolated from the sea hare *Dolabella auricularia*, is the successful example for the identification of its target protein and the mode of action.

To add more successful cases identifying the target proteins and the modes of actions, we turned to three cyclic peptides including kapakahine A, kulolide, and apratoxin A that contain the functional sites of an amino group, a terminal acetylene, or an α, β-unsaturated carbonyl group, respectively. In fact, kapakahine A, a cytotoxic cyclic peptide isolated from the sponge *Cribrochalina olemda*, was recently reported to be derivatized at the amino group with the fluorescent coumarin to visualize its localization in the cells.

In this study, we investigated the preparation procedures of chemical probes using two cyclic peptides, kulolide and apratoxin A. Kulolide contains a terminal acetylene can be the counterpart of the azide group in the "click reaction", which is the simple and efficient way to prepare the chemical probes. Apratoxin A isolated from the cyanobacterium *Moorea bouillonii* as the inducer of differentiation to cardiomyocyte, contains the α, β-unsaturated carbonyl group, an acceptor of Michael reaction by thiol group.

In this presentation, preparation of fluorescent probes from marine cyclic peptides utilizing their functional groups as well as their application to the cell imaging will be presented.



PS1-E-014

Isolation, separation and structure elucidation of natural products from marine endophytes with potential activity towards bacterial infections and cancer cell lines

Gaia Burgio[1], Mostafa E. Rateb[1,2]

[1]University of The West of Scotland, Paisley, United Kingdom, [2]University of Aberdeen, Aberdeen, United Kingdom

This project consists in the extraction, isolation and structure determination of compounds obtained from marine sources, with the aim of identifying new drug leads against bacterial infections and cancer cell lines. The choice of the marine environment as focus for the research derives from the myriad of opportunities hidden in the biochemistry of sea organisms belonging to diverse kingdoms and phyla, and the opportunities given to chemists by the modern analytical methods and techniques.
The samples object of the study are endophytes from plants, twigs and algae collected from the shores of the Red Sea and Sicily. The extraction process involved surface sterilisation – in order to remove epiphytes – and isolation on agar plates of fungi and bacteria by means of Nystatin and Nalidixic Acid. Large scale fermentation of the sample of choice takes place on malt and yeast media – with the addition of mannitol for fungi and MOPS, casaminoacids and glycerol for the bacteria. Separation into fractions is obtained through liquid-liquid fractionation and various chromatographic methods, including: thin layer chromatography (TLC), normal phase chromatography (flash column and VLC), high pressure liquid chromatography (with reversed phase C_{18} prep and semi-prep columns). The isolated compounds thus yielded are identified by means of nuclear magnetic resonance (NMR) spectra, both 1-D and 2-D, HRMS, and the aid of databases such as Reaxys and Antimarine 2012.
Crude fractions and pure compounds are to undergo bioassays to test their biological activity toward bacterial strains such as *Pseudomonas aeruginosa*, *Escherichia coli*, *Straphylococcus aureus*, and different tumoral cells, such as HeLa cells and breast cancer cell lines.

PS1-E-015

How the culture conditions affect the production of a potent anticancer metabolite by *Nostoc*

Alexandros Polyzois[1,2], Quoc Dang Thai[2], Elie-Bertrand Duran[2], Sylvie Michel[1]

[1]Laboratoire de Pharmacognosie, UMR CNRS 8638 COMETE, Faculté de Pharmacie de Paris, Université Paris-Descartes, Sorbonne Paris Cité, 4, av. de l'Observatoire, 75006, Paris, France, [2]Phycosource S.A., 13 Boulevard de l'Hautil, 95000, Cergy, France

Cryptophycin-1 (Cry-1) is the first circular depsipeptide which was isolated in 1990 from the species *Nostoc* sp. Since then it is the subject of numerous researches which focus especially on its potential anticancer activity. Responsible for that activity is its ability to inhibit the polymerization of tubulin. Indeed, cryptophycin represents the most powerful tubulin-inhibitor. However, it faces high toxicity issues. Leaded by its potential therapeutic activity cryptophycin-1, and its analogues, have been extensively studied the last 25 years. Though, both natural isolation and total synthesis were not successful in terms of industrial production. On the one hand, the isolation of Cry-1 from *Nostoc* sp. provides a yield of 0.1% [1]. On the other hand, the total chemical synthesis way has multiple steps with a not sufficient final outcome [2].

In that way, an alternative solution could be the modification of environmental conditions of culture in order to obtain an overproduction of the metabolite of interest.

Firstly, our research focused on the effect of light in culture. Light wavelength, intensity and light photoperiod were tested. The media composition was also examined. According to the conditions, a great variation of the production of Cry-1 was observed.

We describe here the first results of the stress on the environmental conditions. Further biotic and abiotic stress would contribute to determine the optimal conditions and finally lead on the overproduction of the metabolite.

Acknowledgments: The author would like to acknowledge funding support from European Union's Horizon 2020 research program #grant 675006

Keywords: Cryptophycin, *Nostoc* sp.

References:

[1] Trimurtulu G et al., J. Am Chem Soc 1994;116: 4729–4737

[2] Lu Liu W et al., Arch. Pharm., Pharm Med Chem, 2009; 342: 577–583

PS1-E-016

Chemical profile of indole alkaloids from the nudibranch *Tambja stegosauriformis*

Douglas Siqueira Almeida Chaves[1], Rosiane Silveira[1], Marcin Ozarowski[2], Piotr Kachlicki[3], Hoffgan Félix[4], Raquel Rennó Braga[4]

[1]Department of Pharmaceutical Sciences, Universidade Federal Rural Do Rio De Janeiro, Rio De Janeiro, Brazil, [2]Department of Pharmacology and Phytochemistry, Institute of Natural Fibers and Medicinal Plants, Poznan, Poland, [3]Department of Pathogen Genetics and Plant Resistance, Metabolomics Team, Institute of Plant Genetics of the Polish Academy of Science, Poznan, Poland, [4]Pharmacy School, Federal Institute of Education, Science and Technology of Rio de Janeiro, Rio de Janeiro, Brazil

Several compounds with biological activity may be found in nudibranchs, which with the indole alkaloids as a particularly group. The aim of this study was to identify indole alkaloids from ethanolic and methanolic extracts of *T. stegosauriformis* (TS) from the Brazilian coast. Ten individuals were collected on the coast of Cabo Frio/Rio de Janeiro/Brazil, preserved in ethanol followed by methanol extraction, and rotary evaporator drying. The extracts were solubilized in methanol (1mg mL-1) and analyses were performed using HPLC-DAD-MS^n with a PDA detector and an ion trap mass spectrometer (operating in the positive ion mode) using the X-Bridge C_{18} column (2.1 × 150 mm). The injection volume was 10 µL and elution was carried out using water (0.1% formic acid) and acetonitrile gradient. The most important MS parameters were as follows: the ion source ESI voltage 4 kV; nebulization with nitrogen at a pressure of 30 psi at a gas flow rate 9L/min. Ion source temperature at 310 ° C, skimmer 1: -10 V. The spectra were scanned in the range of 50-3000*m/z*. Ten TS indole alkaloids were identified between 13-22 min retention time. All alkaloids are derived from the base structure with two interconnected pyrrolic rings. Tambjamycin A (*m/z* 189), B (*m/z* 267 and 269), C (*m/z* 246), D (*m/z* 324 and 326), G (*m/z* 297), J (*m/z* 260), and 4-methoxy-2,2'-bipyrrole-5-carboxaldehyde (*m/z* 191) were previously identified and reported in the literature. A new alkaloid, derived from tambjamycin G, has been identified (*m/z* 376 and 378) for the first time during this study. This compound has two bromine atoms in its structure, which could be confirmed by their loss (*m/z* 216 [M-2Br]). A tambanmycin G dimer was also identified (*m/z* 595). This is the first report of two new indole alkaloids isolated from the *Tambja* genus.

PS1-E-017

Bioactive 3,7–cyclized cembranes and cembranes from the soft coral *Klyxum flaccidum*

Chiung–Yao Huang Huang[1], Atallah F. Ahmed[2], s. Wan–Ru Tseng[1], Yi–Ying Tsai[1], Tsong–Long Hwang[3], Jyh–Horng Sheu[1,4]

[1]Department of Marine Biotechnology and Resources, National Sun Yat–sen University, Kaohsiung 80424, Taiwan, [2]Department of Pharmacognosy, College of Pharmacy, King Saud University, Riyadh 11451, Saudi Arabia, [3]Graduate Institute of Natural Products, College of Medicine, Chang Gung University, Taoyuan 333, Taiwan, [4]Frontier Center for Ocean Science and Technology, National Sun Yat–sen University, Kaohsiung 80424, Taiwan

In order to discover bioactive secondary metabolites, we have investigated the chemical constituents from the organic extracts of the soft coral *Klyxum flaccidum*, which was collected from the coast of the island of Pratas, Taiwan. This study led to the isolation of six natural compounds, including four new cembrane–based diterpenoids, flaccidenes A and B (1 and 2), flaccidodioxide (3), and flaccidodiol (4), and three known compounds 5–7 [1–3] (Figure 1). The structures and relative configurations of all metabolites have been established by detailed spectroscopic analyses, including 1D and 2D NMR (COSY, HSQC, HMBC and NOESY) data in association with experiments. The anti–inflammatory activities of compounds 1–7 on neutrophil pro–inflammatory responses were evaluated by measuring their ability in suppressing fMLF/CB–induced superoxide anion (O_2^- •) generation and elastase release in human neutrophils. From the results, 14–*O*–acetylsarcophytol B (7) showed significant anti–inflammatory activity through inhibiting elastase release (59.7 ± 0.8 %) at 10 μM.

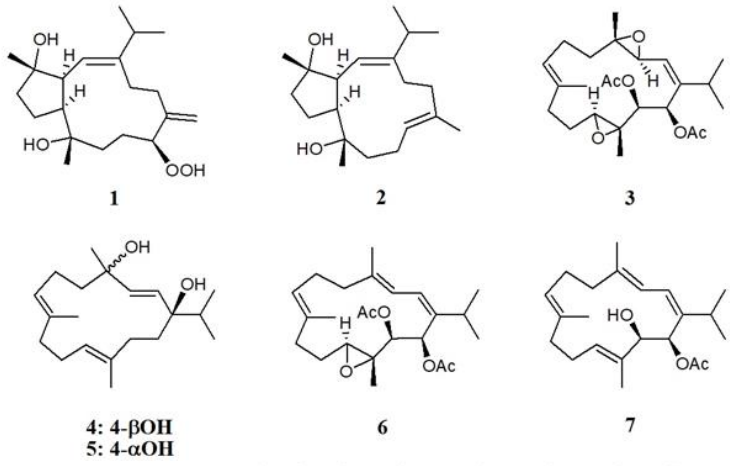


Figure 1. Structures of isolated cembranes from *Klyxum flaccidum*

Acknowledgements: Ministry of Science and Technology of Taiwan (MOST104–2113–M–110–006 and 105–2113–M–110–002) is acknowledged. The authors extend their appreciation to the International Scientific Partnership Program ISPP at King Saud University, Saudi Arabia, for funding this work through ISPP–116.

Keywords: soft coral, *Klyxum flaccidum*, cembrane

References:

[1] König G M, Wright AD. J Nat prod 1998; 61: 494–496.
[2] Gray CA, Davies–Coleman MT, Schleyer MH. J Nat prod 2000; 63: 1551–1553.
[3] Iguchi K, Shimura H, Yamada Y. J Nat prod 1992; 55: 1779–1782.

PS1-E-018

Metabolites from a bioactive organic fraction of an unidentified Red Sea sponge

Atallah F. Ahmed[1,2], Chi–Jen Tai[2], Raha S. Orfali[1], Walied M. Alarif[3], Jyh–Horng Sheu[1,4]

[1]Department of Pharmacognosy, College of Pharmacy, King Saud, University, Riyadh 11451, Saudi Arabia, [2]Department of Marine Resources and Biotechnology, National Sun Yat–sen University, Kaohsiung 80424, Taiwan, [3]Department of Marine Chemistry, College of Marine Sciences, King Abdul Aziz University, Jeddah 21589, Saudi Arabia, [4]Frontier Center for Ocean Science and Technology, National Sun Yat–sen University, Kaohsiung 80424, Taiwan

The dichloromethane–soluble fraction of the organic extract of an unidentified Red Sea sponge has been shown to exhibit *in vitro* cytotoxic and anti–inflammatory activities and thus selected for further chemical Investigation to discover the active compounds. Chromatographic fractionation of this fraction afforded a novel seco–spongian diterpenoid (1), an isoprenoid–derived amide (2) and an isoprenoid–derived carboxylic acid (3), along with known metabolites: cholest–7–ene–3β,5α–diol–6–one (4) [1], 18–nor–3,17–dihyoxyspongia–3,13(16),14–trien–2–one (5) [2], irciformonin G (6) [3] (figure 1). The structures of new compounds were elucidated on the basis of spectroscopic analyses including 2D NMR (COSY, HSQC, HMBC, and NOSEY) correlations. The absolute configuration of 1 and 6 were determined by Mosher's method. The sponge is currently under taxonomical study.

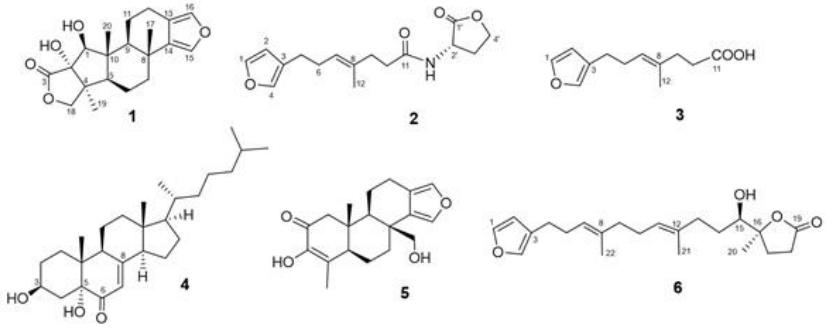


Figure 1. Structures of compounds from a Red Sea sponge

Keywords: Red Sea sponge, *Hyrtios* sp., seco–spongian diterpenoid, isoprenoid derivatives.

References:

[1] Aiello A, Fattorusso E, Magno S, Menna M. Steroids 1991; 56: 337–340.
[2] Parrish SM, Yoshida WY, Konatyuk TP, Park EJ, Pezzuto JM, Kelly M, Willia PG. J Nat prod 2014; 77: 1644–1649.
[3] Shen YC, Shih PS, Lin YS, Lin YC, Kuo YH, Kuo YC. Helv Chim Acta 2009; 92: 2101–2110.

PS1-E-019

Extraction of chlorophylls and phycobiliproteins from Indonesian red seaweed

Hakiki Melanie[1,2], Ambo Tuwo[3], Marcos A. Neves[1,4,5], Mitsutoshi Nakajima[1,4,5]

[1]Tsukuba Life Science Innovation (T–LSI), University of Tsukuba, Tsukuba, Japan, [2]Research Center for Chemistry, Indonesian Institute of Sciences (LIPI), South Tangerang, Indonesia, [3]Center of Excellence for Development and Utilization of Seaweed, Univ. Hasanuddin, Makassar, Indonesia, [4]Life and Environmental Science, University of Tsukuba, Tsukuba, Japan, [5]Food Research Institute, NARO, Tsukuba, Japan

In recent years, seaweeds have increasingly attracted interest in the search of bioactive natural compounds and biomaterials utilized in food, pharmaceuticals and industrial development. The market trend and consumers growing interest in natural and healthy products have forced researches and industry to develop novel products with positive health effects. Seaweed pigments such as chlorophylls and phycobiliproteins have great potential in food industry as functional ingredients. This study was aimed to investigate the potential of seaweed pigments from Indonesian red seaweed. Chlorophylls and phycobiliproteins were extracted from *Euchema cottonii* and *Gracillaria* sp. using combined solvent of ethanol and hexane at ratio 1:10 for chlorophylls, and ethanol and water at ratio 1:20 for phycobiliproteins. The extraction was conducted at room temperature and 50 °C and then the filtrates were collected by centrifugation at 10,000 rpm for 15 mins. Quantitative analysis of chlorophylls and phycobiliproteins were analyzed by UV–Visible spectrophotometry at several numbers of wavelengths. The chemical characteristics of seaweed pigments were structurally verified by Fourier Transform Infrared Spectroscopy (FTIR). *Gracillaria* extract has higher content of chlorophylls and phycobiliproteins than *E. cottonii*. Chlorophylls in both *E. cottonii* and *Gracillaria* extracts were slightly increased at 50 °C, which range from 14.12–26.39 mg/100 g and 44.58–52.49 mg/100 g, respectively. This may be due to the increasing of solubility of the solute in liquid, which is enhanced the molecular diffusion as temperature increasing. However, the phycobiliproteins were comparatively more stable at room temperature and their concentrations slightly decreased at 50°C. The FTIR spectra showed respective bands assigned to the structure of chlorophylls and phycobiliproteins.

PS1-E-020

Antithrombotic and anticoagulant activity of Brazilian and Antarctic brown algae

Ana Cláudia Philippus[1], Gabriele Andressa Zatelli[1], Stephanie Milis Syracuse[1], Nathalia da Silva Moura[1], Bianca Regina Alberton[2], Rubia Karine de Souza[2], Aline Paternostro Martins[3], Paulo Antunes Horta[4], Pio Colepicolo Neto[3], Ana Carolina Rabello de Moraes[1], Miriam de Barcellos Falkenberg[1]

[1]Postgraduation Program in Pharmacy, UFSC, Florianópolis, Brazil, [2]Graduation in Pharmacy, UFSC, Florianópolis, Brasil, [3]Department of Chemistry, USP, São Paulo, Brasil, [4]Department of Botany, UFSC, Florianópolis, Brasil

Cardiovascular diseases are highly prevalent all over the world. Anticoagulant and antiplatelet drugs are frequently used to reduce the risk of thromboembolic events, but they are known to cause adverse effects and drug interactions. Many natural products were reported to present antithrombotic activity, including algal sulphated polysaccharides and terpenes, so on our ongoing search for new drugs modulating hemostasis, we tested 49 extracts of brown algae collected off the Brazilian coast (mainly oceanic islands) and in the Antarctic. The dried algae were exhaustively extracted with CH_2Cl_2/MeOH 2:1. Selected extracts were fractionated by subsequent chromatographic columns (VLC, gravity column and HPLC with *n*–hexane and increasing EtOAc). Pure compounds were identified by NMR (^1H, ^{13}C, HSQC, COSY, HMBC), HRMS and comparison to literature. Extracts and isolated compounds were tested for anticoagulant and antiplatelet activities in concentrations ≤600 µg/mL and 100 µM, respectively. The anticoagulant activity was evaluated by activated partial thromboplastin time (aPTT) and protrombin time (PT) assays1 (kit HemosIL®, using platelet–poor plasma pool). The platelet aggregation assay2 was performed using platelet–rich plasma pool. From 32 extracts of Brazilian algae, platelet aggregation induced by ADP (PAA) or epinephrine (PAE) were inhibited respectively by 18 and 13 extracts; 3 extracts also increased aPTT values over 50%. From 12 extracts of algae from Antarctic, 8 inhibited PAA and 1 inhibited PAE, without significant changes in aPTT or PT. Fractionation of selected extracts led to the isolation of active metabolites: 2 disulfides (1 and 2) from Brazilian *Dictyopteris jolyana* and hydroxysargaquinone (3) from Antarctic *Desmarestia menziesii*. Metabolites 1 and 2 were moderate inhibitors of PAA, while 3 was a potent inhibitor of PAA and PAE. Compound 3 represents a new antithrombotic candidate for further study regarding to its mechanism of action and in vivo studies.

PS1-E-021

Agaclathratosides A and B, two new monogalactosylacyl glycerols from the brown alga *Agarum clathratum* subsp. *yakishiriense*

Yukyoung Jeon[1], Hwan Su Yoon[2], Jong Hwan Kwak[1]

[1]School Of Pharmacy, Sungkyunkwan University, Suwon, South Korea, [2]Department of Biological Sciences, Sungkyunkwan University, Suwon, South Korea

Agarum clathratum subsp*. yakishiriense* Yamada ex G.H. Boo & P. C. Silva is a perennial brown alga in the family Costariaceae and is widely distributed in the coastal area of the East Sea in Korea [1]. Previous studies on *A. clathratum* subsp. *yakishiriense* have reported neuroprotective effect [2]. The EtOH extract of *A. clathratum* subsp. *yakishiriense* was suspended in water and then partitioned consecutively with CH_2Cl_2, EtOAc, and *n*-BuOH. Among these fractions, the EtOAc fraction was subjected to column chromatographic separation. Three monogalactosylacyl glycerols(1-3) and a degraded terpene were isolated from the EtOAc fraction. The structures of two new monogalactosylacyl glycerols, named agaclathratosides A and B, were determined as (2*S*)-1-*O*-(6*Z*,9*Z*,12*Z*,15*Z*-hexadecatetraenoyl)-3-*O*-*β*-D-galactopyranosyl glycerol and (2*S*)-1-*O*-(6*Z*,9*Z*,12*Z*-hexadecatrienoyl)-3-*O*-*β*-D-galactopyranosyl glycerol from spectral data and chemical evidence. The degraded terpene, 6-hydroxy-isololiolide was identified by comparing its spectral data with literature values. The isolated compounds were investigated anti-diabetic activity in zebrafish models for type 1 and 2 diabetes.

Acknowledgements: This work was supported by a grant from Marine Biotechnology Program (Genome Analysis of Marine Organisms and Development of Functional Applications) funded by Ministry of Oceans and Fisheries.

Keywords: *Agarum clathratum* subsp. *Yakishiriense*, monogalactosylacyl glycerol, agaclathratosides A and B, 6-hydroxy-isololiolide, anti-diabetic activity

References:

[1] Boo SM, Ko YD. Marine Plants from Korea. Seoul: Junghaeng-Sa; 2012; 114.
[2] Kim IH *et al*. Pharm Biol (Abingdon, U. K.) 2014; 52: 335–343

PS1-E-022

A new and seven known phlorotannins from *Eisenia bicyclis* and their anti-diabetic activity in Zebrafish model

Eunbin Kim[1], Yukyoung Jeon[1], Youn Hee Nam[2], Tong Ho Kang[2], Jong Hwan Kwak[1]

[1]School of Pharmacy, Sungkyunkwan University, Suwon, South Korea, [2]Department of Oriental Medicinal Biotechnology, College of Life Sciences, Kyung Hee University, Yongin, South Korea

Eisenia bicyclis (Kjellman) Setchell is an edible perennial brown alga in the family Lessoniaceae and is widely distributed in the coastal area of Ulleung and Jeju islands, Korea, and in Japan [1,2]. Previous studies on *E. bicyclis*, have reported several biological activities including anti-inflammation [3], and antioxidative effect [2]. Type 1 diabetes zebrafish model was induced by alloxan, which cause pancreatic *β*-cell necrosis. In addition, type 2 diabetes zebrafish model was induced by exposure of excess insulin. The EtOH extract of E.bicyclis was consecutively partitioned with CH_2Cl_2, EtOAc and *n*-BuOH to give four fractions. Among these fractions, the EtOAc fraction which showed antihyperglycemic activity was subjected to activity-guided fractionation and isolation. A new and seven known phlorotannins were isolated from the EtOAc fraction. The structure of a new compound (eiseniacyclol) was determined as 4,9-bis(3,5-dihydroxyphenoxy) dibenzo[b,e][1,4]dioxine-1,3,6,8-tetraol from spectral data and chemical evidence. The seven known phlorotannins, phloroglucinol, eckol, dieckol, 8,8'-bieckol, eckstolonol, phlorofucofuroeckol-A and -B, were identified by comparing their spectral data with literature values. The isolated compounds revealed anti-diabetic activity for type1 and 2 in the zebrafish model. The isolated compounds were investigated inhibitory activities on *α*-glucosidase.

Acknowledgements: This work was supported by a grant from Marine Biotechnology Program (Genome Analysis of Marine Organisms and Development of Functional Applications) funded by Ministry of Oceans and Fisheries.

Keywords: *Eisenia bicyclis*, phlorotannin, eiseniacyclol, anti-diabetic activity, zebrafish

References:

[1] Boo SM, Ko YD. Marine Plants from Korea. Seoul: Junghaeng-Sa 2012; 119.
[2] Kwon TH *et al*. J Food Sci 2013; 78: 679–684.
[3] Jung HA *et al*. Food Chem Toxicol 2013; 59: 199–206

PS1-E-023

6-Bromoindole derivatives from the marine sponge *Geodia barretti*: isolation and anti-inflammatory activity

Xiaxia Di[1,2], Caroline Rouger[3], Ingibjorg Hardardottir[2,4], Jona Freysdottir[2,4], Tadeusz F. Molinski[5], Deniz Tasdemir[3], Sesselja Omarsdottir[1]

[1]Faculty of Pharmaceutical Sciences, University Of Iceland, Reykjavik, Iceland, [2]Department of Immunology and Centre for Rheumatology Research, Landspitali-The National University Hospital of Iceland, Reykjavik, Iceland, [3]GEOMAR Helmholtz Centre for Ocean Research Kiel, Kiel, Germany, [4] Faculty of Medicine, Biomedical Center, University of Iceland, Reykjavik, Iceland, [5]Department of Chemistry and Biochemistry and Skaggs School of Pharmacy and Pharmaceutical Sciences, University of California, San Diego, USA

The development of new natural compounds with anti-inflammatory effects has become one focus in research aiming for treatment of chronic inflammatory diseases. This study was undertaken to isolate new compounds from the marine sponge *Geodia barretti* and to determine their potential anti-inflammatory activity.

Crude extract of *G. barretti* was chemically profiled by UPLC-qTOF-MS to obtain molecular formulas of detected peaks and to track the compounds following isolation. The results indicated three new 6-bromoindole derivatives, geobarettin A-C (1-3), and four known compounds, barettin (4) [1], 8,9-dihydrobarettin (5) [2], 6-bromoconicamin (6) [3], and *L*-6-bomohypaphorine (7) [4]. The structures of the isolated compounds were elucidated by combination of extensive NMR spectroscopy and HRESIMS data. The stereochemistry was assigned by spectroscopic data, ECD analysis and Marfey's method, which derivatized with a new chiral reagent, 5-fluoro-2,4-dinitrophenyl-Na-L-tryphanamide [5], after total hydrolysis and then followed by HPLC analysis.

The isolated compounds were screened for anti-inflammatory activity using human dendritic cells (DCs). Of the new compounds, compounds 2 and 3 decreased DC secretion of the pro-inflammatory cytokine IL-12p40 and compound 3 increased DC secretion of the anti-inflammatory cytokine IL-10. DCs treated with compounds 2 or 3 and then co-cultured with allogeneic CD4+ T cells led to a decrease in T cell secretion of IFNγ (Th1). Barettin (4) decreased DC secretion of both IL-12p40 (IC50 = 21.04 μM) and IL-10 (IC50 = 11.80 μM). However, DCs treated with barretin (4) did not affect their ability to induce T cell secretion of either IFNγ (Th1) or IL-17 (Th17).

The results indicate that among the three new 6-bromoindole derivatives may be anti-inflammatory drug leads with potential therapeutic effects for the treatment of inflammation, mainly of the Th1 type, which is linked to many chronic inflammatory diseases [6].

Acknowledgements: We are grateful for the financial support received from the University of Iceland Research Fund (Doctoral Grant and Project Grant), AVS R&D Fund of Ministry of Fisheries and Agriculture in Iceland, the Landspitali University Hospital Research Fund and the Memory Fund of Helga Jonsdottir and Sigurlidi Kristjansson.

Keywords: *Geodia barretti*, 6-bromoindole derivatives, anti-inflammatory, barettin

References:

[1] Solter S, Dieckmann R, Blumenberg M, Francke W, Barettin. Tetrahedron Lett 2002; 43(18): 3385–3386.

[2] Sjogren M, Goransson U Johnson AL, Dahlstrom M, Andersson R, Bergman J, Jonsson PR, Bohlin L. J Nat Prod 2004; 67(3): 368–372.

[3] Takahashi Y, Tanaka N, Kubota T, Ishiyama H, Shibazaki A, Gonoi T, Fromont J, Kobayashi J. Tetrahedron 2012; 68(41): 8545–8550.
[4] Kasheverov IE, Shelukhina IV, Kudryavtsev DS, Makarieva TN, Spirova EN, Guzii AG, Stonik VA, Tsetlin VI. Mar Drugs 2015; 13(3): 1255–1266.
[5] Salib MN, Molinski TF. J Org Chem 2017; 82(19): 10181–10187.
[6] Romagnani S. Clin Immunol Immunopathol 1996; 80(3 Pt 1):225–235.

PS1-E-024

AlgaeCeuticals: Development of microalgae-based natural carotenoids as cosmeceuticals and nutraceuticals

Stefan Martens[1], Panagiotis Madesis[2], Nikolaos Labrou[3], Leonardo Ceracino[1], Nico Salmaso[1], Luisa Palmieri[1], Silvia Fluch[4], Pablo F. Ruiz[5], Ángel M. San Martin[5], Jose L. Mullor[6], Natalie Vagioni[7], Marina Chatzikonstantinou[7]

[1]Edmund Mach Foundation, San Michele All'Adige (tn), Italy, [2]Centre for Research and Technology Hellas – CERTH, Thessaloniki, Greece, [3]Agricultural University of Athens, Athens, Greece, [4]Ecoduna AG, Bruck an der Leitha, Austria, [5]Centro Tecnológico Nacional de la Conserva y Alimentación CTNC, Spain, [6]Bionos Biotech S.L., Valencia, Spain, [7]Fresh Line Company, Koropiou, Greece

Microalgae biomass represents a rich source for discovery. The potential for algae-based ingredients in the industry relies on the manipulation and targeting of ingredients to fit increasingly niche product specifications. Algae are exposed to extreme environment and thus have developed unique mechanism for protection. Furthermore, algae produce for the same reason different metabolites, which we need to identify and exploit in a sustainable way for the production of food, drugs and cosmetics.

The AlgaeCeuticals project will take advantage of the native algae strains producing high added value products such as carotenoids and through the application of novel -omics technologies (genomics, transcriptomics, proteomics, enzymomics and metabolomics) as well as algae culture technologies and production of novel products. AlgaeCeuticals will screen and characterize algae carotenoid biodiversity, develop and optimize algae culture systems, develop -omics resources for algae and downstream processing strategies and also novel products based on these natural compounds. For this reason and in order to achieve its object three academic and research centers from Greece (CERTH/INAB; AUA) and Italy (FEM) will collaborate for four years with four industrial R&D partners from Greece (Fresh Formula), Spain (Bionos Biotech ND and Centro Tecnológico Nacional de la Conserva y Alimentación) and Austria (Ecoduna AG). Through this collaboration the academic partners will work closely with the industrial R&D and form a complementary and highly competitive team that will promote transfer of knowledge and excellence to industrial partners. This will strengthen the industrial competitiveness in the field of food and cosmetics in the process of the design, development, testing of the products proposed by the project.

PS1-E-025

Chemical profiles of *Hypoxylon monticulosum* fungal strains isolated from marine red algae *Acanthophora spicifera* and *Dichotomaria marginata*

Dulce Silva[1], Alana Honorio[1], Beatriz Barcellos[1], Rebeca Medina[1], Iata Mendonca[1], Victor Rufino[1], Marcos Soares[2]

[1]Sao Paulo State University - UNESP, Araraquara, Brazil, [2]Federal University of Mato Grosso, Cuiaba, Brazil

Endophytic fungi isolated from marine sources have been increasingly investigated due to their promising chemical and biological profiles for bioprospection purposes [1]. A strain of *Aspergillus* isolated from marine alga *Halimeda copiosa*, for instance, has afforded cytotoxic diketopiperazines halimide and plinabuline, with the latter currently in phase III clinical studies [2]. This work explored two strains of *Hypoxylon monticulosum*, isolated from marine red algae *Acanthophora spicifera* (AS) and *Dichotomaria marginata* (DM), which were grown in Malt liquid media using sea water or ultrapure (MilliQ) water. After micelia separation, each solution was partitioned with EtOAc to yield the crude extracts. Comparison of their chemical profiles by ^{1}H NMR and HPLC-UV evidenced higher yield and metabolic variability for the extracts from cultures using ultrapure water: AS09MQ and DM02MQ. Moreover, the strain isolated from *A. spicifera* disclosed twice the yield of *H. monticulosum* isolated from *D. marginata*, evidencing a putative host-dependent metabolism. Both extracts were further fractionated and yielded two major compounds from each extract, with dihydrosporotriolide (S2) as a common constituent in both extracts. This class of compounds belongs to the furofurandione family, widely reported in genus *Hypoxylon*. From DM02MQ extract, a novel cyclohexadienone derivative (S3) was isolated. The extracts were also tested against multi-resistant bacterial strains and biofilm-forming bacteria, as well as for their anti-cholinesterase potential. Both extracts exhibited strong anticholinesterase activity, with higher AChE inhibition (98% for AS09MQ and 96% for DM02MQ) than tacrine (84%), used as positive control. In addition, AS09MQ inhibited biofilm formation (52%) when tested against the Methicillin-resistant *Staphylococcus epidermidis* strain ATCC 35984. Such results highlight the chemodiversity and biological potential of marine-derived fungal strains in bioprospection studies.

Acknowledgements: CNPq CAPES FAPESP

Keywords: marine endophytic fungi, algae, *Hypoxylon monticulosum*

References:

[1] Blunt JW, Copp BR et al., Nat Prod Rep 2016; 33(3): 382–431.
[2] https://www.cancer.gov/about-cancer/treatment/clinical-trials/intervention/plinabulin, accessed Sep.2018.

PS1-E-026

2,5-Diketopiperazines from marine-derived bacteria isolated from marine sediments collected in the Eastern Mediterranean Sea

Maria Harizani, Panagiota Georgantea, Eleni Katsini, Vassilios Roussis, Efstathia Ioannou

Section of Pharmacognosy and Chemistry of Natural Products, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, Greece

2,5-Diketopiperazines (DKPs) are the smallest possible cyclic peptides and, therefore, are among the most common peptide derivatives found in nature. They derive from the condensation of two α-amino acid forming a bis-lactam. Although they are relatively simple and low molecular weight compounds, they can be highly substituted resulting in very complex structures. DKPs have been reported so far from a variety of sources, from microorganisms (bacteria, fungi, lichens) to higher organisms (algae, plants, marine sponges, gorgonians, tunicates, mammals).

DKPs have recently attracted attention due to their diversity and remarkable bioactivity. They are associated with activities such as cytotoxic, antibacterial, antifungal, antiparasitic, insecticidal, antiviral, antiprion, antifouling, antioxidant, anti-inflammatory, antihyperglycemic, neuroprotective etc, thus making them potential drug candidates. Moreover, they are involved in quorum sensing and ion-transport, and exhibit high-affinity binding to a large variety of receptors.

In the framework of our investigations on the chemistry of marine microorganisms, several bacterial strains isolated from marine sediments collected in the Eastern Mediterranean Sea were cultivated in large scale and the obtained organic extracts were submitted to a series of chromatographic separations, leading to the isolation of a number of DKPs. The structures of the isolated compounds, among which two are new natural products, were determined on the basis of extensive analysis of 1D and 2D NMR and MS data.

Acknowledgements: The authors thank Special Account for Research Grants and National and Kapodistrian University of Athens for funding to attend the meeting.

Keywords: marine-derived bacteria, 2,5-diketopiperazines, isolation, structure elucidation

PS1-F-001

Comparative analysis of antidiabetic, anti-obesity and antioxidant activities of leaves and flowers of *Ocimum basilicum* and identification of chemical constituents in their essential oils and extracts

Dildar Ahmed, Zoy I Noor, Muhammad Tariq Qamar

Forman Christian College Lahore, Lahore, Pakistan

Diabetes, obesity, and oxidative stress are associated metabolic disorders having long-term health implications, and demand safe and affordable remedies. The objective of the present study was to explore porcine pancreatic *α*-amylase (PPA) and lipase (PPL) inhibitory activities and antioxidant potential of polar and nonpolar extracts of the leaves and flowers of *Ocimum basilicum* and phytochemicals of their essential oils and extracts. Fresh leaves and flowers were used to extract their essential oils. The dried parts were used to obtain hexane extract (HE) and hydro-ethanolic extract (EE) sequentially, which were used to study total phenolic content (TPC), total flavonoid content (TFC), PPA and PPL inhibitory and antioxidant activities. Constituents of the essential oils and HE were determined by GC-MS. In PPA inhibitory activities, IC_{50} (µg/mL) of the extracts were 0.27-0.37, which were close to 0.24 of the standard Acarbose. In PPL inhibitory activities, IC_{50} (µg/mL) of the extracts were 278.40-399.65, and that of Orlistat was 145.72. The EE were more active than HE; the flowers EE being the most active. In the DPPH assay, the flowers EE was most potent followed by leaves EE. The TPC and TFC of leaves EE were highest followed of flowers EE. The essential oil of flowers had higher estragole (55%) than linalool (37%), while the essential oil of the leaves had higher linalool (42%) than estragole (38%). The HE of the flowers contained higher estragole (42%) than linalool (23%), while of the HE of the leaves too had higher estragole (65%) than linalool (18%). The PPA and PPL inhibitory activities of *O. basilicum* extracts and their notable antioxidant potential propose the herb as a multi-target complimentary medicine for diabetes, obesity and oxidative stress.

PS1-F-002

Identification of antioxidant and hypoglycemic compounds in aqueous-methanol fraction of methanolic extract of *Ocimum canum* leaves

CJ Ononamadu[1], MS Sule[2], AJ Alhassan[2], GO Ihegboro[1,2], TA Owolarafe[1,2], FC Nwachukwu[1]

[1]Nigeria Police Academy, Wudil, Kano, Nigeria, Wudil, Nigeria, [2]Bayero University Kano, Kano City, Nigeria

Background: *Ocimum canum* belongs to a genus of aromatic annual and perennial herbs and shrubs in the family of Lamiaceae. It is native to Africa and cultivated commonly for its culinary and medicinal values. Previously, we had reported the antioxidant and hypoglycemic potential of the methanol crude extract of *Ocimum canum* leaves and its solvent partitions.
Aim: This present study was aimed to partially isolate and identify some bioactive compounds in the most active fraction (aqueous-methanol partition) of methanol extract of *Ocimum canum* leaves.
Method: Dried *Ocimum canum* leaves was macerated in methanol and the resulting crude extract was partitioned into Hexane, ethylacetate and aqueous-methanol sequentially. The aqueous –methanol partition was fractionated using silica gel column and solvents of varying polarity (chloroform, methanol and water) to yield many fractions which was pooled using analytical TLC. The pooled fractions were screened for antioxidant activity (DPPH scavenging assay and Ferric Reducing Power) and ability to reduce fasting sugar. The most active fraction was analysed with LC-MS and GC-MS to identify the active compounds.
Result: A total of 197 fractions were collected and pooled to seven (7) subfractions. Fraction OC4 showed the best activity with IC_{50}s 68.4±8.1ug/ml and 15.21±3.9ug/ml for DPPH scavenging and Ferric Reducing Power assays respectively and a 27% reduction in FBG in treated STZ-induced diabetic rats. The LCMS analysis on positive ionization mode revealed tentatively the following compounds: Dihydrotoshimone, Proanthocyanidin B, 3-hydroxylgabrol, Taxifolin, Digalloylglucose, Ananaflavosides, Embinim, Orietin. GCMS analysis also revealed the presence of the following low-polarity compounds: hexadecanoic acid, 9,12-octadecanoic acid, octa decanoic acid, γ-sitosterol and eugenol.
Conclusion: The study corroborates the previously reported antioxidant activity and hypoglycemic potential of the leaves of *Ocimum canum* and further more identified some bioactive compounds that can be isolated and further characterised.

Keywords: *Ocimum canum*, hypoglycemic, antioxidant, LCMS, GCMS

PS1-F-003

Automated metabolomic analysis of LC-MS data

Jiri Gruz**,** Andrea Luterova, Miroslav Strnad

Laboratory of Growth Regulators, Centre of the Region Haná for Biotechnological and Agricultural Research, Institute of Experimental Botany ASCR & Palacky University, Olomouc, Czech Republic

Metabolomic analysis of natural extracts typically relies on non-targeted analysis of all metabolites by using GC-MS or LC-MS. The resulting 3D-chromatogram contains thousands of features that need to be properly analysed and processed. In our laboratory, a software for automated data processing was developed in Matlab programming language. Besides basic chromatogram alignment, various postprocesses were designed and developed. One of them is grouping of so called “friendly ions” that consist of isotopic, adduct and fragment ions (originating from a single metabolite). Another postprocessing approach is the mass correction which is based on known background ions or identified metabolites. This allows lockmass-like correction with no measured lockmass data. The third postprocessing method is an algorithm for automated molecular peak identification. This approach is based on identification of typical fragments and adducts under defined experimental conditions. As it relies on correct determination of friendly ions, its effectivity is also affected by retention time stability and separation effectivity. The developed algorithms can help to process high density metabolomic data with improved accuracy and effectivity.

Keywords: LC-MS, metabolomic analysis

PS1-F-004

Metabolism of isotopically labeled benzoic and cinnamic acids in oat

Klára Supíková, Andrea Luterová, Miroslav Strnad, Jiří Grúz

Laboratory of Growth Regulators, Centre of the Region Haná for Biotechnological and Agricultural Research, Institute of Experimental Botany ASCR & Palacky University, Šlechtitelů 11, 783 71 Olomouc, Czech Republic

Phenolic acids are abundant plant secondary metabolites that form an important part of human diet. Surprisingly, the biosynthesis of benzoic acids is far from being resolved, especially in monocots, similarly as in the case with cinnamic acids. In this study, we used isotopically labeled precursors of benzoic acid (^{2}H5-BA) and trans-cinnamic (^{13}C6-tCA) to follow their metabolism in oat (*Avena sativa*). Precursors were applied to seedlings that were analyzed by both non-targeted and targeted UHPLC-HR-MS/MS. Firstly, the analysis of benzoic and cinnamic acids in control plants was performed, resulting in the detection of 5 derivatives of BA and tCA, including sulfated derivatives of BA and tCA. Free acids were not detected. Secondly, non-target analysis of plants feeded with precursors resulted in the detection of 18 metabolites of applied precursors from which 5 metabolites were also detected by targeted method. BA was incorporated in 10 metabolites, and tCA in 8 metabolites. Incorporation of precursors in sulfated derivatives of BA and tCA was not detected. The metabolites of tCA were predominantly hydroxy/methoxy derivatives and conjugates with other substances, whereas hydroxylation of BA was not observed. In conclusion, the biosynthesis of benzoic acids in oat seems to be different from the current knowledge of dicots, e.g. Arabidopsis, due to the absence of C2-chain shortening in cinnamic acids.

PS1-F-005

Probing lipid catabolism in cyanobacteria

Sandra Figueiredo, Teresa Martins, Kathleen Abt, Pedro Leão

Interdisciplinary Centre of Marine and Environmental Research (CIIMAR/CIMAR), Matosinhos, Portugal

Cyanobacteria, often regarded as the ancestors of chloroplast, are an important source of bioactive natural products [1,2]. The lipids that constitute their membranes are identical to the lipid composition of chloroplast membranes but distinct from that of most bacteria. Although the biosynthesis of lipids in cyanobacteria has been studied for some time, the specific characterization of these metabolic pathways has not yet been fully disclosed. Indeed, a key pathway involved in lipid metabolism, the β-oxidation metabolic pathway, responsible for the degradation of the fatty acids, seems to be lacking in cyanobacteria [3]. In this communication we report the strategies that we applied to support this hypothesis. These experiments included the heterologous expression of cyanobacteria acyl-ACP synthetase (Aas) in *E. coli*. This enzyme binds free fatty acids (FA) to ACP to form acyl-ACPs and allows exogenous FA to enter the *E. coli* FA biosynthetic pathway [4]. Thus, exogenous FA can be previously elongated or directly incorporated into complex lipids by acyltransferases specific for acyl-ACPs, allowing the study of lipid catabolism. This mutant *E. coli* was used as a positive control for the experiments with phylogenetically diverse cyanobacterial strains. The results obtained by feeding of labelled fatty acids and mass spectrometry analysis were important to give a deeper understanding of the biochemical aspects of lipid metabolism in cyanobacteria.

References:

[1] Burja AM, Banaigs B, Abou-Mansour E, Wright PC. Marine cyanobacteria - a prolific source of natural products, in: S. Kim (Ed.), Mar. Microbiol., Wiley-VCH Verlag GmbH & Co. KGaA, 2001: pp. 9347–9377.
[2] Tan LT. Phytochemistry 2007; 68: 954–979.
[3] Taylor G, Fatty acid metabolism in cyanobacteria, 2012.
[4] Kaczmarzyk D, Fulda M, Plant Physiol 2010; 152: 1598–1610.

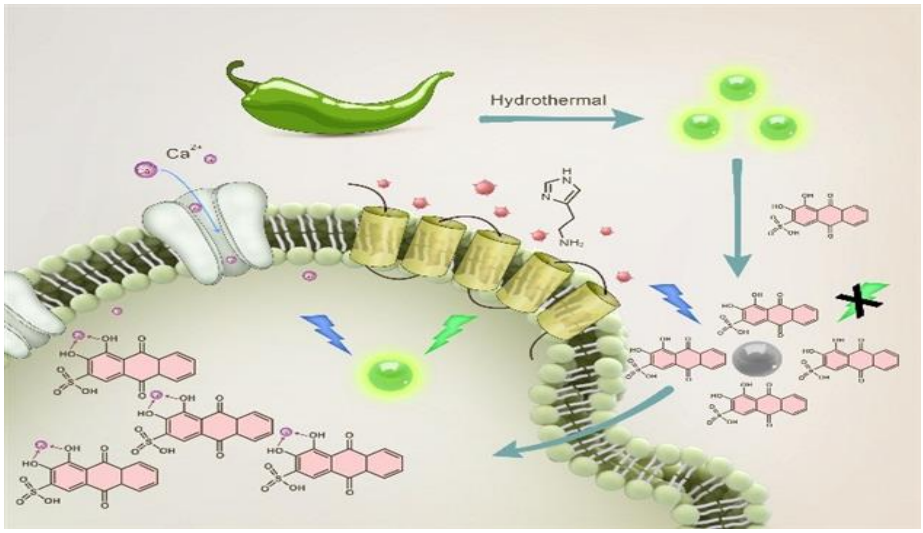
PS1-F-006

Capsicum-derived biomass quantum dots coupled with alizarin red S as an inner-filter-mediated illuminant nanoprobe for imaging of intracellular calcium ion dynamics

Shu-lin Zhao

State Key Laboratory for the Chemistry and Molecular Engineering of Medicinal Resources, Guangxi Normal University, guilin, China

The calcium ion (Ca^{2+}) plays a crucial role in signal transduction pathways associated with neurotransmitter release. Monitoring intracellular Ca^{2+} and studying its dynamics changes are of great significance for cell biology and biomedicine research. In this work, we report the use of biomass quantum dots (BQDs) derived from capsicum, as a specific fluorescent nanoprobe based on inner-filter-mediated emission for imaging of intracellular Ca^{2+}. In this imaging system, the BQDs were prepared by hydrothermal heating of capsicum. The as-prepared BQDs exhibit a maximum excitation wavelength at 405 nm, and a maximum emission wavelength at 520 nm. Alizarin red S (ARS), a Ca^{2+}chelator, exhibits a maximum absorption wavelength at 435 nm which overlaps with the excitation wavelength of the BQDs. When BQDs and ARS coexist, the BQD fluorescence is quenched by an inner-filter effect, and the quenched fluorescence is restored in the presence of Ca^{2+} due to the formation of a Ca-ARS complex, which exhibits a maximum absorption wavelength at 560 nm. Thus, BQD-ARS can be used as a specific fluorescent nanoprobe with inner-filter-mediated emission for imaging of intracellular Ca^{2+}. The BQD fluorescence intensity increased with an increase in the Ca^{2+} concentration. This novel nanoprobe was successfully used to image intracellular Ca^{2+}, and examine the changes in the intracellular Ca^{2+} concentration under histamine stimulation. It is found that the BQD-ARS respond within seconds to Ca^{2+} changes, which suitable for monitoring extracellular calcium signaling processes. This result indicate that BQD-ARS could become the primary fluorescent probe for Ca^{2+} in a wide range of biological areas, such as neuroscience.



PS1-F-007

Lipid and polyphenol composition in *Vaccinium* wild berries and cultivars

Ilona Vanaga[1,2], Egija Kitija Meijere[3], Linards Klavins[4], Maris Klavins[4], Anete Rateniece[2], Zane Greiza[2], Ugis Kletnieks[2]

[1]University of Latvia, Faculty of Medicine, Riga, Latvia, [2]Silv EXPO, SIA, Riga, Latvia, [3]University of Latvia, Faculty of Chemistry, Riga, Latvia, [4]University of Latvia, Laboratory of Natural Compounds Research, Riga, Latvia

Cultivated and wild berries from forests and bogs of Northern Europe are an excellent source of biologically active natural substances such as antioxidants, vitamins and fatty acids. Since berries are an essential part of diet and their processing is a major direction in food industry, it is necessary to analyse these valuable phytochemicals in berries.

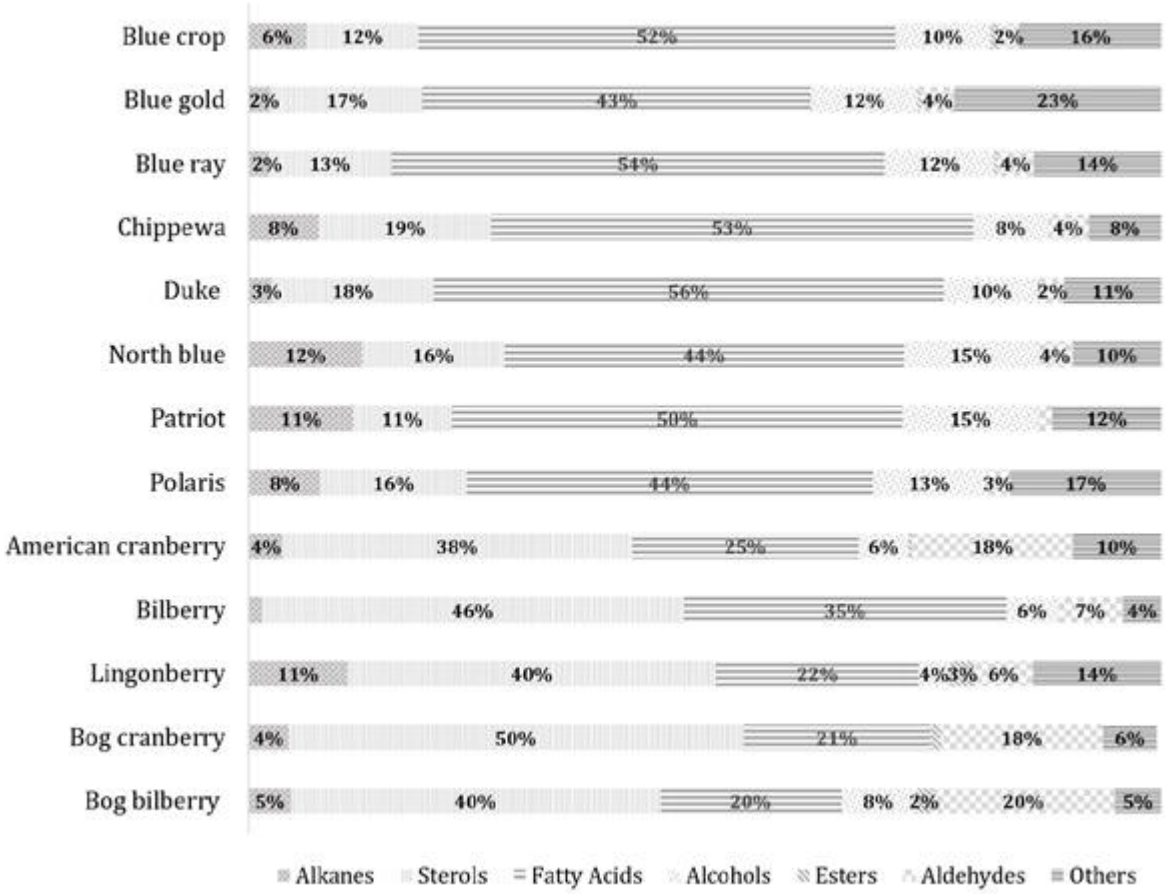
The aim of this study was to perform a comparative analysis of lipids, total polyphenols, flavonoids and other groups of extractives in genus *Vaccinium* wild berries and cultivars.

During the summer and autumn of 2017, the following berries were collected in Latvia and analysed: blueberry (*Vaccinium myrtillus* L.), highbush blueberry (*Vaccinium corymbosum* L.) cultivars, i.e., North blue, Duke, Chippewa, Blue ray, Blue gold, Blue crop, Polaris and Patriot, bog bilberry (*Vaccinium uliginosum* L.), lingonberry (*Vaccinium vitis-idaea* L.), American (*Vaccinium macrocarpon* L.) and bog cranberry (*Vaccinium oxycoccos* L.). Berry lipids were extracted with chloroform, derivatized with *N,O*-bistrifluoroacetamide and analysed using gas chromatography - mass spectrometry. Polyphenols and anthocianins, extracted using 70% ethanol and formic acid, were analysed by high performance liquid chromatography - mass spectrometry.

In the 6 types of the analysed berries 76 lipid compounds and 70 polyphenol compounds were identified and quantified. The lipid fraction contained such compound classes as fatty acids, sterols, triterpenoids, alkanes, phenolic and carboxylic acids, and carotenoids. All fresh berries contained high amounts of C18 unsaturated fatty acids and phytosterols. Apart from that, amino acids and vitamins (mostly B group and vitamin C) were quantified as well.

Overall, the cultivated highbush blueberries (*Vaccinium corymbosum* L.) were found have the highest amounts of lipids, while being much less rich in polyphenols than other *Vaccinium* berries. Bog bilberry (*Vaccinium uliginosum* L.) had the highest amount of polyphenols, whereas blueberries (*Vaccinium myrtillus* L.) had the least amount of lipids.

Fig. 1 *Vaccinium* berry extractives by compound classes, % (GC-MS analysis)



PS1-F-008

Quality control of cultivated greek thyme (*Thymus vulgaris*)

Anastasia Karioti[1], Anna Rita Bilia[2]

[1]Aristotle University of Thessaloniki, School of Pharmacy, Laboratory of Pharmacognosy, Thessaloniki, Greece, [2]Università degli Studi di Firenze, PHYTOLAB, Departimento di Chimica, Sesto Fiorentino (Firenze), Italy

Thymus vulgaris (Lamiaceae), is an important culinary herb and medicinal plant. Thyme extracts have a long use as expectorants, for soothing mouth and throat in common cold [1]. These activities are mainly attributed to the essential oil, especially to thymol and carvacrol, which are reported to have important antibacterial, spasmolytic and antioxidant activities. However, the use of hydroalcoholic or glycerol extracts suggests that non–volatile constituents might contribute to the efficacy of the herb. In the framework of quality control studies on cultivated Greek medicinal plants a method for the quality control of herba thymi was developed and validated. Three commercial samples were used, including a glycerol extract, together with four samples of Varico 3 hybrid collected at four different development stages. A simple and effective method based on ultrasonication and HPLC–DAD–MS was developed and accordingly validated. The method was optimised to ensure separation and identification of the components. Overall, 32 constituents were identified belonging to flavonoids, depsides and terpene derivatives. Rosmarinic acid and luteolin–7–*O*–glucoside were the most abundant representative metabolites and thus selected for the method validation. The HPLC method was validated for linearity, LOD and LOQ, precision (intra and inter–day variations between 1.03 and 3.78 expressed as %RSD) and accuracy (recovery was 95–106%). The overall assay was finally validated for precision (%RSD ranged from 2.07 to 3.48) and accuracy (recovery ranged from 99 to 106%) in three different concentration levels. The proposed protocol is proven suitable for the quality control of herba thymi and preparations thereof. To the best of our knowledge this is the first detailed report on the non–volatile content of *Thymus vulgaris* hybrid Varico 3.

Acknowledgements: Research was financed by grants of the Aristotle University of Thessaloniki (code 93267).

References:

[1] EMA Assessment Report on *Thymus vulgaris* L., *vulgaris zygis* L., herba. EMA/HMPC/342334/2013

PS1-F-009

Targeted & untargeted profiling of Muscat of Alexandria grapes

Maria Marinaki[1], Miss Konstantina Liva[1,2], Christina Virgiliou[2], Anastasia Ketsetzi[2], Helen Gika[3], Georgios Theodoridis[2], Dimitrios Christofilos[1], Andreana Assimopoulou[1]

[1]School of Chemical Engineering, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [2]Department of Chemistry, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [3]Department of Medicine, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece

Profiling of wine and grapes provide useful knowledge to improve the winemaking process and ensure compliance with labelling, safety requirements, traceability and authenticity. Regulatory agencies are demanding improved methods offering wider coverage and enhanced analytical performance. Several analytical techniques have been developed to determine constituents in wine, must and grape. In this study, the profiling of grapes and must constituents was performed. Grapes from the Muscat of Alexandria variety (Lemnos Island) and grape must, sampled at different days of alcoholic fermentation, were analysed by untargeted UHPLC–HRMS profiling method and Raman spectroscopy. Furthermore, a targeted LC–MS/MS method (HILIC mode) was applied to profile the polar metabolic content.

The aim of this work was to obtain a comprehensive profile of the metabolites present in the samples and monitor their differentiation among vineyards, grape ripening periods and throughout alcoholic fermentation. Multivariate analysis was performed to reveal trends in relation to sampling location or ripeness. UHPLC–Orbitrap MS analysis identified several secondary metabolites, while HILIC–MS/MS indicated differences in several primary metabolites such as aminoacids.

Raman spectroscopy was applied to investigate its potential in wine analysis with its signals indicating specific fingerprints of sugars (fructose and glucose), ethanol and water. Using standard compounds, the conversion of sugars to ethanol was monitored and a semi–quantification of these compounds was performed.

Acknowledgements: ANA and DC acknowledge support of this work by the project "Upgrading the plant capital" (MIS 5002803) implemented under the Action "Reinforcement of the Research and Innovation Infrastructure", funded by the Operational Programme "Competitiveness, Entrepreneurship and Innovation" (NSRF 2014–2020) and co–financed by Greece and the European Union (European Regional Development Fund). The authors acknowledge the Center of Interdisciplinary Research and Innovation of Aristotle University of Thessaloniki, for access to the Instrumentation of the LC–MS Laboratory (School of Chemical Engineering) and to Raman instrumentation (Faculty of Engineering).

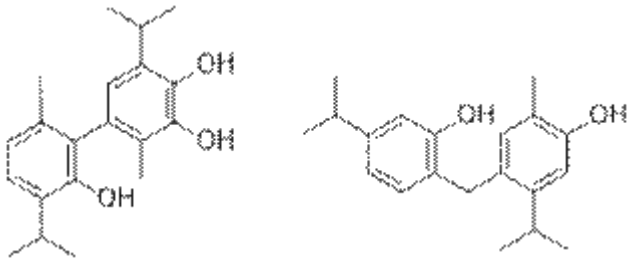
PS1-F-010

HPLC–DAD–MS guided investigations on the Greek cultivated medicinal plants *Origanum dictamnus* and *Thymus vulgaris* (Lamiaceae) reveal new natural products

Anastasia Karioti, Sofia Govari, Charikleia Paloukopoulou, Ilias Stefanis, Athina Soulioti

Aristotle University of Thessaloniki, Thessaloniki, Greece

In the framework of quality control studies on cultivated Greek medicinal plants, two widely used herbs were selected: *Thymus vulgaris* and the endemic species *Origanum dictamnus*. Besides long–standing traditional use, supported by Community herbal Monographs [1], both plants are important for the local economy. Previous studies on *T. vulgaris* have revealed antioxidant biphenyl derivatives, while *O. dictamnus* seems to be particularly rich in phenolic content [2,3]. Targeted isolations guided by HPLC–DAD–MS on extracts of both plants not only enabled us to create an in house chemical library, but also revealed the presence of new constituents which we report for the first time in the literature. Fractionations were based on classical isolation protocols including CC on Sephadex LH–20 and silica gel and RP HPLC. Structure elucidations were carried out by means of 1D and 2D NMR methods (COSY, HSQC, HMBC, ROESY, NOESY) and HPLC–DAD–MS analysis. Quite interestingly, both plants contain dimeric monoterpene derivatives, some of which are shown below. Antityrosinase activity experiments are underway.



Acknowledgements: Research was financed by grants of the Aristotle University of Thessaloniki (code 93267). Authors are grateful to ELGO–DIMITRA Thessaloniki for the plant material

References:

[1] EMA/HMPC/200429/2012 Corr. and EMA/HMPC/342334/2013
[2] Miura *et al.* ChemPharmBull 1989; 37:1816.
[3] Chatzopoulou *et al.*, JAFC 2010;58: 6064.

PS1-F-011

Simultaneous analysis of flavonoids and artemisinin with its analogues in *Artemisia annua* and real–time monitoring of its interaction with Bcl–2 with in–cell NMR spectroscopy

Ioannis Gerothanassis, Vassiliki Kontogianni, Alexandra Primikyri, Marianna Sakka

University of Ioannina, Ioannina, Greece

Artemisia annua is a promising and potent antimalarial herbal drug. This activity has been ascribed to its component artemisinin, a sesquiterpene lactone [1]. The ability to detect artemisinin and its known analogues in plant extracts is an especially difficult task since the compounds are present in very low concentrations, are thermolabile, and lack UV or fluorescent chromophores [2]. As a follow–up of our studies on the use of NMR spectroscopy in mixture analysis of plant extracts [3, 4] we report herein a facile and rapid NMR method for the simultaneous determination and quantification of both flavonoids and artemisinin and its analogues in *Artemisia annua* extracts. The analytical results were confirmed with HPLC/DAD/MS measurements. Qualitative and quantitative results obtained using an NMR method are described. Finally, in–cell NMR spectroscopy was employed to probe the binding mode of standard artemisinin and the *Artemisia annua* extracts to the unlabelled Bcl–2 anti–apoptotic protein in living human cancer cells.

Acknowledgements: This project has been co–financed by the Operational Program "Human Resources Development, Education and Lifelong Learning" and is co–financed by the European Union (European Social Fund) and Greek national funds.

Keywords: NMR spectroscopy, mixture analysis, artemisinin, *Artemisia Annua*

References:

[1] Callaway, E, Cyranoski D. Nature 2015;526:174–175.
[2] Christen P, Veuthey JL. Curr Med Chem 2001; 8: 1827–1839.
[3] Charisiadis P, Primikyri A, Exarchou V, Tzakos A, Gerothanassis IP. J Nat Prod 2011; 74: 2462–2466.
[4] Charisiadis P, Kontogianni VG, Tsiafoulis CG, Tzakos AG, Gerothanassis IP. Phytochem. Anal 2017; 28: 159–170

PS1-F-012

Metabolomic analysis of micromolecular diversity from Caatinga using LC–ESI–MS/MS

Danielle Rocha Pinho, Alan Cesar Pilon, Norberto Peporine Lopes

University São Paulo, Ribeirao Preto, Brazil

The Caatinga is an endemic and few studied Brazilian biome defined to the south by the Cerrado (savannah) and to the east and west by the Atlantic and Amazonian forests, respectively. Considering the lack of knowledge about the micromolecular diversity of the Caatinga, a biome in which it survives with water scarcity during most of the year, this work aims to understand how the metabolic profiles can explain the possible adaptation mechanisms of plants. Thus, the metabolomic approach – qualitative and quantitative analysis of organisms submitted to different conditions (genetic, abiotic and environmental) – combined with the molecular network tool, available on the Global Natural Product Network (GNPS) platform, was used to visualize MS spectral data through grouping charts. With this combination it is possible a discriminant evaluation allowing the systematic creation of models, resulting in statistical interpretations of the chemical, biological and ecological variations of the studied systems. In this work it is proposed to integrate this information in the attempt to find metabolic patterns that may help in understanding the mechanisms involved in the survival of the plants present in the Caatinga.

PS1-F-013

Dereplication by ^{13}C NMR in the presence of high boiling point solvents

Marine Canton[1,2], Stéphane Poigny[2], Richard Roe[2], Jean–Hugues Renault[1], Jean–Marc Nuzillard[1]

[1]Reims Institute of Molecular Chemistry, UMR CNRS 7312, Reims, France, [2]Laboratoires Pierre Fabre Dermocosmétique, Centre de Recherche et Développement, Toulouse, France Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771 Athens, Greece

Natural extracts represent a rich source of biologically active new ingredients of potential interest for the cosmetic industry. The increasing occurrence of these ingredients in marketed products created a need for rigorous regulations in order to prove their biological activity and to ensure their safety and stability. These fulfilling of these constraints require the thorough chemical profiling of the natural ingredients, a difficult task owing to their usually high composition complexity.

An innovative analytical workflow for the dereplication by Nuclear Magnetic Resonance (NMR) spectroscopy has been developed at the university of Reims in order to quickly identify known compounds present in natural extracts. This dereplication procedure combines Centrifugal Partition Chromatography (CPC) fractionation, ^{13}C NMR analyses, Hierarchical Clustering Analysis on the NMR data and a ^{13}C NMR chemical shift databasing.

In practice, many natural extracts of industrial interest are produced or stabilized in high boiling point solvents such as glycerol (BP=286 °C) or propanediol. These solvents interfere with the analytical procedure because solute concentrations are most often less than 10% in mass. After CPC fractionation, several fractions contain a high proportion of the support solvent which complicates the analysis by ^{13}C NMR for two reasons: (i) the receiver is saturated by the resonances of the support solvent due to its high concentration, and (ii) the strong solvent resonances produce decoupling artifacts that can be confused with the signals of interest.

Two solutions are proposed as remedies to these problems. The first one is the simultaneous presaturation of all the ^{13}C NMR resonances of the support solvent. The second solution consists in removing the decoupling artifacts using bi–level adiabatic ^{1}H pulse decoupling during ^{13}C signal acquisition. First results were obtained using model solutions of sucrose and glycerol in DMSO–d_6. Future works will deal with real extracts, possibility prepared in other solvents than glycerol.

PS1-F-014

Targeted and untargeted UHPLC-HRMS-based metabolomics of Boraginaceae roots

Nebojša Rodić[1], Angeliki Vlachou[1], Euaggelia Michailidou[1], Evangelos Tzimpilis[1], Helen Gika[2], George Theodoridis[3], Vassilios Papageorgiou[1], Andreana Assimopoulou[1]

[1]Laboratory of Organic Chemistry, School of Chemical Engineering, Aristotle University of Thessaloniki, 541 24, Thessaloniki, Greece, [2]School of Medicine, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [3]School of Chemistry, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece

Alkannins and Shikonins (A/S) are enantiomeric naphthoquinone natural products biosynthesized in the roots of at least 150 species mainly of the *Boraginaceae* plant family. Intense investigations over the past 40 years have shown a wide spectrum of biological activity of A/S, such as strong wound healing, antimicrobial, antioxidant, anti-inflammatory, tissue regenerative and antitumor ones. A/S comprise the active pharmaceutical ingredients (API) of several approved pharmaceutical and cosmeceutical preparations (HELIXDERM®, Histoplastin Red®, Histoplastin Junior®) invented by Prof. Papageorgiou of our group. In the frame of "MICROMETABOLITE" EU H2020 project, we have developed and optimized a workflow for the metabolic profiling of plant roots. Specifically, a fast and reliable extraction procedure was achieved for the comprehensive profiling and identification of a broad range of metabolites biosynthesized in plant roots, including the API A/S by UHPLC-HRMS.
Due to the small amounts of plant material available, aiming to protect biodiversity, and the large number of samples, every step of the workflow (drying, grinding, extraction, UHPLC-HRMS/MS) was optimized based on commercial samples. Solvents of varying polarities, neat or in mixtures were investigated for the extraction of metabolites. An UHPLC gradient method in hyphenation with Ion Trap-Orbitrap hybrid mass spectrometer in positive and negative ESI mode was applied. Several metabolites were detected and A/S derivatives were quantified. This methodology will be applied to all root samples collected and cultivated during the ''MICROMETABOLITE'' project with the aim to investigate metabolome variations.

Acknowledgements: This study was supported by the project "MICROMETABOLITE" that has received funding from the European Union's Horizon 2020 research and innovation programme, under the ITN Marie Skłodowska-Curie grant agreement No721635. We are thankful to the Center of Interdisciplinary Research&Innovation of Aristotle University of Thessaloniki, Greece, for access to the Large Research Infrastructure of the Liquid Chromatography and Mass Spectrometry Laboratory (School of Chemical Engineering).

PS1-F-015

Interaction between blue light and ABA during tomato seed germination

Pavla Šimurová (Pokorná)[1], Veronika Turečková[2], Miroslav Strnad[2], Martin Fellner[2]

[1]Department of Chemical Biology and Genetics, Centre of the Region Haná for Biotechnological and Agricultural Research, Palacký University, Olomouc, Czech Republic, [2]Laboratory of Growth Regulators, Centre of the Region Haná for Biotechnological and Agricultural Research, Palacký University in Olomouc and Institute of Experimental Botany ASCR, Olomouc, Czech Republic

Light, as important environmental signal, mostly stimulates seed germination via phytochromes by induction of absisic acid (ABA) catabolism. In tomato (*Solanum lycopersicum*), seeds can germinate well in total darkness (D) and their germination is sensitive to red light (RL), far-red light (FR) and blue light (BL) conditions. It was observed that BL alters the level of endogenous ABA [1] and reduces tomato seed germination [2]. However, the molecular mechanism of this BL-induced inhibition of seed germination is not well understood. Using genetic approach and hormone profiling in photomorphogenic mutants we are investigating the mechanisms by which blue light reduces seed germination in tomato. As a model plant, we are using tomato mutant 7B-1, which is an ABA over-producer and it is defective in several phototropin-mediated pathways. This mutant shows higher tolerance to the BL-induced inhibition of seed germination compared to corresponding WT (cv. Rutgers). So far, the gene expression analyses of 8 PYL ABA receptors revealed that 6 hours after sowing, BL significantly increases transcript level of PYL5 in 7B-1 seeds, but not in WT. Contrary, the expression of PYL3 in 7B-1 is significantly lower than in WT. Further gene expression analyses will be focused on BL photoreceptors, phototropins and cryptochromes. The targeted ultra-high performance liquid chromatography-tandem mass spectrometry (UHPLC-MS/MS) analysis revealed that the germinating 7B-1 seeds contain less ABA than those of WT and that the decrease of ABA level during first 24 hours of incubation in BL is faster in 7B-1 than in WT.

Acknowledgement: This work was supported by Operational Programme Research, Development and Education Support of Academic mobility at Palacký University Olomouc CZ.02.2.69/0.0/0.0/16_027/0008482.

Keywords: blue light, tomato, seed germination, abscisic acid, 7B-1

References:

[1] Humplik JF, Bergougnoux V, Jandova M, Simura J, Pencik A, Tomanec O, Rolcik J, Novak O, Fellner M. PLoS One 2015; 10: e0117793
[2] Fellner M, Sawhney VK. Planta 2002; 214: 675–682

PS1-F-016

Aqueous ethanolic extracts from arid halophyte species *Arthrocnemum macrostachyum* and *Tetraena qatarensis*

Samar Al-Jitan[1], Saeed Ahmed Alkhoori[1], Michael Ochsenkühn[2], Shady A. Amin[2,3], Lina F. Yousef[1]

[1]Khalifa University of Science and Technology, Department of Chemistry, Abu Dhabi, United Arab Emirates, [2]New York University Abu Dhabi, Biology Division, Abu Dhabi, United Arab Emirates, [3]New York University Abu Dhabi, Chemistry Division, Abu Dhabi, United Arab Emirates

The plant species *Arthrocnemum macrostachyum* and *Tetraena qatarensis* can grow on desert saline soil and can be a valuable source of phytochemicals in sustainable agricultural systems. In this study, various concentrations of ethanol in water (25%, 50%, and 75% v/v) were used in microwave-assisted extraction of biomass (1g in 30 mL solvent; 30 second at 231W). Total phenolic content expressed as mg Gallic acid equivalent (GAE) per g extract was determined using the Folin-Ciocalteu assay. Antioxidant activity was measured using the half-maximal inhibitory concentration (IC_{50}) of DPPH free radical. Qualitative analysis of extracts was carried out using spectral analysis and metabolomics profiling via UHPLC-Q-ToF-MS. Extracts acquired from both plants using 75% ethanol solvent exhibited the highest DPPH scavenging activity (IC_{50} = 62.7 ± 0.4 and 67.9 ± 4.8 µg/mL) and total phenolic content (45.6 ± 1.0 and 54.4 ± 0.8 mg GAE/g extract) for *A. macrostachyum* and *T. qatarensis* respectively. Qualitative analysis of spectral profiles and metabolomics data suggest the composition of crude extracts from the 50% and 75% ethanol extracts are similar in *A. macrostachyum*, whereas the 25% and 50% ethanol are similar in *T. qatarensis*. In conclusions, increasing the concentration of ethanol in water decreases the extraction yield, but the extracts have higher phenolic content, DPPH scavenging activity and greater chemical complexity in extracted compounds.

Keywords: antioxidant; DPPH; metabolomics; phenolic; UV-VIS

PS1-F-017

An LC/HRMS method for the determination of naturally occurring saturated hydroxy fatty acids

Maroula Kokotou, Christiana Mantzourani, George Kokotos

National and Kapodistrian University of Athens, Athens, Greece

Hydroxylated derivatives of saturated fatty acids have been scarcely studied for their bioactivities. Saturated 2-hydroxy and 3-hydroxy fatty acids have been found in foodstuffs, in particular in milk and cheese, as demonstrated using gas chromatography coupled with electron-capture negative ion mass spectrometry [1]. In addition, monounsaturated fatty acids, such as 2-hydroxy oleic acid, have been identified in the oil of *Salvia nilotica* seed [2] or in *Thymus vulgaris*. Recently, it has been shown that hydroxy fatty acids are better ligands for the GPR84 receptor than the corresponding non-hydroxylated fatty acids. Thus, a method for the rapid determination of 2- and 3-hydroxy fatty acids in foodstuffs and plant extracts is of high interest. Here, we describe the first LC/HRMS method for the simultaneous determination of 2- and 3-hydroxy fatty acids. An AB SCIEX TripleTOF® 4600 system coupled with a micro-LC Eksigent was used. Electrospray ionization (ESI) –negative mode– was used for the Full Scan MS experiments, while Halo C_{18} 2.7 μm 90Å 0.5x50 mm from Eksigent was employed as a column and the mobile phase consisted of a gradient (A: acetonitrile/0.01% formic acid-isopropanol 80/20 v/v; B: H_2O/ 0.01% formic acid). The data acquisition was carried out with MultiQuant™ from AB SCIEX. A variety of 3-hydroxy fatty acids (capric, lauric, myristic, palmitic, stearic) as well as 2-hydroxy fatty acids (palmitic and stearic) were studied. The method allows the simultaneous determination of saturated hydroxy fatty acids in a 10-min run and may find applications in food and phytochemical analysis.

Acknowledgements: The research presented was carried out within the framework of a Stavros Niarchos Foundation grant to the National and Kapodistrian University of Athens

Keywords: Hydroxy fatty acids, LC/HRMS

References:

[1] Jenske R, Vetter W. 2009.
[2] Bohannon MB, Kleiman R. Lipids 1975; 10: 703–706

PS1-F-018

Study of glucosinolate-hydrolysis products in broccoli and brussels sprouts by gas chromatography-mass spectrometry and liquid chromatography-mass spectrometry

Irene Mesimeri**,** Maroula G. Kokotou, Violetta Constantinou-Kokotou

Department of Food Science and Human Nutrition, Chemical Laboratories, Agricultural University of Athens, Iera odos 75, Athens 11855, Greece, ATHENS, Greece

Cruciferous vegetables are a rich source of phytochemicals, such as glucosinolates, that have been reported as precursors of human health promoting agents. Glucoraphanin and glucobrassicin represent the major groups of glucosinolates in broccoli, while other glucosinolates are less abundant. Glucosinolates, upon hydrolysis, produce various products, including isothiocyanates and indole-3-carbinol, which have been reported to exhibit antimicrobial, antioxidative, antitumor and antiviral activity [1]. Due to the growing interest in their potential health protective effects, dietary isothiocyanates have attracted much attention. Limited number of isothiocyanates could be simultaneously investigated using a single chromatographic method, because they possess different polarities and volatilities. Our aim was the development of complementary gas chromatography-mass spectrometry and liquid chromatography-mass spectrometry methods allowing the detailed study of isothiocyanates in cruciferous vegetables. Florets and roots of fresh broccoli (*Brassica oleracea* L. var. Marathon, Parthenon, Monrello) and Brussels sprouts (*Brassica oleracea L*. var. Trafalgar, Abacus, Franklin), originated from Greece (Athens, Argos, Evia, Vonitsa, Agrinio), Holland and Belgium, were used for the preparation of extracts. Samples were lyophilized, immediately after purchase, ground to a fine homogenous powder using a mortar and pestle and extracted with dichloromethane. Erucin (4-methylthiobutyl isothiocyanate), 3-methylthiopropyl isothiocyanate, allyl isothiocyanate and phenethyl isothiocyanate were studied employing GC/MS, while sulforaphane and indole-3-carbinol by LC/HRMS, according to a method recently developed by us [2]. All the isothiocyanates were detected in higher concentrations in roots than in florets. Allyl isothiocyanate was detected only in Brussels sprouts. 3-Methylthiopropyl isothiocyanate was detected only in the root samples.

Keywords: *Brassica oleracea*, Brussels sprouts, isothiocyanates, LC/HRMS

PS1-F-019

Improving the definition of maca products' quality using NMR, HPTLC and HPLC

Francesca Scotti[1], Pinelopi Nika[1,2], Raman Suthar[1], Michael Heinrich[1]

[1]UCL School of Pharmacy, London, United Kingdom, [2]Faculty of Pharmacy, University of Athens, Athens, Greece

In Peru Maca (*Lepidium meyenii* Walp.*)* has been used medicinally and for its nutritional value. It recently saw a surge in popularity due claims as a male fertility enhancer, an aphrodisiac and a sport performance booster. Food supplements of maca are now readily available in stores and online and China has risen as the biggest producer of maca. This has led to the registration of 'Maca Junín-Pasco' as an Appellation of Origin at the World Intellectual Property Organization (WIPO) in 2012.
No quality standard is currently available as the USP only has a pending monograph in need of analytical information. The compounds responsible for the claimed activity are unknown, resulting in uncertainty about the best analytical approach.
Here we looked at a collection of commercial products (powders, tablets, capsules, whole or sliced hypocotyls) sourced in a number of countries, including Peru and China. They were analysed using a combination of ^{1}H-NMR-based PCA metabolomics and HPTLC to compare different types of formulations, raw and processed material of different origin with HPLC being used for fatty acids, macamides and macaenes content.
A certain degree of variability was found among the raw material. Unfortunately, it was not possible to compare Chinese and Peruvian raw material given the legal bounds involved in acquiring Peruvian material. Commercial unprocessed powders purchased from other countries generally claimed to contain Peruvian material. Such samples seemed of consistent quality, while processed products, especially extracts, showed extreme variability.
HPTLC has previously been shown to be particularly suitable for pharmacopoeial monographs and further investigation for an optimal method is of relevance for the USP monograph. This study calls for a clearer definition of good quality maca product and of clarifying the alleged differences between Chinese and Peruvian produce. This would help to deliver high-quality supplements to customers.

Acknowledgements: PN worked on this project while being a visiting Erasmus student at SoP. FS's position has been in part supported by a charitable donation by Dr. Willmar Schwabe GmbH, Karlsruhe, Germany. However, the donor has not had any input into the design and interpretation.

Keywords: Maca, *Lepidium meyenii*, Intellectual Property, Quality Control, HPTLC, NMR, HPLC, extracts, macamides, chemical fingerprint, sample variability

References:

[1] Beharry S, Heinrich M. J Ethnopharmacol 2018; 211: 126–170

PS1-F-020

High resolution mass spectrometry studies of sulforaphane and indole-3-carbinol in broccoli

Maroula G. Kokotou, Panagiota-Kyriaki Revelou, Christos Pappas, Violetta Constantinou-Kokotou

Department of Food Science and Human Nutrition, Chemical Laboratories, Agricultural University of Athens, Iera odos 75, Athens 11855, Greece, Athens, Greece

Vegetables in the Brassicaceae (Cruciferae) family are a rich source of bioactive compounds, such as glucosinolates, flavonoids, and vitamins. In particular, two bioactive compounds present in broccoli namely sulforaphane and indole-3-carbinol, have attracted a lot of attention, because their consumption is associated with reduced risk of cancer [1]. Here, the development of an efficient and direct method for the simultaneous determination of sulforaphane and indole-3-carbinol in broccoli using UPLC–HRMS/MS is described. A Q-TOF mass analyzer (Bruker Maxis Impact) allowed high resolution and accuracy, sensitivity and selectivity, offering rapid and effective analysis. Electrospray ionization (ESI) –negative mode– was used. For the chromatographic study of sulforaphane and indole-3-carbinol, a Chromolith RP column (100 x 4.6 mm, Merck) was used. The mobile phase was a mixture of MeOH/H_2O (80/20, v/v) at a flow rate of 0.3 mL/min. An auto-MS method was developed for the determination of sulforaphane and indole-3-carbinol in broccoli samples. Data acquisition was carried out with Data Analysis from Bruker Daltonics (version 4.1). The correlation coefficient and limits of detection (LOD) and quantification (LOQ) were 0.993, 0.77 mg/L and 2.35 mg/L for sulforaphane and 0.997, 0.42 mg/L, 1.29 mg/L for indole-3-carbinol, respectively. The content of sulforaphane and indole-3-carbinol varied between 72 ± 9–304 ± 2 mg and 77 ± 1–117 ± 3 mg per 100 g of fresh florets, respectively [2]. Taking into consideration the differences in cultivar, geography, season and environmental factors, the results agreed with values published in the literature using other techniques. In conclusion, a rapid and accurate analytical method for the simultaneous quantification of sulforaphane and indole-3-carbinol in broccoli was developed using high resolution mass spectrometry.

Keywords: sulforaphane, broccoli, LC/HRMS

References:

[1] Liang H, Yuan Q. Crit Rev Biotechnol 2012; 32: 218–234
[2] Kokotou MG, Revelou P-K, Pappas C, Constantinou-Kokotou V. Food Chem 2017; 237: 566–573

PS1-F-021

Variability of phytohormones in Brassicaceae determined by high resolution mass spectrometry

Panagiota-Kyriaki Revelou, Maroula G. Kokotou, Violetta Constantinou-Kokotou

Department of Food Science and Human Nutrition, Chemical Laboratories, Agricultural University of Athens, Iera odos 75, Athens 11855, Greece, Athens, Greece

Phytohormones are natural products that regulate all physiological and developmental processes in plants. They are grouped in classes on the basis of their structure and function and include auxins, jasmonates, cytokinins, abscisic acid, salicylic acid (SA), gibberellins, ethylene, brassinosteroids and polyamines [1,2] Auxins are signaling molecules involved in multiple stages during growth and development of plants. The levels of the most important auxin, indole-3-acetic acid (IAA), are regulated by the formation of amide and ester conjugates with amino acids and sugars. In this work, 27 compounds including auxins and related metabolites, IAA amide conjugates, salicylic acid and jasmonic acid, were studied using high resolution mass spectrometry. Previously, undescribed conjugates of IAA with L-serine were identified and quantified in vegetables of the Brassicaceae family for the first time as a result of using a new LC-HRMS. The HRMS spectra were recorded on an Agilent 6530 Quadrupole Time of Flight LC-MS system (QTOF LC-MS), with an ESI source (negative mode), coupled with Agilent 1290 Infinity UHPLC system and an autosampler. The data acquisition was carried out with Agilent MassHunter software (version B.06.00). Chromatographic study of plant hormones was performed with an Agilent Zorbax C_{18} (50 x 2.1 mm, 1.8 μm) column and the mobile phase consisted of a gradient (A: ultrapure water-formic acid 0.1% and B: MeOH-formic acid 0.1%). The method was validated and characterized by excellent linearity and detection limits that varied from 2.2 ng/mL (IAA conjugate with glycine) to 97.8 ng/mL (IAA). This is the first report employing high resolution mass spectrometry for the study and quantification of plant hormones and IAA conjugates in cruciferous vegetables.

Keywords: *Brassica oleracea*, LC/HRMS, auxins

References:

[1] Santner A, Estelle M. Nature 2009; 459: 1071–1078
[2] Cai W-J, Ye T-T, Wang Q, Cai B-D, Feng Y-Q. Plant Methods 2016; 12: 47

PS1-F-022

New strategies to strigolactone determination in complex sample matrices

Kristyna Flokova[1,2], Mahdere Z. Shimels[3], Beatriz Andreo Jimenez[3], Yanting Wang[2], Harro J Bouwmeester[2]

[1]Laboratory of Growth Regulators, Centre of the Region Haná for Biotechnological and Agricultural Research, Institute of Experimental Botany AS CR & Faculty of Science, Palacký University, Slechtitelu 27, 78371 Olomouc, The Czech Republic, [2]Plant Hormone Biology, Swammerdam Institute for Life Sciences, Sciencepark 904, 1098 XH Amsterdam, The Netherlands, [3]Laboratory of Plant Physiology, Wageningen University, Droevendaalsesteeg 1, 6708 PB Wageningen, The Netherlands

Strigolactones (SLs), chemically sesquiterpene lactones, are the most recently described group of plant hormones. These root-exuded compounds can be recognized by parasitic plants as a host plant signal resulting in enormous negative impact on yield of agriculturally important crops [1]. Detailed insight to biosynthesis and metabolism of SLs is hampered by their limited stability and extremely low quantities in complicated plant/soil matrix. Conventional approaches to SL determination by mass spectrometry-based techniques require large initial volumes of root exudate wash (≥ 500 ml), eventually gram-amounts of plant material followed by multi-step and low specific purification procedures [2,3]. We present highly sensitive and validated UHPLC-MS/MS method for simultaneous profiling of ten selected SL compounds. In order to minimize strong matrix effect, sample size of root exudate extract and root tissue was reduced to 15 ml and 50 mg, respectively. The combination of rapid extraction with water miscible aprotic solvents and single-step purification using macroporous polymer-based sorbents with both hydrophilic and lipophilic retention characteristics simplified current sample preparation procedures, notably improved stability and extraction recovery of selected SLs (≥ 85%). A new ion source, UniSpray (Waters), was employed as the interface of mass spectrometer to improve the signal of SL analytes, lacking ionizable residues in the structure. The novel atmospheric pressure ionization showed the average intensity gain of factor 4.5 compared to electrospray (ESI).

Acknowledgements: We thank to Swammerdam Institute of Life Sciences and Waters Corporation for use of the instrumentation; and the project CZ.02.2.69/0.0/0.0/16_027/0008482 for financial support.

Keywords: Strigolactones, Sample Preparation, UHPLC-MS/MS analysis, UniSpray

References:

[1] Al-Babili S, Bouwmeester HJ. Annu Rev Plant Biol 2015; 66: 161–186
[2] neyama K, Xie X, Sekimoto H, Takeuchi Y, Ogasawara S, Akiyama K, Hayashi H, Yoneyama K. New Phytol 2008; 179: 484–494
[3] Boutet-Mercey S, Perreau F, Roux A, Clave G, Pillot J-P, Schmitz-Afonso I, Touboul D, Mouille G, Rameau C, Boyer F-D. Phytochem Anal 2018; 29: 59–68

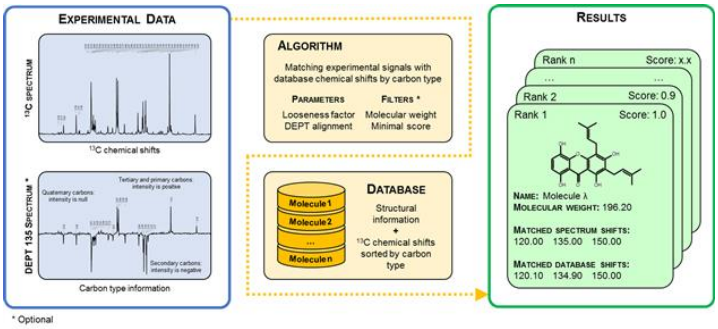
PS1-F-023

Custom-made algorithm: a powerful tool for ^{13}C-NMR dereplication of complex mixtures. Proof of concept on a *Garcinia mangostana* fruit peel extract

Antoine Bruguiere, Joel Dietsch, Derbre Severine, Sorphon Suor-Cherer, Dimitri Breard, Pascal Richomme

SONAS/SFR QUASAV - University of Angers, Angers, France

Isolation and identification of natural products is often a long and difficult task that can unfortunately lead to the characterization of already known compounds, resulting in a considerable waste of time. Over the past few years, so-called “dereplication techniques” have emerged, allowing to quickly identify the major compounds in a mixture with, ideally but depending on the extract or fraction complexity, a minimum of purification/separation steps.
However, dereplication analysis through LC-MS^n profiling is sometimes unable to distinguish stereoisomers. Recently, Hubert *et al*. implemented ^{13}C NMR dereplication methods either after CPC fractionation [1] or, in a faster way, directly on crude extracts [2]. Nevertheless, in our experience, this process was not discriminative enough to characterize xanthone derivatives in a *Garcinia mangostana* (*Clusiaceae*) fruit peel extract. A new method was thus developed. Such process requires chemical shifts databases (DBs) as well as a dedicated algorithm to search for carbon chemical shifts (δC) matching. Two DBs containing the δC associated with 382 xanthones were created. The first one (DB1) was built up from the JEOL CH-NMR-NP [3], based on reported experimental data. The second one (DB2) contains theoretical δC [4] calculated through HOSE predictions with the ACD/Labs software [5]. A Python algorithm was then designed, using both ^{13}C{^{1}H} and ^{13}C-DEPT-135 spectra to produce a ranked score list of candidates with δC assignments (Fig. 1).
Based on DB1 or DB2, using DEPT results or not, the dereplication analysis of *G. mangostana* fruit peel extract will be presented and discussed.



Keywords: NMR, dereplication, algorithm

References:

[1] Hubert J, Nuzillard J-M, Purson S, Hamzaoui M, Borie N, Reynaud R, Renault J-H. Anal Chem 2014; 86: 2955–2962
[2] Bakiri A, Hubert J, Reynaud R, Lanthony S, Harakat D, Renault J-H, Nuzillard J-M. J Nat Prod 2017; 80: 1387–1396
[3] Hayamizu K. Natural Product NMR-DB "CH-NMR-NP". (2017). Available at: https://www.j-resonance.com/en/nmrdb (accessed: May 2018)
[4] Bruguière, A. *et al*. Fitoterapia, Under review.
[5] ACD/Labs (2014 Release)

PS1-F-024

Targeted isolation of *trans*-crocin 4 (TC4) from *Crocus sativus* L. employing step gradient Countercurrent Chromatography (CPC), followed by a metabolome study in mice after i.p. administration of TC4

Evangelia Karkoula[1,2], Apostolis Angelis[3], Ioanna Dagla[3], Nikolaos-Stavros Koulakiotis[2], Evagelos Gikas[2,3], Nikolaos Kokras[1,4], Christina Dalla[1], Maria Halabalaki[3], Alexios-Leandros Skaltsounis[3], Anthony Tsarbopoulos[1,2]

[1]Medical School, National and Kapodistrian University of Athens, Department of Pharmacology, 115 27, Athens, Greece, [2]The Goulandris Natural History Museum GAIA Research Center, Bioanalytical Department, 145 62, Kifissia, Greece, [3]Department of Pharmacy, National and Kapodistrian University of Athens, 157 71, Athens, Greece, [4]Medical School, National and Kapodistrian University of Athens, First Department of Psychiatry, 115 27, Athens, Greece

Saffron, the dried stigmas of *Crocus sativus* L. is the most valuable spice. Saffron's main constituents, crocins, exhibit anti-inflammatory and anti oxidant activity while offering enhancement of memory capacity and promising results for neuroprotection. [1] The initial aim of this study was to develop a simple and effective method for one-step isolation of the main secondary metabolites of a Methanol-Water 50:50 v/v extract of saffron by using step-gradient countercurrent partition chromatography (CPC). CPC analysis performed both in dual mode and elution extrusion mode, led to the isolation of 15 compounds while a scaled up, fit for purpose, targeted extraction procedure led to the recovery of significantly high quantities of pure TC4, the major crocin of saffron. Subsequently, an untargeted UPLC-HRMS metabolomic study was performed in order to estimate the metabolic fingerprint and its associated alterations following i.p. administration of the TC4 in male and female mice. Hence, 58 mice (control and treated) have been administered with TC4 (50 and 150 mg/kg) and have been sacrificed at a time range of 0-240 min. The obtained results were subjected to multi level multivariate analysis, i.e. PCA, PLS DA. The statistically important features contributing to the discrimination were annotated with comparison to online databases e.g. HMDB aided by chemometric processing (adduct and fragment identification, covariance searching etc). The high variability of the studied groups imposed by various factors e.g. sex and time points was overcome by a ML PLS DA enabling the split of variation to each individual component and allowing a clear depiction of the metabolites contributing to each discrimination. A distinct sex-related effect on the metabolome has been made apparent as well as internal differences among the treated populations.

Keywords: saffron, step-gradient Countercurrent Chromatography, *trans*-crocin 4, UPLC HRMS, metabolomics

References:

[1] Srivastava R *et al*., Pharmacogn Rev 2010; 4(8): 200–208.

PS1-F-025

Untargeted metabolomics study reveals Greek propolis novel anti-tyrosinase agents using ultrahigh-performance liquid chromatography-hybrid quadrupole-orbitrap mass spectrometry

Aikaterini Termentzi[1], Maria-Ioanna Stavropoulou[2], Konstantina-Georgia Stathopoulou[2], Kyriaki Machera[1], Evagelos Gikas[3], Nektarios Aligiannis[2]

[1]Laboratory of the Toxicological Control of Pesticides, Benaki Phytopathological Institute, Kifissia, Greece, [2]Laboratory of Pharmacognosy, School of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [3]Department of Pharmaceutical Chemistry, School of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece

Propolis has been used since the ancient times as an antibacterial, antifungal and anti-inflammatory agent. Many scientific studies have proved those activities, as well as the anti-oxidant, anti-viral, immunostimulating, even the anti-tumour activity of propolis. Those biological properties are associated with the phytochemical composition, which is connected with the botanical and geographical source. There are very few references on the tyrosinase inhibitory activity of propolis, attributing the effect to the phenolic and flavonoidic content. Tyrosinase is an enzyme that triggers melanogenesis. Thus, compounds that inhibit this enzyme can be considered as candidates for developing new skin bleaching agents in cosmeceutical or medical applications.

In the present study forty four propolis samples were collected from eleven different regions of Greece (mainland and island areas). All samples were extracted following the same protocol and extracts were evaluated for their tyrosinase inhibitory activity. In parallel their UHPLC-HRMS profiles were recorded on a Q-Exactive Orbitrap HRMS/MS apparatus. The phytochemical analysis of the extracts revealed the presence of phenolic acids, flavonoids, diterpenic and triterpenic acids. PCA score plots from MS data in negative and positive ion modes (Compound Discoverer), showed a clear separation between the mainland and island samples. The Variable Importance in Projection (VIP) values were calculated for the PLS-DA approach, in order to pinpoint the most influential parameters to the classification of the two regional groups (SIMCA P+ 11.5). The coefficient plots of the spectrometric features in combination to the anti-tyrosinase activity of the samples revealed that it is not the phenols, but specific resin acids that are responsible for the activity. Those features match with the most influential for the PLS-DA clustering VIPs. Results showed that specific propolis extracts from the Greek islands, rich in resin acids, may serve as raw material for novel anti-tyrosinase agents.

PS1-F-026

A comparative and integrated study for the extraction and determination of phenolic compounds in olive fruits and stones

Georgia Sarikaki, Konstantina Touvleliou, Maria Halabalaki, Sofia Mitakou, Leandros A Skaltsounis

Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771, Athens, Greece

It is widely known that the polar fraction of olive fruits and stones (*Olea europaea*) contains a variety of phenolic compounds with a huge range of bioactivities such as antioxidant, cardioprotective, antimicrobial and anticancer [1]. Many different approaches for the analysis of these compounds have been published, which have led to different results and the need for a thorough study on the extraction methods is more important than ever [2,3]. Thus, we attempted to shed light into the controversial data reported in the literature, by examining different extraction procedures (different solvents, different extraction temperatures) and plant forms (lyophilized fruits and stones, pulp and whole fruits). The work has been carried out on olive fruits and stones prepared from one cultivar. The obtained extracts were chemically characterized using liquid chromatography-mass spectrometry (LC-MS). Interestingly, the results exhibited a higher yield after successive extraction, while phenolic profiles were mainly influenced by the plant form. More specifically, the lyophilized fruits and stones were more favorable as they had a richer polyphenolic content as identified and quantified using LC-MS and high-performance liquid chromatography-diode array detection chromatography (HPLC-DAD). These data provide a detailed analysis regarding qualitative and quantitative differences in phenolic composition among different extraction methods and plant form.

Acknowledgments: The work is funded by the Hellenic National Scholarships Foundation (IKY).

Keywords: extraction method, olive fruit, olive stones

References:

[1] Taamalli A et al. Food Sci 2012; 77(4):.R83–92.
[2] Talhaoui N, Gómez-Caravaca MA, León L et al. Int J Mol Sci 2016; 17
[3] Romero C, Garcia P, Brenes M et al. Eur Food Res Technol 2002; 215: 489

PS1-F-027

Assessment of the effect of extraction solvent on cannabinoid yield and phytochemical profile of fibre-type Cannabis sativa L. using UPLC-PDA and HPTLC

Petros S. Tzimas, Eleftherios A. Petrakis, Apostolis Angelis, Maria Halabalaki, Leandros A. Skaltsounis

Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771, Athens, Greece

C. sativa L. of fibre type (hemp) and related phytocannabinoids represent an area of rigorous research owing to reported evidence of numerous health-promoting attributes [1]. Solvent selection is a critical step in the development of an efficient extraction protocol for hemp quality assessment but equally important in phytochemical profile studies. The present work aimed to investigate the effect of different extraction solvents on cannabinoid recovery and extraction yield as well as to evaluate the chemical profile of the extracts obtained. To this end, ultra-high performance liquid chromatography coupled with photodiode array detection (UPLC-PDA) and high performance thin-layer chromatography (HPTLC) were utilized.

Samples of hemp inflorescences of Greek origin were subjected to ultrasound-assisted extraction (UAE), applying the method proposed by UNODC with slight modifications [2]. Different extractants covering a range of polarities relevant to cannabinoids were examined. Their selection was based on official methods [2,3] as well as on recent literature [4,5]. Alcohols (e.g. ethanol, methanol) along with less polar solvents such as acetonitrile, acetone, ethyl acetate, and n-hexane, employed either alone or as binary systems, were used among others. To determine the recovery of principal cannabinoids (e.g. CBDA and CBD) achieved by each extractant, quantification was performed using a newly developed and validated UPLC-PDA method. HPTLC methodology was followed to compare the metabolite profiles of the obtained extracts, allowing for a rapid and cost-effective fingerprint analysis. The differences in extract profiles were indicative of the respective solvent's selectivity and efficiency and the observed patterns can be attributed to the preferential affinity of each extraction solvent to specific metabolites.

Acknowledgements: The research presented was carried out within the framework of a Stavros Niarchos Foundation grant to the National and Kapodistrian University of Athens. The "Kannavis Producers Cooperative" (To Kannavi) is acknowledged for kindly providing hemp samples.

Keywords: Cannabis sativa L., phytocannabinoids, HPTLC, UPLC-PDA, solvent selection, fingerprinting

References:

[1] Abrams D. Eur J Intern Med 2018; 49: 7–11.
[2] UNODC, Recommended Methods for the Identification and Analysis of Cannabis and Cannabis Products. United Nations 2009; pp. 25–46.
[3] Commission Delegated Regulation (EU) 2017/1155 of 15 February 2017. Off J Eur Union L 167: 1–15.
[4] Brighenti V, Pellati F, Steinbach M, Maran D, Benvenuti S. J Pharm Biomed Anal 2017; 143: 228–236.
[5] Wang YH, Avula B, ElSohly MA, Radwan MM, Wang M, Wanas AS, Mehmedic Z, Khan IA. Planta Med 2018; 84(4): 260–266.

PS1-F-029

Development of a GeLC-MS/MS based strategy for the identification of novel drug or diagnostic targets against Breast and prostate cancer

Ioanna-Maria Orfanou[1], Theodoros Karampelas[1], Manousos Makridakis[2], George Mermelekas[2], Konstantinos Vougas[2] and Constantin Tamvakopoulos[1]

[1]Pharmacology-Pharmacotechnology Division, Center for Clinical, Experimental Surgery and Translational Research, Biomedical Research Foundation of the Academy of Athens, Athens, Greece, [2]Proteomics Laboratory, Division of Biotechnology, Biomedical Research Foundation of the Academy of Athens, Athens, Greece

The goal of this study was the identification and quantification of membrane receptors (known and new) which are overexpressed in aggressive subtypes of Breast Cancer (BC) and Prostate Cancer (CaP), such as Triple Negative BC and castration resistant CaP. These novel membrane target receptors can be used either as biomarkers or for the design of natural products based drugs. In order to achieve this goal, we have developed and applied a GeLC-MS/MS based proteomic approach using specific BC and CaP subtypes.

For our study, three well characterized BC cell lines: HCC-1954 (HER2 overexpressing), MDA-MB-231 (TNBC), MCF-10A (benign control) and one CaP cell line: DU-145 (androgen-independent) were used. Our strategy included the combination of a subcellular fractionation and membrane enrichment protocol with the Discovery-based GeLC-MS/MS technique for the identification of key membrane receptor targets. The GeLC-MS/MS strategy was based on the initial separation of the isolated protein fractions by 1D-gel electrophoresis followed by In-Gel trypsin digestion and the subsequent Mass spectrometric peptide analysis using a high resolution Orbitrap mass analyser.

Through this approach, we succeeded to identify the most established receptors linked with BC, EGFR and HER2, as well as other potential protein targets in concordance with the literature (e.g. TFR1, EPHA2, GPCR5A). We further confirmed these results by Western blot and Immunofluorescence analysis.

Our results indicate that the strategy of developing and applying the GeLC-MS/MS technique into selected cell lines for the identification of new protein targets will add more information to the genetic and histological classification of the tumor as well as aim to the development of the most effective and targeted treatment of aggressive subtypes of cancer using either natural products or newly designed based drugs.

Keywords: GeLC-MS/MS, membrane receptors, cancer, drug discovery.

References:

[1] Tamvakopoulos C. Mass Spectrom Reviews 2007; 26 (3): 389–402.
[2] Katsila T, Siskos AP, Tamvakopoulos C. Mass Spectrom Reviews 2012; 31: 110–133.
[3] Limonta P, Marelli MM, Mai S, Motta M, Martini L, RM M. Endocrine Reviews 2012; 33(5): 784–811.

PS1-F-030

An optimized analytical methodology for the determination of olive oil biophenols, based on IOC recommended method

E. Bata[1], P. Stathopoulos[1], A. Rodi[2], M. Halabalaki[1], A-L Skaltsounis[1]

[1]Laboratory of Pharmacognosy and Natural Products Chemistry, Department of Pharmacy, University of Athens, Panepistimioupoli Zografou, 15771, Athens, Greece, [2]PharmaGnose S.A., Papathanasiou 24, 34100, Chalkida, Euboea, Greece

Olive oil is a complex and multifaceted mixture of compounds and a source of valuable nutrients. In 2012, the European Food Safety Authority (EFSA), assessing the biological properties of olive oil phenolic components issued a scientific opinion in favor of the specific health claim, pointing out that olive oil polyphenols contribute to the protection of blood lipids from oxidative damage (Regulation No 432/2012 of EC) [1]. Characteristic compounds of olive oil are hydroxytyrosol, tyrosol as well as the secoiridoids oleacein, oleocanthal, oleuropein and ligstroside aglycons. Many analytical methods have been proposed in literature for the determination of the phenolic components as well as the verification of the EFSA's health claim. However, the only method recognized by the International Olive Oil Council (IOC) is the COI / T.20 / Doc No 29 method by HPLC-DAD. Although the IOC method is robust, reproducible and suitable for laboratory workflow analysis, it suffers from strong co-elution phenomena related especially to secoiridoids, flavonoids and lignans resulting to inaccurate quantification. Here we purpose an optimized analytical method, based on the IOC recommended which improves significantly the quantification procedure. It is worth noting that in the suggested method an array of reference compounds have been used which enhance also the identification of olive oil biomarkers.

Acknowledgements: The present work was funded by the EU and project OliveNet-H2020-MSCA-RISE-2016. "The author would like to thank the Special Account for Research Grants and the National and Kapodistrian University of Athens for funding their participation in this meeting"

References:

[1] http://www.efsa.europa.eu/en/efsajournal/doc/2033.pdf

PS1-F-031

Development and validation of an HPTLC densitometric method for the rapid quantification of bioactive lignans in sesame seeds: comparison with HPLC-PDA

Eleni V. Mikropoulou, Eleftherios A. Petrakis, Aikaterini Argyropoulou, Sofia Mitakou, Maria Halabalaki and Leandros A. Skaltsounis

Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou 15771, Athens, Greece

Sesame seeds (*Sesamum indicum* L., Pedaliaceae) have been found to contain a group of bioactive compounds named lignans, which are well-known for their potent health-promoting properties. Sesamin and sesamolin, the major secondary metabolites of sesame seeds, have in recent years gained considerable attention due to their numerous biological properties [1]. In the present work, an HPTLC-based method was developed in compliance with the requirements of the European Pharmacopoeia [2] for the quantification of these two lignans in the methanolic extract of sesame seeds. A comparative study was simultaneously performed through HPLC-PDA for assessing the sesamin and sesamolin content of diverse samples. The results were subsequently subjected to statistical analysis in order to compare the methods as well as to investigate possible correlations. The methods were shown to be adequately correlated in terms of performance, as revealed by Pearson's rank correlation coefficient (>0.99 for sesamin and >0.98 for sesamolin) and Bland-Altman analysis (relative method bias 0.06-0.21, SD of bias 0.05-0.07). HPTLC densitometry is thus proposed as a valid and reliable tool for the rapid determination of the major lignans in sesame seed samples.

Acknowledgements: The authors wish to thank the European Directorate for the Quality of Medicines & Healthcare (EDQM) and especially Mrs. M. Bald & Dr. E. Reich for kindly providing the sesame seed samples. This research is implemented through IKY scholarships programme and co-financed by the European Union (European Social Fund - ESF) and Greek national funds through the action entitled "Reinforcement of Postdoctoral Researchers", in the framework of the Operational Programme "Human Resources Development Program, Education and Lifelong Learning" of the National Strategic Reference Framework (NSRF) 2014 – 2020.

Keywords: *Sesamum indicum*; sesame seed; sesamin; sesamolin; HPTLC densitometry; HPLC-PDA

References:

[1] Grougnet R, Magiatis P, Mitaku S, Terzis A, Tillequin F & Skaltsounis AL (2006). J Agric Food Chem 2006; 54(20): 7570–7574.
[2] Methods in pharmacognosy. Ph Eur 9.0 (2017); pp. 295–296.

PS1-F-032

Essential oil composition of *Juniperus oxycedrus* ssp. *macrocarpa* from Greece

Nikolaos Armenis, Maria Couladis

Section of Pharmacognosy and Chemistry of Natural Products, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, Athens 15771, Greece

The genus *Juniperus* L. comprises about 140 species many of which are cultivated as ornamental or for wood. These species are dioecious or monoecious evergreen shrubs or trees. The main genera of this family are *Cupressus, Thuja Chamaecyparis, Tetraclinis* and *Juniperus*. Essential oils of the genus Juniperus have been used since ancient times as perfumes, additives in cosmetics, insecticides, medicines and flavorings in food. Aerial parts of *Juniperus oxycedrus* ssp. *macrocarpa* were collected from the Greek islands Chryssi and Elafonissos. The essential oils obtained from different plant parts were analyzed by means of gas chromatography-mass spectrometry (GC-MS). Eighty seven components were identified from the female cones, representing 91.7% of the total oil. The major compound was α-pinene (30.5%), followed by α-cedrol (10.4%), myrcene (10.1%) and germacrene D (9.7%). One hundred thirteen components were identified, representing 97.3% of the total oil. The main constituent was α-pinene (28.2%), followed by manool oxide (12.5%) and α-cedrol (9.8%). One hundred and seven compounds were identified from the branches, amounting to 92.5% of the total oil. The major compounds were manool oxide (20.5%), α-cedrol (14.4%) and α-pinene (11.2%). Comparison of our results to the literature data showed mainly quantitative differences.

Acknowledgements: The authors thank the Special Account for Research Grants of the National and Kapodistrian University of Athens for funding to attend the conference.

Keywords: Juniperus; essential oil; GC-MS

PS1-F-033

Validation and implementation of an HPLC-DAD method for the simultaneous quantification of chlorogenic acid, flavonoids and flavonolignans in *Cecropia* species

Andrés Rivera-Mondragón[1], Sebastiaan Bijttebier[1], Tania Naessens[1], Deborah Custers[1], Catherina Caballero-George[2], Sandra Apers[1], Luc Pieters[1], Kenn Foubert[1]

[1]Natural Products & Food Research and Analysis (NatuRA), Department of Pharmaceutical Sciences, University of Antwerp, Universiteitsplein 1, 2610, Antwerp, Belgium, [2]Group of Pharmaceutical Research, Centre of Innovation and Technology Transfer, Institute of Scientific Research and High Technology Services (INDICASAT-AIP), Building 219, City of Knowledge, Panama, Republic of Panama.

Some plant species of the genus *Cecropia* (Urticaceae) are traditionally used in Latin America as anti-diabetic, anti-hypertensive and anti-inflammatory agents [1] and are commercially available as food supplements. However, until now there is a lack of quality control for these herbal products. An HPLC-DAD method was validated for the quantification of chlorogenic acid (CA), total flavonoids (TF) and flavonolignans (FL) following the ICH guidelines [2]. Fourteen authentic *Cecropia* leaf samples collected in Panama and three commercial products were analysed. The chemical composition of different species was compared by principal component analysis (PCA).

Validation parameters of the method for CA, TF and FL were satisfactory. Specificity was defined for each analyte. An adequate linear response was obtained for all analytical curves (r^2 >0.999). All relative standard deviations (%RSD) for repeatability and intermediate precision were lower than 2 and 5%, respectively. Accuracy, expressed as the recovery, varied from 98 to 102% for all concentration levels. The limit of detection and limit of quantification were on the scale of nanogram per milliliter (ng/mL). Analysis of the samples showed qualitative and quantitative differences in their chemical composition. Both analytical results and PCA revealed chemical similarities for *C. obtusifolia*, *C. peltata*, *C. insignis* and *C. hololeuca*, which contains mostly *C*-glycosyl flavones and *C*,*O*-glycosyl flavones. Additionally, *C. obtusifolia* differentiated from other species due to its relative high concentration of flavonolignans. Moreover, *C. hispidissima* showed to be different due to the high content of *O*-glycosyl-flavonols and low content of *C*-glycosyl flavones. This study serves as a useful tool for the quality control of herbal supplements of *Cecropia* species and the subsequent interpretation of their related pharmacological effects.

Acknowledgements: The authors would like to thank the National Secretariat for Science, Technology and Innovation (SENACYT) and the Institute for Training and Development of Human Resources (IFARHU) of the Republic of Panama for acknowledging a scholarship to Mr. Rivera to carry out his doctoral studies (scholarship No. 670 and resolution No. 270-2015-025).

Keywords: *Cecropia* species, chlorogenic acid, flavonoids, flavonolignans, validation, HPLC-DAD

References:

[1] Rivera-Mondragón A, Ortiz O, Bijttebier S, Vlietinck A, Apers S, Pieters L, Caballero-George C. Pharm Biol 2017; 55: 1500–1512

[2] International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use, Validation of Analytical Procedures: Text and Methodology Q (R1). 200

PS1-F-034

Cannabis-related Bioanalysis Applications Using a Compact Mass Spectrometer (CMS)

Ben Nie[1], Jack D Henion[1], George Maylin[2]

Advion Inc., Ithaca, NY[1]
New York Equine Drug Testing and Research Program, Morrisville State College, Morrisville, NY[2]

The legalization of marijuana and hemp provides commercial opportunities as well as analytical challenges. Accurate and precise quantitative analysis of plant and medicinal products is needed in this new industry to document composition and assure safety of cannabis products. LC/MS techniques can provide unparalleled selective and sensitive measurements for these purposes. Here we describe the use of a relatively inexpensive compact mass spectrometer for SIM LC/MS analysis of cannabis- related applications.

PS1-G-001

Gasca D as potent diabetic weight and blood glucose reduction formulation

Salihu Ismail Ibrahim[1], Gezawa Ibrahim[2], Fakhradden Yahya M[3], Chedi Basir[4], Nafisatu Kabir[5], A A Imam[6]

[1]Department of Biochemistry, Federal University Dutse, Dutse, Nigeria, [2]Department of Medicine, Aminu Kano Teaching hospital, KANO, Nigeria, [3]Department of Medicine, Abdullahi Wase Specialist Hospital, KANO, Nigeria, [4]Department of Pharmacology, Bayero University Kano, KANO, Nigeria, [5]Department of Biochemistry, Federal University Dutse, Dutse, Nigeria, [6]Department of Biochemistry, Bayero University Kano, KANO, Nigeria

Research in the treatment of diabetes has centered on attempts to normalize fasting and postprandial blood glucose levels by aggressively controlling hyperglycemia. However, intensive glycemic control is not without risks, such as hypoglycemia, weight gain and possible cardiovascular events and mortality in high-risk individuals. This constitutes the major force for finding alternatives, mainly from plant kingdom that are of less severe or even no side effects.

Gasca D is a herbal derived formulation organically synthesized from the combination of *Adansonia digitata*, *Gum acacia* and *Hyphaenae thebaica* for the management of diabetic condition, the formulation was found to be safe up to a dose of 5,000mg/kg bw and the rats did not show any sign of morbidity or mortality 24 days after acute administration. The level of some toxic heavy metals (Cadmium, Arsenic, Mercury, and Lead) and essential elements were determined using microwave plasma-atomic emission spectrometry (MP-AES) and the result showed no traces of the toxic heavy metals in the formulation. Iron was found to have highest concentration among the essential elements 67.16 + 7.5 µg/g followed by Manganese 7.72 + 0.9µg/g while chromium had the lowest concentration of 0.72 + 0.04 µg/g. A very important element of the transition metals whose organic derivatives have been found to have insulin-mimetic and antidiabetic properties is Vanadium with a concentration of 2.6 + 0.7µg/g has been quantitatively determined in Gasca D herbal formulation. GC-MS analysis of Gasca D on GC-MS Agilent 6890N series gas chromatograph (Agilent, Santa Clara, CA), revealed the presence of 14 biologically active compounds which include *N*-Formyl-beta-alanine, Paromomycin, 3,4-Altrosan, Benzamide, 1,3,4-Thiadiazol-2-amine, Carbamodithioic acid, Carbonic acid, alpha-D-Glucopyranoside, Ethyl isocyanide, 2-Propanesulfinic acid, Propanamide, 2-Butenenitrile, Dicyclopropyl carbinol, Isoxazolidine, 1,5-Hexadiene 10-Azido-1-decanethiol. Gasca D herbal formulation can be useful in the management of diabetes.

PS1-G-002

Effective HPLC methods for the detection of *Sophora japonica* adulteration in *Ginkgo biloba* leaf extracts

Evangelia Bampali[1], Zarko Kulic[1], Stefan Germer[2]

[1]Preclinical Research, Dr Willmar Schwabe GmbH & Co. KG, Karlsruhe, Germany, [2]Pharmaceutical Development, Dr Willmar Schwabe GmbH & Co. KG, Karlsruhe, Germany

Background: *Ginkgo biloba* L. (Ginkgoaceae) leaf is the raw material of widely used herbal medicinal products [1]. Being one of the most widely sold medicinal plants; it is of significant economic value. There have been reports claiming poor quality and adulteration (either accidental or intentional and economically motivated) of Ginkgo leaf extracts with *Sophora japonica* fruit, which is rich in flavone glycoside content [1, 2].
Aims: Our key aim is to investigate whether *Ginkgo biloba* genuinely contains genistein and its glucoside sophoricoside, since they can be used as marker substances to detect *Sophora japonica* adulteration in *G. biloba* extracts.
Methods: 33 samples of dried *Ginkgo biloba* leaves were sourced from Dr Willmar Schwabe ginkgo plantations in China (n=12), USA (n=13), and France (n=8). After extraction, the samples were analyzed using two HPLC methods (for genistein's and sophoricoside's investigation, respectively). Chromatograms were compared to three standard reference materials: genistein, sophoricoside and apigenin.
Results and Conclusion: All of the *G. biloba* samples tested in this study were found to contain neither genistein nor sophoricoside. Their presence in commercially available *Ginkgo biloba* products, thus always indicates an (intentional) adulteration with *S. japonica*. The new HPLC method for the detection of genistein (which has been claimed to be a genuine constituent in Ginkgo leaves [3]), combined with high-resolution mass spectrometry was able to discriminate genistein from apigenin (a native constituent of Ginkgo leaves, which is present in traces). Furthermore, the method for the detection of sophoricoside allows easily identifying the adulteration with *Sophorae Fructus* without prior hydrolysis. In addition, both new HPLC methods are able to detect an adulteration of ≥ 2% *S. japonica* in *G. biloba* extract. Consequently, the new methods will detect adulterations with *S. japonica* without any doubt which is important to unanimously distinguish regular and adulterated Ginkgo extracts.
Keywords: *Ginkgo*, genistein, sophoricoside, adulteration, marker compounds, *Sophora japonica*

References:
[1] Wohlmuth H *et al*., Phytomedicine 2014; 6: 912–8.
[2] Booker A *et al*. J Herb Med 2016; 6: 79–87.
[3] Yao JB *et al*. Nat Prod Commun 2017; 12(8): 1241–1244.

PS1-G-003

Hepatoprotective potentials of *Picralima nitida* against *in vivo* carbon tetrachloride-mediated hepatotoxicity

Macdonald Idu[1], Oghale Ovuakporie-Uvo[2], Gerald Eze[3]

[1] Department of Plant Biology and Biotechnology, University of Benin, PMB 1154, Benin City, Edo State, Nigeria, [2]Department of Plant Biology and Biotechnology, University of Benin, PMB 1154, Benin City, Edo State, Nigeria, [3]Department of Anatomy, University of Benin, PMB 1154, Benin City, Edo State, Nigeria

Picralima nitida when administered orally has a wide broad spectrum in the management of several systemic diseases in African folk medicine. This research was aimed at investigating the *in vivo* Carbon tetrachloride (CCl_4) - mediated hepatotoxicity of methanolic seed extract of *Picralima nitida* (*P. nitida*). Twenty five (25) Wistar rats randomly selected into five groups of five animals were used in this research. Group 1 was administered Normal saline (Negative control); Group II was administered 1 ml of Carbon tetrachloride only (Positive control/ Reference drug); Group III- 10 ml *P. nitida* extract + 1 ml Carbon tetrachloride; Group IV- 100 ml *P. nitida* extract + 1 ml Carbon tetrachloride while Group V received 1000 ml *P. nitida* extract + 1 ml Carbon tetrachloride. Results show that treatment with *P. nitida* extract had no adverse effect on the body weight of Wistar rats. Biochemical analyses show increase in antioxidant agents; CAT and GSH which are positive responses to treatment. Photomicrographs display moderate amelioration from steatosis caused by Carbon tetrachloride in the treatment groups. It is recommended that further studies be carried out to ascertain possible total reverse fat degeneration of liver cells induced by Carbon tetrachloride.

Keywords: *Picralima nitida*, Hepatotoxicity, CCl_4

PS1-G-004

Correlating carob pods (*Ceratonia Siliqua* L.) aroma profile with geographical origin

Andriana Krokou[1], Agapios Agapiou[1], Marinos Stylianou[1], Rebecca Kokkinofta[2]

[1]Department of Chemistry, University of Cyprus, Nicosia 1678, Cyprus, [2]State General Laboratory, Nicosia 2081, Cyprus

Carob is an evergreen long-lived drought-tolerant tree widely cultivated mostly in the eastern Mediterranean region and later distributed across the globe (e.g. Australia, USA). It is well known for the sweetness of its fruit and the related traditional food products, as well as for its health benefits (multi-purpose food crop). A strong persistent and characteristic odor associates not only carob fruit, but also its food and beverages products. In a pilot study, Solid Phase Microextraction/Gas Chromatography-Mass Spectrometry (SPME/GC-MS) analysis was performed in grounded carob pods from 9 different cultivars in order to decode the released aroma. The method was applied to *Ceratonia Siliqua* L. cultivars from Cyprus (Kountourka, Koumpota, Tilliria), Spain (Negra, Rojal, Matalafera) and Italy (Saccarata, Racemosa, Gibiliana). The most abundant Volatile Organic Compounds (VOCs) evolved from the grounded carob pods were propanoic acid-2-methyl, acetic acid, butanoic acid, furfural and propanal, 2-methyl. The detected VOCs peaks were integrated and the results were further analyzed with multivariate data analysis, which included Principal Component Analysis (PCA), Hierarchical Analysis (HA) and Partial Least Squares-Discriminant Analysis (PLS-DA). PCA showed great similarity between cultivars from the same country, HA classified carob samples in clusters supporting discrimination based on geographic origin and PLS-DA technique distinguished the samples according to their origin and revealed the characteristic compounds for every origin. In addition, PLS discriminant analysis (with "leave-one-out cross-validation") showed that the performed analytical method can distinguish carobs according to their origin with 100% correct classification. It appears that VOCs determination can be very helpful for identifying the unique geological and climatic conditions existing in the island of Cyprus that lead to an authentic product. More carob samples from varieties cultivated in other countries are needed in order to strengthen the results of the method output.

Acknowledgements: The research was financially supported by UCY "Black Gold" project.

PS1-G-005

Combinatory chromatographic and spectroscopic methods-approach for honeys botanical origin authentication based on the volatile fractions

Ewa Makowicz[1], Izabela Jasicka-Misiak[1], Dariusz Teper[3], Paweł Kafarski[2]

[1]Deparmtment of Analytical and Ecological Chemistry, Faculty of Chemistry, Opole University, Opole, Poland, [2]Department of Bioorganic Chemistry, Faculty of Chemistry, Wrocław University of Science and Technology, Wroclaw, Poland, [3]Research Institute of Horticulture, Apiculture Division, Puławy, Poland

Honey is natural food product made by honeybee (*Apis melifera*) from plant, honeydew or both. It is used not only in food industry but also is popular product in the cosmetics as well as medical industry. Honey is a complex mixture of a variety of different chemical classes of compounds. The chemical composition of honeys is very variable and significantly dependent on huge number of varied factors, such as botanical origin, geographical area of origin, bee species, honey processing, age of honey and storage method. Alongside with constantly increasing interest of consumers of natural and healthy food products, growing popularity of bee products is also observed, mostly honeys. In addition to flavor and nutritional values, therapeutic effect in one of the most valuable qualities of honey.

The purpose of presented study was evaluation of volatiles chemical content of honeys of different botanical origin, which are characterized by high flavour and nutritional values and construction of chemical profiles based on the different extraction methods. For extracts analysis GC-MS and ^{1}H NMR measurements were performed. Moreover, for analysis of volatiles extracts of honeys HPTLC analysis was applied. This method allows us to construct something like cod-bars useful to differentiate honeys of different botanical origin.

Additionally, based on the obtained results we could determine and identify the markers, for different unifloral honeys and show that combination of those three techniques create a specific fingerprint of honeys with different botanical origin and that those methods are powerful techniques for differentiation origin of honeys what was confirmed by use of chemometric approaches.

Acknowledgement: This study was carried out with financial support from the National Science Centre in Poland project No. 2014/15/B/NZ9/02182 "Chemical markers of unifloral honeys"

Keywords: volatile compounds, chemical profiles, HPTLC, GC-MS, unifloral honeys

PS1-G-006

HPTLC – fast and accurate technique for determination of botanical origin of honeys based on the fluorescent compounds

Ewa Makowicz[1], Izabela Jasicka-Misiak[1], Paweł Kafarski[2]

[1]Deparmtment of Analytical and Ecological Chemistry, Faculty of Chemistry, Opole University, Opole, Poland, [2]Department of Bioorganic Chemistry, Faculty of Chemistry, Wrocław University of Science and Technology, Wroclaw, Poland

Honey is a natural, sweet, aromatic and complex food product produced by honey bees (*Apis melifera*). Valuable nutritional and therapeutic properties of honeys are resulting from presence of rich cocktail of organic compounds derived from flower nectar or honeydew.
Unfortunately, it is very disturbing that honey is one of the most common falsified food products. This dealing is done by admix honeys with glucose-fructose syrup or by mixing honey with cheaper and inferior quality honeys. All this leads to a significant decrease of the unique qualities of honey; moreover, methods commonly used to control its quality are not sufficient.
The aim of presented study was to used HPTLC (high performance thin layer chromatography) technique as a fast method for differentiation of honey of different botanical origin based on the analysis of obtained extracts by HPTLC method at a wavelength of 366 nm and 254 nm, using both whole honeys (water extracts) and certain honey extracts. For separation purposes, various techniques were used: ultrasound-assisted extraction (USE), solid-phase extraction (SPE) and liquid-liquid extraction. For each of these methods, optimization of different parameters was performed. Next, in order to construct profiles of fluorescence compounds that can create a unique fingerprint for selected honeys HPTLC were performed.
The obtained results along with chemometric analysis show that HPTLC is a very sensitive visual method for the rapid and precise differentiation of honeys of different botanical origins, since they generate bar-code-like images. This is because the obtained extracts by different extraction procedures create a specific pattern of separated compounds for each type of monofloral honey.

Acknowledgement: This study was carried out with financial support from the National Science Centre in Poland project No. 2017/25/N/NZ9/00623 "Fluorescence compounds as specific markers of Polish honeys with different botanical origin"

Keywords: HPTLC, fluorescent compounds, honey, honey authenticity

PS1-G-007

HPTLC fingerprints as useful tools for profiling of honeys to determine authenticity

Natalia Stanek[1], Izabela Jasicka–Misiak[1], Paweł Kafarski[2]

[1]Faculty of Chemistry, University of Opole, Oleska 48, 45–051 Opole, Poland, [2]Faculty of Chemistry, Wrocław University of Science and Technology, wyb. Wyspiańskiego 27, 50–370 Wrocław, Poland

Phenolic compounds are one of the most important compounds occurring in honeys. They exhibit many biological effects, for example: antioxidant, antibacterial, anti–inflammatory, anti–allergic activities. Analysis of phenolic compounds is a very promising way of determining floral and geographical origins of honeys. In recent decades, there have been implemented different methods for the authentication of honey. This study presents the phenolic profiles of various honey samples using high performance thin layer chromatography (HPTLC) as a detection method. Although, HPLC remains the most dominant techniques for the separation and identification of phenolic compounds, HPTLC seems to be a very promising alternative to the analysis of these compounds in honeys. This method is simpler, more flexible, more accessible, and cheaper than other commonly used methods. Furthermore, HPTLC allows a parallel separation and quantitative determination of many samples at the same time [1]. We used HPTLC fingerprint analysis to look for a characteristic pattern of honey samples (willow, buckwheat, heather, pine honeydew and manuka honey). HPTLC chromatograms determine the differences in botanical origin of honey samples and show common bands. The entire chromatogram is treated as a unique, distinctive fingerprint. Therefore, the HPTLC fingerprint characteristic for the selected type of honey could thus complement quality–control tool for the honeys [2].

Acknowledgements: This work was supported by The National Science Centre of Poland in the frame of Grant NO: 2014/15/B/N29/02182.

Keywords: HPTLC fingerprint, honey, phenolic compounds, high–performance thin–layer chromatography

References:

[1] Ristivojević P, Trifković J, Vovk I, Milojković–Opsenica D. Talanta 2017; 72–79.
[2] Stanek N, Jasicka–Misiak I. Food Anal Methods 2018

PS1-G-008

Characterization of *Abies Sibirica* L. needle polyprenols and safety of their liposome product in marathoners

Ilona Vanaga[1,2], Ilva Nakurte[3], Sandra Rozenstoka[4], Simona Steina[4], Dace Suna[4], Ugis Kletnieks[2], Baiba Jansone[1]

[1]University Of Latvia, Faculty of Medicine, Riga, Latvia, [2]Silv EXPO, SIA, Riga, Latvia, [3]University Of Latvia, Faculty of Chemistry, Riga, Latvia, [4]Sporta laboratorija, SIA, Riga, Latvia

Polyprenols (PPs) are long–chain linear polymers consisting of isoprene units that are abundant in conifers. It has been shown in both animal model and patients that PPs act as protectors of atorvastatin–induced muscle weakness (Jansone *et al.*, 2016; Latkovskis *et al.*, 2016). PPs also improve oxygen consumption efficiency and increase high intensity load tolerance in well–trained amateur floorball players. There is no data, however, on the PP influence on the performance of marathon and half marathon runners.

Study objective was to investigate the structure of *Abies Sibirica* L. PPs and to examine safety aspects of PP product's use in marathoners.

PPs (purity ~ 80%) were extracted from the Siberian spruce (*Abies Sibirica* L.) needles using 2–step CO_2 extraction. Molecular structure and quantification of PPs was done by UHPLC–DAD method. PP liposomes were prepared via co–solubilisation and modified ethanol injection of PPs in an ethanol–phospholipid system (ratio of ~ 1:17 w/w). The resulting multilamellar liposomes (vesicle size ~ 1.36 μm) were further encapsulated into soft gelatin capsules (1 capsule contains 30 mg of PPs). A total of 22 runners from Latvia, aged 25–42 years, were divided into experimental (16 men that took 30 mg of PPs 3 times/day for 4±1 days before the run) and control (6 men) groups. Several blood biochemical parameters were determined before and after the use of PPs.

The obtained Siberian spruce needle PPs consisted of a mixture of PP homologues with isoprene unit number n = 11 to 20 (from $C_{55}H_{90}O$ to $C_{100}H_{162}O$), but mainly from Pren14 to Pren17 (from $C_{70}H_{114}O$ to $C_{85}H_{138}O$). The use of PP liposome softgel caps was deemed safe, as they did not significantly or adversely influence the full bloodwork results or blood biochemistry parameters (CFK, AST, GGT, Mb, IL–2,6, TNF–α etc.) of the marathoners.

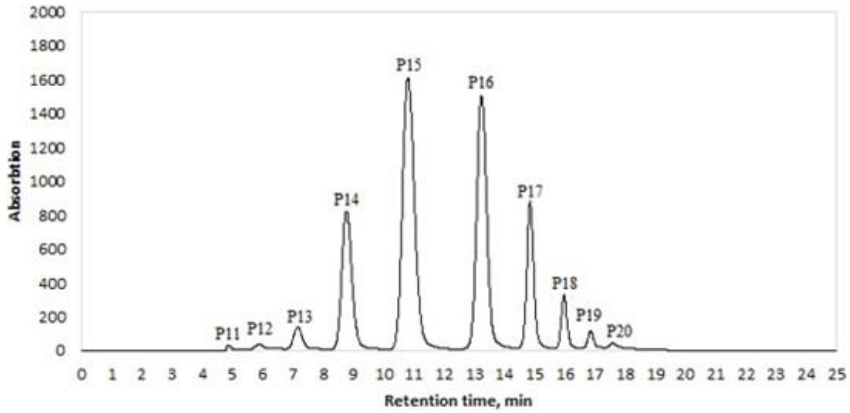


Fig. 1. UHPLC-DAD chromatogram of extracted polyprenols from Siberian fir *(Abies sibirica)*

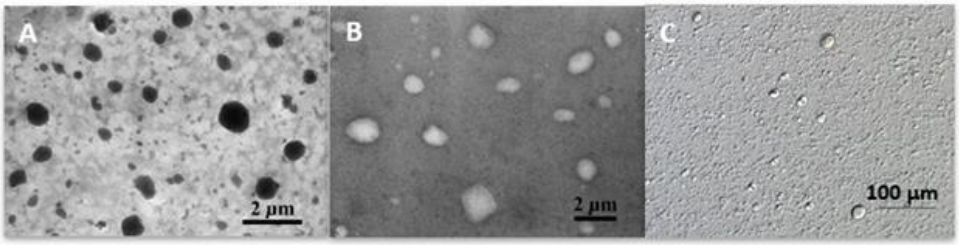


Fig. 2. Images of polyprenol liposomes by TEM (A – positive staining and B – negative staining) and light microscope (C)

Keywords: polyprenols, *Abies Sibirica* L., liposomes, marathoners

PS1-G-010

Identification of quality marker compounds in Greek EVOOs using integrated LC-HRMS & FIA FTICR MS platforms and chemometrics

Theodora Nikou[1], Matthias Witt[2], Panagiotis Stathopoulos[1], Aiko Barsch[2], Leandros A Skaltsounis[1], Maria Halabalaki[1]

[1]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapostrian University of Athens, Panepistimiopolis Zofrafou, 15771, Athens, Greece, [2]Bruker Daltonik GmbH, Bremen, Germany

Extra virgin olive oil (EVOO) consumption has globally increased due to its superior nutritional and sensory properties. In combination with its importance for European Union's economy [1], it has been established as a product of high economic priority and the need for its quality and authenticity control is of outmost importance. Its chemical complexity and variability enhances the hassle in investigating the most suitable methodology and consequently numerous analytical methods have been suggested [2]. However, a reliable methodology to ensure authenticity and quality of EVOO is still unavailable. In this study, Fourier Transform High Resolution Mass Spectrometry (FTHRMS) techniques were integrated for metabolomics analysis of Greek EVOO and their corresponding biophenolic extracts in order to reveal the distinctive metabolites of significant quality parameters of EVOO production. In particular, Ultra High Performance Liquid Chromatography coupled with orbitrap analyser (UPLC-Orbitrap-MS) and Fourier Transform Ion Cyclotron Resonance Mass Spectrometer (FT-ICR-MS) using Flow Injection Analysis (FIA) method were incorporated providing novel data for EVOO chemical discrepancy and classification. More than 300 EVOO samples were collected from the main Greek olive oil producing regions, for two harvesting years. After pre-treatment, data were subjected to multivariate data analysis (MDA), assisted by advanced chemometric tools, for clustering consideration and identification of certain quality markers. Until now, more than 20 metabolites from various chemical classes have been identified as statistical significant biomarkers for selected discriminant factors in the EVOO and their biophenolic extracts. To our knowledge this is the first time that two FT MS platforms combining LC and FIA methods were integrated, analysing both lipophylic and polyphenolic components, to give solutions to quality control aspects of EVOO.

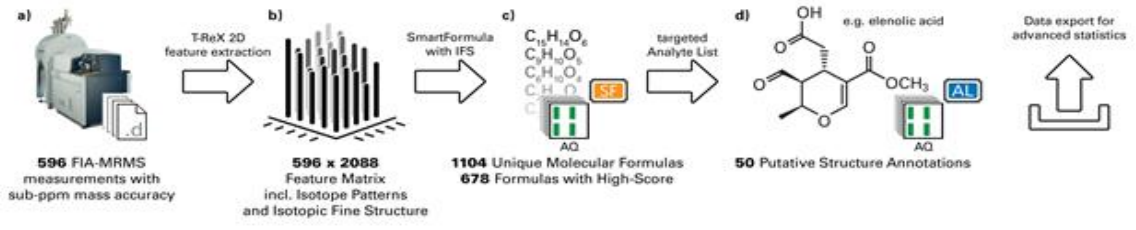
Acknowledgements: The author would like to thank IKY institute for the financial support and the Greek olive oil producers for their kind offer of EVOO samples.

Keywords: EVOO, biophenols, quality control, FIA-FT-ICR-MS, UPLC-Orbitrap-MS, biomarkers, authentication.

References:

[1] Carrasco-Pancorbo A, Cerretani L, Bendini A, Segura-Carretero A, Gallina-Toschi T, Fernndez-Gutirrez A. J. Sep. Sci 2005; 28: 837–858

[2] Kalogeropoulos N, Tsimidou M. Antioxidants 2004; 3: 387-413.



PS1-G-011

Isotopic traceability (^{13}C and ^{18}O) of Greek olive oils

Karalis P.[1,*], Halabalaki M. [2], Skaltsounis L.A.[2], Diamantopoulos G.[1], Dotsika E.[1]

[1] Laboratory of Stable Isotopes and Radiocarbon, Institute of Nanoscience & Nanotechnology, NCSR "Demokritos", 153 10, Greece
[2] Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece
**Corresponding author: p.karalis@inn.demokritos.gr*

Olive oil is one of the most important ingredients in the Mediterranean diet due to its nutritional value but also to organoleptic properties (taste, flavorand odor) that it presents. Its quality is related to the environmental conditions (temperature, precipitation, altitude) in which the olive tree was cultivated. Two regulations of the European Community, No. 2081/91 and 2082/91 (Denomination of Protect Origin) have been issued in order to safeguard the quality of olive oils originating from areas particularly suitable for olive growing due to the better organoleptic characteristics. As a result, there is an increased consumer awareness and interest in products with specific geographical origin and varieties as this information are related to the quality and authenticity.Thehigh demand for olive oil with such quality and authenticity characteristics in addition with the high cost of production leads producers to adulterate olive oil by using mixtures of lower quality oils or olive oils from different countries/areas.
To protect the authenticity of high nutritional food, like olive oil, the isotope ratio mass spectrometry (IRMS) analytical method has been introduced. The carbon isotope ratio $^{13}C/^{12}C$ (expressed as $\delta^{13}C$) reflects the different varieties and the oxygen isotope ratio $^{18}O/^{16}O$ (expressed as $\delta^{18}O$) reflects the water sources of the plant (usually rain water) with minor deviations due to the contribution of atmosphere oxygen O_2 and CO_2. Thus the $\delta^{18}O$ of local water is related to the climatic and geographical features of the region (mean temperature, precipitation, relative humidity of rain) while the $\delta^{13}C$ is related to the virginity/quality of the samples. Consequently, the combine isotopes of ^{13}C and ^{18}O can contribute to information related to the geographical origin and quality of olive oil.
In order to investigate the geographical origin and qualityof oil samples, measurements of $\delta^{13}C$and $\delta^{18}O$of the whole oil and some of its fractions have been performed on samples coming from fruits of *Olea europaea* produced in Greece. The results after statistical analysis provide evidence that the oil samples tend to cluster according to the different climatic areas of growing environment. Some overlapping has been observed for samples coming from neighboring countries with similar climates.

PS1-G-012

Isotopic model for detecting original wine vinegar

Diamantopoulos G.[1,*], Karalis P.[1], Gougoura S.[1], Palaigeorgiou E.[1], Dotsika E.[1]

[1] *Laboratory of Stable Isotopes and Radiocarbon, Institute of Nanoscience & Nanotechnology, NCSR "Demokritos", 153 10, Greece*
** corresponding author: p.karalis@inn.demokritos.gr*

The stable isotope ratios of bio-elements have been used for many years for the authentication of different food but only the last years are used in the case of vinegar. The applications of stable isotope analysis of the hydrogen and carbon isotope ($D/^{1}H$, ^{13}C, ^{14}C isotope) of acetic acid are used to identify synthetic vinegars and distinguish the three biological cycles that plants follows in order to incorporate CO_2 during photosynthesis (C3, C4 and CAM cycles) thus allowing detection of vinegars adulterated with raw fermentation materials cheaper than those declared on the label and no complying with the regulations of the European Union.
In addition, the oxygen stable isotope ratio $^{18}O/^{16}O$ (expressed as $\delta^{18}O$) of vegetal water, determined by IRMS, is used in order to distinguish wine vinegar from vinegars made from dried grapes. These analysis is very important because according to European Regulations, wine vinegar is the product obtained exclusively from the acetous fermentation of wine, which is in turn defined as the product exclusively obtained from the alcoholic fermentation of fresh grapes, whether crushed or not, or of grape must (EC 479/2008). So, the "raisin vinegar", commonly produced by fermenting dried grapes and rehydrating with tap water, is not considered as "wine vinegar".
We performed isotopic analysis of 120 different brands labeled as "wine vinegars" originating fromthe Greek market and according to the analysis; more than 80% of the 120 brands were shown not to be authentic, but rather obtained by diluting a concentrated source such as dried grapes with water and alcoholic addition.

PS1-H-001

Phenolic content of the fruit of *Ceratonia siliqua* L. (Carob) determined by HPLC-DAD-MS analysis

S Schwaiger[1], Alexandru Vasincu[2], Christiana Neophytou[3], S Aigner[1], Hermann Stuppner[1], Andreas Constantinou[3]

[1]Institute of Pharmacy/Pharmacognosy, University of Innsbruck, Innsbruck, Austria, [2]Department of Pharmacodynamics and Clinical Pharmacy, "Grigore T. Popa" University of Medicine and Pharmacy, Faculty of Pharmacy, Iaşi, Romania, [3]Department of Biological Sciences, University of Cyprus, Nicosia, Cyprus

Introduction: Various components of the carob fruit have shown health benefits. Our objective is to compare and contrast the phenolic content of extracts obtained from different parts of the carob varieties that are common in Cyprus.
Methods: Extracts were prepared from seeds, pods and the whole fruit (consisting of a mixture of seeds and pods). For the preparation of extracts from pods, the seeds were removed mechanically and the former were extracted with Diethyl Ether (DE) in Soxhlet, followed by Ethyl Acetate (EAc). For the preparation of extracts from seeds and for the mixture of seeds and pods the following procedure was followed: dried and powdered vegetal material was extracted with DE at ambient temperature and the vegetal residue was extracted with EAc. All extracts were dissolved in methanol and analyzed either with an HPLC-DAD-System or LC-MS-System.
Results: Large differences were detected between the seeds, the pods and the mixtures, both with respect the quantity and quality of phenolic compounds. The HPL-chromatograms of mixtures showed two major peaks with retention times (RT) of 6.2 and 15.4, representing about 61% of the DE extract and about 49% of the EAc extract. The major peak at RT 6.2 was identified as gallic acid, while the compound with RT 15.4 was identified as tricin. Extracts from seeds were very poor in phenolic content while the extracts from pods contained phenolic products, but (25X) less than the extracts from mixtures. The content of Gallic acid was determined to 2.9% (w/w) in the DE extract and 0.75 (w/w) in the EAc extract.
Conclusions: The differences in phenolic compounds could be due to different extraction methods used for extracting the seeds and the whole fruit (mixture) and the methods used for extracting the pods.

Keywords: *Ceratonia siliqua*, phenolic content, gallic acid.

PS1-H-002

Innovative extraction techniques for the recovery of phenolic compounds from olive oil by-products

Sofia Chanioti, Paraskevi Siamandoura, Constantina Tzia

NTUA, Athens, Greece

Olive oil production is an important agricultural activity and one of the primary driving engines of the economy of Greece. Olive pomace and olive leaves are the by-products of the olive oil production process. Olive pomace and leaves can reach up to 30% and 10% of the total weight of olives arriving to the mill, respectively. It is well known that these residues are rich in phenolic compounds such as oleuropein, hydroxytyrosol and rutin, which have shown antiviral, antimicrobial, antioxidant, anti-inflammatory and anti-carcinogenic activities. Therefore, ways to valorize these by-products are needed and most welcome.

Different extraction techniques have been used to extract bioactives from olive pomace and leaves; among them, conventional extraction with ethanol:water. Nowadays, new assisted extractions methods using homogenization, microwave heating, ultrasound and high hydrostatic pressure are currently investigated in the extraction of phenolic compounds from various plant resources. These methods can offer high reproducibility in shorter time, simplified manipulation, reduced solvent consumption and lower energy input without decreasing the extraction yield of the target species.

The aim of this study was to investigate the effect of the innovative extraction assisted methods: homogenization, microwaves, ultrasounds, and high hydrostatic pressure, for the extraction of phenolic compounds from olive pomace and olive leaves by using different processing parameters (temperature, time etc.) and various solvent systems such as ethanol:water, water and natural deep eutectic solvents (NADESs). The antioxidant potential of extracts prepared from olive pomace and leaves in terms of their total phenolic content and their antioxidant radical scavenging was evaluated. Moreover, the simultaneous determination of individual polyphenols of extracts by HPLC was performed. The proposed methods represent excellent environmental friendly alternatives for the extraction and quantification of phenolic compounds in olive oil by-products. The results of the present study may be helpful to further exploit and utilize these resources.

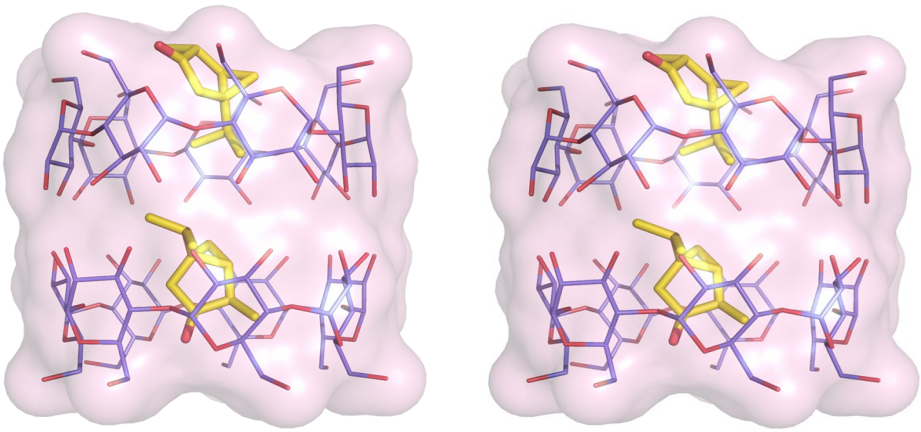
PS1-H-003

Inclusion of borneol, camphor and thujone monoterpens from sage infusion in native α- and β-cyclodextrin

Nefeli Sofia Sotiropoulou[1], Katerina Fourtaka[2], Elias Christoforides[2], Christos Pappas[1], Kostas Bethanis[2], Petros Tarantilis[1]

[1]Laboratory of Chemistry, Department of Food Science and Nutrition, Agricultural University of Athens, Iera Odos 75, 118 55, Athens, Greece, [2]Physics Laboratory, Department of Biotechnology, Iera Odos 75, 118 55, Athens, Greece

Sage consumed widely as an infusion. Infusion is a greener, flexible and cost effective extract. Borneol, camphor and thujone found in abundance in sage's infusion possess beneficial for health properties however potential toxicity has also been reported [1]. A quick, safe and economical way to control their toxicity in infusions is via their encapsulation in Cyclodextrins (CDs) which have the unique ability to act as molecular containers by entrapping guest molecules in their cavity. The removal of borneol, camphor and thujone from the infusion was quantified by GC-MS before and after infusion being processed with CDs. According to the results, the concentrations of the three compounds were significantly lowered although the sage infusions retained their volatile profile. In addition, phase solubility studies, single-crystal X-ray analysis and Molecular Dynamics (MDs) simulations were performed, giving a detail description of the crystal structure, the host-guest interactions, the dynamic behavior and the binding affinity of the examined inclusion complexes. The crystal structures of thujone in α- and β-CD presented in this study, complement the structural studies of borneol and camphor inclusion complexes in α- and β-CDs that have been previously determined [2] and reveal the formation of stable CD dimers encapsulating one and two thujone molecules respectively. Phase solubility and MD studies showed that these inclusion complexes are also stable in aqueous solution with relatively high binding affinities.



Acknowledgements: This research has been supported by the Anthir A.B.E.E., Medicinal and Aromatic Plants (Agrinion, Greece).

Keywords: Sage infusion, borneol, camphor, thujone, α- & β-cyclodextrin, GC-MS, X-ray crystallography

References:

[1] EFSA 2009; 7.
[2] Christoforides E, Mentzafos D, Bethanis K. J Incl Phenom Macrocycl Chem 2015; 81: 193-203.

PS1-H-004

HPLC-DAD-ESI-TOF-MS analysis and antiulcer effects of some extracts from *Vernonia kotschyana* Sch. Bip. ex Walp.

Alexandru Vasincu[1], Christiana Neophytou[2], Luca-Simon Vlad[3], Anca Miron[3], Ioana-Mirela Vasincu[4], Bogdan-Gabriel Şlencu[5], Daniela Carmen Ababei[1], Oana-Dana Arcan[1], Delia Bulea[1], Monica Neamţu[1], Veronica Bild[1]

[1]Discipline of Pharmacodynamics and Clinical Pharmacy, University of Medicine and Pharmacy "Grigore T. Popa" Iasi, Romania, [2]Department of Biological Sciences, University Of Cyprus, Nicosia, Cyprus, [3]Discipline of Pharmacognosy, University of Medicine and Pharmacy "Grigore T. Popa" Iasi, Romania, [4]Discipline of Pharmaceutical Chemistry, University of Medicine and Pharmacy "Grigore T. Popa" Iasi, Romania, [5]Discipline of Environmental and Food Chemistry, University of Medicine and Pharmacy "Grigore T. Popa" Iasi, Romania

Introduction. *Vernonia kotschyana* Sch. Bip. ex Walp is used to treat a number of ailments across Central and West Africa. In traditional medicine of Mali, the plant is highly valued to treat gastric ulcer disease and wounds. Our objective was to investigate the phenolic and saponin profile and antiulcer effect of two extracts obtained from the roots of this African plant.
Material and methods. Ethyl acetate (V-EA) and aqueous (V-A) extracts were analyzed on a Phenomenex Gemini C18 column. Gradient elution was adopted using H_2O/acetonitrile with 0.1% formic acid. ESI-TOF-MS analysis was performed in positive ionization mode. Antiulcer effect was *in vivo* evaluated using indomethacin as ulcerogenic agent. Extracts were administrated orally in the following sequence: 200, 400 and 800 mg/kg body weight. Experiments were conducted in accordance with the international bioethic rules.
Results and discussions. HPLC-DAD-ESI-TOF-MS profiling of *V. kostchyana* extracts revealed the presence of phenolic acids, mostly derivatives of caffeoyl- and dicaffeoylquinic acids. V-EA contained additionally feruloylcaffeoyl- and tricaffeoylquinic acids. Moreover, ten rare stimagastane-type steroid saponins, such as vernoniosides D1–D3, vernoniosides F1–F2 and vernocuminosides I–J were tentatively identified in both extracts. V-EA presented a marked inhibition of ulcers, starting with a dose of 400 mg/kg bw and a maintenance of effect for interval 400-800 mg/kg bw. V-A (800 mg/kg bw) decreased the damage score by 64.5%. Antiulcer activity was also partially confirmed by histologic analysis of gastric tissues.
Conclusions. A number of compounds with potential biological activity were found in the obtained extracts following this phytochemical screening. Sub-fractionation of the plant extracts is further necessary to identify novel compounds. Also, these preliminary evaluations underlined the antiulcerous potential of the two extracts.

Acknowledgements: Scientific research funded by "Grigore T. Popa" University of Medicine and Pharmacy Iaşi, based on contract no 29029 / 28.12.2016.

Keywords: *Vernonia kotschyana*, LC-MS, vernoniosides

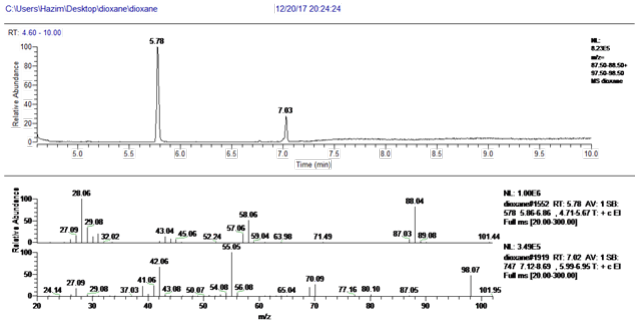
PS1-H-005

Development a highly sensitive and selective GC-MS method for the determination of Dioxane in cosmetic products

Ibrahim Alsohaimi[1], Hazim Ali[1,2], Tarek Seaf Elnasr[1,3]

[1]Jouf University, Sakaka, Saudi Arabia, [2]Forensic Medicine Authority, Egypt, [3]Al-Azhar University, Assiut, Egypt

1,4-Dioxane is a contaminant of emerging concern that has been found widespread in different environmental samples. It's a flammable liquid having a faint pleasant odor and the vapors are harmful [1,2].
A potential byproduct of the sulfation process, 1,4-dioxane is generated in the manufacture of alcohol ether sulfates and may be created in the ethoxylation process using ethylene oxide. The FDA has requested manufacturers of sodium laurel sulfate remove a substantial amount of 1,4-dioxane by vacuum stripping [3,4].
The objective of the present work is to development a simple, fast and sensitive new method for the determination of 1,4-dioxane traces in the cosmetic products using gas chromatography mass spectrometry (GC-MS). The procedure included the extraction of 1,4-dioxane from different environmental samples by dichloromethane, separation from other components by GC, and identification by MS. The quality parameters of the developed method (GC-MS) has been established and very good precision. The detection limit was 0.65 µg/kg, the linear range was large, relative standard deviations (R.S.D.) < 3%), and the correlation coefficient was 0.999. The total sample analysis time was found to be 5 min. The method is suitable for quality control in cosmetic products and surveillance and wide applications.



Keywords: 1,4-dioxane, gas chromatography mass spectrometry, cosmetic products, extraction

Acknowledgements: The authors are thankful chemistry department at Jouf University for access to analytical equipment.

References

[1] National Cancer Institute. In: Carcinogenicity technical report. Bethesda: National Cancer Institute, (1978)
[2] Lundberg I, Hogberg J, Kronevi T, Holmberg B. Cancer Lett. 1987; 36: 29
[3] Birkel T., Warner C., Fazio T., Journ. of the AOAC. 1979; 62: 931
[4] Environmental Health Association of Nova Scotia, Guide to Less Toxic Products (EHANS, 2004).

PS1-H-006

Extraction method development for the creation of a bank of extracts

Thibault Lefebvre, Sandrine Zubrzycki, Manon Bunout, Chloé Catan, Eric Lesellier, Emilie Destandau

ICOA (Institute of Organic and Analytical Chemistry) UMR7311, Orléans, France

During the last decades, pressurized fluids are major actors in plant extraction field. The increase of pressure allows the enhancement of the extraction by: reducing extraction time, using new extraction solvents, improving extract composition, being environmentally friendly and so on.
The objective here, is to create a library of extract that stores different plant fingerprints which represent the phytochemical profile of the plant. To do so, methods need to be developed to get the largest variety of compounds in term of polarity.
In this study, two different extraction techniques are investigated: Supercritical Fluid Extraction (SFE) using pure or mixed CO_2 and Accelerated Solvent Extraction (ASE) using liquid solvents under pressure. Also, different plants and parts of plant have been selected with different metabolic compositions.
Some conditions have been tested. These two techniques are different and also their parameters are. Conditions related to the instrument have been studied and the majority of possibilities have been tested for both instruments.
In the case of a bank extract designed to contain various extracts, the most important result is the profile of the extract. The best conditions are achieved for the extract contains the widest variety of molecules. Other results are also studied such as the mass yield of the extract, the concentration of the main extracted compounds and, from a more practical point of view, the duration of all the extraction process: from the cell filling to the storage of the dried extract, or the total volume of solvent used and collected before the extract concentration. The final method will be applied successively on varied plant sample chosen for their diversity in term of matrix and compound composition. Two strategies have been selected and they all have balanced advantages and drawbacks.

PS1-H-007

Experimental archaeology: The scent of antiquity reborn by KORRES for the National Archaeological Museum

Iordanis Samanidis, Lena Philippou

KORRES NATURAL PRODUCTS S.A., Metamorfosi, Greece

The production of fragrances used to be a common practice in ancient civilizations around the Mediterranean region, while fragranced oils were extensively used as a trade commodity. According to archaeological sources, as the Mycenaean civilization flourished (1600-1100 BC) ingredients like aromatic vegetable oils became indeed quite popular. The deciphering of Linear B script ideograms found on clay tablets that were uncovered during excavations at Mycenaean palaces in Pylos, Mycenae, Thebes, and Knossos revealed a list of natural ingredients used for both culinary and cosmetic purposes in antiquity. Olive oil, roses, coriander, cyperus, and wine are part of this list. Later studies focusing on the techniques of ancient perfumers, confirm that these raw materials were widely used for the production of aromatic oils like the ones described in the ancient texts of Pedanius Dioscurides and Theophrastus of Eresus. Following an invitation by the National Archaeological Museum and based on these findings, KORRES lab worked on reviving the scent of antiquity for the first time. The process involved several stages, starting with the ancient extraction technique of "stypsis"; a type of hot oil extraction to prepare a semi-finished oil. In this context two distinct "stymmata" were used, coriander and cyperus, mixed with olive oil, water and wine. Rose petals (referred to as "hedysma") were then used to add perfume to the treated oil, while the deep red alkanna root extract provided the fragranced oil with an alluring red color. ROSE is a primordial, one-dimensional scent, offering a unique sensory experience of the past. It has now taken its place among invaluable archaeological treasures, as well as the prevailing image of Goddess Aphrodite in the temporary exhibition of the National Archaeological Museum The countless aspects of Beauty that focuses on a presentation of the approach on Beauty in the various periods of antiquity.

PS1-H-008

Optimization of ultrasound–assisted extraction of polyphenolic compounds from *Plantago lanceolata* using response–surface methodology

Jelena Zivkovic, Katarina Savikin, Nada Cujic, Teodora Jankovic

Institute for Medicinal Plants Research "Dr Josif Pancic", Belgrade, Serbia

In folk medicine *Plantago lanceolata* (ribwort plantain) is used for the treatment of diseases of the upper respiratory tract, wound healing, and as a diuretic and anti–inflammatory drug [1]. In Mediterranean countries it is consumed as a part of the human diet, mainly as ingredient for fresh salads [2]. *P. lanceolata* contains iridoid glycosides, phenylpropanoid glycosides, flavonoids and phenylcarboxylic acids [2]. In the present study, we have determined optimal conditions for ultrasound–assisted extraction (UAE) of polyphenols from plantain herb using response surface methodology (RSM). The influence of extraction time (X1: 5-65 min), ethanol concentration (X2: 10–90 %), solid to solvent ratio (X3: 1:10–1:50) and extraction temperature (X4: 20–80 °C) on total polyphenols content (TPC) as well on content of dominant individual polyphenolic compounds (acteoside and luteolin-7-*O*-glucoside) was investigated. RSM was used to evaluate the relationship between process variables (independent parameters) and responses (dependent parameters) and to determine the optimum process conditions. The optimal extraction process conditions were as follows: extraction time of 64 min, ethanol concentration of 45%, solid to solvent ratio of 1:49, and extraction temperature of 40 °C. Experimentally obtained values agreed with those predicted by RSM model, indicating suitability of the employed model and the success of RSM in optimizing the extraction conditions.

Keywords: extraction optimization, plantain, acteoside, luteolin-7-*O*-glucoside

References:

[1] Gonçavles S, Romano A. Ind. Crops Prod 2012; 83:213–226.
[2] Hadjichambis A, Hadjichambi–Paraskeva D, Della A, Giusti ME, Pasquale C, Lenzarini C, et al. Int J Food Sci Nutr 2008; 59: 383–414.
[3] Rønsted N, Gobel E, Franzyk H, Rosendal Jensen S, Olsen CE, Phytochem 2000; 55: 337–348.

PS1-H-009

Simultaneous HPTLC densitometric estimation of acetyl-11-keto-*β*-boswellic acid and 11-keto-*β*-boswellic acid from *Boswellia serrata*

Meenu Mehta, Saurabh Satija

Lovely Professional University, Phagwara, India

Boswellic acids (BAs) are extracted from oleo gum of *Boswellia serrata* and are utilized as the potential anti-inflammatory, hypolipidemic, immunomodulatory and antitumor specialists. The present examination was meant to assess KBA and AKBA in *Boswellia serrata* separate by High-Performance Thin Layer Chromatography (HPTLC). The separation of bioactive compounds was performed utilizing mobile phase glacial acetic acid, *n*-hexane, ethyl acetate and toluene (0.3: 1: 8: 2) (v/v/v/v) and distinguished at wavelength 254 nm. The technique was approved for linearity, precision, accuracy, limit of detection (LOD), limit of quantification (LOQ), and so forth by International Conference on Harmonization guidelines. The calibration range was observed to be 2– 14 µg/band for both the bioactive compounds. KBA was isolated with an R_f estimation of 0.39 ± 0.02 and AKBA with an R_f estimation of 0.42 ± 0.02. The accuracy was seen to be as high as 99.17% and 97.42 for KBA and KBA respectively. The percentage RSD value for intra-day and between day varieties were under 2%. The system indicated high affectability and specificity. The developed HPTLC method was simple, precise, robust, specific, rapid, and cost-effective and could be used for quality control analysis and quantification of KBA and AKBA in different herbal formulations containing the plant species.

PS1-H-010

Development of novel HPTLC fingerprint method for simultaneous estimation of berberine and rutin in medicinal plants and their pharmaceutical preparations followed by its application in anti-oxidant assay

Saurabh Satija, Meenu Mehta

Lovely Professional University, Punjab, India, Phagwara, India

A straightforward, delicate and fast privileged thin layer chromatographic system (High performance thin layer chromatography; HPTLC) has been produced and validated for quantitative simultaneous determination of berberine and rutin in *Tinospora cordifolia* extract and their pharmaceutical preparations. The chromatographic development was completed on HPTLC plates precoated with silica gel 60 F_{254} utilizing a blend of ethyl acetate, glacial acetic acid, formic acid and methanol (10: 1.1: 1.1: 2.5) (v/v/v/v) as mobile phase. Detection was completed densitometrically at 254 nm. The Rf estimation of Berberine and rutin was observed to be 0.67 ± 0.02 and 0.47 ± 0.02 respectively. The system was validated according to ICH guidelines regarding linearity, precision, accuracy, robustness and so forth. The calibration curve was observed to be straight over a scope of 0.2–1.4 µg/spot with a regression coefficient of 0.988 for berberine and 2-14 µg/spot with a regression coefficient of 0.991 for rutin. The accuracy was observed to be as high as 96.30% and 94.89% for Berberine and Rutin respectively. The percentage RSD values for intra-day and between day variations were under 2%. The system showed high affectability and specificity. A new procedure has been used to separate and quantify the free radical-scavenging activity of berberine and rutin in *Tinospora cordifolia* plant extract based on the combination of HPTLC with diode array detector (DAD) and post-chromatographic DPPH radical derivatization.

PS1-H-011

Isolation and identification of endophytic fungi from *Piper nigrum* L. and investigation of their chemical constituents

Pragya Paramita Pal

Birla Institute of Technology and Sciences- Hyderabad Campus, Bits- Pilani, Hyderabad, Shameerpet, India

Endophytes largely contribute in the formation of active metabolites in the host medicinal plant. With an interest of exploring such endophytes, the study was conducted on *Piper nigrum*. It was chosen as the target for the isolation of the endophytic fungi due to its widespread pharmacological activities and its unexplored reports of its endophytes.

Surface sterilization of fresh ripened fruits was done by immersion in 70% ethanol for 1min, 5% NaOCl for 5min, 96% ethanol for 0.5min, followed by three rinses in sterile distilled water. Later, the sample tissues were incubated on Potato-Dextrose-Agar. The distinct colonies were then sub-cultured to obtain pure culture. [1] The isolated fungi was subjected to identification by DNA isolation from the fungal culture, followed by PCR to amplify the ITS region. The results of DNA sequencing were compared with NCBI data base. The fungus was identified to be *Penicillium polonicum* through BLAST search and phylogenetic tree analysis.

To perform preliminary phytochemical analysis, *P. polonicum* was cultured in a media of Potato Dextrose Broth (supplemented with 0.1% antibiotic solution) for 30 days at 28 ± 2^{0}C in Erlenmeyer Flasks with intermittent shaking. The broth was then extracted using ethyl acetate and concentrated under reduced pressure to obtain the crude extract.

The was analysed to contain 120 µg/ml of total phenolics (Folin Ciocalteu method), 113 µg/ml of total flavonoids ($AlCl_3$ Colorimetric method), 10.21 µg/ml of total alkaloids (Bromocresol Green method) and 359 µg/ml of total Sterols (Liebermann-Buchard method). The LC-PDA-ESIMS fingerprint of the culture broth extract revealed the presence of 7 major compounds having molecular weight in the range of 64 – 785 daltons, which can be further isolated and explored for themedicinal effects of *P. nigrum*.

Keywords: endophytic fungi, isolation, LC-MS.

PS1-H-012

Oil and fatty profile on the film from the *Pistacia lentiscus,* region of Collo. east in Algeria

Nassima Boudiaf

Faculté De Médecine, Annaba, Algeria

The objective of this study is the determination of the analytical parameters and the fatty acid composition of the film from *Pistacia lentiscus*, the extraction was carried out by soxhlet using an apolar solvent which is hexane, the chemical composition of fatty acids was performed by chromatography alone and coupled to mass spectroscopy (CGC, GC / MS) this study identified 7 constituents representing 86.81% the compounds the major compounds are palmitic acid 28.15%, oleic 26.56% and linoleic 24.57% .

Keywords: *Pistacia*, extraction, oil, fatty acids, GC/MS

PS1-H-013

N-doped magnetic hydrochar to toxic metals from water: synthesis, characterization, and adsorption studies

Ayoub Alqadami[1], Zeid Alothman[1], Moonis Khana[1], Mohammad Algamdi[2]

[1]King Saud University, Riyadh, Saudi Arabia, [2]King Abdulaziz City for Science and Technology, Riyadh, Saudi Arabia

Efforts to immaculate water quality have led to the development of green and sustainable water treatment approaches [1-2]. Herein, nitrogen-doped magnetized hydrochar (mSBHC-*N*) was synthesized, characterized, and used for the removal of post-transition and transition heavy metals, viz. Pb^{2+} and Cd^{2+} from aqueous environment. mSBHC-*N* was found to be mesoporous (BET surface area – 62.5 m2 /g) and paramagnetic (saturation magnetization – 44 emu/g). Both, FT-IR (with peaks at 577, 1065, 1609 and 3440 cm-1 corresponding to Fe – O stretching vibrations, C – N stretching, N – H in-plane deformation and stretching) and XPS analyses (with peaks at 284.4, 400, 530, 710 eV due to C 1s, N 1s, O 1s, and Fe 2p) confirmed the presence of oxygen and nitrogen containing functional groups on mSBHC-*N*. The adsorption of Pb^{2+} and Cd^{2+} was governed by oxygen and nitrogen functionalities through electrostatic and co-ordination forces. 80% of Pb^{2+} and Cd^{2+} adsorption, either from deionized water or humic acid solution was accomplished within 15 min. The data was fitted to pseudo-second-order kinetic and Langmuir isotherm models, with maximum monolayer adsorption capacities being 323 and 357 mg/g for $Cd^{2+}$and Pb^{2+} at 318K, respectively.

Keywords: Sugarcane bagasse, hydrothermal carbonization, magnetization, nitrogen-doping; water treatment

PS1-H-014

Extraction and purification of metabolites from natural substance or biotechnology by Centrifugal Partition Chromatography

Sébastien Chollet[1], Luc Marchal[2], Jean-Hugues Renault[3]

[1]Capacites Sas, Nantes, France, [2]University of Nantes, Nantes, France, [3]University of Reims, Reims, France

Centrifugal Partition Chromatography is a solid support-free separation process based on the partitioning of solutes between two partially miscible liquid phases. A CPC column consists of a series of partition cells linked in cascade by ducts and arranged in a centrifuge (one axis, two rotary seals), which provides a centrifugal acceleration responsible for stationary phase retention and mobile phase dispersion. This process allows realizing different modes of purification: partition chromatography, displacement chromatography, selective and intensive extraction and also intensified reaction. For each kind of mode, a suitable column can be size.
After 25 years of research and developments, we can consider that knowledge and tools are nowadays available for CPC method development (solvent, elution, displacement, operating conditions choice and column engineering). CPC Engineering team gathers this knowledge to perform metabolites purification, process intensification and scale changes.
This versatile process, without any solid support, was and still to be promising for the separation of complex mixtures. This technology, initially used for natural substances, proves to be also promising for the field of biotechnology.
In this presentation, diverse way to purify metabolites from plant extracts and blue (or marine) biotechnology and columns selection will be presented.
The examples will be:

- The purification of alkaloids from plant extract by displacement chromatography. On a 300mL column (laboratory scales), this mode allows treating 780g of crude extract per day.
- The fractionation of carotenoids from microalgae extract by partition chromatography (figure 1). On a 200mL column (laboratory scales), this mode allows treating few grams of lipid extract per day.
- The selective extraction of proteins from microalgae extract using Aqueous Two Phases Systems. On a 50mL prototype column, this mode allows treating 450 g of microalgae extract per day.

Feasibility at lab-scale will be presented and scalability discussed.

PS1-H-015

Cannabis Process Separation of Cannabinoids

Mouroutis-Prionistis, Yianns

HELLAMCO

Introduction:
The separation of cannabinoids is an important process necessary to obtain pure compounds for clinical studies. The aim of these studies is to investigate the effect of the isolated cannabinoids on patients suffering from chronic illnesses and side effects caused by chemotherapy [1]. Recent modernisation of laws concerning medical use of cannabis all around the world, especially in the United States, has increased the interest in determining the effect of cannabinoids on the human body [2]. The research emphasis lies in treating the side effects of chemotherapy, inflammatory diseases like multiple sclerosis or degenerative illnesses such as Parkinson's disease [3].
Aim of the Study:
Creating of pure isolated cannabinoids and formulate them in a controlled workflow to supply the high demand for clinical research.
Materials and Methods:
BUCHI Reveleris, BUCHI Rotavapor, BUCHI Spray Dryer, BUCHI Encapsulator, BUCHI Freeze Dryer.
Results and Discussion:
After the extraction of the plant material, various processes were performed to prepurify the extract. Subsequently, the extract is concentrated gently under vacuum in a rotary evaporator to prevent degeneration of the active compounds. By means of preparative chromatography the all the different molecules are separated using C_{18} modified silica. The collected fraction containing the isolated compounds were evaporated to dryness. During the entire process only food grade aqueous solvents are used. Further steps for involve formulating the isolated compounds with either spray dryers, freeze dryers or encapsulators, to vary the intake and the release of the medication into the organism and modulate.
Conclusions: Clinical studies require large amount of highly pure separated cannabinoids. This can easily be achieved applying Buchi's products for the cannabis workflow.

References

[1] Sutton IR, Daeninck P. J Support Oncol 2006; 4: 531–535
[2] State Medical Marijuana Laws. National Conference of State Legislatures. (16.3.2015). http://www.ncsl.org/research/health/state-medical-marijuana-laws.aspx
[3] Kogan NM, Mechoulam R. Dialogues Clin Neurosci 2007; 9: 413–43

PS1-H-016

A combination of liquid phase microextraction and spectrophotometric determination of ascorbic acid in foods

Yaroslav Bazel[1], Tetiana Riabukhina[2]

[1]Pavol Jozef Šafárik University in Košice, Kosice, Slovakia, [2]Uzhhorod National University, Uzhhorod, Ukraine

Ascorbic acid (Asc) is a water-soluble vitamin, and it participates in redox processes in human organism. Ascorbic acid enters the human body with foods and pharmaceuticals. The methods used for determination of Asc often based on its reduction properties. A variety of analytical methods have been previously used for Asc determining at trace levels in foods and other samples, but spectrophotometry is the most commonly used method.
Our research is based on the redox-reaction between Asc with triiodide anion in the presence of polymethine dye Astra Phloxine FF with next combination of liquid-liquid microextraction and spectrophotometric determination of Asc. This was used as a basic of developed spectrophotometric method for determining Asc in some foods.
An analysis of various samples of food products (fruits, berries and beer) was conducted.
The linear regression equation was A = 2.208xC(Asc) – 0.004 in the range 0.003 – 0.53 mg/L Asc, the LOD (n=10, P=0.95) was found to be 0.89 μg/L of Asc. As the extractant, we used carbon tetrachloride, the quantity of which is minimized (500 μL) a preconcentration factor of 10 can be obtained. The suggested procedure was successfully applied for the determination of Asc in food samples (RSD 2.1 – 3.9%, recovery 98.0 – 102.3%) and could play a useful role in checking the level of Asc in foods. The data showed satisfactory convergence with the reference spectrophotometric method. The proposed method is more sensitive, and satisfactory metrological characteristics indicate its practical use.

Acknowledgements: this work has been supported by the Scientific Grant Agency of the Ministry of Education of the Slovak Republic and the Slovak Academy of Sciences VEGA (grant No. 1/0253/16).

PS1-H-017

Application of the liquid phase microextraction to separation and extraction of biological active compounds from plant samples

Vasil Andruch

University of P.J. Safarik, Slovakia

The analysis of plant materials is a challenging task due to complex matrix simultaneously with low concentration of the analytes. The task of sample pre-treatment is isolation of the target analytes, clean up, and pre-concentration which in an ideal state should able to perform in a single step. In recent years, the liquid-phase microextraction techniques (LPME) coupled with variety of analytical equipments more and more are coming to the fore for determination of both organic and inorganic analytes. These microextraction approaches involve dispersive liquid–liquid microextraction (DLLME), dispersive liquid-phase microextraction (DLPME), and single-drop microextraction (SDME). The earlier papers devoted to these techniques focused mainly on the analysis of aqueous samples. To analysis of solid samples, especially to analysis of plants have paid much less attention. However, over the past few years, a growing number of publications devoted to the use LPME on analysis of plant samples can be observed.
Based on LPME technique, the procedures for analysis of various parts of plant samples, such as root, stem, leaf, fruit, leaves, and flowers were reported. Various organic and inorganic analytes were determined using LPMEs procedures. Before application of the microextraction procedure, the analytes to be transferred from raw solid plant samples to the aqueous phase. After sample pretreatment step, variety microextraction procedures for analytes preconcentration were applied. In addition to conventional DLLME procedures, modalities of this technique were also described.

Acknowledgements: This work was supported by the Scientific Grant Agency of the Ministry of Education, Science, Research and Sport of the Slovak Republic (VEGA 1/0010/15).

PS1-H-018

Environmentally-friendly enzyme-assisted extraction of Naphthoquinones from *Alkanna tinctoria* roots

Elodie Bossard[1], Angelos Argyriadis[2], Anthi Karnaouri[3], Nikos Tsafantakis[1], Nektarios Aligiannis[1], Andreanna Assimopoulou[2], Evangelos Topakas[3], Nikolas Fokialakis[1]

[1]Faculty of Pharmacy, Laboratory of Natural Product Chemistry and Pharmacognosy, School of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [2]Organic Chemistry Laboratory, School of Chemical Engineering, Aristotle University of Thessaloniki, Thessaloniki, Greece, [3]Industrial Biotechnology & Biocatalysis Group, Biotechnology Laboratory, School of Chemical Engineering, National Technical University of Athens, Athens, Greece

Natural products derived from plant sources, also known as plant secondary metabolites (SMs), have an important role in pharmaceutical, cosmeceutical and food supplements industries. The increasing demand of SMs is leading to the over-exploitation of plant resources, the over-consumption of organic solvent and also poses an important risk of making the species rare or even extinct. For this reason, the design of green and sustainable extraction of naturals products and cultivation methods is currently a key research topic in the multidisciplinary area of applied chemistry, biology and technology. Green extraction processes allow to reduce organic solvent consumption and allow the use of alternative solvents and renewable natural products.

Within the frame of the EU H2020 "MICROMETABOLITE" project, eco-friendly and innovative technologies, such as semi-hydroponic, *in vitro* cultivation systems integrating microorganisms and green extraction technologies, are developed. This project is focused on Boraginaceae plants, especially on *Alkanna tinctoria* roots and in the study of the enantiomeric naphthoquinones, Alkannin and Shikonin.

Within the frame of this project we employed enzyme treatment as a tool to enhance the release of naphtoquinones that are linked to the protein/glycosylated-protein structures of *Alkanna tinctoria* root cells. Various enzyme combinations, such as proteases and carbohydrate-acting enzymes, are used to loosen the structural integrity of the root cells and disrupt the protein-naphtoquinone bonds. Micro enzymatic-assisted extractions of commercial *Alkanna tinctoria* roots were investigated either as an enzymatic pretreatment step or in combination with sonication. The percentage of carbohydrates, ash and protein was calculated to determine the enzymatic hydrolysis parameters, like the type of enzyme, optimum enzyme concentration and incubation time. The obtained naphtoquinone was analyzed with methods based on HPTLC and HPLC.

Acknowledgments: This project has received funding from the European Union's Horizon 2020 research and innovation program under the ITN - Marie Skłodowska-Curie grant agreement No 721635.

PS1-H-019

A holistic green strategy for the isolation of bioactive compounds from fennel seed oil and the corresponding by-products, with pilot scale potentials

Dimitris Michailidis, Apostolis Angelis, Alexios-Leandros Skaltsounis

University of Athens School of Pharmacy Laboratory of Pharmacognosy and Natural Products Chemistry, Panepistimioupoli, Zografou, 15771, Athens, Greece

Foeniculum vulgare, commonly known as fennel, is an aromatic plant natively grown in the Mediterranean countries. Although, nowadays it is worldwide cultivated due to its extensive use and its globally high economic importance [1]. Our study focuses on the development of a holistic framework for the extraction and separation of high added value compounds from fennel seed oil and byproduct paste, using technologies of applied science and characterized as totally green, swift and functional for industrial applications.

The experimental process started with the extraction of both seed oil and paste. For the fennel seed oil adsorption resins XAD-7 were used, while for the paste, were employed two different green technologies to cover the whole polarity range of the raw material. In particular, fennel seed paste was extracted with three successive Ultrasound Assisted Extractions (UAE) (a: food grade *n*-Hexane for the defatting, b: EtOH and c: EtOH/H_2O 1:1 v/v). Moreover, Supercritical Fluid Extraction (SFE) was conducted with three successive steps as well (a: CO_2 for the defatting, b: CO_2/EtOH 10% and c: CO_2/EtOH 20%). The second step was the fractionation of the extracts with Centrifugal Partition Chromatography (CPC) which is a liquid-liquid solid support free technique providing the flexibility of various solvent range [2]. For this purpose, the selected system was *n*-Hex/EtOAc/EtOH/H_2O in different ratios, depending the chemical profile of each extract. Selected middle polar and non-polar CPC fractions were treated with Supercritical Fluid Chromatography (SFC) coupled to MS and PDA detectors for further purification, giving noteworthy results and achieving separation even in different isomeric forms of compounds.

The designed experimental procedure aims to give applied solutions for the obtainment of bioactive natural products through the application of green and smooth handling of extracts, capable on pilot and industrial level up with predictable results.

Acknowledgements: The research work was supported by the Hellenic Foundation for Research and Innovation (HFRI) and the General Secretariat for Research and Technology (GSRT), under the HFRI PhD Fellowship grant (GA. no. 14498).

Keywords: fennel seed, byproducts, green technology, SFE, UAE, CPC, SFC.

References:

[1] Al-Snafi A., Int J Pharmacol Toxicol, 2015; 5: 76–81.
[2] Berthod A, Spivakov B, Shpigun O, Sutherland IA, Pure Appl Chem, 2009; 81: 355–387.

PS1-H-020

Ultrasound-Assisted Extraction (UAE): induced physicalimpacts on Rosemary (*Rosmarinus officinalis* L.) and Artichoke (*Cynara scolymus* L.) leaves

Boutheina Khadhraoui[1], El Maâtaoui Mohammed[2], Isabelle Bornard[3], Rémi Imbert[1], Philippe Robinet[1], Anne-Sylvie Fabiano-Tixier[4], Njara Rakotomanomana[4], Farid Chemat[4]

[1]Laboratoires Arkopharma, Carros, France, [2]Avignon University, Qualisud UMR95, F-84000 Avignon, France, [3]Inra PACA, Pathologie végétale, F84143 Montfavet, France, [4]Avignon University, UMR 408, Green Extraction Team, 84000 Avignon, France

The ultrasound positive contribution on the extraction performances such as reduction of extraction time, diminution of required solvent quantity and enhancement in yields were widely proved [1]. This performances' gain is generally attributed to the cavitation phenomena [2]. However, ultrasound mechanisms were poorly investigated [3].
Our study aims at better understanding ultrasound-induced mechanical effects on complex plant matrices in the particular case of Rosemary and Artichoke leaves.
Effect of ultrasound has been assessed by submitting a single leaf, fixed in a perforated disk, to an ultrasonic field (US probe, 20 kHz) at different treatment durations. Extraction was performed in demineralized water. Conventional process was carried out at the same conditions without ultrasound.
The surfaces of untreated and treated leaves were examined by Scanning Electron Microscopy (SEM) and Environmental Scanning Electron Microscopy (e-SEM 3D). Cyto-Histochemical study was also conducted to analyze ultrasound-induced alterations on the inner structures. Mechanisms of action were then concluded on the basis of these different observations
Our findings proved that conventional process preserved structural properties and the integrity of both rosemary and artichoke leaves compared to the initial leaves. In contrast, ultrasound induced noticeable alterations on the two studied leaves. It is crucial to note that ultrasound impacted Rosemary and Artichoke leaves by different mechanisms. In the case of rosemary leaf, ultrasound seemed to act through chain detexturation mechanism respecting a sequential steps: local erosion, shear forces, sonoporation, fragmentation, capillary effect and detexturation: local erosion, shear forces, sonoporation, fragmentation, and detexturation. As for Artichoke leaf, only four mechanisms were noticed: shear forces, sonoporation, erosion and fragmentation.

Keywords: Ultrasound, extraction, microscopic observations, e-SEM 3D, mechanisms

References

[1] M. Virot, V. Tomao, C. Le Bourvellec, C.M.C.G. Renard, F. Chemat. Towards the industrial production of antioxidants from food processing by-products with ultrasound-assisted extraction UltrasonicsSonchemistry. 17 (2010) 1066-1074.
[2] M. Toma, M. Vinatoru, L. Paniwnyk, T. Mason, 2001, Investigation of the effects of ultrasound on vegetal tissues during solvent extraction, Ultrasonics Sonochemistry, 8, pp 137-142.
[3] F. Chemat, N. Rombaut, A-G. Sicaire, A. Meullemiestre, A-S. Fabiano-Tixier, M. Abert-Vian. Ultrasound assisted extraction of food and natural products. Mechanisms, techniques, combinations, protocols and applications. A review. UltrasonicsSonochemistry. 34 (2017) 540-560.

PS1-H-021

Oleocanthalic and oleaceinic acids: Isolation, identification and semi-synthesis of new secoiridoids compounds of extra virgin olive oil

Lemonia Antoniadi, Apostolis Angelis, Panagiotis Stathopoulos, Maria Halabalaki, Leandros A. Skaltsounis

Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimioupolis Zografou, Athens, Greece

Extra Virgin Olive Oil (EVOO) utilization in dietary patterns world-wide is increasing due to its particular flavor, aroma, nutritional and health beneficial effects. However, despite the high number of studies available in the literature regarding EVOO constituents its complete characterization still remains challenging due to the high complex nature and the variation of factors affecting its composition. An ongoing phytochemical investigation of Extra Virgin Olive Oil (EVOO) secondary metabolites, led to the isolation of two new secoiridoid acids, Oleocanthalic acid and Oleaceinic acid. The isolation procedure included the liquid-liquid extraction of EVOO by Centrifugal Partition Extractor (CPE) [1], the fractionation of the recovered TPF by Centrifugal Partition Chromatography (CPC) and finally, the analysis of selected CPC fractions using preparative TLC and preparative HPLC-DAD methodology. The structure elucidation of purified compounds was based on NMR (1D and 2D) and HRMS and HRMS/MS analysis.

Their presence in diverse EVOOs was investigated by UHPLC-HRMS analysis. 10 EVOOs representative of diverse olive oil producing areas of Greece were randomly selected and analyzed for this purpose. Based on the derived data the two newly isolated compounds were detected in all analyzed oils and only quantitative differences were observed. This fact indicates that the above secoiridoid acids are characterized as standard ingredients of olive oil.

In order to confirm the structures of oleocanthalic and oleacein acid as well as to support *in vitro* and *in vivo* bioassays , the semi-synthesis of these compounds was conducted, using pure oleocanthal and oleacein as starting material. The reaction was based on a previously described method by using Oxone reagent diluted in DMF [2]. The selective oxidation of C-3 aldehydic moiety to carboxylic acid proceeded with high efficiency in yields greater than 60%. The spectroscopic data of oleocanthalic and oleaceinic acids obtained by this semi synthetic route are consistent with the corresponding purified compounds.

Key Words: EVOO, polyphenols, oleocanthal, oleacein, oleocanthalic acid, oleaceinic acid, semi-synthesis

References:

[1] Angelis A *et al*. J. Chromatogr A 2017; 1491: 126–136.
[2] Travis B *et al*. Organic Letters 2003; 5 (7): 1031–1034.

PS1-H-022

CORNET Project SaliChem: Halophytes cultivation for sequential biomass valorisation

Marie-Caroline Jonville[1], Job Tchoumtchoua[1], Stefan Dröge[2], Cécile Beauve[1], Patrick Ballmann[2], Michael Müller[2]

[1]CELABOR, Biomass Valorisation Platform - Extraction Department, Avenue du Parc 38, 4650 Herve, Belgium, [2]Prüf- und Forschungsinstitut (PFI), Marie-Curie-Str. 19, 66953 Pirmasens, Germany

About 2,000 square metres of farmland is being destroyed everyday worldwide due to salination. This mainly due to artificial irrigation and over-cultivation of crops. Conventional cultivated plants do not survive salination. Moreover, the need of biomass for the production of food, chemicals and energy is apparent, considering the increase in costs and unknown reserves of fossil petroleum and the increase in world population. However, due to the food vs. fuel debate, only biomass which cannot be used for food can be considered for chemical and energy production.

The CORNET Project SaliChem focused on finding a solution to these problems by doing research on the use of salt-tolerant plants also called halophytes, which are naturally adapted to salinity, can grow under these conditions, and are able to provide biomass in order to generate energy after extraction of bioactive molecules.

Therefore, screenings of 6 halophytes namely *Aster tripolium*, *Crambe maritima*, *Beta maritima*, *Suaeda maritima*, *Ammophila littoralis*, and *Spartina maritima* were conducted based on secondary metabolites profiling, biological activity (antimicrobial, anti-oxidant and anti-aging potency), lignocellulosic composition for fermentation, and anaerobic digestion for methane production. *Spartina maritima*, giving the most promising results, was selected to maximize the valorization potential by applying a biorefinery concept. This work focuses on the conversion of the lignocellulosic content to fermentable sugars and the anaerobic digestion of the *S. maritima* biomass to methane with a prior supercritical CO_2 extraction of a lutein-enriched lipophilic fraction having anti-elastase and anti-collagenase potential.

Pretreatment using Thermal Pressure Hydrolysis[1] gave a composition of glucan, xylan, and lignin comparable to typical feedstocks considered for lignocellulosic biorefineries, such as wheat straw and corn stover[2]. In terms of biogas production, *S. maritima* showed similar results compared to grass silage, a typical feedstock for agricultural biogas plants.

In conclusion, *S. maritima* is a promising candidate as feedstock for lignocellulosic biorefineries with the additional benefits of being cultivatable on saline soil, accumulating valuable extractable compounds to be used in anti-aging cosmetics, and being usable as substrate for biogas production.

Acknowledgements: We are grateful to the AiF and the Federal Ministry for Economic affairs and Energy of Germany, and the Walloon region of Belgium for supporting this research through the Cornet Program. We wish to thank Serra Maris bvba (Belgium) who generously provided the plant material.

Keywords: Bioraffinery, Anti-aging, plant extraction, *Spartina*, secondary metabolite.

References:

[1] Gasser E, Ballmann P, Dröge S, Bohn J, and König H. J Appl Microbiol 2014; 117: 1035–1044

PS1-J-001

HS-SPME analysis of seven truffles (*Tuber* sps) growing wild in Greece. Nutritional value & biological activity

Eleni Stavraki[1], Konstantia Graikou[1], Olga Gortzi[2], Ioanna Chinou[1]

[1]Sect Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, National & Kapodistrian University of Athens, Athens, Greece, [2]Department of Food Technology, Technological Educational Institution (TEI) of Larissa, Karditsa, Greece

The analysis of seven selected species of *Tuber* truffles (*T. aestivum, T. melanosporum, T. mesentericum, T. magnatum, T. borchii, T. brumale and T. uncinatum*) is reported in this study, for the first time. Truffles, known since antiquity as «ύδνον», are the fruiting bodies of mychorrhizal filamentous fungi, well-known and valuable as food. The aim of this study was to qualify their aroma profile, as well as, to evaluate their total phenolic content. The volatile organic compounds (VOC's) were analyzed by HS-SPME with two different polarity fibers (PDMS and CAR-PDMS) and led to the identification of 82 VOCs. The chemical profile of *T. melanosporum*, *T. brumale* and *T. magnatum* is characterized by aldehydes, while in the latter, 2,4-dithiapentane is served as fingerprint marker. Moreover, *T. magnatum* is the only among the studied truffles containing amine and nitrogen derivatives. *T. uncinatum*, *T. aestivum* and *T. borchii* showed an abundant presence of ketones and alcohols, while *T. borchii* presented the highest concentration of sulfur derivatives, which is in accordance with literature. *T. mesentericum*, was dominated by the aromatic compound 3-methylanisole, well known as its chemical marker.

Additionally their nutritional value (content of protein, fat, fiber, carbohydrates, sugar, saturated and unsaturated fatty acids and energy) was determined, their antimicrobial activity was tested against a panel of six Gram positive and Gram negative bacteria and three human pathogenic fungi and their antioxidant activity was estimated using the Rancimat method.

Moreover, all studied truffles were evaluated for their total phenolic content, where *T. mesentericum* and *T. borchii* revealed as the richest sources of phenolics (7.8 and 7.4 mg GAE/g of sample, respectively).

Acknowledgements: The author would like to thank the Special Account for Research Grants and the National and Kapodistrian University of Athens for funding their participation in this meeting.

PS1-J-002

Characterization of volatiles, phenolics and antioxidant properties of different populations of *Sideritis clandestina*

Ioannis Bakratsas, Virginia Dimaki, Lygeri Papaioannou, Gregoris Iatrou, Fotini Lamari

University of Patras, Patras, Greece

S. clandestina (Bory & Chaub.) Hayek is an endemic taxon of Peloponnisos and has two subspecies *S. clandestina* ssp. *clandestina* (SCC) and *S. clandestina* ssp. *peloponnesiaca* (Boiss. & Heldr.) Baden (SCP).
Five SCP populations from Dourdouvana, Chelmos, Gaidourorahi and of two SCC ones from Parnonas mountains in Peloponnisos as well as cultivated *S. scardica* Griseb were analysed. Plant material was extracted with a citrate buffer (75 min, 37°C) and then with petroleum ether in a sonicator bath [1]. The volatiles were analysed with GC-MS. Total phenol and flavonoid content of the aqueous extracts was estimated with Folin-Ciocalteu and $AlCl_3$ assays, respectively. Their antioxidant capacity was assessed using the FRAP and DPPH methods.
Alpha-pinene (4.07-28.63%) was the predominant component in nearly all populations; β-pinene (4.85-10.29%), β-caryophyllene (3.04-7.47%), linalool (0.77-18.28%) and α-bisabolol (0.98-7.28%) were main components. Linalool was at a surprisingly high percentage (18.3%) in a Parnonas population. In the leaves of both *S. clandestina* and *S. scardica*, monoterpenes were lower than in the flowers, while sesquiterpenes were more abundant. Significant variation of total polyphenol and flavonoid content (6.65-64.19 mg GAE/g and 4.58-14.85 mg QE/g) was recorded. In both SCP and *S. scardica*, total polyphenols were at least two times higher in the leaves than in the flowers. The IC_{50} values estimated with the DPPH method were equal or lower than that of butylated hydroxytoluene. A significant variation of the FRAP values was recorded and a high correlation of those with polyphenol and flavonoid content. The antioxidant properties of leaves were better than those of the flowers.
We herein record for the first time with the use of a miniaturised methodology the volatiles and polyphenols/flavonoid content and show the great diversity of the wild populations of *S. clandestina.*

References:

[1] Dimaki VD, Iatrou G, Lamari FN. J Chromatogr A 2017; 1524: 290-297.

PS1-J-003

Phenolic compounds profile and antioxidant properties of different sweet cherry (*Prunus avium* (L.) L.) varieties from Spain

Dolores Muñoz Mingarro[1], Antonia García[2], Associate professor Nuria Acero[3], Associate professor Ana Gradillas[2]

[1]Chemistry and Biochemistry Department. Pharmacy Faculty. San Pablo-ceu University, Madrid, Spain, [2]Centre for Metabolomics and Bioanalysis (CEMBIO), Chemistry and Biochemistry Department, Pharmacy Faculty. San Pablo-CEU University., Madrid, Spain, [3]Pharmaceutical and Health Science Department. Pharmacy Faculty. San Pablo-CEU University, Madrid, Spain

Sweet cherries are a source of phenolic substances which are actively involved in reducing reactive oxygen species content (ROS) in our bodies. ROS are closely related with inflammatory processes, cardiovascular problems, or generation and development of tumors, among other pathologies. The consumption of fresh cherry fruits or other related products are highly appreciated by consumers due to their taste and nutritional qualities [1]. These aspects are related with the fruit's phytochemical composition, which depends on genotype and other external factors [2].

In the present work, three Spanish local varieties of *Prunus avium* (L.) L.: Navalinda, Jarandilla, and Pico Colorado; as well as two foreign varieties: Van and Sunburst, all grown in the Jerte Valley, were studied. A phytochemical profile of these five cultivars was performed by UHPLC-qTOF-MS. The employed chromatographic method allowed a clear and rapid separation of hydroxycinnamic acids, anthocyanins and flavonoids, the three main phenolic compound groups present in the extracts [3]. *In vitro* scavenging capacity of the extracts was analyzed through DPPH and xanthine oxidase/xanthine assays. Finally, the effect in ROS intracellular concentrations in HepG2 cell line cultures were measured. Differences between *in vitro* and in cell culture antioxidant results evidence the interaction among the phenolic compounds of the extract.

Acknowledgements: Fundación Universitaria San Pablo-CEU and Banco de Santander are acknowledged for financial support (PPC 20/2015).

Keywords: *Prunus avium*, cherries, phenolic compounds, UHPLC-qTOF-MS, antioxidant

References:

[1] Nawirska-Olszańska A, Kolniak-Ostek J, Oziembłowski M, Ticha A, Hyšpler R, Zadak Z, Židová P, Paprstein F. Food Chem. 2017; 228: 136–142.
[2] Faniadis D, Drogoudi P D, Vasilakakis M. Sci. Hortic. 2010; 125: 301–304.
[3] Martini S, Conte A, Tagliazucchi D. Food Res. Int. 2017; 97: 15–26.

PS1-J-004

Development of a novel functional goats' milk yoghurt enriched with *Pistacia atlantica* resin extracts and *Saccharomyces boulardii*: Stability and organoleptic effects

Elena Hadjimbei[1], George Botsaris[1], Vlasios Goulas[1], Eleni Alexandri[2], Vassilis Gekas[1], Ioannis Gerothanassis[2]

[1]Department of Agricultural Sciences, Biotechnology and Food Science, Cyprus University of Technology, Limassol, Cyprus, [2]Nuclear Magnetic Resonance Center, Department of Chemistry, Section of Organic Chemistry and Biochemistry, University of Ioannina, Ioannina, Greece

Yoghurt is an ideal vehicle for providing functional ingredients as it is widely consumed by young adults and already contributes to good health because of probiotics, calcium and other minerals and vitamins. Already several attempts have been undertaken to produce yoghurt fortified with various plant extracts. The present study describes the development of a novel functional dairy product using *Pistacia* resin extracts and *Saccharomyces boulardii*. Different fractions of *Pistacia* resin extracts were incorporated into goats' milk yoghurt alone or in combination with *S. boulardii*. Five different yoghurt formulations were prepared: Yoghurt 1– Control, with only the starter culture added; Yoghurt 2– Acidic, with acid fraction of mastic extract; Yoghurt 3– Neutral, with neutral fraction; Yoghurt 4– Total, with total mastic extract; and Yoghurt 5– Combined, with total mastic extract and yeast. Enumerations of total lactic acid bacteria (LAB) were performed every three days for 25 days. Stability of *Pistacia* resin extracts and fatty acids was examined by NMR analysis. Finally, the samples were also assessed organoleptically. Results demonstrated that all *Pistacia* resin extracts promoted the growth of LAB at least until day 18. LAB numbers remained significantly higher and steady at about 8 log10cuf/g in Yoghurt 5 for 25 days. Four fatty acids were identified and quantified in yoghurts: linoleic, linolenic, caproleic and conjungated linoleic acid (CLA) and keep intact during shelf life. All the bioactive compounds were maintained during shelf life in yoghurt. Based in group organoleptic assessment there were not significant differences in flavor between different yoghurts. When *Pistacia* resin extracts alone or in combination with *S. boulardii* have been added in goat's yoghurt the growth of LAB was promoted and very good organoleptic properties were demonstrated. This new product could be marked as a potential innovative functional food, offering an alternative way of disease prevention and wellbeing.

PS1-J-005

Chemical characterization of seed oils obtained from different apple cultivars

Jelena Zivkovic[1], Mihailo Ristic[1], Josephine Kschonsek[2], Anna Westphal[2], Angelika Malarski[2], Volker Bohm[2]

[1]Institute for Medicinal Plants Research "Dr Josif Pancic", Belgrade, Serbia, [2]Institute of Nutrition, Friedrich Schiller University Jena, Jena, Germany

Numerous reports state that introduction of vegetable oils in the diet has beneficial effects in prevention of cardiovascular diseases, preserving healthy cholesterol levels, improving brain function and fighting with free radical species. [1] Recently, the interest in finding newer sources of oils for edible and non–edible applications such as unconventional plant seeds increased. The aim of the present study was to explore the composition (fatty acids, carotenoids and tocols) of fatty oils obtained from apple seeds. The seeds were recovered from two Balkan autochthonous cultivars (Šumatovka and Kablarka) and one commercial apple cultivar (Melrose). The oil yield was in the range from 9.9% d.w (for Šumatovka cultivar) to 18.0% d.w. (for Melrose cultivar). The polyunsaturated fatty acids were the predominant group of fatty acids and all of the investigated samples can be classified as oleic–linoleic oils. Fatty oils obtained from Melrose cultivar showed higher amount of linoleic acid and lower amount of oleic acid compared to investigated autochthonous cultivars. The concentration of tocopherols ranged from 26.07mg/g in Melrose seed oil to 40.05 mg/g in Šumatovka seed oil. While the main isomers in Šumatovka oil was β–tocopherol, followed by α–tocopherol, their contents were almost equal in oils obtained from Melrose and Kablarka cultivars. Among tocotrienols dominant compound was γ–tocotrienol. Concerning carotenoids, their concentration ranged from 0.07 mg/100 g of oils in Melrose oil to 0.32 mg/100 g of oil in oil obtained from Šumatovka cultivar, with a predominance of lutein. The obtained results demonstrated the potential of use of apple seeds as new alternative sources of vegetable oils.

Keywords: apple seed, waste, tocol, carotenoid

References:

[1] Costa T, Jorge N Nutr Food Sci 2012; 42: 279

PS1-J-006

Deciphering bacterial composition in the rhizosphere of *Baphicacanthus cusia* (NeeS) Bremek

Meijuan Zeng[1,2], Yongjia Zhong[3,4], Shijie Cai[5], Yong Diao[1]

[1]Huaqiao University, China, [2]Zhangzhou Health Vocational College, China, [3]Root Biology Center, Fujian Agriculture and Forestry University, China, [4]Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, China, [5]Nuffield Division of Clinical Laboratory Sciences, University of Oxford, UK

Rhizobacteria is an important ingredient for medical herb growth, health and synthesis of pharmacological effective substances. In this study, we investigated the community structure and composition of rhizobacteria in *Baphicacanthus cusia* (NeeS) Bremek via 16S rRNA amplicon sequencing. We obtained an average of 3,370 and 3,730 OTUs for bulk soil and rhizosphere soil samples respectively. Beta diversity analysis suggested that the bacterial community in the rhizosphere was distinctive from that in the bulk soil, which indicates that *B.cusia* can specifically recruit microbes from bulk soil and host in the rhizsosphere. Within these specially enrich bacteria, we found that Burkholderia was significantly enriched in the rhizosphere, which was a potential beneficial bacteria and had been reported to play a major role in the synthesis of major effective substances indigo in *B. cusia*. In addition, we found that Bacilli or Bacillus were depleted in the rhizosphere that was a beneficial microbe use for biocontrol soil–borne disease, and this may explain the continuous cropping obstacles in *Baphicacanthus cusia* planting. Our results revealed the structure and composition of bacterial diversity in *B. cusia* rhizosphere, and may provide clues for improving medical value of *B. cusia* in the future.

Acknowledgements: This work was supported by the Natural Science Foundation of China (Association of Science and technology cooperation across the Taiwan straits, Project No U1405215).

Keywords: *Baphicacanthus cusia* (NeeS) Bremek, Rhizosphere bacteria, 16S rRNA, Diversity

PS1-J-007

Investigation of sea fennel nutritional value under the effect of iodine biofortification using different metabolomics approaches

Eirini Sarrou[1], Anastasios S. Siomos[2], Samantha Riccadona[3], Pavlos Tsouvaltzis[2], Paschalina Chatzopoulou[1], Stefan Martens[4]

[1]Hellenic Agricultural Oranisation DEMETER, Thessaloniki, Greece, [2]Aristotle University, Department of Horticulture, Laboratory of Vegetables, Thessaloniki, Greece, [3]Fondazione Edmund Mach, Centro Ricerca e Innovazione, Department of Computational Biology, San Michele all'Adige (TN), Italy, [4]Fondazione Edmund Mach, Centro Ricerca e Innovazione, Department of Food Quality and Nutrition, San Michele all'Adige (TN), Italy

Sea fennel (*Crithmum maritimum L.*), also known as rock samphire, is a fleshy aromatic, perennial littoral and edible halophyte, that naturally thrives on maritime cliffs and sometimes in sand. Its succulent leaves and young branches are consumed as salads or pickled in vinegar. Besides its use as a gastronomic ingredient, sea fennel has been considered as a functional food largely used for nutritional and medicinal purposes, due to its high content in vitamins, carotenoids, flavonoids and other bioactive constituents. Considering that sea fennel is distributed in maritime areas, where atmospheric iodine is available in higher concentrations, the aim of this study was to evaluate whether sea fennel has the potential to accumulate iodine, through a biofortification experiment in a floating system, by the use of two iodine forms (KI, KIO_3) in concentrations of 20, 40 and 80 μM. In parallel, our main scope was to determine important substances that characterize the nutritional value of sea fennel, under the effect of iodine biofortification treatments. For this reason sea fennel tissues obtained from the hydroponic cultivation, subjected to various chromatographic analyses (LC-MS/MS, UPLC-DAD, GC-MS) in order to identify their iodine content, polyphenolic, volatile and carotenoid profile. According to our data, both iodine forms were able to be assimilated by sea fennel plants indicating a successful iodine biofortification potential. Seventeen polyphenolic constituents were identified in leaf and stems tissues through a targeted LC-MS/MS (MRM) analysis, with chlorogenic acid, cryptochlorogenic acid, neochlorogenic acid and 1,5-dicaffeoylquinic acid being the major ones. Three carotenoids were identified: lutein, neoxanthin and violaxanthanin while the main pigment constituents were chlorophyll a and b. Twenty-seven volatile compounds were detected belonging in four groups: hydrocarbons monoterpenes, oxygenated monoterpenes, sesquiterpenes and phenylpropenes. The main compounds characterizing sea fennel essential oil were sabinene, limonene and *γ*-terpinene.

PS1-J-008

A holistic approach to sustainable use of Greek native medicinal/aromatic plants' by-products to produce innovative feeds for domestic animals

Katerina Grigoriadou[1,4], Ilias Giannenas[2], Eftyxia Agapidou[4], Panagiotis Bagatzounis[3], Nektarios Aligiannis[5], Efterpi Christaki[2], Panagiota Florou-Paneri[2]

[1]Institute of Plant Breeding and Genetic Resources, Hellenic Agricultural Organization – Demeter, Thermi - Thessaloniki, Greece, [2]Laboratory of Nutrition, School of Veterinary Medicine, Faculty of Health Sciences, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [3]M. Bagatzounis & Sons S.A, Vatero, 50100, Kozani, Greece, [4]ELVIZ Hellenic Feedstuff Industry S.A., Plati-Imathia, 59300, Greece, [5]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, 15771, Athens, Greece

Cultivation of Greek Medicinal and Aromatic plants (GAMP)s is increasing in recent years at a rate of 15-20% annually. Among the plants grown are many native/endemics of the Greek flora with unique properties used to produce dry material for food seasoning, essential oils or plant extracts. Their processing produces by-products, some of which, although containing very same ingredients and properties with the final product they are considered unacceptable by the market. Up to date, those materials are treated as "waste" and discharged into the environment. The by-products constitute a huge pool of valuable metabolites with significant biological properties. An extensive assessment of the by-products derived from the processing of GMAPs was performed. Four cultivated species were studied *Origanum vulgare* subsp. *vulgare*, *Sideritis scardica*, *Thymus* spp. and *Matricaria chamomilla*. Mechanical separation divided plant material in different fractions depended on the species. Results showed that "wastes" were more than 60% for the first two species and over 40% for thymus and chamomile. These non-marketable raw materials were used for the development of packaged novel feeds for different categories of domestic animals (chickens/rabbits). The use of those materials in animal nutrition was based on their phenolic content which is positively related to their antioxidant and antimicrobial activity and on their fiber content; these by products can be considered as raw fiber concentrate products with high content in cellulose, hemicellulose and lignin and they can be used as supplementary feeds for animals. Several by-products of GAMPs will be studied whether they can be used to improve the economic efficiency of animal production, due to their low or zero cost. Due to their phenolic content, they will be investigated and classified as new sources of concentrates with high biological and functional value, for improving animal nutrition and sustainable production.

PS1-K-001

Green propolis: how resin collected by honey bees from *Baccharis dracunculifolia* could be influenced by the gender, number of galls and chemical components of this plant?

Debora Munhoz Rodrigues[1], Caroline Arruda[1], Jairo Kenupp Bastos[1], Rodrigo Augusto Santinelo Pereira[2]

[1]Faculty of Pharmaceutical Sciences of Ribeirão Preto, University of São Paulo, Ribeirão Preto, Brazil, [2] Faculty of Philosofy, Sciences and Letters of Ribeirão Preto, University of São Paulo, Ribeirão Preto, Brazil

Baccharis dracunculifolia is the main botanical source for the production of green propolis by honey bees. Green Propolis contains a complex and varied chemical composition rich in prenylated p-coumaric acid derivatives, which display important biological activities, and has drawn international interest. Thus, considering the increasing demand of green propolis, we investigated the interaction between bees, secondary metabolites of *B. dracunculifolia* and galling insects, aiming to increase the quality and production of green propolis. For that, 400 individuals of *B. dracunculifolia* were cultivated and 48 individuals, being 24 females and 24 males, were investigated for the degree of galls infestation, number of visiting bees and the time of resin collection by the bees. Volatile and phenolic compounds were analyzed by gas chromatography and liquid chromatography, respectively. Statistically significant differences were observed between male and female plants, with males showing higher infestation by galling insects and females' higher number of visiting bees, which also spent more time collecting resin. Correlations between *trans*-caryophyllene with number of galls and the time of resin collected were also observed. *Trans*-caryophyllene concentration was higher in females, contributing to the differences found in the field. The cultivation of *B. dracunculifolia* was successful in attracting high bee visitation and increasing the quality and green propolis production in a small area, as well as reducing the environmental impacts. In addition, it is suggested to increase the percentage of female *B. dracunculifolia* in the field, which would attract more bees for propolis production.

PS1-K-002

Natural products diversity of *Tithonia diversifolia* (Asteraceae) and its specialist insect herbivore *Chlosyne lacinia* (Nymphalidae)

Marilia Gallon[1], Eduardo Silva-Junior[1], Juliano Amaral[2], Norberto Lopes[1], Leonardo Gobbo-Neto[1]

[1]University Of São Paulo, Ribeirão Preto, Brazil, [2]Universidade Federal da Bahia, Vitória da Conquista, Brazil

Interactions between plants and insects are the most common type of plant-herbivore interactions and they shape community dynamics and ecosystem evolution. *Chlosyne lacinia* (Lepidoptera: Nymphalidae) is an olygophagous insect herbivore, which uses mainly host plants of the tribe *Heliantheae* of Asteraceae. Herein, plant-insect interaction in *Tithonia diversifolia* (tribe *Heliantheae*) and *Chlosyne lacinia* was investigated by means of chemical diversity and insect performance bioassays. Samples of *T. diversifolia* and *C. lacinia* were analyzed by untargeted LC-MS/MS based metabolomics and molecular networking. Additionally, the performance of *C. lacinia* feeding on leaves of *T. diversifolia* was measured. The survive rate for larvae of *C. lacinia* fed with *T. diversifolia* was 100% and larvae developed until fifth instar completing metamorphosis to the adult phase. Chemical profiles of *T. diversifolia* (leaves and non-consumed abaxial surface) and *C. lacinia* (eggs, larvae, larvae feces, pupae and butterflies) were clearly distinct. Molecular networking indicated metabolites that were unique to larvae feces and also metabolites that were shared between larvae feces and leaves of *T. diversifolia*. Therefore, *C. lacinia* larvae were able to metabolize some plant metabolites, while other metabolites were excreted intact. Moreover, eggs, butterflies and pupae of *C. lacinia* shared some metabolites. Our results highlight the natural products diversity in this plant-insect interaction and may contribute for a better understanding of their ecological roles.

PS1-K-003

Oregano spices in the European market: Taxonomic identification and quality control

Dimitrios Mertzanidis[1], Dimitrios Koureas[1,2], Andreana Assimopoulou[3], Stella Kokkini[1]

[1]Laboratory of Systematic Botany and Phytogeography, School of Biology, Aristotle University of Thessaloniki, Thessaloniki, Greece, [2]Naturalis Biodiversity Center, 2300RA, Leiden, the Netherlands, [3]Organic Chemistry Laboratory, School of Chemical Engineering, Aristotle University of Thessaloniki, Thessaloniki, Greece

Oregano is a very popular culinary herb widely used in everyday cooking of European citizens. Commercial samples of dried oregano sold in supermarkets, flea markets and traditional shops were purchased from different European countries. Based on micro–morphological diagnostic characters found in the ground samples, such as calyx form, bract colour, non–glandular hairs, density and size of sessile glands, two species of the genus *Origanum* (Lamiaceae family) have been identified, *O. onites* L. and *O. vulgare* L. The first is a native species of East Mediterranean and it is widely collected from the wild, whereas the latter has a much wider distribution all over Europe which it is also extended to Asia. Two distinct subspecies of *O. vulgare* have been to date identified in the commercial oregano sold in the European market, *O. vulgare* subsp. *vulgare* and *O. vulgare* subsp. *hirtum* (Link) Ietsw. Subsp. *hirtum* commercially known as Greek oregano, grows wild in SE Europe where it is also cultivated. It is clearly distinguished from subsp. *vulgare* by the numerous conspicuous sessile glands, easily seen using a stereoscope. The total essential oil content of the commercial oregano plants was measured using a Clevenger type apparatus. A high variation of the total essential content was recorded, with a minimum value of 0,2 mL $100g^{-1}$ d.w. in *O. vulgare* subsp. *vulgare*, purchased in Netherlands and a maximum of 5,2 mL $100g^{-1}$ d.w. in *O. vulgare* subsp. *hirtum* from Greece. Our results may contribute to the quality control of the commercial oregano.

Acknowledgements: This research has been supported by the project "Upgrading the Plant Capital (PlantUp)" (MIS 5002803) which is implemented under the Action "Reinforcement of the Research and Innovation Infrastructure", funded by the Operational Programme "Competitiveness, Entrepreneurship and Innovation" (NSRF 2014–2020) and co–financed by Greece and the European Union (European Regional Development Fund)

PS1-K-004

Volatile metabolomic analysis of the heartwood of *Pinus heldreichii* H. Christ: Chemodiversity insights

Elena Zioga, Olga Tsiftsoglou, Diamanto Lazari

Laboratory of Pharmacognosy, School of Pharmacy, Faculty of Health Sciences, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece

So far, most of the research on the chemodiversity of the species *Pinus heldreichii* H. Christ is concentrated on the needles and a certain chemotype has been created (Nikolić *et al.* 2015; Petrakis *et al.* 2001). Noteworthy differentiations in the smell of the heartwood from different trees growing in the same area in Greece were observed in terms of an ethnobotanic study in Pindos mountain range. Hence, the chemical profile of the essential oils from the heartwood of trees growing in the certain area was investigated. The composition of the essential oils was analyzed by Gas Chromatography–Mass Spectroscopy (GC–MS) and 60 volatile compounds were identified in total. Significant differences among the main chemical constituents of the samples were observed, which was presumably the reason why they smelled differently. The most abundant constituent was limonene ($\bar{x}$ = 28.4%), while the presence of *β*–longipinene was reported for the first time in the essential oil of the species. The ratios of the compounds *δ–3*–carene, limonene, terpinolene and terpinen–4–ol in the samples, seem to play an important role. Thus, in order to evaluate the relationship among some of the main compounds detected in the heartwood samples, a nonparametric correlation test was applied on the data and both negative and positive correlations were observed between those compounds. In conclusion, the results of the present work could be of great taxonomic significance, suggesting that the essential oil of the heartwood should also be considered when it comes to the evaluation of the chemotaxonomic markers for this species.

Keywords: *Pinus heldreichii*, Pinaceae, Chemodiversity, Wood, Essential oils

PS1-K-005

Evaluation of silymarin content and flavonolignans composition in native *Silybum marianum* populations from Greece

Dimitrios Arampatzis, Anestis Karkanis, Nikolaos Tsiropoulos

Department of Agriculture Crop Production and Rural Environment, University of Thessaly, Volos, Greece

Silybum marianum (L.) Gaertn, commonly known as milk thistle, is usually an annual species native to the Mediterranean basin and in Greece it is considered a weed. The fruits extract, known as silymarin, contains flavonolignans that display antioxidant, hepatoprotective and anticancer properties. The content of silymarin most often ranges from 1.5% to 3.0% of the seed dry weight. Milk thistle is cultivated as a medicinal plant in several countries in Europe and Asia mainly to produce silymarin, although it can be used for oil and bioenergy production [1].

Mature fruits collected from milk thistle plants growing in diverse areas of Greece with different environmental conditions in order to evaluate seventeen Greek milk thistle populations and to estimate the chemical diversity regarding the silymarin content and the flavonolignans composition. Constituents' analysis was carried out by HPLC after methanolic extraction of defatted fruits. Fruits oil content and antioxidant activity assays were also carried out. Mean oil content was 26.21±1.19%. The DDPH• radical scavenging activity ranged from 3093 to 4015 μmol ascorbic acid/100g dw. With respect to silymarin content, the results show a relatively high amount of variation among the studied populations. Highest silymarin content recorded in two populations (about 5%), while the plurality of populations ranged between 2.80% and 3.50%. Regarding flavonolignans composition, significant differences observed among the studied populations. Three chemotypes were observed regarding the dominant regioisomer; in type I the dominant flavonolignan was silybin (36-42%), while in type II and type III was isosilybin (38-40%) and silydianin (44% to 57%), respectively.

Keywords: Milk thistle; medicinal plant; oil; silymarin; flavonolignans.

References:

[1] Karkanis A, Bilalis D, Efthimiadou A. Ind Crops Prod 2011; 34: 825–830

PS1-K-007

An ethnobotanical study and phytochemical analysis of medicinal plants in the Greek islands of North Aegean region

Evangelos Axiotis, Maria Halabalaki, Sofia Mitakou, Leandros A. Skaltsounis

National and Kapodistrian University of Athens, Athens, Greece

Greek islands of the North Aegean Region are a group of 9 inhabited islands (Lemnos, Agios Efstratios, Lesvos, Chios, Psara, Oinousses, Samos, Ikaria and Fourni) in the northern part of the Aegean Sea, close to Asia Minor. Given the rich biological [1] and cultural heritage of each island, we have tried to evaluate the present status concerning the medicinal flora of the area and its uses in the everyday life of the inhabitants. We analyzed 109 wild plant species of medicinal importance, from 52 families, listing their uses for therapeutic purposes and galenic preparations provided by local medical doctors and pharmacists. Furthermore, we determined the phytochemical profile and the antioxidant activities of the extracts of specific plant species. This study confirmed the extensive indigenous knowledge on medicinal plants and their key role in folk medicine for this region. This ethnopharmacological survey in parallel with the phytochemical analysis of specific plants is a fundamental step for the preservation of the local knowledge, which could be of critical importance both for further scientific research and for the protection of endangered endemic medicinal plants of the area.

Acknowledgements: The research presented was carried out within the framework of a North Aegean Region specific grant, Greece NSRF 2014-2020 (National Strategic Reference Framework)

Keywords: ethnopharmacology, North Aegean Sea, ethnobotany, phytochemistry

References:

[1] Strid, A. (2016). Atlas of the Aegean Flora. Part 1: Text & plates. Part 2: Maps. Berlin: Botanic Garden and Botanical Museum Berlin.

Topic B: Bioactive Natural Products

PS2-B-001
Activated charcoal promotes surgical wound healing effects of *Musa sapientum* and *Citrus limon* peel gel in *Rattus novergicus*
James Omale

PS2-B-002
Antibacterial, anti-inflammatory and peroxidase-mediated cyclooxygenase-1 inhibitory properties of *Fusarium Solani* extract
Kenneth Ngwoke, Tochichukwu Nwalusiuka, Chinechem Ekwealor, Valerie Nwankwo, Uju Obi-okafor, Chisom Izundu, Festus Basden Okoye, Charles Esimone, Peter Proksch, Chen Situ

PS2-B-003
A new benzophenone glycoside from the leaves of *Mitracarpus villosus*
Kenneth Ngwoke, Njideka Orame, Shuai Liu, Festus Okoye, Georgios Daletos, Peter Proksch

PS2-B-004
Bioactive compounds from *Achyranthes aspera* (chirchitta)
Rashmi Sehrawat

PS2-B-005
Anti-inflammatory activity of the indonesian propolis and its molecular marker
Muhamad Sahlan, Andrea Davina, Herbert Situmorang

PS2-B-006
Chemical composition of essential oil from *Schinus molle* L and their antithrombotic properties
Douglas Chaves, Rosiane da Silveira, Alessandra Guedes, Flavia Frattani, Marco Andre Santos

PS2-B-007
Gelatin-based films additivated with mind essential oil and red propolis ethanolic extract as active material: Microbiological and antioxidant activity
Guillermo Salamanca Grosso, Laura Maria Reyes Mendez, Mónica Patricia Osorio Tangarife, Eng Paola Andrea Mayorquin Súarez

PS2-B-008
Food phytochemicals modulating autophagy in human iPS cells
Izumi Sasanuma, Takashi Tsukahara, Ayato Ohashi, Kyouya Shigeta

PS2-B-009
Enhanced cell adhesion and proliferation of skin fibroblasts by ethanolic extracts of two genotypes of *Ugni molinae* Turcz. leaves
Paula Valenzuela-Bustamante, Pablo Parra, Rubén Veas, Ivette Seguel, Guillermo Díaz-Araya, Carla Delporte

PS2-B-010
Metabolic profiling and wound-healing activity of *Boerhavia diffusa* leaf extracts
Kriti Juneja, Rutusmita Mishra, Partha Roy, Debabrata Sircar

PS2-B-011

Exploring CADD strategies for identification of natural agonists against PPAR-gamma for breast cancer

Pawan Kumar Gupta, Aishwarya Laxmiaraya, Jeena Gupta, Aleksandrs Gutcaits, Prabha Garg

PS2-B-012

Anti-inflammatory properties of betulinic acid and xylopic acid in the carrageenan-induced pleurisy model of lung inflammation in mice

Edmund Ekuadzi, Robert Peter Biney, Charles Kweku Benneh, Bismark Osei Amankwaa, Jonathan Jato

PS2-B-014

Spice-derived bioactive compounds: potential agents or food adjuvants in the management of diabetes mellitus?

Shahidul Islam, Aminu Mohammed

PS2-B-015

***In vitro* alpha-glucosidase and glycogen phosphorylase an inhibition by chilean propolis ethanolic extracts**

Ruben Veas, Paula Valenzuela-Bustamante, Gabriela Valenzuela-Barra, Carla Delporte

PS2-B-016

Modulation of doxorubicin-induced renal and hepatotoxicity by *Calligonum comosum* extract via dual mechanism

Doaa Abdelhady, Emad Ghazi, Walied Abdo, Zizy Elbialy, Mustafa Shukry, Engy Mahrous, Sendker Jandirk, Essam Abdel-Sattar

PS2-B-017

Neurotrophic compounds of Javanese ginger bangle, *Zingiber purpureum*

Yoshiyasu Fukuyama, Miwa Kubo, Megumi Nakai, Kenichi Harada, Nobuaki Matsui, Midori Suenaga, Yoichi Matsunaga, Mitsuhiko Miyamura, Eishin Kato

PS2-B-018

The effects of gamisoyo-san decoction, a traditional chinese herbal medicine, on the pacemaker potentials in murine small intestinal interstitial cells of cajal

Doeun Kim, Jung Nam Kim, Joo Hyun Nam, Jong Rok Lee, Sang Chan Kim, Byung Joo Kim

PS2-B-019

Amaryllidaceae alkaloids from *Narcissus pseudonarcissus* cv. Dutch Master as potential drugs in treatment of Alzheimer´s disease

Daniela Hulcová, Jana Maříková, Lubomír Opletal, Angela De Simone, Lucie Cahlíková

PS2-B-020

From traditional use to standardized neuroprotective green-extract of *Huperzia serrata*

Elnur Garayev, Laurent Boyer, Philippe Poindron, Maud Combes, Noëlle Callizot

PS2-B-021

Phytosterol stability to thermal treatments in Bronte's Pistachio (*Pistacia vera*, L.) and in Pistachio processed products

Massimo Lucarini, Laura D'Evoli, Gabriella Di Lena, Ginevra Lombardi Boccia

PS2-B-022

Antifungal and cytotoxic activity of commercially-available essential oils

William Setzer, Chelsea Powers, Jessica Osier, Emily Olsen, Brianne Brazell, Prabodh Satyal, Robert McFeeters, Debra Moriarity

PS2-B-023

Ecoextraction of Mediterranean natural resources to develop innovative cosmetic ingredients

Pauline Burger, Hortense Plainfossé, Grégory Verger-Dubois, Xavier Fernandez

PS2-B-024

Development of an innovative natural cosmetic ingredient derived from *Teucrium lucidum* for skin repair

Hortense Plainfossé, Pauline Burger, Stéphane Azoulay, Anne Landreau, Grégory Verger-Dubois, Xavier Fernandez

PS2-B-025

Anti-neuroinflammatory compounds from ethnic medicinal plant *Tinospora sinensis* in Guangxi province of China

Haibing Liao, Dong Liang

PS2-B-026

Chemical interactions between *Fusarium oxysporum* and an endophytic fungus *Xylaria* sp. with antifungal activities

Morgane Barthélemy, Laurent Intertaglia, Didier Stien, David Touboul, Véronique Eparvier

PS2-B-027

Wound healing effect of some tropical fruits

Linda Chularojmontri, Khwandow Kunchana, Orapin Wongsawatkul, Wattanased Jarisarapurin, Suvara Wattanapitayakul

PS2-B-028

Anti-proliferative effects of *Ceratonia siliqua* L. (Carob) fruit extracts on breast cancer cells

Christiana Neophytou, Alexandru Vasincu, Eve Ioannou, Antonia Matsentidou, Atalanti Christou, Haria Hadjipakkou, Andreas Constantinou

PS2-B-029

Effects of *Carica papaya* seeds on acetaminophen-induced hepatotoxicity in male albino Wistar rats

Edisua Itam, Isaac Atakpa, Eneji Egbung

PS2-B-030

A natural oxadiazine isolated from cyanobacteria kills cancer cells in multicellular culture systems by impairing cellular respiration

Maria Lígia Sousa, Marco Preto, Rosário Martins, Vítor Vasconcelos, Stig Linder, Ralph Urbatzka

PS2-B-031

Analyses of the biological activities and the mode of action of marine lipopeptide jahanyne and its analogs

Shizuka Hoshina, Arihiro Iwasaki, Takafumi Kudo, Toshiaki Teruya, Kiyotake Suenaga

PS2-B-032

Lignans in male and female *Schisandra rubriflora* soil-grown plants and in their *in vitro* cultures

Agnieszka Szopa, Michał Dziurka, Marta Klimek-Szczykutowicz, Paweł Kubica, Angelika Warzecha, Halina Ekiert

PS2-B-033

***In vitro* chrysin nanoparticles primary hippocampal cells protection under Cu(II) neurodegeneration conditions**

Christiane M. Nday, Graham Jackson, Athanasios Salifoglou

PS2-B-034

Naringin magnetic silica nanoparticles against amyloid-induced oxidative stress

Christiane M. Nday, Graham Jackson, Athanasios Salifoglou

PS2-B-035

Clavariopsins C-H, antifungal cyclic depsipeptides from the aquatic hyphomycete *Clavariopsis aquatica*

Thin Wut Soe, Chunguang Han, Tomohiko Tomura, Ryosuke Fudou, Makoto Ojika

PS2-B-036

Discovery of new GPBAR1 agonists by ligand-based pharmacophore modeling and virtual screening

Benjamin Kirchweger, Jadel M Kratz, Angela Ladurner, Ulrike Grienke, Verena M. Dirsch, Thierry Langer, Judith M. Rollinger

PS2-B-037

Butanol fraction of *Ficus carica* fruit enhances pancreatic *β*-cell functions, stimulates insulin secretion and ameliorates other type 2 diabetes-associated parameters in rats

Shahidul Islam, Ramgopal Mopuri, Chika I. Chukwuma, Balaji Meriga, Dowlathabad Muralidhara Rao

PS2-B-038

An antifungal substance from the endophytic fungi *Epichloë festucae* and its effective production by gene modification

Enkhee Purev, Jennifer Niones, Daigo Takemoto, Tatsuhiko Kondo, Hitomi Isobe, Rika Miura, Takushi Hashikawa, Kazuhito Kawakita, Makoto Ojika

PS2-B-039

***Caenorhabditis elegans* as model to study natural products affecting metabolism and lifespan**

Theresa Lehner, Benjamin Kirchweger, Julia Zwirchmayr, Ammar Tahir, Judith M. Rollinger

PS2-B-040

Evaluation of Mistletoe (*Viscum album* L.) callus and plant protein extracts against gynecological cancer cell lines

Vasileios Tsekouras, Georgia Moschopoulou, Sophia Mavrikou, Maria-Argyro Karageorgou, Spyridon Kintzios

PS2-B-041

Essential oil characterisation, antimicrobial and antioxidative activity of extracts of *Erodium cicutarium* (L.) L'Hér. ex Aiton (Geraniaceae) from Croatia

Vanja Ljoljić Bilić, Ivan Kosalec, Dario Kremer, Valerija Dunkić, Jadranka Vuković Rodríguez

PS2-B-042

Chemical-biological study of ent-kaurane and ent-trachylobane diterpenes from *Psiadia punctulata*

Lorenzo Fiengo, Antonio Vassallo, Roberta Cotugno, Nunziatina De Tommasi, Fabrizio Dal Piaz, Ammar Bader

PS2-B-043

Sameuramide A, a new cyclic depsipeptide isolated from an ascidian of the family Didemnidae

Koshi Machida, Daisuke Arai, Ryosuke Katsumata, Satoshi Otsuka, Jun K. Yamashita, Tao Ye, Shoubin Tang, Nobuhiro Fusetani, Yoichi Nakao

PS2-B-044

Narciclasine of *Cyrtanthus contractus* has *in vitro* anti-inflammatory properties identified by correlation-based metabolomics

Lucie Rarova, Bhekumthetho Ncube, Johannes Van Staden, Robert Fürst, Miroslav Strnad, Jiri Gruz

PS2-B-045

Antiplasmodial activity and interaction between cannabidiol and artemisinin

Lúcia Mamede, Gilles Degotte, Allison Ledoux, Olivia Jansen, Pauline Desdemoustier, Michel Frédérich

PS2-B-046

Physicochemical characterization and stability assessment of oil-in-water nanoemulsions as delivery system of bioactive compounds: Conjugated linoleic acid (CLA) as model compound

Maria Katsouli, Virginia Giannou, Constantina Tzia

PS2-B-047

Assessment of phytopathogenic fungi diseases removal and plant protection induction by plant-associated-bacteria of *Vicia faba* L. plant: Broad spectrum of bioactive metabolites for antifungal and plant growth promoting activities

Imen Haddoudi, Isabel Mora, Jordi Cabrefiga, Emilio Montesinos, Moncef Mrabet

PS2-B-048

Toxicity, antioxidant and anti-asthmatic studies of *Anchomanes difformis* (Blume) Engl. leaf extract

Oghale Ovuakporie-uvo, MacDonald Idu

PS2-B-049

***Erythrina* alkaloids as source of valuable chemicals**

Marcos Soto-Hernandez, Rosario Garcia-Mateos, Ruben San Miguel-Chavez, Geoffrey Kite

PS2-B-050

Two types of lipid elicitors with different functions from the potato pathogen *Phytophthora infestans*

Makoto Ojika, Ryo Murata, Shiho Tenhiro, Mohammad S. Monjil, Kentaro Matsuda, Daigo Takemoto, Kazuhito Kawakita

PS2-B-051

Propolis ethanolic extract induces apoptosis and cell cycle arrest on certain cancer cell lines in a p21 and cyclin D1 dependent manner

Başak Aru, Etil Güzelmeriç, Aslı Akgül, Gülderen Yanıkkaya Demirel, Hasan Kırmızıbekmez

PS2-B-052

Antidiabetic and antimicrobial properties of three Sri Lankan medicinal plants: *Phyllanthus emblica*, *Cassia auriculata* and *Hemidesmus indicus*

Dhanushki Wickramarachchi, Rizliya Visvanathan, Amriya Nizar, Majed Bawazeer, Bathini Tissera, M. Mallique Qader, Mostafa E. Rateb, Ruvini Liyanage

PS2-B-053

Stilbenes of *Yucca gloriosa* L. and their antioxidant, pro-apoptotic and anti-proliferative activities

Aleksandre Skhirtladze, Mariam Benidze, Ether Kemertelidze

PS2-B-054

Therapeutic effect of total phenolic fraction of extra virgin olive oil against murine experimental cutaneous leishmaniasis

Kalliopi Karampetsou, Olga Koutsoni, Nektarios Aligiannis, Maria Halabalaki, Leandros Alexios Skaltsounis, Eleni Dotsika

PS2-B-055

Dynamic of phenolic acids production in *Aronia* × *prunifolia* (Marshall) Rehder agitated shoot cultures during the growth cycles

Agnieszka Szopa, Paweł Kubica, Joanna Żywko, Halina Ekiert

PS2-B-056

Oleocanthal exerts *in vivo* antileishmanial and immunomodulatory effect against murine experimental cutaneous leishmaniasis

Kalliopi Karampetsou, Olga Koutsoni, Apostolis Angelis, Nektarios Aligiannis, Maria Halabalaki, Leandros Alexios Skaltsounis, Eleni Dotsika

PS2-B-057

Sensory perception and bitterness masking of olive polyphenols in fortified mayonnaise

Petroula Tsitlakidou, Giorgos Nasioudis, Ioannis Mourtzinos, Costas G. Biliaderis

PS2-B-058

Dynamic of flavonoids production in agitated shoot cultures of three *Hypericum perforatum* L. cultivars 'Elixir', 'Helos' and 'Topas' during the growth cycles - preliminary results

Inga Kwiecień, Natalia Więckowska, Ludger Beerhues, Halina Ekiert

PS2-B-059
Discovery and characterization of new natural products from promising Cyanobacteria
Sara Freitas, Teresa P Martins, Pedro N Leão

PS2-B-060
Evaluation of cytotoxic potential of *Jurinea macrocephala* DC. on A-549 and MCF-7 cell lines
Perihan Gürbüz, Şengül Dilem Doğan, Ebru Öztürk, Mükerrem Betül Yerer Aycan

PS2-B-061
Chemical composition of a nephroprotective ethanolic extract of *Pistacia lentiscus* L. fruits
Nassima Cheraft-Bahloul, Cécile Husson, Meriam Ourtioualous, Sébastien Sinaeve, Cédric Delporte, Djebbar Atmani, Caroline Stévigny, Joëlle L. Nortier, Marie-Hélène Antoine

PS2-B-062
Phytochemical analyses of three endemic Boraginaceae plants from Turkey: *Phylocarra aucheri*, *Symphytum anatolicum*, *Cynoglottis barrelieri*. Biological activities
Elisavet Varvouni, Konstantia Graikou, Gokhan Zengin, Christos Ganos, Tomasz Mroczek, Ioanna Chinou

PS2-B-063
Bioassay-guided isolation, identification and cytotoxicity of diterpenoids from *Justicia insularis*
Idowu Eniafe Fadayomi, Okiemute Rosa Johnson-Ajinwo, Alexander Kagansky, Ted Hupp, Nick Forsyth, Wen-Wu Li

PS2-B-064
Ethnomedicinal survey of plants used in the treatment of malaria in Southern Nigeria
Macdonald Idu, Pass Iyamah

PS2-B-065
Synthesis and evaluation of novel hybrid antioxidant peptides
Athina Lykoura, Nicky Panousi, Virginia Dimaki, Nikolaos Assimomytis, Fotini Lamari, Vassiliki Magafa

PS2-B-066
Assessment of tyrosinase inhibitory effects and antioxidant properties of *Glaucium corniculatum* (L.) Rud. subsp. *refractum* (Nab.) Cullen and *Glaucium Ieiocarpum* Boiss. growing in Turkey
Fatma Ayaz, Yavuz Bağcı, Nuraniye Eruygur, Esra Maltaş, Hamide Filiz Ayyıldız, Cengizhan Ceylan

PS2-B-067
***In vitro* biological effects of hydroxytyrosol on human hepatoma HepG2 cells**
Georgia Gogou, Andriana Kavallari, Maria Lypiridou, Haralambos Stamatis, Constantinos Pantos

PS2-B-068

Anticancer effects of extracts from aerial part of *Glinus oppositifolious* (L.). Aug. DC.

Alexandru Vasincu, Veronica Bild, Christiana M Neophytou, Christiana Charalambous, Ioana-Mirela Vasincu, Bogdan-Gabriel Şlencu, Andreas Ioannou Constantinou, Anca Miron

PS2-B-069

Acaricidal activity of constituents from *Gleditsia japonica* against *Dermanyssus gallinae*

Jin ah Kim, Yun-Hyeok Choi, Ji Eun Lee, Yeon Woo Jeong, Changon Seo, Jin Gwan Kwon, Jin Kyu Kim, Seong Su Hong, Wonsik Jeong, Chun Whan Choi

PS2-B-070

Biomonitored chemical study of extracts from *Pouteria ramiflora* for *in vitro* cytotoxic activity in human prostate cancer cells

Sarah Cardoso, Matheus Andrade, Vinícius Cunha, João Victor Gomes, Cristian Silva, Mayra Leão, Christopher Fagg, Eliete Guerra, Francisco Neves, Luiz Simeoni, Dâmaris Silveira, Adriana Lofrano-Porto

PS2-B-071

Antifungal activity of *Sapindus saponaria* L.

Cristian Aldemar Gasca Silva, Marline Dassoler, Guilherme Brand, Christopher W Fagg, Pérola O. Magalhães, Yris M Fonseca-Bazzo, Sueli Maria Gomes, Dâmaris Silveira

PS2-B-072

Ent-clerodanes and other constituents from the bark of *Croton oligandrus* (Euphorbiaceae) and evaluation of their cytotoxicity

Stephanie T Guetchueng, Lutfun Nahar, Kenneth J Ritchie, Fyaz M D Ismael, Nicola M Dempster, Andrew R Evans, Satyajit D Sarker

PS2-B-073

Potential anti-hyperglycemic effects and chemical characterization of *Senecio clivicolus*

Immacolata Faraone, Flavio Prinzo, Dara Kirke, Lucia Chiummiento, Eloy Fernandez, Dilip K Rai, Luigi Milella

PS2-B-074

Studies on flavonoids in *Schisandra rubriflora* male and female plant raw material and in their *in vitro* cultures

Agnieszka Szopa, Marta Klimek-Szczykutowicz, Paweł Kubica, Angelika Warzecha, Sebastian Granica, Halina Ekiert

PS2-B-075

Verbascoside neuroprotective potential through enzyme inhibition and radical scavenging activity

Nuria Acero, Inmaculada Zafra, Dolores Muñoz Mingarro, Carmen Martón Cordero

PS2-B-076

Insulin secretory, antibacterial and antiproliferative activity of flavonoidal isolates from *Pseudarthria hookeri* Wight & Arn.

Joseph Tchamgoue, Simeon Kouam F, Jean Paul Dzoyem, Muhammad H Rahman, Udo Bakowsky, Muhammad Iqbal Choudhary

PS2-B-077

Stability of phenolic compounds and antioxidant capacity of carob pulp products subjected to simulated *in vitro* digestion

Vlasios Goulas, Andriani Hadjisolomou

PS2-B-078

Natural phthalides as mono and dimer in *Kelussia Odoratissima Mozaff*.: insight from chemistry to biological activity

Mahdi Ayyari, Mohammad Rezaei, Safiollah Raeissi, Maxime Le Bot, Aniseh Alimohammadpour, Shahram Eisa-Beygi, Sara Pahlavan, Gilda D'urso, Mahya Hosseini, Mehrnaz Namiri, Alexander D Crawford, Dimitri Bréard, Yaser Tahamtani, Seyed Hamzeh Hossieni Kahnouj, Ingrid Freuze, Sonia Piacente, Pascal Richomme, Hossein Baharvand

PS2-B-079

Traditional herbal remedy revisited - the example of bioactive compounds from carnivorous plants combined with silver nanoparticles used to combat *Staphylococcus aureus*

Marta Krychowiak, Anna Kawiak, Magdalena Narajczyk, Agnieszka Borowik, Eng. Aleksandra Krolicka

PS2-B-080

Biological properties of *Tasmannia lanceolata* CO_2 extract

Marina Humbert, Emmanuelle Gaillard

PS2-B-081

Response of *Bradyrhizobium japonicum* nodule variables to cucurbitacin-containing phytonematicides in cowpea (*Vigna unguiculata*) on N-deficient soil

Kola M. E., Mashela P. W., Lukhele-Olorunju P.

PS2-B-082

Actiscent®: boosting cosmetic activity with a fragrance

Anthony Pegard

PS2-B-083

Cucurbitacin A affects mobility of *Meloidogyne incognita*

Zakheleni Dube

PS2-B-084

Effects of fermented *Cucumis africanus* and *Momordica balsamina* extracts on *Pseudococcus citri*

Zakheleni Dube, Foster Mokhoelele

PS2-B-085

The effect of natural hydroxynaphthoquinones on 3T3-L1 cells and adipogenic differentiation

Athanasios Arampatzis, Olga Tsave, Vassilios P Papageorgiou, Andreana N Assimopoulou

PS2-B-086

Anti-fouling potential from the LEGE culture collection (LEGE-CC)

Joana Azevedo, Jorge Antunes, Pedro Leão, Vítor Vasconcelos

PS2-B-087

α-glucosidase inhibitory activity of isolated compounds from the endophytic fungi *Annulohypoxylon stygium* derived from the leaves of *Pandanus* simplex

Roberth Riggs Rondilla, Thomas Edison de la Cruz, Fang-Rong Chang, Maribel Nonato

PS2-B-088

***Physalis angulata* calyces fraction promoted mice recovery in AOM/DSS-induced colon cancer model**

Luis Franco, Yanet Ocampo, David Rivera, Daneiva Caro, Johana Cuadro, Jenny Castro, Coralia Osorio

PS2-B-089

Cytotoxic activity of an enriched fraction of *Physalis angulata* calyces against two human colon cancer cell lines

Daneiva Caro, Yanet Ocampo, Johana Cuadro, David Rivera, Guillermo Schinella, Ruben Salas, Luis Franco

PS2-B-090

Cape gooseberry (*Physalis peruviana*) modulates the release of inflammatory mediators in LPS-activated RAW 264.7 macrophages

Jenny Castro, Daneiva Caro, Nely Mejia, Jaime Salgado, Daniela Amaris, Jesus Cortez, Luis Franco, Ruben Salas

PS2-B-091

Synthetic cannabinoids AM–251 and AM–1241 induce apoptosis in prostate cancer cells

Sotia Louka, Christiana Neophytou, Andreas Constantinou

PS2-B-092

***In vitro* anti–proliferative and apoptosis–inducing activity of *Rhodiola rosea* L. against human glioblastoma cells**

Aney Marchev, Iliyan Manoylov, Gabriela Boneva, Silviya Bradyanova, Ivanka Koycheva, Aney Tchorbanov, Milen Georgiev

PS2-B-094

Field assessment of a carvacrol rich essential oil as a multitask mosquito control agent

Epameinondas Evergetis, Romeo Bellini, George Balatsos, Marco Carrieri, Rodolfo Veronesi, Arianna Puggioli, Vassiliki–Nafsika Kapsaski–Kanelli, Serkos Haroutounian, Dimitrios Papachristos, Antonios Michaelakis

PS2-B-095

Evaluation of *Stevia rebaudiana* extracts as potential active ingredients for natural cosmetics

Katarzyna Gaweł–Bęben, Wirginia Kukuła–Koch, Marcelina Strzępek, Uliana Hoian, Paweł Osika, Kazimierz Głowniak

PS2-B-096

Magnetic nano–formulations for drug delivery applications of natural polyphenol–metal complexes against cancer

Eleftherios Halevas, Maria Pelecanou, Marina Sagnou, Barbara Mavroidi, Anastasia Pantazaki, Antonios Hatzidimitriou, George Katsipis, Theodore Lialiaris, Dimitrios Chronopoulos, Athanasios Salifoglou, George Litsardakis

PS2-B-097

Valorization of citrus juicing industries by–products

Eleni Myrtsi, Epameinondas Evergetis, Grigor Kheiranov, Sofia Koulocheri, Serkos Haroutounian

PS2-B-098

Exploitation of plant derived essential oils as potent means for the control of *Sitophilus oryzae*

Apostolia Tzimopoulou, Dimitrios Papachristos, Antonios Michaelakis, Epameinondas Evergetis, Serkos Haroutounian

PS2-B-099

Smell the Roses: Rosaceae family plants extracts as potent natural antiviral agents against influenza

Anna Apostolou, Olympia Vangelatou, Sandra Liekens, Serkos Haroutounian

PS2-B-100

Exploitation of Rosaceae Family plants extracts as potent natural antiviral agents

Anna Apostolou, Sandra Liekens, Sofia Koulocheri, Serkos Haroutounian

PS2-B-101

PlantUp – Upgrading the plant capital | Aristotle University of Thessaloniki (AUTh) Core Node

Andreana Assimopoulou, Vassilios Papageorgiou, Stella Kokkini, Maria Tsimidou, Assoc. Diamanto Lazari, Dimitrios Vlachos, Dimitrios Christofilos

PS2-B-102

The roles of triterpenes and phytosterols in Talang Mamaks' traditional medicines of Indonesia

Hilwan Yuda Teruna, Adel Zamri, Jasril Karim, Titania T. Nugroho, Yum Eryanti, Muhamad Almurdani, Kamal Rullah

PS2-B-103

Antifungal alkaloids from the aerial parts of *Glaucium corniculatum* subsp. *refractum* against *Candida albicans*

Mohsen Bagheri, Samad Nejad Ebrahimi, Ali Nazarirad, Atousa Aliahmad, Emerson F. Queiroz, Pierre–Marie Allard, Laurence Marcourt, Abdulelah Alfattani, Jean–Luc Wolfender

PS2-B-104

The potential of macroalgae to be used as a functional ingredient in meat products

Ana Rita Circuncisão, Catarina Marçal, Carla Monteiro, Artur Silva, Susana M. Cardoso

PS2-B-105

Impact of *Fucus vesiculosus* aqueous extracts as ingredients in a vegetarian "alheira"

Catarina Marçal, Ana Rita Circuncisão, Carla Monteiro, Artur Silva, Susana Cardoso

PS2-B-106

Nutritional and phytochemical content of hemp seeds (*Cannabis sativa* L.) from Finola cultivar grown in Greece

Maria Irakli, Eleni Tsaliki, Apostolos Kalivas, Eirini Sarrou

PS2-B-107
Variation of phenolic compounds in Greek oregano (*O. vulgare* L. subsp. *hirtum*) accessions
Eirini Sarrou, Paschalina Chatzopoulou, Maria Irakli

PS2-B-108
Identification of piperine based P-gp inhibitors with decreased cross reactivity with CYP3A4
Safiulla Basha Syed, Hemant Arya, Hsing-Pang Hsieh, Mohane Coumar Selvaraj

PS2-B-109
Isolation and characterization of terpenoids from the root extracts of *Euphorbia grandicomita*
Douglas Magozwi, Douglas M. Kemboi, Jacqueline Tembu, Xolani Peter, Moses Langat

PS2-B-110
Chemical characterization, biological assessment and molecular docking studies of essential oil of *Ocimum viride* for potential antimicrobial and anticancer activities
Monica Sangral, Madhulika Bhagat

PS2-B-111
Effects of the *Thumbergia laurifolia* Lindl. extract on gastric ulceration in rats
Linda Chularojanamontri

PS2-B-112
Strategies for the sustainable production of novel high-value terpenoids
Norbert Mehlmer, Wolfgang Mischko, Thomas Brück

PS2-B-113
Phytochemical and pharmacological investigation on medicinal plants of different origin
Syed Imran Hassan, Majekodunmi Fatope, Sabira Begum, Essam Ads, Saravanan Rajendrasozhan

PS2-B-114
Tissue specific effects of sheep/goat whey protein on antioxidant enzymes in rats
Efthalia Kerasioti, Maria Gasdrogka, Ioannis Taitzoglou, Demetrios Kouretas

PS2-B-115
Effects of feed supplemented with olive oil mill wastewater on detoxification enzymes in lamb tissues
Sotiria Makri, Sofia Raftopoulou, Demetrios Kouretas

PS2-B-116
LC-MS fingerprinting and chemical analysis of TNF-α, IL-6 and IL-1β inhibitory culture broth extract of *Pseudomonas* species
Kirti Hira, Pragya P Pal, A Sajeli Begum

PS2-B-117
Coffee improves rat redox status by increasing GSH biosynthesis
Alexandros Alexandros, Vasiliki Soursou, Anthi-Styliani Makiou, Fotis Tekos, Ioannis Taitzoglou, Dimitrios Kouretas

PS2-B-118
Vasodilating activity of *Hypericum revolutum* through nitric oxide (NO) synthase-dependent pathway
Noha Timraz, Hossam M. Abdallah, Hany A. El-Bassossy, Sabrin R. Ibrahim, Ali M. El-halawany, Ibrahim A. Shehata

PS2-B-119
Expanding the scope of biosynthetic cooperativity between polyketide megasynthases through genome mining
Michio Sato, Yi Tang, Kenji Watanabe

PS2-B-120
The mechanism of action of zingerone in the pacemaker potentials of interstitial cells of Cajal isolated from murine small intestine
Jeong Nam Kim, Hyun Jung Kim, Iksung Kim, Yun Tai Kim, Byung Joo Kim

PS2-B-121
On the relevance of DPPH, ABTS and FRAP assays for the evaluation of antioxidant properties of natural extracts
Radu Claudiu Fierascu, Alina Ortan, Irina Fierascu, Cristina Elena Dinu-Pirvu, Ioana Catalina Fierascu

PS2-B-122
Application of gamma radiation for the development of "green" products
Irina Fierascu, Radu Claudiu Fierascu, Eduard Marius Lungulescu, Nicoleta Nicula, Raluca Somoghi, Sorin Marius Avramescu, Camelia Ungureanu, Anca Nicoleta Sutan, Liliana Cristina Soare

PS2-B-123
Labdane diterpenoids from *Leonotis ocymifolia* with potential anticancer activity
Jane Ncongwane, Comfort Nkambule, Vuyelwa Tembu, Gerda Fouche, Nyeleti Vukea, Adrienne Edkins, Jo-Anne de la Mare

PS2-B-124
***In vitro* antitumor evaluation of different extracts of medicinal plants**
Majed Bawazeer, Bathini Thissera, Mallique Qader, Mostafa Rateb E., Mohammed Yaseen

PS2-B-125
Assessment of the antioxidant and antimutagenic profile from a total polyphenolic olive oil extract and its polyphenols separately
Paraskevi Kouka, Fotios Tekos, Alexios-Leandros Skaltsounis, Demetrios Kouretas

PS2-B-126
Anti-inflammatory and antiproliferative compounds from *Sphaeranthus africanus*
Huyen Thi Tran, Xuehong Gao, Nadine Kretschmer, Eva-Maria Pferschy-Wenzig, Olaf Kunert, Loi Huynh, Rudolf Bauer

PS2-B-127
Dual enzymes inhibitor of tiger milk mushroom (*Lignosus rhinocerus*) extracts on HIV-1 protease and reverse transcriptase
Chanin Sillapachaiyaporn, Siriporn Chuchawankul

PS2-B-128
Isolation of a new bi-flavonoid from *Rhus leptodictya* leaves extract
Tshifhiwa Matamela

PS2-B-129
***M. charantia*: short review on its phytochemicals and potential therapeutic use**
Alina Ortan, Daniela Ionescu, Simona Spinu

PS2-B-130
Anti-proliferative activity of melampolide-type sesquiterpene lactones from *Enhydra Fluctuans* on cancer cell lines
Thanh Triet Nguyen, Nadine Kretschmer, Eva-Maria Pferschy-Wenzig, Olaf Kunert, Rudolf Bauer

PS2-B-131
Secondary metabolites from the leaves and seeds of *Centaurea vlachorum* (Asteraceae) and their biological activity
Entela Çeliku, Olga Tsiftsoglou, Erjon Mamoci, Sokol Abazi, Lulezim Shuka, Dimitra Hadjipavlou-Litina, Diamanto Lazari

PS2-B-132
Antioxidant properties and safety use of goutweed
Karolina Jakubczyk, Katarzyna Janda, Katarzyna Watychowicz, Justyna Kałdyńska, Daniel Styburski, Karolina Dec

PS2-B-133
Lifespan extending and oxidative stress resistant properties of *Anacardium occidentale* L. leaf extract in *Caenorhabditis elegans*
Chatrawee Duangjan, Tewin Tencomnao, Micael Wink

PS2-B-134
Goutweed infusions as a source of antioxidants
Katarzyna Janda, Karolina Jakubczyk, Kamila Dębia, Katarzyna Watychowicz, Justyna Kałduńska, Daniel Styburski, Karolina Dec

PS2-B-135
Dietary quercetin impacts the concentration of tau-fluvalinate in honey bees (*Apis mellifera*)
Hamidreza Ardalani, Nanna Hjort Vidkjær, Per Kryger Inge S. Fomsgaard

PS2-B-136
Antioxidant and lifespan extension effects of *Caesalpinia mimosoides* extracts in nematode *Caenorhabditis elegans*
Panthakarn Rangsinth, Anchalee Prasansuklab, Michael Wink, Tewin Tencomnao

PS2-B-138

The use of innate microflora of honey in fermented milk drink

Hosam Baksamawi, Izlem Haktanir, Dimitrios Kafetzopoulos, Georgios Kafetzopoulos, Kostas Gkatzionis

PS2-B-139

Synthesis of novel artemisinin dimers and hybrids with other antimalarial agents

Dionissia Pepe, Dimitra Toumpa, Christophe Menendez, Christiane André-Barrès, Christina Koumpoura, Michel Baltas, Constantinos Athanassopoulos

PS2-B-140

Abietane diterpenes - high value natural precursors for the development of new antimicrobial agents

Antonia Antoniou, Macarena Funes Chabán, Catherine Karagianni, Claudia Solá, Despina Palla, Myrsini-Irene Tachliabouri, Maria C. Carpinella, Constantinos Athanassopoulos

PS2-B-141

Investigation of the *in vivo* oral acute toxicity and genotoxicity of Chios mastic gum in male Wistar rats

Eirini-Christina Psarou, Katerina Kyriakopoulou, Aikaterini Termentzi, Pelagia Anastasiadou, Marios Meidanis, Kyriaki Machera

PS2-B-142

Xanthone derivatives from NPs library as potential UPR inhibitors for alternative crop protection: Molecular modelling and biological activity

G. Viault, D. Bréard, CP Dinh, N. Blon, AM Le Ray, N. Bataillé-Simoneau, T. Guillemette, P. Simoneau, P. Richomme

PS2-B-143

Escalating low-dose Δ^9-THC treatment during adolescence affects motor function and dopaminergic activity in adult male rats

Foteini Delis, Nafsika Poulia, Charalampos Brakatselos, Nikolaos Kokras, Christina Dalla, Katerina Antoniou

PS2-B-144

Escalating low-dose Δ^9-THC treatment during adolescence affects cognitive functions and neuroplasticity markers in adult rats

Nafsika Poulia, Foteini Delis, Charalampos Brakatselos, Alexia Polissidis, Katerina Antoniou

PS2-B-145

Ficusnotins, diarylbutanoids from the Philippine endemic plant *Ficus nota* (Blanco) Merr.

Mylene Uy, Felmer Latayada, Shinji Ohta

PS2-B-146

The impact of chronic low-dose Δ^9-THC administration during adolescence on psychotic-like behaviour of adult rats

Charalampos Brakatselos[1], Nafsika Poulia[1], Foteini Delis[1], Alexia Polissidis[2], Katerina Antoniou[1]

PS2-B-147
On the dual role of natural compounds in crop protection
Valeria Terzi, Caterina Morcia, Roberta Ghizzoni, Nesrine Salhi

PS2-B-148
Phytochemical profiling using LC–PDA–MS and determination of the antioxidant activity of date palm (*Phoenix dactylifera* L.) seed extracts (Kentichi cultivar) from Tunisia
Sahar Ben Youssef, Olfa Taktak, Kenn Foubert, Luc Pieters, Noureddine Allouche

PS2-B-149
The cytotoxicity and phytochemical characterization of *Ipomoea pileata* Roxb. extracts
Piotr Gorski, Idowu Eniafe Fadayomi, Fidelia I Uche, Elisabete Pires, James McClaugh, Wen-Wu Li

PS2-B-150
Role of plant polyphenols in intervening aging and associated events
Kanti Bhooshan Pandey

PS2-B-151
Antiplasmodial, antimicrobial and antiviral compounds from South African plants
Fanie Van Heerden, Nasir Tajuddeen, Wonder Mpapane, Jude Chukwujekwu

PS2-B-152
Kushenol E, a Prenylated Flavonoid, Attenuated Autophagy Maturation and Impaired Lysosome Localization through VCP/p97 Inhibition
Min Cheol Kwon, Sung-Kyun Ko, Jong Seog Ahn

PS2-B-153
Natural Products with Antioxidant and Anticancer Potentials from Philippine's Biodiversity
Mylene Uy

PS2-B-154
Punicalagin quantification by CZE in extracts of *Combretum aculeatum* used traditionally for TB treatment
Assane E. Diop

PS2-B-155
Exploring the Traditional Medicine of Atacama: an inestimable source of bioactive compounds
M. Sergio Ortiz, Marylin Lecsö-Bornet, Christine Bonnal, Sandrine Houze, Ali Tahrioui, Emeline Bouffartigues, Sylvie Chevalier, Aikaterini Gioti[5] Roxane Tenta, Naveen Kumar, Thorsten Rose, Bernd Fiebich, Konsyantina Stathopoulou, Maria Makropoulou, Nektarios Aligiannis, Sylvie Michel, Raphaël Grougnet, Sabrina Boutefnouchet

PS2-B-156
Cyanidin-3-*O*-*β*-glucoside combined with its metabolite protocatechuic acid attenuated the activation of mouse primary hepatic stellate cells *in vivo* and *in vitro*
Xinwei Jiang

PS2-B-157

Evaluation of antioxidant and anti-inflammatory potential of various extracts from different parts of *Tanacetum cilicicum* (Boiss.) Grierson

Ali Şen, Aybeniz Yıldırım, Ahmet Dogan, Leyla Bitis

PS2-B-158

A study on the antioxidant activity, total phenolic and flavonoid content of *Calepina irregularis (Asso)* Thell. (Brassicaceae)

Sevda Deniz Dalğın, Ali Şen, Gizem Bulut, Leyla Bitis

PS2-B-159

Evaluation of anti-inflammatory activities of some medicinal plants

Leyla Bitiş, Ali Şen

PS2-B-160

Insecticidal potential of aqueous extracts from *Dittrichia viscosa*, *Cistus villosus*, *Rosmarinus officinalis* and *Lavandula stoechas* from the Tlemcen area (Algeria)

Ahmed Taibi, Choaib Lokbani, Mohammed Zerriouh

PS2-B-161

Insecticidal effects of some *Sinapis* aqueous extracts on the control of *Aphis fabae* and *Myzus percisae*

Ahmed Taibi, Amel Bechta

PS2-B-162

Activation of silent natural product biosynthetic pathways in fungi using epigenetic modifiers

Mohammed Aldholmi, A Ganesan

S2-B-163

The *Crocus sativus* compounds *trans*-crocin 4 and *trans*-crocetin modulate the amyloidogenic pathway and tau misprocessing in Alzheimer disease neuronal cell culture models

Ioanna Chalatsa, Demetrios A. Arvanitis, Nikolaos Stavros Koulakiotis, Athina Giagini, Alexios Leandros Skaltsounis, Zeta Papadopoulou-Daifoti, Anthony Tsarbopoulos, Despina Sanoudou

PS2-B-164

***Sideritis scardica* and *Cichorium spinosum* reduce amyloidogenesis and tau aggregates in Alzheimer's disease models**

Demetrios A. Arvanitis, Ioanna Chalatsa, Eleni V. Mikropoulou, Athina Giagini, Nektarios Aligiannis, Maria Halabalaki, Zeta Papadopoulou-Daifoti, Anthony Tsarbopoulos, Alexios Leandros Skaltsounis, Despina Sanoudou

PS2-B-165

Screening of plants extracts from Reunion Island as biopreservatives for cosmetic applications

Laure-Anne Peyrat, Gwenn Atheaux, Nicolas Gaboriaud-Kolar
Bioval Ocean Indien, Montpellier, France

PS2-B-166
Quantitative determination of the prodrug of the naturally occurring antibiotic colistin in formulations
Ioanna Dagla, Evangelia Karkoula, Eirini Baira, Anthony Tsarbopoulos, Evagelos Gikas

PS2-B-167
Ethnopharmacological study of plants from the greek flora for the discovery of cytotoxic agents and inhibitors of acetylcholinesterase, hyaluronidase and phospholipase
Rozalia Michalea, Vasiliki Dedousi, Panagiotis Polychronopoulos, Konstantina Stathopoulou, Dimitra Benaki, Emmanuel Mikros, Nektarios Aligiannis

PS2-B-168
Chemical Constituents from the Stem Bark of *Pentaclethra macrophylla* Benth (Fabaceae)
Augustine Ahmadu, Chinaka Nnennaya, Sunday Garba, Nikolaos Tsfantakis, Nikolas Fokaliakis

PS2-B-169
***Dendropanax morbifera* extract ameliorates thioacetamide-induced hepatic fibrosis via TGF-β1/Smads pathways**
Kyeong Seok Kim, Hae Ri Kim, Jong Seung Im, In Su Kim, Hyung Sik Kim

PS2-B-170
Afrocyclamine A, a triterpenoid saponin, induces apoptosis and autophagic cell death via the PI3k/Akt/mTOR pathway in human prostate cancer cells
Sachan Richa, Ji Yeon Son, Hae Ri Kim, Hyung Sik Kim, In Su Kim

PS2-B-171
The beneficial effect of natural compounds in the treatment of post-menopausal osteoporosis
A. Vontzalidou, M. Makropoulou, A. Meligova, D.J. Mitsiou, S. Mitakou, M.N. Alexis, N. Aligiannis

PS2-B-172
Exacerbative effect of *Paullinia pinnata* methanol leaf extract on ethylene glycol monomethyl ether-induced testicular dysfunction in male Wistar rats
Oluwatoyin Adeyemo-salami

PS2-B-173
Investigation of potential cytotoxicity and genotoxicity of olive secondary metabolites and an olive oil polyphenols extract
Agathi Charistou, Anastasia Spyropoulou, Effrosyni Katsanou, Apostolis Angelis, Dimitris Michailidis, Maria Halabalaki, Alexios-Leandros Skaltsounis, Kyriaki Machera

PS2-B-174
Identification of additional anti-microbials and anti-oxidants in South African *Hypoxis hemerocallidea* Lam (African potato) extracts
Kokoette Bassey, Noel-David Nogbou, Andrew Musyoki

PS2-B-175
Characterization of traditionally used medicinal plants in the treatment of HIV
Babalwa Tembeni, Dashnie Naidoo-Maharaj, Thomas Klimkait, Vinesh Maharaj

PS2-B-176
Halogenated metabolites from the red alga *Laurencia majuscula* collected in the Red Sea
Mohamed Tammam, Efstathia Ioannou, Vassilios Roussis

PS2-B-179
Purification of Chios Mastic active constituents and development of analytical methods for quality control purposes
Vasiliki K. Pachi, Ekaterini Arguropoulou, Efi Thoma, Sofia Mitakou, Maria Halabalaki, Leandros-Alexios Skaltsounis

PS2-B-180
Investigation of the absorption and colonic biotransformation of olive oil polyphenols in the GIDM-Colon, a validated *in vitro* Gastrointestinal Dialysis Model with Colon phase
Maria Eleni Sakavitsi, Annelies Breynaert, Apostolis Angelis, Luc Pieters, Maria Halabalaki, Nina Hermans

PS2-B-181
Olive oil's biophenols quantitative study using NMR, LC-HRMS & MS/MS methods
Stavros Beteinakis, Sotirios Katsikis, Emmanouel Mikros, Leandros A Skaltsounis, Maria Halabalaki

PS2-B-182
Bio-guided isolation of volatile compounds with repellent properties against *Aedes albopictus* (Diptera: Culicidae) using CPC technology
Anastasia Liakakou, Apostolos Angelis, Nikolaos Fokialakis, Antonios Michaelakis, Dimitrios Papachristos, Leandros.A Skaltsounis

PS2-B-183
Mediterranean herb extracts inhibit oral microbial growth and biofilm formation of *Streptococcus mutans*
Lamprini Karygianni, Joachim Hickl, Aikaterini Argyropoulou, Maria-Eleni Sakavitsi, Maria Halabalaki, Ali Al-Ahmad, Elmar Hellwig, Nektarios Aligiannis, Alexios Leandros Skaltsounis

PS2-B-184
Indirubin analogues are promising compounds for the treatment of Leishmaniasis and Human African Trypanosomiasis
Antonia Efstathiou, Nicolas Gaboriaud-Kolar, Vassilios Myrianthopoulos, Emmanuel Mikros, Agallou Maria, Dimitrios Bouziotis, Karagouni Evdokia, Alexios-Leandros Skaltsounis, Despina Smirlis

PS2-B-185
Antiviral properties of the leafs extract of succulent plant *Graptopetalum paraguayense* E. Walther against sensitive and resistance Herpes Simplex virus type 2 (HSV-2) strains
Petia Genova-Kalou, Nadezhda Markova, Stefka Ivanova, Kamelia Yotovska, Venelin Enchev

PS2-B-186
***Glycyrrhiza glabra* enhanced extract and Adriamycin anti-proliferative effect against PC-3 prostate cancer cells**
Katerina Gioti, Anastasia Papachristodoulou, Dimitra Benaki, Apostolos Beloukas, Argyro Vontzalidou, Nektarios Aligiannis, Alexios-Leandros Skaltsounis, Emmanuel Mikros, Roxane Tenta

PS2-B-187
Identification of Novel Natural Bioactive Compounds using in Vivo Zebrafish Phenotypic Assays
Adamantia Agalou

PS2-B-188
Aqueous ethanol extracts of red grape pomace exert potent *in vitro* anti-platelet and anti-oxidant properties
Maria Choleva, Vasiliki Boulougouri, Smaragdi Antonopoulou, Elizabeth Fragopoulou

PS2-B-189
Anti-platelet and anti-inflammatory properties of wines from Ionian Islands
Elizabeth Fragopoulou, Maria Choleva, Maria Detopoulou, Filio Petsini, Olga Arvaniti, Efi Kallinikou, Eleni Sakantani, Aggeliki Tsolou, Yiannis Samaras

PS2-B-191
Development of anti-ageing natural products based on biodiversity of the Greek flora by employing environmentally friendly technologies and anti-ageing biological research
Vasiliki-Ioanna Boka, Aimilia Sklirou, Maria Angelopoulou, Aristidis Konstantas4, Aikaterini Argyropoulou, Haris Pratsinis3, Dimitris Bilalis, Eleftherios Kalpoutzakis, Nektarios Aligiannis, Dimitris Kletsas, Ioannis P. Trougakos, Marianna Ralli, Alexios-Leandros Skaltsounis

PS2-B-192
Effects of Mastiha (*Pistacia lentiscus*) supplement on oxidative stress and on plasma free amino acid in Inflammatory Bowel Disease; A randomized, double blind, placebo-controlled trial
Efstathia Papada, Charalampia Amerikanou, Ljilja Torović, Nick Kalogeropoulos, Chara Tzavara, Ilias Smyrnioudis, Alastair Forbes, Andriana C. Kaliora

PS2-B-193
Utilizing compact mass spectrometry for detection and quantification of chemicals related to cannabis
Daniel Eikel, Simon J Prosser, Vijay Gupta, Jack D Henion

TOPIC D: Natural products in material science

PS2-D-001
Valorisation of natural resourses: polymerization of *β*-pinene by natural montmorillonite clay
Amine Harrane, Malika Akeb, Mohammed Belbachir

PS2-D-002
Utilization of lignin from a novel bio-refinery process as a co-component in 3D-printing
Lucas Lagerquist, Patrik Eklund

PS2-D-003
Miscibility of cellulose with synthetic polymers in ionic-liquid based solvent systems
Jiri Dybal, Nikolay Kotov, Vladimir Raus

PS2-D-004
Development of herbal sunscreen formulations from *Ophiorrhiza mungos*
Mayuri Napagoda, Chamika Liyanaarachchi, Shamila Malkanthi, Kaumadhi Abayawardana, Sudhara De Soyza, Sanjeeva Witharana, Lalith Jayasinghe

PS2-D-005
Biofunctional coating inspired by carob polyphenols for potential application in food packaging
Vlasios Goulas, Loukas Chadjivasiliou, Christodoulos Michael, George Botsaris

PS2-D-006
Thin film composite polyamide membranes embedded with Acacia gum: Properties and performance
Viktor Kochkodan, Yehia Manawi, Muataz Ali Atieh

PS2-D-007
Magnetically-responsive hydrogels obtained from marine plants
Tetsu Mitsumata, Mika Kawai

PS2-D-008
Electric properties for Kapton and biobased polyimide films
Mika Kawai, Tatsuo Kaneko, Tetsu Mitsumata

PS2-D-009
Production of an innovative dairy product using plant bioactive compounds
E-M Bata, P. Stathopoulos, L. Skaltsounis, Charalampos Proestos

PS2-D-010
Development of children's toothpaste gel with natural alternative preservatives
Spyridon Papageorgiou, Athanasia Varvaresou, Maria Giannakou, Irene Panderi, Fotini Mellou

PS2-D-011
Electrospun micro/nanofibrous drug delivery systems for the modified release of melatonin
Stefanos Kikionis

Topic I: Bioengeenering and combinatorial biosynthesis

PS2-I-001
Insights into the bifunctional aphidicolan–16–*ß*–ol synthase through rapid biomolecular modeling approaches
Monika Fuchs**,** Max Hirte, Thomas Brück

PS2-I-002
Fungal biotransformation of 7,8-dihydro-*β*-ionone
Özge Özşen Batur, Ismail Kiran, Fatih Demirci, K. Hüsnü Can Baser

PS2-I-003

Fabrication of porous hybrid scaffolds for tissue engineering from gelatin and the marine polysaccharide ulvan

Leto-Aikaterini Tziveleka, Andreas Sapalidis, Niki Chondrogianni, Efstathia Ioannou, Vassilios Roussis

PS2-I-004

***Camelina sativa* press cake and oil for production of vegan omega 3 and bioactive glucosinolates**

Chiara Magoni, Francesco Saliu, Sofia Cavini, Lorenzo Guzzetti, Ilaria Bruni, Massimo Labra

PS2-I-005

Artichoke waste as a source of inuline: a strategy to valorize the agricultural supply chain and its by-products

Sofia Cavini, Francesca Givoia, Lorenzo Guzzetti, Ilaria Bruni, Chiara Magoni, Massimo Labra

PS2-I-006

Metabolically engineered *E. coli* able to produce the highly valuable hydroxytyrosol and derivatives

Emmanouil Trantas, Theodora Nikou, Maria Halabalaki, Leandros Skaltsounis, Filippos Ververidis

PS2-I-007

Determination of urease and catalase activities in soil samples and their incidence in the significant learning of enzymatic kinetics

Jairo Granados, Dolffi Rodríguez

PS2-I-007

Fungal biotransformation of 7,8-dihydro-β-ionone

Özge Özşen, Ismail Kıran, Fatih Demirci, K. Hüsnü Can Başer

PS2-B-001

Activated charcoal promotes surgical wound healing effects of *Musa sapientum* and *Citrus limon* peel gel in *Rattus novergicus*

James Omale

Department of Biochemistry, Kogi State University, Anyigba, Nigeria

Musa sapientum known as banana is used as source of staple food for millions world over. Similarly, lemon, *Citrus limon* (L) is a specie of small evergreen tree in the family Rutaceae. The purpose of this study was to assess the effects of combining activated charcoal with banana and lemon peel gel on the healing of surgical wound s in rats. 36 wistar rats were divided into nine groups of 4 rats each. Wound control (Paraffin base), Standard (Povidone iodine), experimental groups (4% w/w *Citrus limon*, *Musa sapientum* gel ointment) and (activated charcoal mixed with *Citrus limon* peel and or *Musa sapientum* peel). Surgical wound of 40mm X 40mm was created dorsally on each rat, cleaned daily with 0.9% saline, treated with the formulated drugs [1,2] Measurement of wound contractions were done on 4, 8, and 12 days of the experiment. Wound contraction rates were found to be higher in wound treated with *Citrus limon* and *Musa sapientum* gel ointment formulated with activated charcoal. Order of increasing wound closure (unripe *M.sapientum* gel…> ripe *M.sapientum* gel …> activated charcoal + ripe *M.sapientum* gel ……..> activated charcoal + unripe *M. sapientum* gel …….> unripe *M.sapientum* gel + *C.limon* gel ….>activated charcoal + *C. limon* gel. Wound healing elicited by the drugs in this investigation following topical application clearly indicates that activated charcoal enhanced wound healing effects of *M. sapientum* and *C. limon* gel.

Keywords: *Musa sapientum*, *Citrus limon*, activated charcoal, rats

References:

[1] Agarwal PK, Singh A, Gaurav K, Goel S, Khanna HD, Goel RK. Indian J Exp Biol 2009; 47(1): 32–40.

[2] Atzingen DA, Gragnani A, Veiga DF, Abla LE, Mendoca AR, Paula CA, Juliano Y, Correa JC, Faria MR, Ferreira LM. Acta Cir Bras 2011; 26(5): 379–82.

PS2-B-002

Antibacterial, anti-inflammatory and peroxidase-mediated cyclooxygenase-1 inhibitory properties of *Fusarium Solani* extract

Kenneth Ngwoke[1], Tochichukwu Nwalusiuka[1], Chinechem Ekwealor[1], Valerie Nwankwo[1], Uju Obi-okafor[1], Chisom Izundu[1], Festus Basden Okoye[1], Charles Esimone[4], Peter Proksch[2], Chen Situ[3]

[1]Department of Pharmaceutical and Medicinal Chemistry, Nnamdi Azikiwe University Awka, Nigeria, Awka, Nigeria, [2]Institute for Pharmaceutical Biology and Biotechnoogy, Heinrich Heine University, Dusseldorf, Germany, [3]Institute for Global Food Security, Queens University, Belfast, Belfast, United Kingdom, [4]Department of Pharmaceutical Microbiology and Biotechnology, Nnamdi Azikiwe University, Nigeria, AWka, Nigeria

Many of the drugs used currently in clinical medicine originated from soil inhabiting micro-organisms owing to the rich biodiversity that exist in various soil species. The soil fungi population of the South-Eastern Nigeria has not been explored and it is hypothesized that they will promise new chemical structures that can be exploited and developed for clinical use. To investigate the antimicrobial and anti-inflammatory properties of soil fungi-derived extract. Soil fungi were isolated, purified and tested for their ability to produce antibiotics using agar overlay technique. One of the soil fungi, F6, identified by molecular biology characterization as *Fusarium Solani* was selected for fermentation based on observed antibiotic activity. The fermentation extract was tested for antimicrobial and anti-inflammatory properties using agar dilution method, rat-paw and xylene induced edema models respectively. Furthermore, acetic acid induced writhing model was used to investigate its analgesic properties. The ability of extract to inhibit cyclooxygenase enzyme was also determined *in vitro*. *F. solani* extract exhibited profound activity with minimum inhibitory concentration against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* at concentrations <12.5 µg/mL. The extract inhibited xylene induced edema by 65% compared with 61% observed for diclofenac used as positive control and was 2-fold better than diclofenac in rat paw edema model within the first phase of inflammation. The extract was also found to potently inhibit cyclooxygenase-1 (COX-1) peroxidase-mediated activities with an IC_{50} below 5 µg/mL.

PS2-B-003

A new benzophenone glycoside from the leaves of *Mitracarpus villosus*

Kenneth Ngwoke[1], Njideka Orame[1], Shuai Liu[2], Festus Okoye[1], Georgios Daletos[2], Peter Proksch[2]

[1]Nnamdi Azikiwe University Awka, Nigeria, Awka, Nigeria, [2]Institute for Pharmaceutical Biology and Biotechnoogy, Heinrich Heine University, Dusseldorf, Germany

A new benzophenone glycoside, mitraphenone A (1), together with three known compounds (2–4) were isolated from the leaves of the traditionally used medicinal plant *Mitracarpus villosus* (Rubiaceae) collected in Nigeria. A combination of one- and two-dimensional NMR spectroscopic and mass spectrometric measurements were carried out to identify the structure of 1. All isolated compounds (1–4) were screened for their antibacterial activity against several Gram-positive and Gram-negative bacteria. Compound 1 exhibited moderate activity against *Enterococcus faecium* (strains ATCC 35667 and ATCC 700221) and *Staphylococcus aureus* ATCC 25923 with MIC values ranging from 25 to 50 μM.

PS2-B-004

Bioactive compounds from *Achyranthes aspera* (chirchitta)

Rashmi Sehrawat

Chemistry And Bio Prospecting Division, Forest Research Institute, Dehradun, India, Dehrdaun, India

Achyranthes Linn (Family: Amaranthacae), a genus of stiff herbs, is distributed throughout the tropical and subtropical regions. It is an erect annual herb (0.3-0.9mm in height) and possesses diuretic, purgative properties. The plant extract is also used as abortifacient and contraceptive. Presence of ecdysterone in its roots causes insect molting activity. Literature survey revealed that triterpene glycosides, aliphatic compounds, steroids, fatty acids, alkaloids and betaine have been characterized from its different parts.

The present investigation describes the isolation of oleanolic acid glycosides from the seeds and steroidal triterpenes from the leaves, stem and roots of *Achyranthes aspera* collected from Dehra Dun. The structures of these compounds were elucidated as 3-*O*-*α*-L-rhamnopyranosyl-(1-4)-*β*-D-(glucopyranosyl uronic acid)-Oleanolic acid [A], 3-*O*-*α*-L-rhamnopyranosyl-(1-4)-*β*-D-(glucopyranosyl uronic acid)-Oleanolic acid-28-*O*-*β*-D-glucopyranoside[B], 3-*O*-*α*-L-rhamnopyranosyl-(1-4)-*β*-D-(glucopyranosyl uronic acid)- Oleanolic acid-28-*O*-*β*-D-glucopyranosyl (1-4)-*β*-D-glucopyranoside [C], Spinasterol [D], Stigmasterol [E], *α*-spinasterol acetate [F] on the basis of ^{1}H NMR, ^{13}C NMR, 2D NMR, MS spectral data. The compounds A, C, D and E are reported for the first time in *Achyranthes* genus. The occurrence of compound A and C have not been reported earlier from nature. Biological activity of *A.aspera* different extracts was carried out on growth, development and maturation in mulberry silkworm, *Bombyx mori* L. Two extracts were showing efficacy against silkworm. Therefore, a formulation was developed by combining the different extracts/fractions and again tested at rearing house against the silkworm. Generally, Silkworm *Bombyx mori* L. takes 24-36hrs. for complete spinning, however on application of formulation, it took only 15-18hrs. Therefore, we could be able to reduce the time period of spinning i.e. just half of the normal, good quality and uniform spinning was obtained. The above formulations will be useful to silkworm farmers in rearing of silk worm with very good cocoon quality. This is an important aspect of the research work not dealt earlier in literature.

PS2-B-005

Anti-inflammatory activity of the indonesian propolis and its molecular marker

Muhamad Sahlan[1,2], Andrea Davina[1], Herbert Situmorang[3]

[1]Department of Chemical Engineering, Faculty of Engineering, Universitas Indonesia, Depok, Indonesia, [2]Research Center for Biomedical Engineering, Faculty of Engineering, Universitas Indonesia, Depok, Indonesia, [3]Faculty of Medicine, Universitas Indonesia, Depok, Indonesia

The aim of this study is to identify anti-inflammatory properties of the Indonesian propolis and its molecular markers. The propolis that used in this research was from South Sulawesi, Indonesia, there are 2 types of propolis that used were smooth and rough propolis. The Sprague Dawley white rats were used as the subject. The inflammation was induced by Carrageenan and 135 mg/Kg diclofenac sodium was used as positive control. Dose of propolis that used in this research were 50 mg/Kg, 100 mg/Kg, 200 mg/Kg for smooth propolis and 25 mg/Kg, 50 mg/Kg, 100 mg/Kg for rough propolis. Edema volume was measured by plethysmometer, for every 1 hour until 5 hours after carageenan injection. Furthermore, the measurement data was analyzed with Kruskall Wallis statistical test in SPSS 23.0. To identified anti-inflammatory molecule marker in Propolis, LC-MS/MS method was used. The result showed that the smooth propolis with dose 50 mg/Kg had the the best inflammatory inhibition and the value was 66,09%. In addition, rough propolis with dose 25 mg/Kg was the best dose after soft propolis with dose 50 mg/Kg and the value was 59,46%. Therefore, based on those result, both propolis had anti-inflammatory effect. Moreover, if soft and rough propolis were compared in the same dose, soft propolis had more significant inflammatory inhibition than rough propolis. From LC-MS/MS result, 7 anti-inflammatory compounds were identified as the potential anti-inflammatory molecular marker in the propolis. [6]-dehydrogingerdione, alpha-tocopherol succinate, adhyperforin, and 6-epiangustifolin were identified in smooth and rough propolis. Deoxypodophyllotoxin and kurarinone were identified in smooth propolis. Meanwile xanthoxyletin was identified in rough propolis

PS2-B-006

Chemical composition of essential oil from *Schinus molle* L and their antithrombotic properties

Douglas Chaves[1], Rosiane da Silveira[1], Alessandra Guedes[2], Flavia Frattani[2], Marco Andre Santos[3]

[1]Departamento de Ciências Farmacêuticas, Universidade Federal Rural Do Rio De Janeiro, BR 465 km47, 23890-000, Seropédica, Brazil, [2]Departamento de análises Clínicas e Toxicológicas, Universidade Federal do Rio de Janeiro, Rio de janeiro, Brasil, [3]Departamento de Química, Universidade Federal Rural Do Rio De Janeiro, BR 465 km47, 23890-000.

Thrombosis is associated to the ischemic heart disease and stroke, which collectively caused one in four deaths worldwide [1]. Essential oils (EO) have great importance in the development of research, which indicates a possible antithrombotic property. The aim was to evaluate the chemical composition of the EO from *S. molle* and evaluate their activity on the hemostasis. The EO was extracted from leaves of *S. molle* by the hydrodestillation. Dry EO was characterized by CG-FID-EM. Anticoagulant assay was performed by aPTT and PT tests. The platelet aggregation assays were performed in aggregometer following the turbidimetric method [2], using ADP and collagen as aggregation inductor. Forty-five compounds were identified in the EO with 88.77% of sesquiterpenes. α-Muurolol and δ-cadinene were the major compounds (22.85% and 9.66 %, respectively). EO of *S. molle* did not show anticoagulant activity in the concentration used (40, 200 and 400 mg.ml-1). The lowest concentration of OE evaluated to the platelet aggregation (5.0 mg.ml-1), using ADP as inducer, had greater inhibitory power (80%), and the higher concentration (40.0 mg.ml-1) inhibited 48% of platelet aggregation. When using collagen as an inducer, EO did not show significant activity suggesting that the antithrombotic action of EO could be related to the ADP inducer, possibly through the inhibition of P2 family receptors, which are activated by ADP. EO of *S. molle* does not interfere in the blood coagulation pathways and does not act on the induction by collagen, but is an excellent anti-platelet. These results are very useful for following the studies with other inducers of platelet aggregation and future *in vivo* studies, in order to promote scientific bases for the development of a herbal medicine that can be used in the prevention or treatment of cardiovascular diseases. This is the first report of antiplatelet action of *S. molle*.

PS2-B-007

Gelatin-based films additivated with mind essential oil and red propolis ethanolic extract as active material: Microbiological and antioxidant activity

Guillermo Salamanca Grosso[1], Laura Maria Reyes Mendez[2], Mónica Patricia Osorio Tangarife[2], Eng Paola Andrea Mayorquin Súarez[2]

[1]Universidad Del Tolima, Ibagué, Colombia, [2]Mellitopalinological and Physicochemical Properties of Foods Research Group, Ibagué, Colombia

Films produced with biopolymers from renewable sources have the ability to carry active compounds, these materials can be used as active packaging for foods protection1,2. Among natural additives. Essential oils and propolis, has been used due its antibacterial and antioxidant properties3,4. The aim in this work focused on color, light barrier and water permeation properties (WVP) of gelatin-based on red propolis ethanolic extract (EERP) and mint essential oil (MEO), antioxidant activity and the antimicrobial activity against Gram () bacteria's. Microbial growth is generally responsible for the spoilage in meats and meat products together with biochemical and enzymatic deteriorations. Activated films were developed using six formulations: T1 (4% Gelatin, as control), T2 (4% Gelatin + 2.0% EERP), T3 (4% Gelatin + 2.0% MEO), T4 (4% Gelatin + 1.0 % MEO + 1.0% RPEE), T5 (4% Gelatin + 0.5 % MEO + 1.5% EERP) and T6 (4% Gelatin + 1.5 % MEO + 0.5% EERP). Diffusional test for microbiological sensibility for Gram (+) [*Staphylococcus aureus* (ATCC 29213), *Listeria monocytogenes* (ATCC 4677)], and Gram (-) [*Escherichia coli* (ATCC 25922) and *Salmonella enteritidis* (ATCC 13076)] was tested. Permeation properties respect to water were measures between 200 and 800 nm, also light barrier properties were determined. Antioxidant activity of films was determined through [DPPH], [ABTS] tests, besides the total phenolic content were measured through Folin-Ciocalteau method. Chromatic properties show decrease values for L and the a* and b* increased as a function of increasing EERP proportion. The WVP depends on both the diffusivity and solubility of water molecules in the polymeric matrix, depend on the cross-links resulting from the interactions between the gelatin, phenolic, amirin, medicarpin and others compounds in EERP. Films were efficient against Gram(+) bacteria (*S. aureus* and *L. monocytogenes*). [DPPH] and [ABTS] test also was probed.

PS2-B-008

Food phytochemicals modulating autophagy in human iPS cells

Izumi Sasanuma, Takashi Tsukahara, Ayato Ohashi, Kyouya Shigeta

National Institute of Technology, Oyama College, Oyama, Japan

Phytochemicals such as polyphenols, steroid saponins, and polysaccharides are consumed as a food and are also widely used in traditional Chinese medicine. It has been reported that phytochemicals have significant activities when ingested by animals, and there is great interest in their potential health benefits. In this work, we define the roles of polyphenols, steroid saponins, and polysaccharides from kanpyo, dioscorea, and barley in the productions of β-glucosidase in iPS cells (iPSCs) because our previous work showed that the phytochemicals positively regulate neural differentiation and the enzyme production. First, we isolated polyphenols, steroid saponins, and polysaccharides from kanpyo, dioscorea, and barley, and iPSCs were treated by the isolated compounds. The polyphenols, steroid saponins, and polysaccharides stimulated the proliferation of iPSCs. The β-glucosidases in iPSCs were induced by the steroid saponins. On the other hand, the induction of β-glucosidases was inhibited by the polysaccharides and polyphenols. Our results indicate that the β-glucosidases involved in the metabolism of sphingolipids such as glucosylceramide are stimulated by the steroid glucosides and are inhibited by the polysaccharides and polyphenols. We conclude that the phytochemicals affect the production of β-glucosidases involved in the autophagy. We have now pointed out that steroid saponins affect the production of β-glucosidases involved in the sphingolipids metabolism. Controlling the enzymes could offer a possible target for curing neurodegenerative diseases. It is hope that the findings that have been presented in this work will contribute to a better treatment of neurodegenerative diseases.

PS2-B-009

Enhanced cell adhesion and proliferation of skin fibroblasts by ethanolic extracts of two genotypes of *Ugni molinae* Turcz. leaves

Paula Valenzuela-Bustamante[1], Pablo Parra[2], Rubén Veas[1], Ivette Seguel[3], Guillermo Díaz-Araya[2], Carla Delporte[1]

[1]Laboratorio de Productos Naturales, Departamento de Química Farmacológica y Toxicológica, Facultad de Ciencias Químicas y Farmacéuticas, Universidad de Chile., Santiago, Santos Dumont 964, PO Box 8380492, Chile, [2]Laboratorio de Farmacología Molecular, , Departamento de Química Farmacológica y Toxicológica, Facultad de Ciencias Químicas y Farmacéuticas, Universidad de Chile, Santiago, Santos Dumont 964, PO Box 8380492, Chile, [3]Instituto Nacional de Investigaciones Agropecuarias (INIA, Carillanca), Temuco, PO Box 58-D, Chile

Ugni molinae Turcz. Myrtaceae, is a native shrub knows as murtilla, murta or uñi which is widely distributed in the south-central regions of Chile. Recently, it was determined that differences in their phenolic composition depend on the genotype. Our laboratory identified hydrolysable tannins in ethanolic extracts (ETEs) [1], a group of polyphenolic compounds that have reported wound healing activity [2]. According to this information, the aim of this study was to evaluate and compare the effect of murtilla leaves ETEs from two genotypes (G. 19-2 and G. ZF-18) in human dermal fibroblasts (HDF) isolated from human skin explants. Total tannin content (TTC) was quantified by Folin-Ciocalteu assay. *In vitro* cytotoxicity and fibroblast proliferation was evaluated by alamarBlue® assay and cell adhesion was determined by crystal violet staining. All tests were performed within 24-hour treatment. Significant differences ($p>0.05$) were obtained in the TTC of both extracts, being higher in G. ZF-18 ETE (201.0 ± 1.6 mg tannic acid equivalent/g dry matter). Cell viability decreased at concentrations higher than 25 µg/mL. Cell proliferation was enhanced by two-fold when cells were treated with 10 µg/mL of both ETEs in presence of 10% FBS, while no effect was observed at the lowest concentration. On the other hand, only the G.19-2 ETE increased cell adhesion after 2 hours compared with the negative control (1 % DMSO) and it was higher at 10 µg/mL. Consequently, these results suggest that ethanolic extracts of *U. molinae* leaves have a potential wound healing activity due to their effect on proliferation and cell adhesion of HDF.

Acknowledgements: Beca Conicyt Nº21150688, Fondecyt N°1170425, Fondecyt N°1130155.

Keywords: Skin fibroblasts, cell proliferation, cell adhesion, *Ugni molinae* leaves, ethanolic extract.

References:

[1] Peña-Cerda M *et al*. Food Chem 2017; 215: 219–227.
[2] Su X *et al*. Burns 2017; 43: 830–838.

PS2-B-010

Metabolic profiling and wound-healing activity of *Boerhavia diffusa* leaf extracts

Kriti Juneja[1], Rutusmita Mishra[2], Partha Roy[2], Debabrata Sircar[1]

[1]Plant Metabolomics and Molecular Biology Lab, Department of Biotechnology, Indian Institute of Technology Roorkee, Roorkee, India, [2]Molecular Endocrinology Lab, Department of Biotechnology, Indian Institute of Technology Roorkee, Roorkee, India

Boerhavia diffusa, a perennial herb belonging to the Nyctaginaceae family, has been widely used in Indian traditional medicinal system to cure many human diseases. However, its traditional use in the treatment and management of wounds has not been validated by any scientific study. The present study was aimed at exploring the *in vitro* and *in vivo* wound healing potential of methanolic extract (ME) and chloroform extract (CE) from *B. diffusa* leaf and identification of active metabolites that may be responsible for its wound healing properties. The study included *in vitro* cell viability, proliferation, and wound scratch test assays as well as *in vivo* excision wound model assay. Both ME and CE were analyzed for their total phenolics, total flavonoids and antioxidant potentials. GC-MS analyses were used for identification and confirmation of bioactive compounds present in both ME and CE. The results of our study demonstrated that ME of *B. diffusa* leaf significantly enhanced proliferation and viability as well as migration of human keratinocyte cells (HaCaT) compared to the untreated control and CE. Topical application of methanolic extract of *B. diffusa* leaf in excision wound model significantly decreased the wound area by the 14th day (91%) as compared to control (22%). The GC-MS studies revealed caffeic and ferulic acids in the methanolic extract and a major bioactive compound D-pinitol. The results suggest that methanolic extract of *B. diffusa* possesses significant wound healing potential.

PS2-B-011

Exploring CADD strategies for identification of natural agonists against PPAR-gamma for breast cancer

Pawan Kumar Gupta[1], Aishwarya Laxmiaraya[2], Jeena Gupta[2], Aleksandrs Gutcaits[3], Prabha Garg[4]

[1]School of Pharmaceutical Sciences & Department of Research and Development,Lovely Professional University, Phagwara, India, [2]School of Biosciences, Lovely Professional University, Phagwara, India, [3]CNS Active Compounds Laboratory, Latvian Institute of Organic Synthesis, Riga , Latvia, [4]Department of Pharmacoinformatics, National Institute of Pharmaceutical Education and Research (NIPER), Mohali, India

Recent studies have demonstrated that PPAR gamma is over-expressed in many tumor types, including breast cancer, suggesting a possible role in tumor development and/or progression and a putative prognostic value [1]. The synthetic agonists (Troglitazone, Efastumab, Pioglitazone and Rosiglitazone) are used initially for the treatment of Type 2 diabetes but are also used later for treating breast cancer [2, 3]. These agonists are associated with serious side effects. Natural products are always an excellent source for identification of new drug leads as it possesses a high chemical scaffold diversity, different biological functions and high drug-likeness [4]. In order to identify the novel scaffolds from natural origin, computer aided drug design [Similarity searching, Pharmacophore, QSAR, Docking] strategies are used. Similarity searching is performed in different natural databases using pharmacophore and shape-based methods. Structural-activity relation is established to understand the important structural features required for PPAR-gamma activity. Selection of the novel screened molecules is achieved by matching of the important structural features required for the activity. Binding mode analysis is exhibited that some of the molecules are having same binding potential as true PPAR gamma agonist. These novel scaffolds can be further exploited for the drug design and discovery.

Keywords: PPARgamma, breast cancer, similarity searching, QSAR, binding mode analysis

References:

[1] Dong JT Am. J Pathol 2013; 182(6): 972.
[2] Hall JM, Robinson ML. J Steroids Horm Sci 2015; 6:2.
[3] Kotta-Loizou I *et al*. Anticancer Agents Med Chem 2012; 12(9): 1025–1044.
[4] Wang, L *et al*. Biochem Pharmacol 2014; 92(1): 73–89.

PS2-B-012

Anti-inflammatory properties of betulinic acid and xylopic acid in the carrageenan-induced pleurisy model of lung inflammation in mice

Edmund Ekuadzi[1], Robert Peter Biney[2], Charles Kweku Benneh[3], Bismark Osei Amankwaa[1], Jonathan Jato[4]

[1]Department of Pharmacognosy, Kwame Nkrumah University Of Science And Technology, Kumasi, Ghana, [2]Department of Pharmacology, University of Cape Coast, Cape Coast, Ghana, [3]Department of Pharmacology, University of Health and Allied Sciences, Ho, Ghana, [4]Department of Pharmacognosy, University of Health and Allied Sciences, Ho, Ghana

Margaritaria discoidea and *Xylopia aethiopica* are plants native in Ghana and the West-African region and used traditionally to treat different pathologies including inflammatory conditions. Betulinic acid (BA) and xylopic acid (XA) extracted from *M. discoidea* and *X. aethiopica*, respectively have been investigated for their anti-inflammatory effects. However, their specific effect on lung inflammation and their ability to modulate oxidative challenge under such circumstances is yet to be reported. The anti-inflammatory effect of BA and XA was established by an *in vivo* assay using the carrageenan-induced pleural inflammation model in mice. Also, the ability of BA and XA to increase catalase, superoxide dismutase, glutathione levels and decrease lipid peroxidation level in reactive oxidative assays was assessed. In addition, the ability of XA and BA to prevent potential lung tissue damage was quantified. Pretreatment with BA and XA reduced significantly, signs of inflammation: neutrophil infiltration, oedema, and alveoli septal thickening in carrageenan-treated lung tissue. Additionally, BA or XA pretreatment lowered the degree of lipid peroxidation in the lung tissue while increasing the levels of catalase, superoxide dismutase, and glutathione *in vivo*. Comparatively, XA was more efficacious than BA in the prevention of lung tissue damage. BA and XA derived from *X. aethiopica* and *M. discoidea* possess anti-inflammatory and *in vivo* antioxidant activities in mice pleurisy model. The effect of these compounds gives credence to the traditional use in the management of inflammatory conditions of the airway.

PS2-B-014

Spice-derived bioactive compounds: potential agents or food adjuvants in the management of diabetes mellitus?

Shahidul Islam, Aminu Mohammed

University Of Kwazulu-natal, Durban 4000, South Africa, Durban, South Africa

Spices possess tremendous therapeutic potential including their activities against a number of non-communicable diseases such as obesity, diabetes and metabolic syndrome, which are attributed to their bioactive ingredients. However, there is no study that critically reviewed the antidiabetic potency, safety and the bioavailability of the spice-derived bioactive compounds (SDBCs). Therefore, the aim of the study was to comprehensively review all published studies regarding the antidiabetic action of SDBCs with the purpose to assess whether the compounds are potential antidiabetic agents or adjuvants. Factors considered were concentration/dosages used, the extent of blood glucose reduction, the IC_{50} values, and the safety concern of the SDBCs. From the results, cinnamaldehyde, curcumin, diosgenin, thymoquinone (TQ) and trigonelline were showed the most promising antidiabetic effects and hold future potential as antidiabetic agents. Conclusively, future studies should focus on improving the tissue and cellular bioavailability of the promising SDBCs in order to maximize their use for the prevention and management of non-communicable diseases, particularly diabetes. Additionally, clinical trials and toxicity studies are with these SDBCs are warranted in order understand their true efficacies in humans.

Acknowledgements: This study was supported partly by a Competitive Research Grant from the Research Office of the University of KwaZulu-Natal, Durban; and Grant Support for Women and Young Researchers from the National Research Foundation (NRF), Pretoria, South Africa.

Keywords: Adjuvant, antidiabetic, diabetes mellitus, *in vitro*, *in vivo*, spices

PS2-B-015

In vitro alpha-glucosidase and glycogen phosphorylase an inhibition by chilean propolis ethanolic extracts

Ruben Veas, Paula Valenzuela-Bustamante, Gabriela Valenzuela-Barra, Carla Delporte

Laboratorio de Productos Naturales, Departamento de Química Farmacológica y Toxicológica, Facultad de Ciencias Químicas y Farmacéuticas, Universidad de Chile, Santiago, Chile

Propolis is a resinous material produced by honeybees (*Apis mellifera*) from resin of buds, leaves and flowers of plants. Almost 50% of that product corresponds to resin, a complex mixture of phenolics and their derivatives. Many of its properties have been studied, including: anti-microbial, antiviral, antifungal, anti-inflammatory, and antioxidant activity [1]. Chilean propolis is classified as Poplar type propolis with few published studies, where none have reported the potential anti-hyperglycaemic or hypoglycaemic effects by enzyme inhibition. The aim of this work is to stablish the activity of Chilean propolis ethanolic extracts (ETE) from three localities (Pirque, Pudahuel and Peñaflor) of Región Metropolitana (Chile) against alpha-glucosidase (AG) and glycogen phosphorylase a (GPa). AG and GPa inhibition analysis were carried out following the methodology of Kim *et al*., 2005 [2] and Zhang *et al*., 2008 [3] respectively, with slight modifications. ETE of Peñaflor propolis showed the higher inhibitory potency ($p<0.05$) against AG with an IC_{50} = 7.6 ± 2.4 µg/mL. All extracts showed more potency than the positive control acarbose (IC_{50} = 259.1 ± 33.4 µg/mL). On the other hand, ETE of Pirque propolis showed the higher inhibitory potency ($p<0.05$) against GPa with an IC_{50} = 9.0 ± 1.4 µg/mL. All extracts showed less potency than the positive control caffeine (IC_{50} = 5.3 ± 0.7 µg/mL). More studies are needed to evaluate the composition of extracts and their *in vivo* anti-hyperglycaemic or hypoglycaemic effects.

Acknowledgment: Thanks to Comisión Nacional de Investigación Científica y Tecnológica, Grand: Beca CONICYT Doctorado Nacional N°21161324

References:

[1] Valenzuela-Barra G *et al*. J Ethnopharmacol 2015; 168: 37–44.
[2] Kim YM *et al*. Nutrition 2005; 21: 756–761.
[3] Zhang L *et al.* Eur J Med Chem 2012; 58: 624–639.

PS2-B-016

Modulation of doxorubicin-induced renal and hepatotoxicity by *Calligonum comosum* extract via dual mechanism

Doaa Abdelhady[1], Emad Ghazi[2], Walied Abdo[3], Zizy Elbialy[4], Mustafa Shukry[5], Engy Mahrous[6], Sendker Jandirk[7], Essam Abdel-Sattar[8]

[1]Department of Clinical Pathology, Faculty of Veterinary Medicine, Kafrelsheikh University, Kafrelsheikh 33516, Egypt,, [2]Department of Clinical Pathology, Faculty of Veterinary Medicine, Kafrelsheikh University, Kafrelsheikh 33516, Egypt, [3]Department of Pathology, Faculty of Veterinary Medicine, Kafrelsheikh University, Kafrelsheikh 33516, Egypt, [4]Department of Fish Processing and Biotechnology, Faculty of Aquatic and Fisheries Sciences Kafrelsheikh University, 33516, [5]Department of Physiology, Faculty of Veterinary Medicine, Kafrelsheikh University, 33516, Kafrelsheikh, Egypt, [6]Department of Pharmacognosy, Faculty of Pharmacy, Cairo University, 11562 Cairo, Egypt, [7]Institute of Pharmaceutical Biology and Phytochemistry, University of Münster, Hittorfstraße 48, 48149 Münster, Münster, Germany, [8]Department of Pharmacognosy, Faculty of Pharmacy, Cairo University, 11562 Cairo, Egypt.

Doxorubicin (DXR) is a highly effective chemotherapeutic agent; however, its multi-organ toxicity has limited its use. In this study, we investigate possible protective effect of *Calligonum comosum* extract (CCE) against DXR induced hepato-renal toxicity in rats receiving a single intraperitoneal (i.p.) injection of DXR (20 mg/kg). Our data indicated that animals that received DXR injection without further treatment displayed hematological and biochemical signs related to liver and kidney injury, oxidative stress, DNA damage, renal and hepatic genotoxicity. However, when animals received a daily oral dose of CCE (100 mg/kg) following DXR injection for two weeks, signs of organ toxicity, DNA damage and genotoxicity were greatly reduced. Moreover, administration of CCE resulted in an increase in total lymphocyte count (TLC) and in phagocytic activity indicating immunostimulant effect of CCE. Chemical investigation of CCE indicated that the extract was rich in natural antioxidants (phenolics, flavonoids, etc) that can reduce oxidative stress especially procyanidins which might be responsible for its immunostimulant activity. Overall, our results suggested that CCE is a promising candidate that can be used to ameliorate the hepatic and nephric damage caused by administration of DXR due to its potent antioxidant effect and immunostimulant activity.

PS2-B-017

Neurotrophic compounds of Javanese ginger bangle, *Zingiber purpureum*

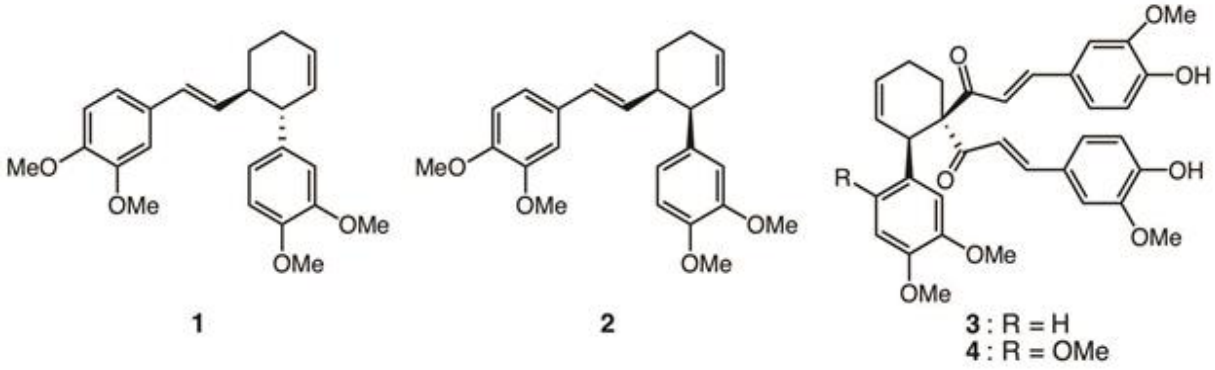
Yoshiyasu Fukuyama[1], Miwa Kubo[1], Megumi Nakai[2], Kenichi Harada[1], Nobuaki Matsui[1], Midori Suenaga[1], Yoichi Matsunaga[1], Mitsuhiko Miyamura[2], Eishin Kato[3]

[1]Faculty of Pharmaceutical Sciences, Tokushima Bunri University, Tokushima, Japan, [2]Kochi Medical School, Nangoku, Japan, [3]Hosoda SHC Co. Ltd., Miyuki, Japan

Neurotrophins (NGF, BDNF etc.) are recognized as important regulatory substance in the nervous system. However, they cannot cross blood -brain barrier because of the properties of their high molecular polypeptides and are easily metabolized by peptidases under physiological conditions. To address this issue, considerable efforts have been made to find small molecules that mimic neurotrophic properties.

Javanese ginger bangle, *Zingiber purpureum*, has been used as a spice as well as an important component of traditional medicine "Jamu" in Indonesia. In the course of our efforts to discover natural products with neurotrophic properties, we found that the MeOH extract of the roots of bangle (*Zingiber purpurenum*) exhibited neuritogenesis activity in PC12 cells. Bioassay-guided fractionation resulted in the isolation of neurotrophic phenylbutenoid dimers 1 and 2, and new compounds 3 and 4. The structure of 3 and 4 was elucidated by analysis of spectroscopic data and comparing the NMR data with cussumunarin A [1]. Compounds 1 and 2 were found not only to significantly induce neurite sprouting of PC12 cells, but also to increase the neurite length and number of neurites in primary cultured rat cortical neurons, and also showed protective activity against cell death caused by deprivation of serum. Furthermore, chronic treatment of these compounds enhanced hippocampal neurogenesis in dementia model OBX mice [2]. Compounds 3 and 4 are the first example of curcumin coupled with phenyl butenoid. In addition, compound 3 was found to accelerate the prevention of Aβ42 aggregation.

These results suggest that compounds 1 and 2 have both neurotrophic effects and neurogenesis, and thus Javanese ginger bangle may be developed as a valuable functional food for potentially protecting neurodegenerative diseases such as Alzheimer disease.



References:

[1] M. Kubo *et al*. Bioorg. Med. Chem. Lett., 2015, 25. 1586–1591.
[2] N. Matsui *et al*. Neuroscience Lett., 2012, 513, 72–77.

PS2-B-018

The effects of gamisoyo-san decoction, a traditional chinese herbal medicine, on the pacemaker potentials in murine small intestinal interstitial cells of cajal

Doeun Kim[1], Jung Nam Kim[1], Joo Hyun Nam[2], Jong Rok Lee[3], Sang Chan Kim[4], Byung Joo Kim[1]

[1]Division of Longevity and Biofunctional Medicine, Pusan National University School of Korean Medicine, Yangsan, South Korea, [2]Department of Physiology, College of Medicine, Dongguk University, South Korea, [3]Department of Pharmaceutical Engineering, Daegu Haany University, , South Korea, [4]College of Oriental Medicine, Daegu Haany University, , South Korea

The Gamisoyo-san (GSS) has been used for improving the gastrointestinal (GI) symptoms. The purpose of this study was to investigate the effects of GSS, a traditional Chinese herbal medicine, on the pacemaker potentials of mouse small intestinal interstitial cells of Cajal (ICCs). ICCs from the small intestines were dissociated and cultured. Whole-cell patch-clamp configuration was used to record pacemaker potentials and membrane currents. GSS depolarized ICC pacemaker potentials in a dose-dependent manner. Pretreatment with 4-diphenylacetoxypiperidinium iodide completely inhibited GSS-induced pacemaker potential depolarizations. Intracellular GDP-β-S inhibited GSS-induced effects, and in the presence of U-73122, GSS-induced effects were inhibited. Also, GSS in the presence of a Ca^{2+}-free solution or thapsigargin did not depolarize pacemaker potentials. However, in the presence of calphostin C, GSS slightly depolarized pacemaker potentials. Furthermore, GSS inhibited both transient receptor potential melastatin 7 and Ca^{2+}-activated Cl^- channel (anoctamin1) currents. GSS depolarized pacemaker potentials of ICCs via G protein and muscarinic M3 receptor signaling pathways and through internal or external Ca^{2+}-, phospholipase C-, and protein kinase C-dependent and transient receptor potential melastatin 7-, and anoctamin 1-independent pathways. The study shows that GSS may regulate GI tract motility, suggesting that GSS could be a basis for developing novel prokinetic agents for treating GI motility dysfunctions.

PS2-B-019

Amaryllidaceae alkaloids from *Narcissus pseudonarcissus* cv. Dutch Master as potential drugs in treatment of Alzheimer´s disease

Daniela Hulcová[1,2], Jana Maříková[3], Lubomír Opletal[1], Angela De Simone[4], Lucie Cahlíková[1]

[1]ADINACO Research Group, Department of Pharmaceutical Botany, Faculty of Pharmacy, Charles University, Heyrovského 1203, 500 05 Hradec Králové, Czech Republic, [2]Department of Pharmacognosy, Faculty of Pharmacy, Charles University, Heyrovského 1203, 500 05 Hradec Králové, Czech Republic, [3]Department of Organic and Bioorganic Chemistry, Faculty of Pharmacy, Charles University, Heyrovského 1203, 500 05 Hradec Králové, Czech Republic, [4]Department for Life Quality Studies, University of Bologna, Corso D'Augusto 237, 47921 Rimini, Italy.

Alzheimer's disease (AD) is the most prevalent neurodegenerative disease worldwide with complex etiology and multifaceted pathophysiology. Data indicate an exponential rise in the number of cases of AD. Based on the various causative factors of AD, several hypotheses, including cholinergic, amyloid, τ protein and calcium dyshomeostasis, have been put forward. AD is characterized by massive deposits of amyloid-β peptide, neurofibrillary tangles of the hyperphosphorylated τ-protein, inflammatory mediators, and reactive oxygen species, leading to neuronal death via a complex array of intertwined pathways. AD is manifested by damage of cognitive and noncognitive functions [1].

The genus *Narcissus* from the Amaryllidaceae family comprising approximately 80-100 wild species, which are mainly distributed in south-western Europe, North Africa and some populations in the Balkans, Italy and France. This family contains special type of isoquinoline substances, called Amaryllidaceae alkaloids (AA), possessing a wide range of pharmacological properties such as antitumor, antiviral, antimalarial and acetylcholinesterase (AChE) inhibitory activity. The well-known AA galanthamine is used for AD therapy.

Twenty-one known Amaryllidaceae alkaloids of various structural types and one novel alkaloid called narcimatuline, have been isolated from fresh bulbs of *Narcissus pseudonarcissus* cv. Dutch Master. The bulbs were processed by extraction, followed by column and preparative chromatography and recrystallization. The chemical structures were elucidated by combination of MS, HRMS, NMR spectroscopic techniques. All isolated compounds were evaluated for their *in vitro* AChE, butyrylcholinesterase (BuChE), prolyl oligopeptidase (POP) and glycogen synthase kinase-3β (GSK-3β) inhibitory activities. The most important biological profile has been demonstrated by narcimatuline (IC_{50},BuChE = 5.9 ± 0.2 μM, IC_{50},POP = 29.2 ± 0.9 μM; IC_{50},GSK-3β = 20.8 ± 2.4 μM).

Acknowledgements: This project was supported by Charles University grants.

References:

[1] Kumar K, Kumar A, Keegan RM, Deshmukh R. Biomed Pharmacother 2018; 98: 297–307.

PS2-B-020

From traditional use to standardized neuroprotective green-extract of *Huperzia serrata*

Elnur Garayev, Laurent Boyer, Philippe Poindron, Maud Combes, Noëlle Callizot

Neuro-sys SAS, Gardanne, France

Some plants belonging to the Lycopodiaceae species such as *Huperzia serrata* (Thunb.) *Trevis, H. squarrosa* (Forst.) *Trevis, Lycopodium complanatum* L. and *L. cernua* (L.) Franco & Vasc have been used in Asiatic folk medicine for thousands of years to treat contusions, strains, schizophrenia and memory dysfunction [1, 2]. *H. serrata* and *H. saururus* are used respectively in Chinese and Argentinian folk medicine, via infusion, for their neuromuscular and memory-improving properties [3, 4].

Huperzine A (HA), one of the constituents of *H. serrata*, was proven to be a reversible inhibitor of acetylcholinesterase (AchE) [5]. The results on more than 100.000 Chinese patients indicated that HA significantly improved cognitive functions [6] and had the potential to become an alternative treatment for Alzheimer's disease (AD). However, clinical trials of HA treatment also indicated that large doses of the molecule were needed to obtain significant effects in patients suffering from AD, and therefore increased the occurrence of negative side effects [7].

Here we describe a manufacturing process for standardized extracts, using a Green technique which allows the conservation of chemical profiles and pharmacological activity.

A "Green" extract (obtained via microwave-assisted extraction) was compared to a "traditional" extract. These extracts were analyzed by UHPLC-DAD-MS. We showed that the chemical profiles of these extracts were similar. The Green extract contained 0.17 % (m/m) of HA vs 0.16 % (m/m) in the traditional extract.

In addition, these extracts were evaluated for their putative neuroprotective activities [8]. A synergistic activity between HA and phenolic acids (Caffeic and Ferulic) was found, allowing for a lesser dose of HA, thus reducing AchE inhibition side effects. A patent describing this discovery is pending [9] and a food supplement containing the synergistic ratio of the 3 compounds is expected to be on the market by the end of this year for neuro-indication.

PS2-B-021

Phytosterol stability to thermal treatments in Bronte's Pistachio (*Pistacia vera*, L.) and in Pistachio processed products

Massimo Lucarini, Laura D'Evoli, Gabriella Di Lena, Ginevra Lombardi Boccia

Crea An, Rome, Italy

Although pistachios are the richest sources of phytosterols among nuts, few information are available about the qualitative and quantitative profile of phytosterols in Bronte's pistachios and derived processed products. Phytosterols can be susceptible to oxidation especially when exposed thermal stress during processing and storage, so their biological activity may be impaired.
This study provides data on phytosterols content (campesterol, stigmasterol, β-sitosterol, Δ^5-avenasterol) in raw Bronte's pistachios, four traditional pistachio products largely consumed in Italy (Flour, Finely chopped pistachio, Ice-cream paste, Oil) and in roasted pistachios. The study also contributes to a greather insight into the effect of processing on the formation of β-sitosterol oxidation products (7β-OH, 7α-OH, 6β-OH, 7-keto-sitosterols and 6-keto-sitostanol). The total sterol content in raw Bronte's pistachio (279 mg/100 g oil) was higher than in the processed pistachios where it varied with the type of processing, from 201 to 229 mg/100g oil detected in Oil and Flour, respectively. Beta-sitosterol represented the 86.5% of total sterol content. As β-sitosterol was found the dominant sterol in all the samples studied, the phytosterol stability after processing was investigated by monitoring the behavior of β-sitosterol. The total β-sitosterol oxides content varied from 4.40 µg/g oil found in the raw sample up to 6.30 µg/g oil in the roasted samples at 150°C. The most abundant β-sitosterol oxide was the 6-keto sitostanol (2.27 µg/g oil) represented 51% of the total oxides, between 90°C and 150°C there was a significant ($p<0.05$) increment in its formation up to 72% on the initial content. During the thermal treatments other unknown oxidation products occurred. The reported phytosterols oxides profile lets us to hypothesize that their synthesis initiates via the epoxide pathway, leading to the final product 6-keto sitostanol. For this reason the 6 keto-sitostanol could be used as molecular marker of thermal stress in pistachio products.

PS2-B-022

Antifungal and cytotoxic activity of commercially-available essential oils

William Setzer[1,2], Chelsea Powers[1], Jessica Osier[1], Emily Olsen[1], Brianne Brazell[1], Prabodh Satyal[2], Robert McFeeters[1], Debra Moriarity[1]

[1]University Of Alabama In Huntsville, Huntsville, United States, [2]Aromatic Plant Research Center, Winston-Salem, US

A total of 60 commercially-available essential oils were screened for antifungal activity (*Aspergillus niger, Candida albicans*, and *Cryptococcus neoformans*) and cytotoxic activity (MCF-7 and MDA-MB-231 human breast adenocarcinoma cells). Of the fungi tested, *C. neoformans* was the most sensitive to the essential oils, and eight oils (*Cinnamomum cassia, C. zeylanicum, Coriandrum sativum leaf, Pogostemon cablin, Santalum album, S. austrocaledonicum, S. paniculatum*, and *Vetiveria zizanoides*) showed MICs of 20 μg/mL against this organism. Ten essential oils (*C. cassia, C. zeylanicum, Commiphora myrrha, Cymbopogon flexuosus, Melissa officinalis, P. cablin, S. album, S. austrocaledonicum, S. paniculatum*, and *V. zizanoides*) showed remarkable cytotoxic activity (IC_{50} < 30 μg/mL) against one or both cell lines. These readily-available materials may add to our treatment options, as agents themselves or as adjuvant therapies, to combat fungal infections. In addition to the antifungal and cytotoxic activities of the essential oils in this study, those essential oil that do not show appreciable cytotoxic activity to human cells may be considered relatively safe for other uses such as cosmetics, flavoring, and aromatherapy.

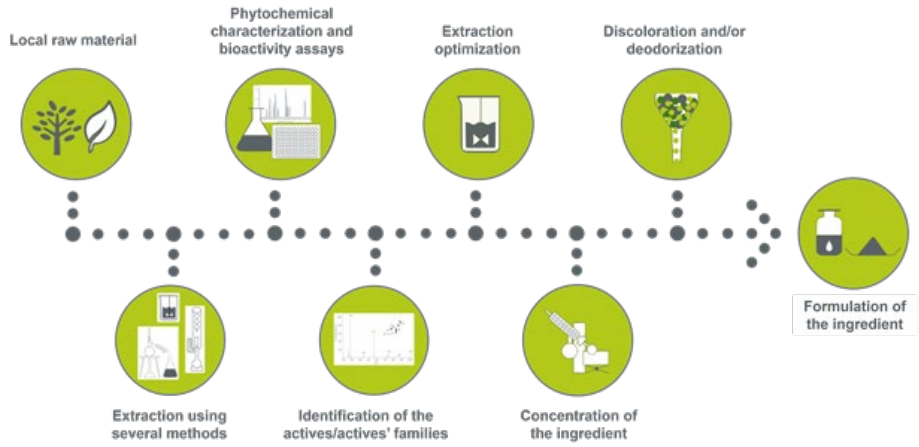
PS2-B-023

Ecoextraction of Mediterranean natural resources to develop innovative cosmetic ingredients

Pauline Burger[1], Hortense Plainfossé[1,2,3], Grégory Verger-Dubois[1,3], Xavier Fernandez[2]

[1]NissActive, Grasse, France, [2]Institut de Chimie de Nice, Nice, France, [3]Laboratoires JYTA, Carros, France

The cosmetic ingredient market is fast-growing world-widely. When it comes to beauty, consumers are increasingly concerned by the efficacy of the cosmetic products they use, and scientifically-substantiated claims are particularly scrutinized. Lately, operating a green U-turn, consumers also primarily focus on the naturality and sustainability of products, without compromising about performance, a phenomenon that has profoundly modified the strategies adopted by cosmetic professionals to innovate in terms of actives, making not only use of renewable raw materials (plant, agricultural by-products, etc.), but also favouring eco-extraction techniques and green solvents. NissActive is investing a lot in developing natural actives to fulfil the formulators' needs in that sense. In collaboration with academic research laboratories from the University Côte d'Azur, NissActive adopted a fully integrated strategy to design natural cosmetic actives, including plant sourcing, eco-extraction technologies and bioguided screening using a combination of *in vitro* assays to satisfy green-minded consumers (Figure 1). The metabolites responsible for the ingredients' bioactivities are then identified by several analytical techniques and their extraction is optimized to potentialize those activities while keeping in mind their further industrial scale-up and formulation.
This objectification methodology enables the development of bio-sourced ingredients intended for cosmetics and personal care formulations, while conveying the very image of high quality and technicity. Specific examples of ingredients developed that way will be given in the poster.



PS2-B-024

Development of an innovative natural cosmetic ingredient derived from *Teucrium lucidum* for skin repair

Hortense Plainfossé[1,2,3], Pauline Burger[3], Stéphane Azoulay[1], Anne Landreau[1,4], Grégory Verger-Dubois[1,2], Xavier Fernandez[1]

[1]Institut De Chimie De Nice, Université de Nice Sophia Antipolis, Nice, France, [2]Laboratoires JYTA, Carros, France, [3]NissActive, Grasse, France, [4]Faculté de santé, Université d'Angers, Angers, France

Dermocosmetics are used to preserve skin's health and beauty while answering to cutaneous issues, being often used after minor operations, or to cure burns, superficial wounds, etc [1]. It is a constantly growing sector driven by new consumers' requests. They pay more and more attention not just to the health and safety aspects of the ingredients entering their dermocosmetics' formulations, but also to their environmental footprint. The efficacy of cosmetic products is particularly scrutinized by consumers, which involves that marketing arguments and claims need to be based on rigorous testing and reliable results. As a result, the increasing demand for natural ingredients with assessed bioactivities has profoundly modified the strategies adopted by cosmetic professionals to innovate in terms of actives. To answer this problematic, studies are carried out to develop innovative biosourced ingredients for wound-healing application. Such a development is a long-term procedure [2] that is thoroughly described via the example of the design of a new innovative active designed based on an extract of aerial parts of *Teucrium lucidum* L., a Mediterranean plant. Together with a strong marketing potential (innovative and locally sourced plant), this extract displays very interesting bioproperties assessed using a combination of *in vitro* assays (anti-inflammatory, anti-collagenase, anti-elastase, anti-hyaluronidase, antioxidant and whitening activities) [3]. So, its phytochemical characterization was performed by bioguided fractionation and isolation of interesting molecules in order to identify the metabolites responsible for the bioactivities evidenced. Some new molecules were isolated using semi-preparative HPLC and are currently being characterized (HMRS and NMR analyses). Only once this is achieved, can the proper ingredient development be undertaken (deposition on cosmetic support, objectification of its formulability, stability testing, etc).

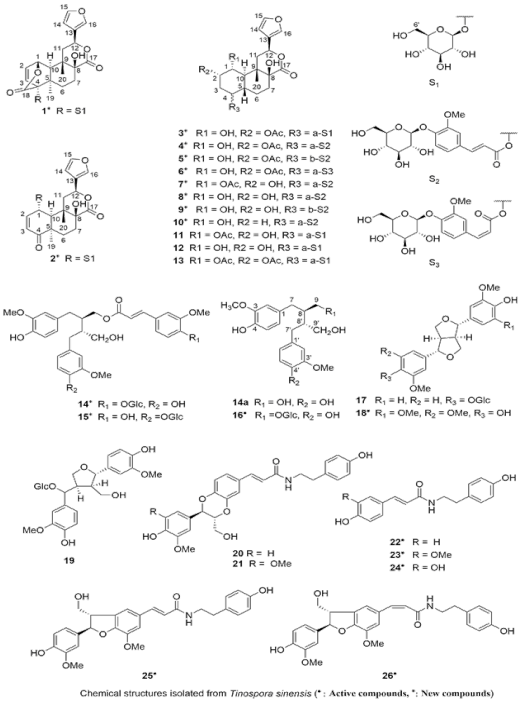
PS2-B-025

Anti-neuroinflammatory compounds from ethnic medicinal plant *Tinospora sinensis* in Guangxi province of China

Haibing Liao, Dong Liang

Guangxi Normal University, Guilin, China

As we know, neuroinflammation, caused by over-activation of microglia, plays an important role in the development of neurodegenerative disease. Finding antineuroinflammation compounds may be help for the treatment of neuroinflammation related ailments.
There are plentiful germplasm resources of Chinese medicinal herbs in Guangxi, and the number of their species is about 4623, which is at the second position in China, just behind Yunnan province. There are also a large population of ethnic minorities such as Zhuang, Yao and alike living in Guangxi. Most of the ethnic minorities are living in mountains with heavy moisture environment. So, many medicinal resources have been used to treat the rheumatism disease in folk.
Tinospora sinensis is widely distributed and cultivated in Guangxi. The stems of this species, commonly known as “Kuan-Jin-Teng” in Chinese, have been used as a traditional Yao medicine to treat rheumatism, lumbar muscle strain, and sciatica. Ten new clerodane diterpenoid glycosides and three known analogues together with six lignan glucosides and seven phenolic amide alkaloids were isolated. The absolute configurations of tinosinenosides A–C were established by using experimental and calculated ECD data. Their cytotoxicity against the human epithelioid cervical carcinoma (HeLa) cell line and the nitric oxide production inhibitory activity of lipopolysaccharide-activated BV-2 microglial cells were tested.
It’s the first time to find anti-neuroinflammatory activities compounds from *T. sinensis*. The activity of compounds 22 and 23 is even better than positive control minocycline. And the preliminary mechanism of the active compounds should be further studied. All the researches will hopefully result in the discovery of good leading compounds for alleviating microglial activation and provide good reference for using *Tinospora* plants resource to treat neuroinflammation and related ailments.



Chemical structures isolated from *Tinospora sinensis* (* : **Active compounds, *: New compounds**)

PS2-B-026

Chemical interactions between *Fusarium oxysporum* and an endophytic fungus *Xylaria* sp. with antifungal activities

Morgane Barthélemy[1], Laurent Intertaglia[3], Didier Stien[2], David Touboul[1], Véronique Eparvier[1]

[1]ICSN, UPR 2301, Université Paris Saclay, CNRS, Gif-sur-Yvette, France, [2]Sorbonnes Université, CNRS, Laboratoire de Biodiversité et Biotechnologies Microbiennes, Banyuls sur Mer, France, [3]Sorbonnes Université, CNRS, Plateforme Bio2mar, Banyuls sur Mer, France

Biopesticides from living microorganisms or compounds from natural substances are used as alternative pest management [1]. In order to find microorganisms that naturally possess weapons against phytopathogens, we investigate the chemical diversity of plant leaf endophytes. Endophytes are microorganisms living in association with plants and can improve host plants resistance against abiotic and biotic stresses, especially phytopathogens aggressions [2]. The palm *Astrocaryum sciophilum* is the host plant model chosen in this work. Due to the longevity and the easy dating of its leaves [3], we expect to find a highly competitive and diverse microbial community.

197 endophytes have been isolated from eight palm specimens. Each strain has been cultured in competition with the phytopathogen *Fusarium oxysporum*. We focused on the chemical study of *Xylaria* sp.: its ethyl acetate crude extract inhibits the growth of *F. oxysporum*. Moreover, the extract activity of *Xylaria* sp. is exacerbated when the strain is cultured in presence of the phytopathogen. Thus, by comparing LC-MS/MS profiles of extracts and with the help of molecular networking, we identified metabolites of *Xylaria* sp. and *F. oxysporum* with an induced production due to the competition.

Molecular networking allowed us to pilot the isolation process: we focused on the identification of molecules with antifungal activity against *F. oxysporum* and molecules that are produced especially during the competition. Isolation of these metabolites and imaging of the competition between *Xylaria* sp. and *F. oxysporum* is at work to follow the chemical interactions between the two microorganisms.

Acknowledgements: This work has benefited from a grant (SECIL, ref ANR-15-CE21-0016) and from an "Investissement d'Avenir" grant (CEBA, ref ANR-10-LABX-0025) managed by Agence Nationale de la Recherche.

References:

[1] Cantrell et al. J of Nat Prod 2012; 75, 1231–1242
[2] Arnold et al. Proc Natl Acad Sci 2003; 100, 15649–54
[3] Charles-Dominique et al. Dissertationes Botanicae 2001; 153–163

PS2-B-027

Wound healing effect of some tropical fruits

Linda Chularojmontri[1], Khwandow Kunchana[2], Orapin Wongsawatkul[2], Wattanased Jarisarapurin[2], Suvara Wattanapitayakul[2]

[1]Department of Pharmacology, Faculty of Medicine, Srinakharinwirot University, Wattana, Thailand, [2]Department of Preclinical Sciences, Faculty of Medicine, Thammasat University, Klonglung, Thailand

Tropical fruits contain a wide range of bioactive compounds which are main players for natural remedy. Here we investigated the wound healing promoting effect of four tropical fruits, including pineapple (*Ananas comosus*, ACL), sugar apple (*Annona squamosa,* AS), longan (*Dimocarpus longan*, DL), and langsat (*Lansium domesticum*, LD). The fruits were selected based on their ability to promote nitric oxide production in endothelial cells. The angiogenic effect was evaluated *in vitro* using endothelial cell line (EA.hy926); the methods were scratch wound, cell migration, and tube formation assay. AS, ACL, and DL at 500 µg/mL showed significant increases in wound closure to 64.93 ± 1.19, 62.08 ± 2.97 and 59.37 ± 1.49, respectively when compared with control (40.43% ± 0.97%), while LD had no significant effect. Only AS could significantly enhance cell migration (138.27 ± 2.56%). In tube formation assay, the tube length was measured in 3D matrigel seeded with endothelial cells. The positive control VEGF at 0.25 ng/mL significantly increased tube length by 61.87% ± 1.59%. AS showed significant increased tube length by 38.39% ± 6.84%. However, ACL and DL shown no significant increase in tube length. In summary, some tropical fruits contain bioactive compounds that can promote wound healing.

PS2-B-028

Anti-proliferative effects of *Ceratonia siliqua* L. (Carob) fruit extracts on breast cancer cells

Christiana Neophytou[1], Alexandru Vasincu[2], Eve Ioannou[1], Antonia Matsentidou[1], Atalanti Christou[3], Haria Hadjipakkou[3], Andreas Constantinou[1]

[1]Department of Biological Sciences, University of Cyprus, Nicosia, Cyprus, [2]Department of Pharmacodynamics and Clinical Pharmacy, "Grigore T. Popa" University of Medicine and Pharmacy, Faculty of Pharmacy, Iaşi, Romania, [3]Department of Chemistry, University of Cyprus, Nicosia, Cyprus

Introduction: *Ceratonia siliqua* L. (Leguminosae) is the carob plant native to Mediterranean area. Various components of the carob fruit are used as food additives. Our objective is to determine if extracts, isolated from the carob varieties indigenous to Cyprus, have antitumor qualities. As a first step we determined the antiproliferative effects of four extracts, obtained from dried and powdered carob pods, in cultured MCF-7 breast cancer cells and normal immortalized MCF-10A breast cells.
Material and methods: We prepared four extracts with solvents of different polarities including diethyl ether (DE), ethyl acetate (EAc), ethanol (EtOH) and water. We evaluated various concentrations of each extract using the MTT viability assay. Their effect on cell cycle distribution was evaluated using flow cytometry. The DCFH-DA assay was applied to measure the pro-oxidant effects of the extracts.
Results: Our results showed that the DE and EAc extracts had the most potent antiproliferative effects. Thus, DE at 0.5 mg/mL reduced MCF-7 cell viability by over 70% and EAc at the same concentration reduced it by over 50%; The EtOH and water extracts were less active. FACS analysis revealed that the DE and EAc extracts induce a significant increase in the subG1 fraction in MCF-7 cells, indicative of the induction of apoptosis. The EAc extract was very potent in inducing the production of ROS in MCF-7 cells. Importantly, the DE and EAc extracts did not have a significant effect in the viability or cell cycle distribution of normal immortalized MCF-10A cells and did not cause the production of ROS in this cell line.
Conclusions: Among four extracts of carob pods from Cyprus tested, the DE and EAc extracts were the most potent against MCF-7 tumor cell proliferation. Further *in vitro* studies are planned to elucidate their mechanisms of action.

Keywords: *Ceratonia siliqua*, proliferation, apoptosis.

PS2-B-029

Effects of *Carica papaya* seeds on acetaminophen-induced hepatotoxicity in male albino Wistar rats.

Edisua Itam, Isaac Atakpa, Eneji Egbung

University of Calabar, Calabar, Nigeria

The liver is a vital organ involved in key metabolic processes as well as a portal for detoxification of drugs. Acetaminophen is a commonly used analgesic with high incidence of abuse in Nigeria. The present study was designed to investigate the effects of *Carica papaya* seeds on acetaminophen - induced hepatotoxicity in male albino Wistar rats. Thirty-six male albino Wistar rats weighing between 100- 150 g were divided into four groups of nine animals each. Hepato-toxicity was induced using 2500 mg/kg b.wt of acetaminophen. At the end of the fourteen days treatment period, the animals were euthanized, blood samples were collected via cardiac puncture. The plasma harvested was assayed for selected biochemical parameters while the liver was collected and used for tissue histology. The result for AST in the 1000 mg/kg b.wt group (induced and treated) showed a significant decrease ($p < 0.05$) compared to the normal control. The 500 mg/kg b.wt (induced and treated) and 1000 mg/kg b.wt (induced and treated) groups showed a significant decrease ($p < 0.05$) in γ-GT level when compared to the normal control. While albumin in the 500 mg/kg b.wt treated group increased significantly ($p < 0.05$) when compared to the normal. In the 1000 mg/kg b.wt treated group, there was a marked increase in SOD concentration and a decrease in MDA level relative to the control group. Therefore, the 1000 mg/kg b. wt extract of *C. papaya* tend to show a probable restoration of the liver damage in acetaminophen treated rats.

Keywords: acetaminophen, hepatotoxicity, *Carica papaya* Linn

PS2-B-030

A natural oxadiazine isolated from cyanobacteria kills cancer cells in multicellular culture systems by impairing cellular respiration

Maria Lígia Sousa[1,2], Marco Preto[1], Rosário Martins[1,5], Vítor Vasconcelos[1,2], Stig Linder[3,4], Ralph Urbatzka[1]

[1]CIIMAR - Interdisciplinary Centre of Marine and Environmental Research, Matosinhos, Portugal, [2]FCUP-Faculty of Sciences of University of Porto, Porto, Portugal, [3]CCK - Department of Oncology and Pathology, Cancer Centre Karolinska, Karolinska Institute, Solna, Sweden, [4]LÄFO - Department of Medical and Health Sciences, Linköping University, Linköping, Sweden, [5]CISA, ESTSP - Centre of Health and Environmental Research, Polytechnic Institute of Porto, Porto, Portugal

Cyanobacteria are versatile microorganisms that ubiquitously inhabit terrestrial and aquatic ecosystems. They adapt to external threats by mainly producing secondary metabolites. Therefore, cyanobacteria have been recognized as producer of natural products with potential biotechnological applications, such as bioplastics, antifouling, antibiotics, antiprotozoal or anticancer treatment.

A known compound with an oxadiazine ring (Nocuolin A) was re-isolated from the cyanobacteria strain *Nodularia* sp. LEGE06071. These heterocyclic rings were already reported as having anticancer properties, namely as telomerase inhibitors, kinase inhibitors, among others. Nocuolin A (NocA) was demonstrated to be cytotoxic against various cancer cell lines, inducing apoptosis.

In the present study, the activity of NocA was analysed on colon carcinoma cells (HCT116) cultured as monolayer or as multicellular culture systems (MTS). Cancer cells within a tumour are hypoxic and nutrient-deprived, and so, commonly chemotherapy treatments fail to treat the inner part of tumours. 3D cultures of cancer cells represent a good model *in vitro* with high physiological relevance.

In monolayer culture, ATP production was decreased after 2 h of exposure in a dose-dependent manner. After analysing cellular respiration rates over 6 h, oxygen consumption was significantly decreased, indicating an impairment of mitochondrial respiration. Moreover, non-mitochondrial respiration rates were also reduced, indicating that both respiration mechanisms are damaged.

In MTS, exposure to NocA induced apoptosis analysed by the M30 Apoptosense assay. Analysis by fluorescence microscopy of MTS stained with Propidium Iodide also confirmed the tumour cell death, and activity of NocA in 3D cultured colon cancer cells.

The imbalance of ATP caused by NocA makes it an interesting candidate to further study its effects on hypoxic cores of tumours.

Acknowledgements: INNOVMAR, reference NORTE-01-0145-FEDER-000035, within the research line NOVELMAR and Project PTDC/MED-QUI/30944/2017, both co-financed by Portugal 2020 and the European Union through the ERDF and by FCT through national funds.

References:

[1] PLOS One 2017, 12(3): e0172850

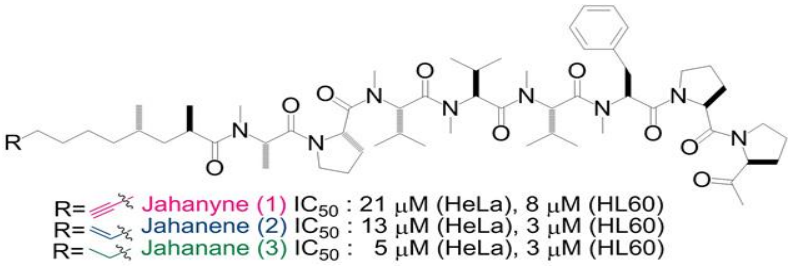
PS2-B-031

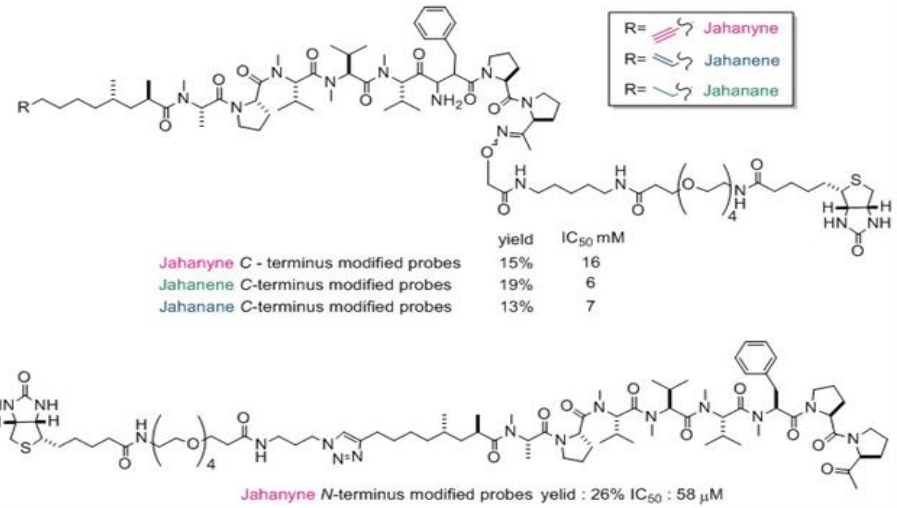
Analyses of the biological activities and the mode of action of marine lipopeptide jahanyne and its analogs

Shizuka Hoshina[1], Arihiro Iwasaki[1], Takafumi Kudo[2], Toshiaki Teruya[2], Kiyotake Suenaga[1]

[1]Keio University, Yokohama, Japan, [2]University of The Ryukyus, Ryukyus, Japan

Marine lipopeptide Jahanyne and its analogs were isolated from marine cyanobacteria by our group (Fig.1). So far, we determined their structures and achieved their total synthesis. Jahanyne and its analogues exhibit the growth-inhibitory activity against human cancer cells such as human cervical cancer cells (HeLa) and human leukemia cells (HL60). To elucidate their mode of action, we investigated the structure-activity relationship and found that the degree of unsaturation of the fatty acid moiety at the N-terminus significantly influenced the cell growth-inhibitory activity. We synthesized two types of jahanyne probes possessing a biotin linker at the N or C-terminus (Fig.2), respectively, and evaluated their activities. As a result, the C-terminus modified probes retained the growth-inhibitory activity against human cancer cells. We carried out affinity purification experiments with HL60 lysate using these probes. As a result, we found a protein specifically bound to the probe retaining the original cell growth-inhibitory activity. Based on the previous works and our experimental results, we hypothesized that this binding protein was Bax, apoptosis-inducing protein, and verified it by western-blotting analyses using Bax antibody.





PS2-B-032

Lignans in male and female *Schisandra rubriflora* soil-grown plants and in their *in vitro* cultures

Agnieszka Szopa[1], Michał Dziurka[2], Marta Klimek-Szczykutowicz[1], Paweł Kubica[1], Angelika Warzecha[1], Halina Ekiert[1]

[1]Chair and Department of Pharmaceutical Botany, Jagiellonian University, Collegium Medicum, 30-688 Cracow, Poland, [2]Polish Academy of Sciences the Franciszek Górski Institute of Plant Physiology, 30-239 Cracow, Poland

Schisandra chinensis is pharmacopoeial plant species in Asia, Europe and USA derived from TCM with well documented e.g. adaptogenic, hepatoprotective, immunostimulant and anticancer properties. They are attributed to specific lignans [1]. Our former studies proved that *S. chinensis in vitro* cultures could be a potential alternative source of lignans [2].
Schisandra rubriflora is dioecious, endemic for China Sichuan province species. There is only poor information about its chemical composition [3].
The aim of the study was the analysis of lignans in: fruit, leaf and shoot extracts of male (♂) and female (♀) specimens collected in different vegetation periods in Poland using UPLC–MS/MS method. The estimations were performed also in microshoot culture extracts (♂,♀ lines) growing on agar MS [4] medium supplemented with 1 mg/l 6-benzylaminopurine and 1 mg/L indole-3-butyric acid.
In the all analyzed methanolic extracts the presence of following groups of lignans were confirmed: dibenzocyclooctadiene lignans (schisantherin A and B, schisandrin, schisandrin C, gomisin A, D, G, J, N, O, 6-*O*-benzoylgomisin O, schisandrin A, rubrisandrin A, epigomisin O, schisanhenol, interiotherin C, angeloylgomisin H and O), aryltetralin lignan (wulignan A1), dibenzylbutane lignans (pregomisin, mesodihydroguaiaretic acid), tetrahydrofuran lignan (fragransin A2), and neolignans (licarin A and B).
The max. total contents (mg/100 g DW) of lignans were as follows: 1055.65 in fruits, 1106.80♂ and 853.33♀ in leaves, and 384.80♂ and 559.97♀ in shoots. The total content (mg/100 g DW) of lignans in microshoot lines extracts were equal 250.92♂ and 220.70♀.
Our results documented for the first time, that both, *S. rubriflora* soil-grown plants and microshoots cultured *in vitro*, could be a rich potential source of lignans of high medicinal value.

Acknowledgements: Funded by National Science Centre, Poland, 2016/23/D/NZ7/01316.

References:

[1] Szopa *et al*. Phytochem Rev 2017; 16: 195–218.
[2] Szopa *et al*. J Biotechnol 2017; 247: 11–17.
[3] Szopa *et al*. Phytochem Rev 2018.
[4] Murashige and Skoog, Physiol Plant 1962; 15: 473–497.

PS2-B-033

In vitro chrysin nanoparticles primary hippocampal cells protection under Cu(II) neurodegeneration conditions

Christiane M. Nday[1], Graham Jackson[2], Athanasios Salifoglou[1]

[1]*Department of Chemical Engineering/Aristotle University of Thessaloniki, 54124 Thessaloniki, Greece,* [2]*Department of Chemistry/University of Cape Town, 7700 Cape Town, South Africa*

Alzheimer disease (AD), affects many people aged 65 years or older, with nearly half of those of age 85 afflicted with this disorder, contributing to 60-70% of dementia cases. Mounting evidence suggests that oxidative stress plays a pivotal role in the development of amyloid peptide and hyperphosphorylated tau pathologoanatomical features.

Prominent among agents inducing oxidative stress are redox active metals such as Cu(II). However, there are natural polyphenol agents, which may be exploited for their antioxidant activity against metal ionic reactivity. For instance, Chrysin ChR can act as an effective anti-oxidant agent, able to improve learning and memory ability.

Developing and using advanced ChR antioxidant forms, exemplified through encapsulation in silica nanoparticles, offers a) advantages of mechanical stability and low toxicity for the encapsulated flavonoid, b) prevents ChR degradation, and c) improves the pharmacokinetic optimization and controls its biodistribution in the body, collectively leading to the efficient permeation of more effective antioxidants through the BBB toward sensitive brain loci. Consequently, a) the synthesis of the base-catalyzed silica gel matrices modified with i) PEG 3000, and ii) CTAB cationic surfactant was pursued and achieved, b) comparison and evaluation of the suitability of these matrices, as potential host-carrier materials for the controlled release of the antioxidant ChR, was made, and c) an investigation of the cytotoxicity and potential protective effects of the ChR-loaded nanoparticles was undertaken under oxidative stress conditions in the presence of Cu(II), notable for its influence on neurodegeneration, in *in vitro* primary hippocampal cultures.

The collective results suggest that the new ChR hybrid nanomaterials contribute to the improvement of therapeutic activity, better protection against degradation, optimization in pharmacokinetics, better control of biodistribution, and decrease of cytotoxicity as a consequence of a slower, more stable ChR release rate, thereby counteracting in a dose-dependent Cu(II)-induced oxidative stress and neurodegeneration.

PS2-B-034

Naringin magnetic silica nanoparticles against amyloid-induced oxidative stress

Christiane M. Nday[1], Graham Jackson[2], Athanasios Salifoglou[1]

[1]Department of Chemical Engineering/Aristotle University of Thessaloniki, 54124 Thessaloniki, Greece, [2]Department of Chemistry/University of Cape Town, 7700 Cape Town, South Africa

Neuronal connectivity, which enables learning and memory, degrades progressively during brain neurodegenerative pathological conditions such as Alzheimer's disease (AD). Cu(II)-mediated oxidative stress has been shown to play a pivotal role in regulating redox reactions leading to formation of RNS/ROS, major culprits in AD.

The antioxidant properties of natural bioactive flavonoids including naringin (NAR) in neurodegenerative processes have been well-documented. However, NAR magnetic encapsulation in nanoparticles (MQNPs) may further protect neuronal survival and morphological connectivity has been poorly demonstrated.

To investigate potential effects of nano-encapsulated naringin (NAR) on neuronal survival and synaptic morphology in primary rat hippocampal neurons, PEGylated silica nanoparticles were synthesized. NAR was loaded on silica nanoparticles in a concentration-dependent fashion, and release studies were carried out using UV-Visible spectroscopy. Further physicochemical characterization of the novel MQNPs included elemental analysis, particle size, z-potential, FT-IR, BET, TGA, and SEM analysis in order to optimize material composition linked to the delivery of loaded NAR in the hippocampal cell milieu.

The findings reveal that, under amyloid induced oxidative stress, the loading ability of the MQNPs was concentration-dependent, based on their NAR release profile. The overall bio-activity profile of the new hybrid nanoparticles a) denoted their enhanced protective activity against oxidative stress as well as hippocampal cell survival in comparison to previous results on NAR, and b) established the basis for in-depth perusal of molecular events underlying synaptic processes, collectively linked to preventive medical nanotechnology in neurodegeneration.

PS2-B-035

Clavariopsins C-H, antifungal cyclic depsipeptides from the aquatic hyphomycete *Clavariopsis aquatica*

Thin Wut Soe[1], Chunguang Han[2], Tomohiko Tomura[1], Ryosuke Fudou[3], Makoto Ojika[1]

[1]Graduate School of Bioagricultural Sciences, Nagoya University, Chikusa-ku, Nagoya, Japan, [2]Research Center for Materials Science, Nagoya University, Chikusa-ku, Nagoya, Japan, [3]R&D Planning Department, Ajinomoto Co., Inc., Chuo-ku, Tokyo, Japan

Aquatic hyphomycetes are a phylogenetically heterogeneous group of fungi occurring mainly in lotic fresh waters. Investigation of the secondary metabolites from aquatic hyphomycetes has become a promising strategy of discovering novel bioactive natural product due to the difficulty of culture. We previously isolated two antifungal cyclic deca-depsipeptides, calvariopsins A and B (1 and 2), from the aquatic hyphomycete *Clavariopsis aquatica* [1]. Afterward, we investigated the constituents of the crude extracts in a large scale and discovered six new analogs. We will present isolation, structure elucidation, and biological evaluation of them.

The fungus was cultured at 25 °C for 18 days. The culture broths (100 flaks) were extracted with acetone and the extract was partitioned between EtOAc and H_2O. The EtOAc fraction was chromatographed by silica gel column, then cyclic peptide fractions were repeating purified by preparative HPLC to afford six new analogs clavariopsins C-H (3-8) together with 1 and 2. Their planar structures were elucidated by 2D NMR and HRESIMS. The absolute configurations of the residues (nine amino acids and one hydroxy acid) were determined by the advanced Marfey' s method and chiral HPLC analysis. Their antifungal and cytotoxic activities were evaluated against six plant pathogenic fungi (*Botrytis cinerea*, *Fusarium oxysporum*, *Alternaria alternata*, *Magnaporthe oryzae*, *Colletotrichum orbiculare*, and *Aspergillus niger*) and a human cancer cell line (HeLa-S3), respectively. In addition, the hyphal swelling induction activity was examined against *A. niger*. Most clavariopsins exhibited potent antifungal activity (minimum inhibition dose = 0.1-10 μg/disk), hyphal swelling induction (minimum effective dose = 0.3-3.0 μg/disk) and moderate cytotoxicity (IC_{50} = 23-48 μM). Based on these results, a brief SAR will be discussed.

Keywords: aquatic hyphomycete, *Clavariopsis aquatica*, cyclic depsipeptide, antifungal activity, cytotoxicity

References:

[1] Suzuki Y, Ojika M, Sakagami Y, Kaida K, Fudou R, Kameyama T. J Antibiot 2001; 54: 22–28.

PS2-B-036

Discovery of new GPBAR1 agonists by ligand-based pharmacophore modeling and virtual screening

Benjamin Kirchweger[1], Jadel M Kratz[1], Angela Ladurner[1], Ulrike Grienke[1], Verena M. Dirsch[1], Thierry Langer[2], Judith M. Rollinger[1]

[1]*Department of Pharmacognosy, Faculty of Life Sciences, University of Vienna, Vienna, Austria,* [2]*Department of Pharmaceutical Chemistry, University of Vienna, Vienna, Austria*

Agonists of the G protein-coupled bile acid receptor (GPBAR1) exert antidiabetic and anti-inflammatory effects *in vitro* and *in vivo*. Previously, it has been shown that a number of herbal remedies and natural small molecule metabolites are able to activate this receptor [1] and thus may exert a therapeutic potential. For a fast-forward discovery of novel GPBAR1 agonists from natural sources we recently elaborated an *in silico* workflow [2]. We employed the software package LigandScout [3] to develop several ligand-based pharmacophore models for GPBAR1 agonists. After theoretical validation, the models were used as queries for virtual screening experiments with open-source and in-house molecular databases. Virtual hits were further processed using shape-focused similarity search and physicochemical clustering. *In vitro* evaluation of 34 promising and chemically diverse predicted hits revealed several potent activators, including new scaffolds from natural and synthetic origin. Triterpenes previously isolated from the South African tree *Burkea africana* and coumarins from the middle-eastern spice *Ferula assa-foetida* showed activities comparable to the endogenous ligands chenodeoxycholic acid and lithocholic acid.

Acknowledgments: The authors thank OpenEye for providing the ROCS software free of charge.

Keywords: GPBAR1, bile acid, pharmacophore, virtual screening, coumarins, triterpenes

References:

[1] Ladurner A, Zehl M, Grienke U, Hofstadler C, Faur N, Pereira FC, Berry D, Dirsch VM, Rollinger JM. Front Pharmacol 2017; 8: 468.
[2] Kirchweger B, Kratz JM, Ladurner A, Grienke U, Langer T, Dirsch VM, Rollinger JM. Front Chem 2018; 6: 242.
[3] Wolber G, Langer T. J Chem Inf Model 2005; 45(1): 160–9.

PS2-B-037

Butanol fraction of *Ficus carica* fruit enhances pancreatic *β*-cell functions, stimulates insulin secretion and ameliorates other type 2 diabetes-associated parameters in rats

Shahidul Islam[1], Ramgopal Mopuri[1,3], Chika I. Chukwuma[1], Balaji Meriga[2], Dowlathabad Muralidhara Rao[3]

[1]Dept. of Biochemistry, University of Kwazulu-Natal, Durban, South Africa, [2]Dept. of Biochemistry, Sri Venkateswara University, Tirupati, India, [3]Dept. of Biotechnology, Sri Krishnadevarava University, Anantapur, India

This study investigated the anti-diabetic activity of the butanol fraction of *Ficus carica* fruits (FCBF) in a type 2 diabetes (T2D) model of rats. T2D was induced by feeding 10% fructose solution ad libitum for two weeks followed by an intraperitoneal (i.p.) injection of streptozotocin (STZ) with a dose of 40 mg/kg body weight) (BW) and the animals were orally treated with 150 or 300 mg/kg BW of the FCBF for five days in a week for 5 weeks intervention period. Another group of non-diabetic rats was similarly administered with 300 mg/kg BW of the FCBF to examine its possible toxicological effects when only vehicle was ingested to the animals in normal and diabetic control groups. Food and fluid intake were measured daily, when body weight changes and blood glucose levels were monitored weekly during the entire intervention period. The 300 mg/kg BW of FCBF treated groups significantly ($p<0.05$) decreased the blood glucose and improved the glucose tolerance ability compared to diabetic rats. The FCBF treatment improved pancreatic *β*-cell function (HOMA-*β*), stimulated insulin secretions, decreased insulin resistance (HOMA-IR), restored liver glycogen, ameliorated serum lipid profiles and decreased liver function enzymes compared to untreated diabetic rats. From the results of this study, it can be concluded that FCBF possesses remarkable anti-diabetic activity which was mediated through the activation of pancreatic *β*-cell function as well as insulin secretion. Some of its identified bioactive compounds might be responsible for these antidiabetic activities.

PS2-B-038

An antifungal substance from the endophytic fungi *Epichloë festucae* and its effective production by gene modification

Enkhee Purev, Jennifer Niones, Daigo Takemoto, Tatsuhiko Kondo, Hitomi Isobe, Rika Miura, Takushi Hashikawa, Kazuhito Kawakita, Makoto Ojika

Graduate School of Bioagricultural Sciences, Nagoya University, Chikusa-ku, Nagoya, Japan

The *Epichloë* endophytes are a group of the Clavicipitaceous fungi family that form symbiotic relationship with a broad spectrum of grasses such as perennial ryegrasses and enhance their resistance against plant pathogens and harmful insects [1]. We have previously revealed that *Epichloë festucae* strain E437 secreted an unknown antifungal substance and it was more effectively produced by a genetically modified strain, E437_vibA, which overexpressed the transcription factor gene vibA. We will present the structure elucidation, heterologous expression of the biosynthetic gene, and biological evaluation of the endophyte antifungal substance.

The mutant strain E437_vibA was cultured in 10 days in PDB medium. The culture filtrate, which showed inhibition of conidia germination of the plant pathogen *Drechslera erythrospila*, was chromatographed on an ion exchange resin. The adsorbed fraction was separated by HPLC with ODS and then HILIC columns to afford a pure active compound, which was determined as ε-poly-L-lysine (εPL) based on NMR, MALDI-TOF MS, advanced Marfey' s method, and the comparison with standard εPL. The polymerization degree judged from MS ranged between 24-36, which is similar to that of the standard isolated from *Streptomyces albulus*. The isolated εPL inhibited the conidial germination of *D. erythrospila* at a minimum dose of 50 μg/mL. Since we found an εPL synthase (pls)-like gene in the *E. festucae* genome, the pls gene was transferred to the different strain *E. festucae* Fl1 to obtain pls-over-expressed strain Fl1_pls, which actually produced εPL. We are trying to establish other genetically modified strains that can effectively produce εPL and evaluating antifungal activity against several important fungal pathogens such as *Magnaporthe oryzae*.

Acknowledgments: We thank JNC Co. (Tokyo, Japan) for providing us with standard ε-poly-L-lysine.

Keywords: endophyte, antifungal, ε-poly-L-lysine, conidia

References:

[1] Schardl CL, Fungal Genet Biol 2001; 33, 69–82.

PS2-B-039

Caenorhabditis elegans as model to study natural products affecting metabolism and lifespan

Theresa Lehner, Benjamin Kirchweger, Julia Zwirchmayr, Ammar Tahir, Judith M. Rollinger

Department of Pharmacognosy, Faculty of Life Sciences, University of Vienna, Althanstraße 14, 1090 Vienna, Austria

Current drug discovery efforts are focused on rationale target directed approaches, but a major part of new drug entities is still discovered by phenotype directed assays [1]. Screening of natural products in phenotypic rodent models is hampered by several disadvantages, e.g. financial efforts, legal and ethical considerations, large quantities of test materials, in particular pure isolates, and a challenging target deconvolution afterwards. The simple roundworm *Caenorhabditis elegans* is considered as a convenient and proficient addition to the set of current preclinical model organisms [2]. We recently established a robust *C. elegans* screening platform using 96-well plates for medium throughput screening of extracts and constituents thereof for the discovery of natural products beneficial to the metabolic syndrome. Herein we present approaches and methods for *C. elegans* based preclinical screening using a combination of (i) optimized extract preparation, (ii) lifespan assay and (iii) fat accumulation assay.

Keywords: *Caenorhabditis elegans*, model organism, metabolic syndrome, lifespan, fat accumulation assay

References:

[1] Swinney DC, Anthony J. Nat Rev Drug Discov 2011; 10: 507–519.
[2] O'Reilly, Linda P. *et al*. Adv Drug Deliv Rev 2014; 0: 247–253.

PS2-B-040

Evaluation of Mistletoe (*Viscum album* L.) callus and plant protein extracts against gynecological cancer cell lines.

Vasileios Tsekouras[1], Georgia Moschopoulou[1], Sophia Mavrikou[1], Maria-Argyro Karageorgou[2,3], Spyridon Kintzios[1]

[1]Faculty of Food Science, Biotechnology and Development, Department of Biotechnology, Agricultural University of Athens, Iera Odos 75, 11855, Athens, Greece, [2]Institute of Nuclear & Radiological Sciences & Technology, Energy & Safety, National Center for Scientific Research "Demokritos, Aghia Paraskevi, 15310, Athens, Greece, [3]Department of Solid-State Physics, NKUA, Athens, Greece

Mistletoe (*Viscum album* L.) is a plant semiparasite growing on a wide range of host plants, traditionally regarded as a natural source of medicinal compounds. Extracts containing alkaloids, viscotoxins, lectins, polysaccharides [1] and triterpene acids are applied for a variety of conditions in complementary and alternative medicine therapies, especially in cancer treatment [2]. Mistletoe tissue culture has been suggested as an approach for standardizing the production of bioactive compounds plus the biosynthesis of novel somaclonal lectins. [3]. This research is focused on highlighting the effect of biotechnologically produced mistletoe lectins against cancer cell lines providing a basic foundation for clinical use.

Callus cultures, derived from plants growing on firs trees in mount Parnitha were used as source of pharmaceutical extracts. Plant cells cultured *in vitro* can undergo a process known as somaclonal variation, affecting the qualitative and quantitative production of proteins. We produced and preserved a number of calluses tested for somaclonal variations in regard of their protein profile. Somaclonal callus lines were selected, propagated and subjected to protein extraction, isolation and purification procedures for further evaluation. The pharmaceutical efficacy of isolated protein molecules, derived from both donor plants and biotechnological produced calluses, was tested by several bioassays. More specific, we investigated the cytotoxic effects of mistletoe lectins and enriched protein extracts, derived from callus tissues, somaclonal callus tissues and donor plants against the selected gynecological cancer cell lines HeLa, MCF-7 and MDA-MB-231.

Mistletoe lectins were radiolabeled with Technetium-99m (^{99m}Tc) to quantify the accumulation of the radiolabeled lectins *in vivo*. Evaluation of their radiochemical purity as well as *in vitro* stability studies in PBS and human serum are demonstrated. *In vivo* biodistribution studies were performed in Swissmice at 30, 60, 120 min post injection.

Acknowledgements: This work is supported by the Operational Program "Human Resources Development, Education and Lifelong Learning", Priority Axes 6, 8, 9 co-financed by European Social Fund entitled "Support of Researchers with Emphasis on Senior Researchers" (NSRF 2014-2020). We thank Dr. P. Bouziotis and Dr. M.Paravatou from the Institute of Nuclear and Radiological Sciences and Technology, Energy and Safety, National Centre for Scientific Research "Demokritos" for assistance in radiochemical studies and the donation of cell lines, respectively. Also, we thank Dr.J.Zoidakis and Dr. M. Makridakis from the Proteomics Laboratory, Biomedical Research Foundation, Academy of Athens who helped with the protein analysis.

Keywords: cancer, cytotoxicity, lectin, mistletoe, protein isolation, radiolabeling, technetium-99m

References:

[1] Seema P, Suryakanta P. 3 Biotech 2014; 4(1): 13–20.

[2] Mulsow K, Enzlein T, Delebinski C, Jaeger S, Seifert G, Melzig MF. PLoS ONE 2016; 11(4): e0153825.
[3] Barberaki M, Dermitzaki E, Margioris AN, Theodosaki M, Grafakos S, Kintzios S. Curr Bioact Compd 2015; 11(2): 104–108.

PS2-B-041

Essential oil characterisation, antimicrobial and antioxidative activity of extracts of *Erodium cicutarium* (L.) L'Hér. ex Aiton (Geraniaceae) from Croatia

Vanja Ljoljić Bilić[1], Ivan Kosalec[1], Dario Kremer[2], Valerija Dunkić[3], Jadranka Vuković Rodríguez[4]

[1]Department of Microbiology, Faculty of Pharmacy and Biochemistry, University of Zagreb, Zagreb, Croatia, [2]Department of Pharmaceutical Botany with Fran Kušan botanical garden, Faculty of Pharmacy and Biochemistry, University of Zagreb, Zagreb, Croatia, [3]Department of Biology, Faculty of Sciences and Mathematics, University of Split, Split, Croatia, [4]Department of Pharmaceutical Analysis, Faculty of Pharmacy and Biochemistry, University of Zagreb, Zagreb, Croatia

The aim of this study was phytochemical characterisation of essential oil and screening of biological effects of plant extracts from aerial parts of *Erodium cicutarium* (L.) L'Hér. ex Aiton from four localities in Croatia (Buzin, Trešnjevka, Plitvice, Podvinje). This native traditional medicinal plant was used for essential oil preparation via hydrodistillation and GC-MS analysis was performed [1]. The screening of biological effects included *in vitro* antioxidant and antimicrobial activity evaluation of plant extracts (aqueous and methanolic). Cupric ion reducing antioxidant capacity assay (CUPRAC) was used to determine antioxidant potency [2]. Standard antimicrobial *in vitro* assays including agar well diffusion assay [3] and serial twofold microdilution assay [4], were used for an antimicrobial screening on *Staphylococcus aureus* ATCC 6538, *Escherichia coli* ATCC 10536 and *Candida albicans* ATCC 90028. GC-MS analysis showed similarity in composition of the major compounds of all four essential oils and, in total, 50 compounds were identified with hydrocarbons as main class of constituents (62.5–65.7%). All analyzed extracts exhibited antioxidant activity in the CUPRAC assay, with higher values for methanol than water extracts. The most active extract was from locality Podvinje (74.6 mg TEs/g extract*). In the well diffusion assay, all investigated extracts show highest activity against *S. aureus*, with zones of bacterial growth inhibition (ZI) ranging from 12±2.5mm (Trešnjevka) to 21±1.41 mm (Podvinje). Activity against *C. albicans* was shown only for methanolic extracts, with highest value of ZI=15±2.55 mm (Podvinje). Within the microdilution assay, minimal inhibitory concentration (MIC) values were obtained only for *S. aureus*. MIC values are within the range from 3.75±1.77 mg/mL (Podvinje, Trešnjevka) to 20±0.00 mg/mL (Buzin). These results contribute to the scientific study of phytotherapeutic potential of the investigated plant species, especially in relations to essential oil composition, antioxidant and antimicrobial activity, and give an impetus for further biological studies.
Note: *TEs/g extract, Trolox equivalents/g extract.

Keywords: *Erodium cicutarium* (L.) L'Hér. ex Aiton, essential oil, antioxidant capacity, antimicrobial activity.

References:

[1] Dunkić V, Kremer D, Jurišić Grubešić R, Vuković Rodríguez J, Ballian D, Bogunić F, *et al*. South African J Bot 2017; 111: 232–41.
[2] Apak R, Güçlü K, Özyürek M, Karademir SE. J Agric Food Chem 2004; 52(26):7970–81.
[3] European Pharmacopoeia, 5th ed, Council of Europe, Strasbourg 2005, pp. 188–191.
[4] Zovko Končić M, Kremer D, Gruz J, Strnad M, Biševac G, Kosalec I *et al*. Food Chem Toxicol 2010; 48(6): 1537–42.

PS2-B-042

Chemical-biological study of ent-kaurane and ent-trachylobane diterpenes from *Psiadia punctulata*.

Lorenzo Fiengo[1], Antonio Vassallo[2], Roberta Cotugno[1], Nunziatina De Tommasi[1], Fabrizio Dal Piaz[3], Ammar Bader[4]

[1]DIFARMA, University of Salerno (Italy), Fisciano, Italy, [2]Department of Science, University of Basilicata, Potenza, Italy, [3]Department of Medicine, Surgery and Odontology, University of Salerno, Fisciano, Italy, [4]Umm Al-Qura University, La Mecca, Saudi Arabia

Recently our research group was involved in the building-up of diterpene libraries from Lamiaceae and Asteraceae family to screen against protein targets by a multidisciplinary approach. Our project is focused on the study of interaction between diterpenes and proteins involved in cancer and inflammation processes, using cell-free and cell-based assays.
One of our target protein is nucleolin (NCL). It is a multilocalized protein implicated in physiological processes such as remodelling of chromatin structure, ribosome biogenesis, DNA transcription, and in cancer, inflammation and viral diseases [2].
In order to discovery molecules able to modulate NCL activity, a kaurane and trachylobane library was screened in Jurkat and HeLa cells against NCL, using Celluar Thermal Shift Assay (CETSA). This screening led us to obtain the diterpene 6,19-dihydroxy-ent-trachiloban-17-oic acid (1) as NCL ligand. To validate compound 1-NCL interaction, the apparent melting curve of NCL (Tm: 55 °C), the EC_{50} of the complex (5 μM) and the time-course experiment observed after 2 h of incubation were determined by CETSA. Subsequently, a chemical proteomic Drug Affinity Responsive Target Stability (DARTS) approach was performed to validate the interaction [4]. DARTS results, analyzed by Mass Spectrometry and Western Blot, confirmed compound 1/NCL interaction. Furthermore, to investigate the diterpene activity in HeLa cells, we carried out: MTT (EC_{50}: 50 μM), Protein Synthesis assays, Cytofluorimetric analysis (sub G0/G1 cell cycle arrest), Real Time PCR and WB (decrease of AKT and Bcl2 levels).

Keywords: diterpenes, drug discovery, cell-free and cell-based assays, nucleolin

References:

[1] Mahadeo K, Grondin I, Kodja H, Soulange J, Jhaumeer Laulloo S, Frederich M, Gauvin-Bialecki, A. J Ethnopharmacol 2018; 210: 48–68.
[2] Berger CM, Gaume X, Bouvet P. Biochimie 2015; 113: 78–85.
[3] Dal Piaz F, Saltos M, Franceschelli S, Forte G, Marzocco S, Tuccinardi T, Ebrahimi S, Hamburgher M, De Tommasi N, Braca A. J Nat Prod 2016; 79: 2681–2692.

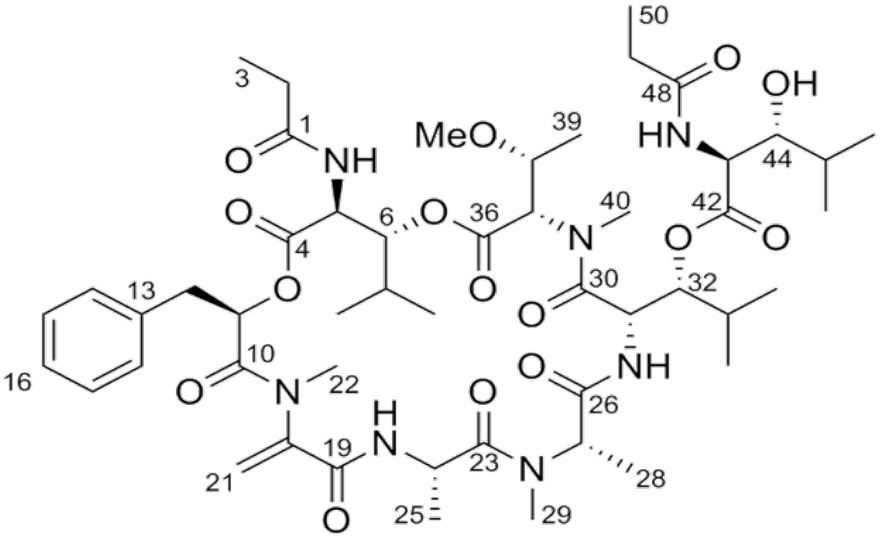
PS2-B-043

Sameuramide A, a new cyclic depsipeptide isolated from an ascidian of the family Didemnidae

Koshi Machida[1,2], Daisuke Arai[2], Ryosuke Katsumata[1], Satoshi Otsuka[1], Jun K. Yamashita[3], Tao Ye[4], Shoubin Tang[4], Nobuhiro Fusetani[5], Yoichi Nakao[1,2]

[1]Department of Chemistry and Biochemistry, Graduate School of Advanced Science and Engineering, Waseda University, 3-4-1 Okubo, Shinjuku-ku, Tokyo 169-8555, Japan, [2]Research Institute for Science and Engineering, Waseda University, 3-4-1 Okubo, Shinjuku-ku, Tokyo 169-8555, Japan, [3]Department of Cell Growth and Differentiation, Center for iPS Cell Research and Application, Kyoto University, 53 Shogoin Kawahara-cho, Sakyo-ku, Kyoto 606-8507, Japan, [4]State Key Laboratory of Chemical Oncogenomics, Peking University Shenzhen Graduate School, Xili, Nanshan District, Shenzhen, 518055, Cina, [5]Fisheries and Oceans Hakodate, 3-1-1 Minato-cho, Hakodate, 041-8611, Japan

Sameuramide A, a new cyclic depsipeptide consisting of alanine, *N*-methyl alanine, *N*-methyl dehydroalanine, *N,O*-dimethyl threonine, phenyllactic acid, *β*-hydroxy leucine, and propionate, was isolated from a didemnid ascidian collected at the northern part of Japan. The planar structure was established based on the interpretation of MS and NMR data. The absolute configuration of the subunits was assigned by the advanced Marfey's method and the chiral LC-MS analysis. Sameuramide A exhibited the activity of maintaining colony formation of murine embryonic stem (mES) cells [1,2] without leukemia inhibitory factor (LIF)[3,4] while down regulating of the gene expression of Krüppel-like transcription factor 4 (Klf4). However, the expression levels of the marker genes (Nestin, T, Sox17) for three germ layers were upregulated in embryoid bodies (EBs) after treatment of sameuramide A together with LIF, suggesting that sameuramide A plays a supportive role for LIF in maintaining the multipotency of mES cells, whereas indicates no effect for undifferentiated state of the mES cells.



PS2-B-044

Narciclasine of *Cyrtanthus contractus* has *in vitro* anti-inflammatory properties identified by correlation-based metabolomics

Lucie Rarova[1], Bhekumthetho Ncube[2], Johannes Van Staden[2], Robert Fürst[3], Miroslav Strnad[4], Jiri Gruz[4]

[1]Department of Chemical Biology and Genetics, Centre of the Region Haná for Biotechnological and Agricultural Research, Faculty of Science, Palacky University in Olomouc, Olomouc, Czech Republic, [2]Research Centre for Plant Growth and Development, School of Life Sciences, University of KwaZulu-Natal, Pietermaritzburg, South Africa, [3]Institute of Pharmaceutical Biology, Biocenter, Goethe University, Frankfurt/Main, Germany, [4]Laboratory of Growth Regulators, Centre of the Region Haná for Biotechnological and Agricultural Research, Institute of Experimental Botany ASCR & Palacky University , Olomouc, The Czech Republic

Treatment of inflammation-related disorders by natural products is still in high demand by pharmaceutical companies due to the lack of effective anti-inflammatory drugs. In this study, we demonstrated that an extract from *Cyrtanthus contractus* has a strong anti-inflammatory activity *in vitro*. To further investigate the active principle of *Cyrtanthus contractus*, we partially separated the extract into 14 fractions, analysed each fraction by non-targeted UHPLC-QTOF-MS and calculated correlation coefficients between biological activities and metabolite levels in each fraction. As a result, the top scoring metabolite from both ionization modes was structurally identified as narciclasine by comparing its MS/MS spectra with those of authentic standard. Due to the known anti-inflammatory activity of this alkaloid, narciclasine was proposed as the active principle of *Cyrtanthus contractus*. This hypothesis was further confirmed by comparing the biological effect of crude extract with that of an authentic standard.

PS2-B-045

Antiplasmodial activity and interaction between cannabidiol and artemisinin

Lúcia Mamede[1], Gilles Degotte[2], Allison Ledoux[1], Olivia Jansen[1], Pauline Desdemoustier[1], Michel Frédérich[1]

[1]Pharmacognosy Laboratory, CIRM, University of Liège, Quartier Hôpital – B36 Tower 4, Avenue Hippocrate 15, 4000 Liège, Belgium, [2]Laboratory for Pharmaceutical Analysis, Center of Interdisciplinary Research on Medicines (CIRM), University of Liège, Avenue Hippocrate 15, 4000 Liège, Belgium

Cannabis sativa, an herbaceous species that originated in Central Asia, has been used in traditional medicine since at least 6000 years. The versality of its applications led to the discovery of phytocannabinoids, terpenophenolic compounds produced by the plant, and the receptors with whom they interact with, cannabinoid receptors. Despite the importance of this discovery, the pharmacological properties of the hundreds of compounds isolated from this plant are still fairly unknown and studied nowadays. In the light of this paradigm, one of the most abundant cannabinoids, cannabidiol, was evaluated *in vitro* on the malaria parasite, *Plasmodium falciparum*, to assess its possible activity. This parasite is considered a priority by the World Health Organization, as it is responsible for thousands of deaths yearly in its endemic regions. Cannabidiol has proved invaluable in its medical applications, e.g. as the treatment for epilepsy or because of its counteractive effect to the psychoactive cannabinoid in *C. sativa*. However, little has been described about its antiparasitic activity. To assess it, growth inhibition tests and isobologram tests were performed with cannabidiol and artemisinin. These were done for two different *P. falciparum* strains, one chloroquine-sensitive and one partially resistant to artemisinin. Cannabidiol showed an interesting activity on the chloroquine-sensitive strain, when compared with the artemisinin-resistant strain. The isobologram tests disclosed synergism between cannabidiol and artemisinin. These tests prove the added interest in the continuation of research on the interaction between cannabidiol and the malaria parasite.

Keywords: *Cannabis sativa*, cannabinoids, malaria, *Plasmodium falciparum*

References:

[1] Welty TE, Luebke A, Gidal BE. Epilepsy Curr 2014; 14: 250–252.
[2] World malaria report 2017. Geneva: World Health Organization; 2017. Licence: CC BY-NC-SA 3.0 IGO.

PS2-B-046

Physicochemical characterization and stability assessment of oil-in-water nanoemulsions as delivery system of bioactive compounds: Conjugated linoleic acid (CLA) as model compound

Maria Katsouli, Virginia Giannou, Constantina Tzia

National Technical University of Athens, Athens, Greece

Conjugated linoleic acid (CLA) is naturally contained in lamb, beef and dairy products and has been reported to present several health benefits (antioxidant and anti-inflammatory properties). Recently, it is gaining recognition as a food supplement due to its various physiological activities and other promising effects in certain allergies. However, it is practically insoluble in water and poorly absorbed from the gastrointestinal tract, thus its oral delivery is challenging. Moreover, the consumption of dietary CLA has decreased because animal fats had been replaced by plant lipids consequently, the enrichment of CLA in food has been gained great interest. Therefore, it has been chosen as the bioactive compound of interest for this study.
There has been growing demand to fortify foods with health-promoting ingredients using food-delivery systems. Nanoemulsions as delivery systems are finding increased use in food, pharmaceutical and cosmetic applications due to their ability to increase the stability of the bioactive compound. Nanoemulsion are non-equilibrium colloidal systems with high kinetic stability, consisting of two immiscible liquids.
The scope of present study was to develop CLA-loaded nanoemulsions using extra-virgin olive (EVOO) or olive-pomace oil (OPO) and Tween 20/Tween 40. It was examined the influence of the incorporated CLA on the physico-chemical properties of nanoemulsions. The effect of the type and ratio of emulsifiers and oils were evaluated based on mean droplet diameter (MDD), polydispersity index, ζ-potential, viscosity, turbidity and the incorporation stability of CLA. Additionally, their physical-chemical stability was evaluated by monitoring of MDD and CLA retention during 90 days storage at 4 ˚C or 25 ˚C.
CLA-loaded nanoemulsions using both vegetable oils presented desirable properties in terms of physical and kinetic stability and both presented satisfactory chemical stability during storage, rendering them excellent delivery systems. Furthermore, all formulations after storage remained in the nanosized range, with high physical and kinetic stability.

PS2-B-047

Assessment of phytopathogenic fungi diseases removal and plant protection induction by plant-associated-bacteria of *Vicia faba* L. plant: Broad spectrum of bioactive metabolites for antifungal and plant growth promoting activities

Imen Haddoudi[1,2,3], Isabel Mora[3], Jordi Cabrefiga[3], Emilio Montesinos[3], Moncef Mrabet[2]

[1]University of Tunis El Manar 2092, Tunis, Tunisia, Faculty of sciences Tunis El Manar, Tunis, Tunisia, [2]Laboratory of Legyme, Centre of Biotechnology of Borj-Cédria (CBBC) BP. 901, Hammam-Lif 2050, Tunisia, Tunis, Tunisia, [3]Laboratory of Plant Pathology, Institute of Food and Agricultural Technology-XaRTA-CIDSAV, University of Girona, Campus Montilivi, 17071, Girona, Spain, Tunis, Tunisia

In this study, we tried to select good candidates produced different bioactive metabolites especially for antifungal activities against different fungal diseases threatening *Vicia faba* L. plant. A collection of 33 bacterial strains was selected from 500 plant-associated-bacterial of *Vicia faba* L. plants based on their high antifungal activity against *Fusarium oxysporum* as first screening, then selected strains tested against 11 different phytopathogenic fungi (Fig1). Analysis of the presence of Cycliclipopeptides (cLPs) genes showed that that fenD and srfAA were the most frequently genes detected 50% and 70%, respectively, then bmyB (40%), when the spaS and dfnM were the less frequent genes (10% and 20%, respectively). A 78% of stains presented at least one of the cLPs genes, although 22% of isolates had not any biosynthetic genes (Fig2). The analysis of the production of the cLPs by HPLC showed that 30% of strains produced Iturin, Fengycin and Surfactin, when 12% only Iturin and Surfactin, 6% Iturin and Fengycin, when others strains were able to produce only one type of Lipopeptide Fengycin and Iturin (18% and 6%, respectively) (Fig3). The correlation among all variables parameters pair-wise was calculated using Pearson correlation coefficient and Bonferroni correction, shows a significantly relationship between antifungal activities and the production of Iturin. We detected some Plant Growth Promoting metabolites such as produced Siderophores, HCN, Auxine and Phosphate solubilization (Fig4.A). Furthermore, the strains were able to produce some enzymes such as Amylase, Protease, Cellulase and Pectinase (Fig4.B). The Biocontrol essay on *Vicia faba* L. plants under greenhouse shows the capacity of selected strains to protect plants from two different phytopathogenic fungi with growth improving.

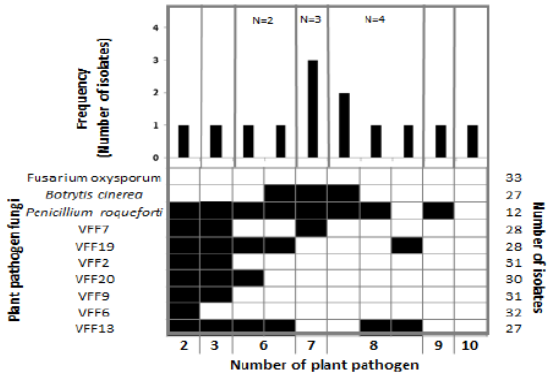


Fig1: Frequency distribution of antifungal activity of selected isolates

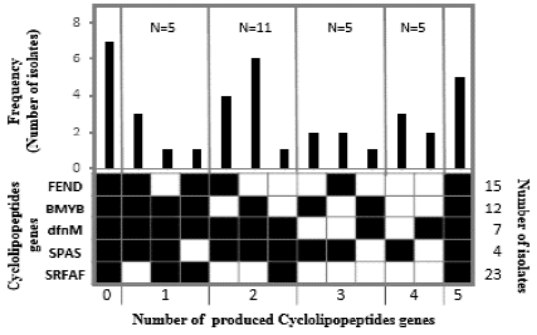


Fig2: Frequency distribution of Cycliclipopeptides genes of the 33 selected bacterial strains

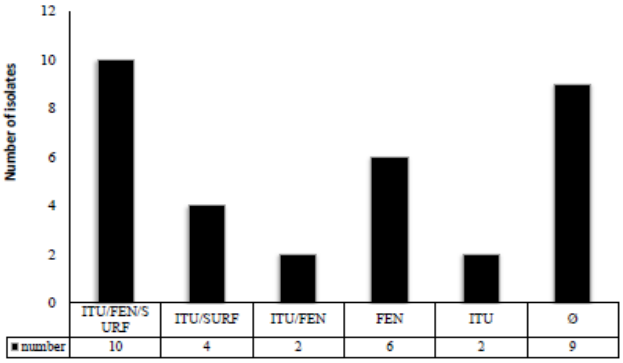


Fig3: Production of Iturin, Fengycin and Surfactin among the 33 fungal isolates

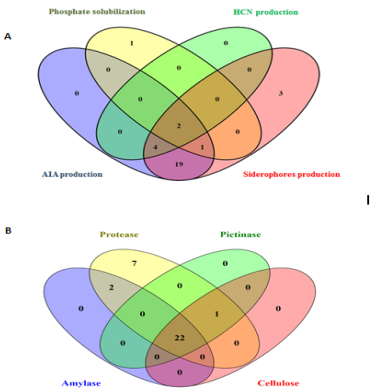


Fig4: **(A)** Venn diagram showing which strains are shared capacity of producing AIA, Siderophores, HCN and phosphate solubilization. **(B)** Venn diagram showing which strains are shared capacity of producing Amylase, Protease, Pectinase and Cellulase enzymes

Acknowledgements: to Dr. Emilio MONTESINOS director of CIDSAV laboratory in Spain for hosting me and Dr. Haythem MHADHBI director of Legumes laboratory in Tunisia

Keywords: plant-associated-bacteria, *Vicia faba* L. plant, antifungal activities, bioactives metabolites.

PS2-B-048

Toxicity, antioxidant and anti-asthmatic studies of *Anchomanes difformis* (Blume) Engl. leaf extract

Oghale Ovuakporie-uvo, MacDonald Idu

University of Benin, Benin City, Nigeria, [2]University of Benin, Benin City, Nigeria

Aim: *Anchomanes difformis* (Blume) Engl is a self-supporting rhizomatous herb reported to have several medicinal uses. This study was undertaken to determine its biosafety and validate its folkloric use in the management of asthma in Delta State, Nigeria.
Materials and methods: For toxicity studies, mice (25-45 g) and Albino rats (120-225 g) were used following a slight modification of Locke's method. *A. difformis* aqueous leaf extract was orally administered to animals for 24 h (Acute) and 21 days (sub-acute). Serum biochemistry, haematology and histopathology of selected vital organs were screened for toxic signs. *In vitro* antioxidant assays; 2, 2-diphenyl-1-picrylhydrazyl (DPPH) and lipid peroxidation assays were carried out following reported procedures. Thirty six (36) guinea pigs (420-451 g) of both sexes were used in the anti-asthmatic study.
Results: Results showed *Anchomanes difformis* had no lethal effect on Albino rats above 5000 mg/kg. Tested organs had mild to no toxic signs compared to control. Percentage protection from asthma of animals administered 400 mg/kg (32.7%) of extract was similar to the response of animals treated with Salbutamol (33.0%) which was the reference drug used in the study. DPPH radical scavenging activity of extract was comparable with ascorbic acid (standard) at 20 µg/ml. However, the extract had lower lipid peroxidation activity compared with the reference (α-tocopherol) at concentrations tested.
Conclusion: *A. difformis* aqueous leaf extract is safe for use as medicine and it possesses positive antioxidant and anti-asthmatic activities as claimed by traditional herbal practitioners in Udu and Ughievwen clans of Delta State, Nigeria

Keywords: *Anchomanes difformis*, Anti-asthmatic, Antioxidant, Ethnomedicine, Toxicity

PS2-B-049

Erythrina alkaloids as source of valuable chemicals

Marcos Soto-Hernandez[1], Rosario Garcia-Mateos[2], Ruben San Miguel-Chavez[1], Geoffrey Kite[3]

[1]Colegio De Postgraduados, Texcoco, Mexico, [2]Universidad Autonoma Chapingo, Texcoco, México, [3]Royal Botanic Gardens Kew, Richmond, United Kindom

Erythrina genus of the Fabaceae family possess one hundred and fifteen species with wide morphological variation and ecological diversity. Worldwide the major distribution of *Erythrina* species is located in Southern México and Central America. The genus has been studied from different point of view as the ornamental or culinary uses to studies of the chemical composition or their ethnomedial applications [1].
This interest due to the content of compounds as flavonoids, isoflavonoids, alkaloids, tripsine inhibitors, hemaglutinins or saponins. The bark, flowers and seeds have been studied intensely with this series of compounds, but are the alkaloids and isoflavonoids [2] who have received special attention by their vary structures or their biological applications. In this presentation it is described details of the recent studies done in Mexico with the alkaloids of several *Erythrina* species.

Keywords: *Erythrina*, alkaloids, isoflavonoids

References:
[1] Soto-Hernández *et at.* 2012
[2] Redko *et al*. 2007

PS2-B-050

Two types of lipid elicitors with different functions from the potato pathogen *Phytophthora infestans*

Makoto Ojika, Ryo Murata, Shiho Tenhiro, Mohammad S. Monjil, Kentaro Matsuda, Daigo Takemoto, Kazuhito Kawakita

Nagoya University, Nagoya, Japan

Elicitors are the chemicals that are derived from pathogens and trigger plant defense reactions. Major members are chitin, glucan, (glyco)proteins, and lipids. On the infection of pathogens, the host plants produce various defense reactions such as reactive oxygen species (ROS) and phytoalexin production, hypersensitive cell death, and the defense signals (e.g., salicylic acid, jasmonic acid) that induce systemic acquired resistance (SAR). The oomycete *Phytophthora infestans* is one of the best known plant pathogen that cause severe damages on important crops such as potatoes. Although several elicitors of *P. infestans* have been studied to develop plant defense inducers, no effective compounds have been identified. Since we have found a methanolic extract of *P. infestans* mycelia (MEM) significantly induced ROS and phytoalexins [1], we investigated the methanol-soluble elicitors of this pathogen.
The MEM was chromatographed on silica gel to give several active fractions. The least polar active fraction induced the production of several phytoalexins (e.g., rishitin) in potato tubers. HPLC purification yielded several diacyl glycerols. On the other hand, the second and third more polar fractions were found to induce an ROS, super oxide anion (O_2^-), in potato culture cells. By using reversed phase HPLC, we isolated 4 ceramides from the second active fraction and the 4 corresponding ceramide aminoethylphosphonates from the third one. The elicitor activity of MEM was found to be attributed to at least two types of lipids with different activities. SAR of these lipids and the mechanism of action will also be presented

Keywords: *Phytophthora*, plant pathogen, elicitor, reactive oxygen species, phytoalexin

References:

[1] Monjil M S, Nozawa T, Shibata Y, Takemoto D, Ojika M, Kawakita K. Compt Rend Biol 2015; 338: 185–196.

PS2-B-051

Propolis ethanolic extract induces apoptosis and cell cycle arrest on certain cancer cell lines in a p21 and cyclin D1 dependent manner

Başak Aru[1], Etil Güzelmeriç[2], Aslı Akgül[3], Gülderen Yanıkkaya Demirel[1], Hasan Kırmızıbekmez[2]

[1]Department of Immunology, Faculty of Medicine, Yeditepe University, TR-34755, Kayışdağı, İstanbul, Turkey, [2]Department of Pharmacognosy, Faculty of Pharmacy, Yeditepe University, TR-34755, Kayışdağı, İstanbul, Turkey, [3]Faculty of Pharmacy, Yeditepe University, TR-34755, Kayışdağı, İstanbul, Turkey

Propolis, a resinous bee product originated from plant buds and exudates, has a variable and complex chemical composition. Previous bioactivity studies on propolis extracts from different geographical and botanical origins revealed their antioxidant, anti-inflammatory, immunomodulatory and anticancer activities [1]. The aim of this study was to evaluate the ethanolic extract of propolis collected from Kastamonu, Turkey for its apoptotic and cell cycle arrest promoting effects on MCF7, HGC27, A549 cancer cell lines and a healthy cell line (HUVEC) in terms of DNA content, morphological features and expression of check point proteins p21, p53, Cyclin D1 and PDL-1. Furthermore, the main constituents of the extract were characterized. The extract showed moderate cytotoxic activity against all tested cell lines (IC50 A549: 58.6, HGC27: 90.3, MCF7: 90.7 HUVEC: 66.8 µg/mL) in MTS assay. Propolis treatment resulted in significant decrease in cell viability and increased apoptosis in all cell lines. Significant increase in necrosis rate was observed particularly in A549 and MCF7. Extract decreased G2/M phases significantly on MCF-7 and HGC27 cell lines, had no effect on A549 while increasing the S phase in HUVEC cells. Also, p21 and Cyclin D1 expressions were increased in all cancer cell lines but HUVEC. Furthermore, p53 was found significantly increased in both MCF7 and HGC27. 3-*O*-methylquercetin, chrysin, caffeic acid, CAPE, galangin and pinocembrin were identified as the main constituents of the extract by HPTLC. Moreover, the amounts of caffeic acid (5.5 mg/g) and CAPE (11.1 mg/g) were quantified by a validated HPLC method. Based on the results, chemically characterized propolis extract exerted apoptotic effect on cancer cell lines, promoted cell cycle arrest through activation of p21 and cyclin D1 while did not show any effect on HUVEC cells.

References:

[1] Sforcin JM. Phytother Res 2016; 30: 894–905.

PS2-B-052

Antidiabetic and antimicrobial properties of three Sri Lankan medicinal plants: *Phyllanthus emblica*, *Cassia auriculata* and *Hemidesmus indicus*

Dhanushki Wickramarachchi[1,2], Rizliya Visvanathan[1], Amriya Nizar[1,3], Majed Bawazeer[4], Bathini Tissera[4], M. Mallique Qader[1,4], Mostafa E. Rateb[4], Ruvini Liyanage[2]

[1]National Institute of Fundamental Studies, Kandy, Sri Lanka, [2]Postgraduate Institute of Science, University of Peradeniya, Peradeniya, Sri Lanka, [3]Faculty of Agriculture, University of Jaffna, Jaffna, Sri Lanka, [4]School of Science and Sport, The University of the West of Scotland, Paisley, United Kingdom

Sri Lanka is a tropical country rich in high floral diversity. Throughout decades, herbal plants play a vital role in the field of traditional Ayurveda medicine. Scientific information which addresses the ethnopharmacological significance of commonly consumed medicinal plants in Sri Lanka is insufficient. The present study is a continuation of determining the antidiabetic activity of ten selected medicinal plants in Sri Lanka. Among them, methanol extracts of *P. emblica* (Pe), *C. auriculata* (Ca) and *H. indicus* (Hi) demonstrated the highest α-amylase and α-glucosidase enzyme inhibitory activity. Their percentage inhibition exceeded 90% for both enzyme assays. IC_{50} values of Pe, Ca and Hi for α-amylase assay was between 3.14±0.30 to 90.91±0.18 µg/mL whereas, for α-glucosidase assay within 1.48±0.05 to 8.51±0.10 µg/mL. Further, hexane, dichloromethane, and ethyl acetate extracts of Pe, Ca and Hi were screened for antimicrobial activity. To identify the phytochemicals present in the extracts of these three plants, qualitative analysis and chemical profiling using GCMS were carried out. Alkaloids, flavonoids, tannins and terpenoids present in all three plants whereas saponins and steroids were found only in Ca and Hi. Hexane extract of Pe showed good inhibitory activity against pathogenic G +ve and G –ve bacteria and antidiabetic assays. The GCMS profile identified the presence of α-amyrin, β-amyrin, sitosterol and stigmasterol metabolites in hexane fraction of Pe which may be responsible for the observed high antimicrobial and antidiabetic activity. This provides the scientific basis for the antidiabetic and antimicrobial activity of these three Sri Lankan medicinal plants. Further LC-HRESIMS profiling of other fractions are underway.

Keywords: antimicrobial, enzyme inhibitory, medicinal plants

PS2-B-053

Stilbenes of *Yucca gloriosa* L. and their antioxidant, pro-apoptotic and anti-proliferative activities

Aleksandre Skhirtladze, Mariam Benidze, Ether Kemertelidze

TSMU Iovel Kutateladze Institute of Pharmacochemistry, Tbilisi, Georgia

From the bark of roots, rhizomes and stems of *Yucca gloriosa* L., cultivated on an industrial scale in eastern Georgia, a sum of phenolic compounds in the yield of 12% was obtained, in which 11 substances of stilbene nature were found. All substances were isolated and their structures were established using high-sensitivity one- and two-dimensional NMR and MS spectroscopy. They are polyphenolic compounds of stilbene series with a rare spirostructure. 6 of them were identified as *trans*-3,3',5, 5'-tetrahydroxy -4'-methoxystilbene and yuccaols A, B, C, D, E, previously isolated from *Yucca schidigera*, and the remaining 5 named gloriosaols A, B, C, D and E - new organic compounds with a more complex spirostructures. Their molecules consist of two C15 rings containing *γ*-lactone connected to each other with *trans*-3,3', 5, 5'-tetrahydroxy -4'-methoxystilbene. Gloriosaols A and B are diastereomers with two C2-C3 centers having a *trans* position; gloriosaol C has *cis*, and gloriosaols D and E have *trans*/*cis* position.

The sum of the phenolic compounds of *Yucca gloriosa* and individual gloriosaols are characterized by high antioxidant activities. Their TEAC is 3.00-5.78, and 2-3 times more that of the comparative drugs - quercetin and *β*-carotene.

The anti-proliferative and pro-apoptotic activities of gloriosaols in relation to tumor cells MCF 7 (thoracic carcinoma), HepG 2 (hepatoblastoma) and U 937 (monocytic leukemia) have been established. Gloriosaol C differs particularly efficiently.

PS2-B-054

Therapeutic effect of total phenolic fraction of extra virgin olive oil against murine experimental cutaneous leishmaniasis.

Kalliopi Karampetsou[1,2], Olga Koutsoni[1], Nektarios Aligiannis[2], Maria Halabalaki[2], Leandros Alexios Skaltsounis[2], Eleni Dotsika[1]

[1]Laboratory of Cellular Immunology, Department of Microbiology, Hellenic Pasteur Institute, Athens, Greece, [2]Division of Pharmacognosy and Natural Product Chemistry, Department of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece

Leishmaniasis, a disease caused by parasites of genus *Leishmania* spp., consists a serious health problem. There is no effective vaccine and chemotherapeutic treatments have drawbacks such as toxicity and parasite resistance [1]. Novel treatment prospects are important to confront the disease. *Olea europea* possesses outstanding position in the Mediterranean area and many biological actions of its derivatives are established [2]. Phenols from Extra Virgin Olive Oil (EVOO) present plethora of bioactivities including antimicrobial, anti-inflammatory and antioxidant. Here, we evaluated the *in vivo* therapeutic effect of Total Phenolic Fraction (TPF) from EVOO in murine experimental cutaneous leishmaniasis. BALB/c mice, subcutaneously infected with *L. major* promastigotes, were intraperitoneally treated with TPF (20 mg/kg body weight) every other day for 28 days. Footpad swelling was monitored for ten weeks post-infection. After treatment termination, we determined; the parasite burden in draining lymph node, the ratio of IgG2a/IgG1 isotypes of Leishmania-specific antibodies and the Th1/Th2 profile of the CD4+ splenocytes. Results demonstrated significant reduction of footpad swelling and parasite burden in TPF-treated mice, compared to untreated. Our data showed an increase of the IgG2α/IgG1 ratio and a significant increase in CD4+ T cells producing IFN-γ with a simultaneous moderate decrease in CD4+ T cells producing IL-4 in TPF-treated mice compared to untreated group. These data indicate immune response polarization towards the Th1-type of response that is required for the effective control of leishmaniasis. Overall, TPF acquires credits of being a source of compounds with antileishmanial and immunomodulatory properties.

Acknowledgements: Thanks are expressed to Hellenic Foundation for Research and Innovation (HFRI) for PhD candidate scholarship.

Keywords: Leishmaniasis, *Olea europea*, immune response

References:

[1] Pace D. J Infect 2014; 69 Suppl 1: S10-8.
[2] Gorzynik-Debicka M *et al*. Int J Mol Sci 2018; 19(3).

PS2-B-055

Dynamic of phenolic acids production in *Aronia* × *prunifolia* (Marshall) Rehder agitated shoot cultures during the growth cycles

Agnieszka Szopa, Paweł Kubica, Joanna Żywko, Halina Ekiert

Chair and Department of Pharmaceutical Botany, Jagiellonian University-Collegium Medicum, Medyczna 9 Str., 30-688 Kraków, Poland

The one subgroup of polyphenols of plant origin, with high antioxidant activity are phenolic acids, the derivatives of cinnamic acid, benzoic acid and also depsides.
Aronia × *prunifolia* is a hybrid of *A. melanocarpa* and *A. arbutifolia* with North American origin, cultivated in Middle Europa and Russia. Native plant and its different types of *in vitro* cultures could be a rich source of phenolic acids, especially depsides with high antioxidant activity.
The aim of the present study was the establishment of agitated shoot cultures and monitoring of dynamic of phenolic acids accumulation during 8-weeks growth cycles.
The shoot cultures (4 series) were maintained on Murashige and Skoog medium (MS) enriched with 1 mg/l BAP (cytokinin) and 1 mg/l NAA (auxin). In the methanolic extracts of biomasses and culture media samples collected at 1-weeks intervals the LC-DAD analysis of 26 compounds was performed.
In the media samples no phenolic acid was detected. In all biomass extracts the presence of the same 11 phenolic acids was confirmed. The quantitatively dominant compounds were: 3,4-dihydroxyphenylacetic acid, 3-phenylacetic acid and two depsides chlorogenic acid and isochlorogenic acid (max. 215.24, 272,72, 157.09 and 214.73 mg/100 g D.W., respectively). After 2 weeks of growth cycles the total content of phenolic acids increased gradually from 723.80 to max. content 1139.12 mg/100 g D.W. (after 4 weeks) and later gradually decreased to 96.29 mg/100 g D.W. (after 8-weeks).
The obtained results are interesting from practical point of view.

PS2-B-056

Oleocanthal exerts *in vivo* antileishmanial and immunomodulatory effect against murine experimental cutaneous leishmaniasis

Kalliopi Karampetsou[1,2], Olga Koutsoni[1], Apostolis Angelis[2], Nektarios Aligiannis[2], Maria Halabalaki[2], Leandros Alexios Skaltsounis[2], Eleni Dotsika[1]

[1]Laboratory of Cellular Immunology, Department of Microbiology, Hellenic Pasteur Institute, Athens, Greece, [2]Division of Pharmacognosy and Natural Product Chemistry, Department of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece

In recent years, natural products from *Olea europea*, especially phenols from Extra Virgin Olive Oil (EVOO), have gained lot of attention about their health beneficial effects. Oleocanthal (OLEO), a phenol that is present in EVOO, has been mainly reported as an anti-inflammatory agent, also possessing anticancer and antioxidant properties [1]. We have previously reported that Total Phenolic Fraction from EVOO, that contains OLEO, has shown antileishmanial properties [2]. In the present study, we evaluated the in vivo antileishmanial effect of OLEO treatment (intraperitoneal administration of 5 and 10 mg/kg body weight 3 days per week) in BALB/c mice with established cutaneous leishmaniasis infection. The progression of the disease was monitored for 10 weeks by measuring footpad swelling. After treatment termination, we determined parasite burden in draining lymph node and the percentage of CD4+ T cells producing IL-12, IFN-γ or IL-4 cytokines in splenocytes. Results demonstrated significant parasite elimination in OLEO-treated mice compared to untreated group, as proved by footpad swelling and parasitic load reduction. Moreover, OLEO treatment exerted significant reduction of CD4+ T cells producing IL-4 while a moderate increase of IL-12-producing CD4+ T cells was determined. These data suggest that the antileishmanial effect induced by OLEO treatment is attributed to its immunomodulatory properties to drive the immune system polarization towards a protective Th1-type of response.

Acknowledgements: Thanks are expressed to Hellenic Foundation for Research and Innovation (HFRI) for PhD candidate scholarship.

Keywords: Leishmaniasis, *Olea europea*, oleocanthal, immune response

References:

[1] Parkinson L, R Keast. Int J Mol Sci 2014; 15(7): 12323–34.
[2] Koutsoni O S *et al*. Phytomedicine 2018 (in press).

PS2-B-057

Sensory perception and bitterness masking of olive polyphenols in fortified mayonnaise

Petroula Tsitlakidou, Giorgos Nasioudis, Ioannis Mourtzinos, Costas G. Biliaderis

Dep. Of Food Science & Technology, School of Agriculture, Faculty of Agriculture, Forestry & Natural

Despite the recently approved EFSA health claim for olive polyphenols, the adverse sensorial effects of these compounds limit their food applications. Preventing interactions between bitter molecules and the humans' taste receptors can be achieved through inclusion complex formation with *β*-cyclodextrin (*β*-CD).
This study aimed at: (a) investigation of inclusion complex formation of *β*-CD with an olive fruit extract powder (OLFE); (b) examination of bitter taste masking of a OLFE-enriched mayonnaise.
Inclusion complex formation (1:1 stoichiometry) was confirmed by nuclear magnetic resonance (^{1}H NMR). The bitterness threshold of OLFE-phenols in aqueous solutions was studied by a trained sensory panel (n=10) and the use of 3-AFC sensory test; i.e. 5 mg OLFE/ 100g H_2O ($p < 0.05$). Then, masking of OLFE-phenols bitter taste was then examined. The panel were asked to compare the bitterness of OLFE added in concentrations ranging from 1 to 50 mg to the samples containing the same concentrations of OLFE being complexed with *β*-CD. A bitterness masking effect ($p < 0.05$) was detected at concentrations ≥ 10 mg OLFE.
Model mayonnaise samples tested were made with: 50% sunflower oil (s.o., reference), 25% s.o. and 25% olive oil, 50% s.o. enriched with OLFE to meet the daily recommended olive phenols uptake/serving, and 50% s.o. enriched with the *β*-CD / OLFE complex. Sensory evaluation was conducted using quantitative descriptive analysis (QDA) and eight trained assessors; fifteen attributes were evaluated in two consecutive sessions. Two-way analysis of variance (ANOVA) revealed significant differences between sample pairs ($p < 0.05$); ratings of 'bitter taste' and 'aftertaste', 'olive tapenade aftertaste' decreased in samples containing the complex of *β*-CD/ OLFE, compared to samples containing free OLFE. Overall, the aforementioned attributes seemed to be directly related to the addition of olive phenols and thus, formation of inclusion complexes with *β*-CD can effectively reduce bitterness.

PS2-B-058

Dynamic of flavonoids production in agitated shoot cultures of three *Hypericum perforatum* L. cultivars 'Elixir', 'Helos' and 'Topas' during the growth cycles - preliminary results

Inga Kwiecień[1], Natalia Więckowska[1], Ludger Beerhues[2], Halina Ekiert[1]

[1]Chair and Department of Pharmaceutical Botany, Jagiellonian University-Collegium Medicum, Medyczna 9 Str., 30-688 Kraków, Poland, [2]Institute of Pharmaceutical Biology, Technische Universität Braunschweig, Mendelssohnstrasse 1, 38106 Braunschweig, Germany

Hypericum perforatum L., famous medicinal plant species, contains among other bioactive compounds, flavonoid aglycones and also glycosides, compounds of polyphenol structure, with high antioxidant activity. Our earlier studies documented, that *in vitro* shoot cultures of three *H. perforatum* cvs. ('Elixir', 'Helos' and 'Topas') cultured in our laboratory could be a rich source of bioactive compounds, e.g. indole compounds, phenolic acids and flavonoids.
The aim of present study was establishment of agitated shoot culture of these three cultivars and evaluation of flavonoids accumulation after 4- and 5-weeks growth cycles.
The shoot cultures of three cultivars were maintained on Murashige and Skoog (MS) medium supplemented with following concentrations of cytokinin – BAP and auxin – NAA [mg/l]: 0.1/0.1, 1/1, 2/2, and 3/3 (3 series). In methanolic extracts of shoots and media samples collected after 4 and 5 weeks of the growth cycles the quantitative analysis of 32 flavonoids (16 aglycones and 16 glycosides) using LC-DAD method was performed.
In media extracts any flavonoids were estimated. In all shoot extracts the presence of the same six compounds were detected, three aglycones – kaemferol, luteolin and quercetin and three glycosids – hyperoside, quercitrin and rutoside. The main compound in all extracts was quercetin (max. 1282.36; 1335.61, 1898.44 mg/100 g D.W. for 'Elixir', 'Helos' and 'Topas', respectively). Higher total content of estimated metabolites was confirmed for 'Elixir' and 'Helos' cvs. after 4-weeks growth cycles (1359.53 and 1401.09 mg/100 g D.W.), for Topas cv. after 5-weeks growth period (1968.93 mg/100 g D.W.). The best productive medium for all cultivars was MS variant containing the lowest (0.1 mg/l) tested amounts of BAP and NAA. The estimated in all cultivars quantities of quercetin were extremely high.
Shoot cultures of Topas cv. was the most productive culture. The results documented differences in biosynthetic potential of three *Hypericum perforatum* cultivars.

PS2-B-059

Discovery and characterization of new natural products from promising Cyanobacteria

Sara Freitas, Teresa P Martins, Pedro N Leão

CIIMAR-Interdisciplinary center of Marine and Environmental Research, 4450-208, Matosinhos, Portugal

The pharmacological and biotechnological industries have been focusing on the application and research of natural products isolated from a variety of organisms. One phylum of organisms that increasingly mark their path on natural product research is cyanobacteria, a group of photosynthetic bacteria spread widely throughout Earth. The adaptation to all source of environments and adverse conditions in this phylum makes the secondary metabolite production/isolation a promising area of research. Many natural products isolated from cyanobacteria are used in a diversity of biological applications and the focus is now directed to their assembly and leading mechanisms responsible for the production.
In this work we focused on the isolation of natural products from a selected group of cyanobacteria with phylogenetic and/or MS potential data for the discovery of new compounds. These cyanobacterial strains were isolated from the Atlantic coast and are present in the BBE culture collection (LEGE cc) [1] from CIIMAR. Information regarding chemodiversity, biosynthetic pathway, enzymology and biotechnological application are also being explored for the new discovered natural products.

References:

[1] Martins J, Regueiras A, Pereira AL, Lopes VR, Frazão B, Gomes D, Moreira C, Costa MS, Brûle S, Faustino S, Martins R, Saker M, Osswald J, Leão PN, Vasconcelos V. J Appl Phycol 2018; 30(3):1437–1451.

PS2-B-060

Evaluation of cytotoxic potential of *Jurinea macrocephala* DC. on A-549 and MCF-7 cell lines

Perihan Gürbüz, Şengül Dilem Doğan, Ebru Öztürk, Mükerrem Betül Yerer Aycan

Erciyes University, Faculty of Pharmacy, Talas, Turkey

The genus *Jurinea* (Asteraceae) with about 250 species is distributed in Asia and Europe. Phytochemical studies on *Jurinea* species led to the isolation of Sesquiterpene lactones (SLS) [1-3]. There are plenty of investigations about the cytotoxic effects of SLS on different cancer cell lines therefore the screening of SLS type terpenoids rich plants remains still importance. The aim of this study is to evaluate the antiproliferative effect of J. macrocephala (JM) extracts, sub extracts and fractions on breast cancer and non-small cell lung cancer cell lines.

Methanol, chloroform and *n*-buthanol extracts were prepared from aerial parts of *J. macrocephala*. To investigate the cytotoxic potential of these extracts real time xCELLigence system was used. A549 and MCF-7 cells were seeded in E-plate then approximately 24 h post-seeding, the cells were treated with the samples. Cell viability was observed during 48 h after treatment and IC_{50} values have been calculated. According to the preliminary screening results, chloroform extract showed significant cytotoxic effects on both two cell lines. After fractionation of this chloroform extract with column chromatography, ten fractions were obtained (JM1-10). These ten fractions were tested again for dose dependent cytotoxic activity; JM7, JM8 and JM10 were selected for further purification with IC_{50} values under 40 µg/mL on both two cell lines.

Acknowledgements: The authors are indebted to Research Foundation of Erciyes University (FCD-2018-7834) for their financial support.

Keywords: Asteraceae, *Jurinea*, MCF-7, A549

References:

[1] Rustaiyan A, M Ganji. Phytochemistry 1988; 27(9): 2991–2992.
[2] Taherkhani M, A Rustaiyan. Nat prod res 2016; 30(23): 2743–2746.
[3] Rustaiyan A *et al*. Phytochemistry 1991; 30(6): 1929–1932.

PS2-B-061

Chemical composition of a nephroprotective ethanolic extract of *Pistacia lentiscus* L. fruits

Nassima Cheraft-Bahloul[1], Cécile Husson[2], Meriam Ourtioualous[2], Sébastien Sinaeve[3], Cédric Delporte[3,4], Djebbar Atmani[1], Caroline Stévigny[3], Joëlle L. Nortier[2], Marie-Hélène Antoine[2]

[1]Laboratoire de Biochimie Appliquée, Faculté des Sciences de la Nature et de la Vie, Université de Bejaia, Bejaia, Algeria, [2]Laboratory of Experimental Nephrology, Faculty of Medicine, Université Libre de Bruxelles (ULB), Brussels, Belgium, [3]RD3-Pharmacognosy, Bioanalysis and Drug Discovery Unit, Université libre de Bruxelles (ULB), Brussels, Belgium, [4]Analytical platform, Faculty of Pharmacy, Université Libre de Bruxelles (ULB), Brussels, Belgium

The world prevalence of kidney stones is increasing and plants are frequently used to treat urolithiasis. *Pistacia lentiscus* L. is widely used in Algeria, as a decoction of aerial parts, in folk medicine for various pathologies. Protective effects of the *Pistacia lentiscus* ethanolic fruit extract (PLEF) has been previously reported in vitro regarding the tubulotoxicity of calcium oxalate monohydrate (COM) crystals with Human Kidney [HK]-2 cells [1].
The aim of the present study was to identify the main secondary metabolites present in the PLEF. For this, a phytochemical screening was performed and a liquid chromatography high resolution mass spectrometry (LC-HRMS) analyses.
The phytochemical screening was performed for alkaloids, anthraquinones, flavonoids, saponins, tannins, and anthocyanins. The LC-MS analysis was made on the PLEF without prior purification. Analysis were performed using a rapid resolution LC (RRLC) system coupled to a electrospray ionization source (ESI)-quadrupole time-of-flight (QTOF).
The results obtained with the phytochemical screening showed the presence of anthocyanins, tannins and flavonoids. Our preliminary results of the LC-HRMS analyses confirmed the compounds identified in the literature such as gallic acid, catechin, quercetin and luteolin [2]. The potential interest comes from the anthocyanins and tannins in urolithiasis. More studies will be held to fully characterize the PLEF, to quantify the identified molecules and to evaluate their biological effect.
In conclusion, considering the protective effect of PLEF against the tubulotoxicity of COM crystals, the identification of the key metabolites, with a specific attention on tannins and anthocyanins, responsible for this beneficial effect is ongoing.

Keywords: *P. lentiscus* L., phytochemical screening, COM crystals, HK2 cells, HPLC-MS.

References:

[1] Cheraft-Bahloul N *et al*. J. Ethnopharmacol 2017; 209: 248–254.
[2] Mehenni C *et al*. J Food Drug Anal 2016; 24: 653–669.

PS2-B-062

Phytochemical analyses of three endemic Boraginaceae plants from Turkey: *Phylocarra aucheri*, *Symphytum anatolicum*, *Cynoglottis barrelieri*. Biological activities

Elisavet Varvouni[1], Konstantia Graikou[1], Gokhan Zengin[2], Christos Ganos[1], Tomasz Mroczek[3], Ioanna Chinou[1]

[1]Sect Pharmacognosy & Chemistry of Natural Products, Department of Pharmacy, National & Kapodistrian University of Athens, Athens, Greece, [2]Department of Biology, Science Faculty, Selcuk University, Turkey, [3]Department of Pharmacognosy with Medicinal Plant Laboratory Unit, Medical University, Poland

In the framework of our chemical analyses on Boraginaceae plants [1,2,3], we report in this study the phytochemical investigation on three endemic plants from Turkey: *Phylocarra aucheri, Symphytum anatolicum* and *Cynoglottis barrelieri*, which to our best of knowledge have never been studied before.

The methanolic extracts were analyzed by LC-MS, where they have been identified eighteen secondary metabolites. Moreover, several of them have been also isolated and structurally determined by modern spectral means.

The flavonoids rutin, isoquercitrin, and quercetin-3-acetyl-glucoside together with caffeic acid derivatives (rosmarinic and salvianolic acids) appeared among the most abundant constituents and can be served as chemotaxonomic markers among Boraginaceae plants.

All extracts were screened for their antioxidant and *in vitro* enzyme inhibitory properties. Their antioxidant capacity was evaluated using free radical scavenging (DPPH, ABTS), reducing power (FRAP, CUPRAC), phosphomolybde- num and ferrous ion chelating assays, while they also evaluated against cholinesterases, α-amylase and α-glucosidase enzymes.

C. barrelieri exhibited the strongest antioxidant activity due probably to its high total phenolic and flavonoid content and it is followed by *S. anatolicum*, *P. aucheri*, while it is noteworthy that *S. anatolicum* showed the highest inhibitory capacity against AChe and BChe.

The phytochemical profiles as well as and bioactivities results suggest that all three herbal materials could be used for further chemical and pharmacological studies as a potential source for phytotherapeutic applications.

Acknowledgements: The author would like to thank the Special Account for Research Grants and the National and Kapodistrian University of Athens for funding their participation in this meeting

References:

[1] Tufa T, Damianakos H, Graikou K, Chinou I. Nat Prod Com 2017; 12:179–180.
[2] Damianakos H, Jeziorek M, Sykłowska Baranek K, Pietrosiuk A, Buchwald W, Chinou I. Phytochem Letts 2016; 15: 234–237.
[3] Tufa T, Damianakos H, Zengin G, Graikou K, Chinou I. S Afr J Bot 2018.

PS2-B-063

Bioassay-guided isolation, identification and cytotoxicity of diterpenoids from *Justicia insularis*

Idowu Eniafe Fadayomi[1,2], Okiemute Rosa Johnson-Ajinwo[1], Alexander Kagansky[3], Ted Hupp[4], Nick Forsyth[1], Wen-Wu Li[1]

[1]Institute for Science and Technology in Medicine, Keele University, Stoke-on-trent, United Kingdom, [2]Department of Biology, Adeyemi College of Education, Ondo, Nigeria, [3]Far Eastern Federal University, Vladivostok, Russia, [4]Institute of Genetics and Molecular Medicine, University of Edinburgh, Edinburgh, United Kingdom

Ovarian cancer stands as the most severe amidst the gynaecological malignancy associated with the highest level of death. The resistance of ovarian cancer cells against some of the available anti-cancer drugs in recent time has exacerbated the problem. Novel therapies are still needed. This study evaluated the anti-ovarian cancer activities of *Justicia insularis* T. Anders (family Acanthaceae), which is a medicinal plant widely used in the treatment of different diseases across Africa. The cytotoxicity of the plant extract was evaluated in human ovarian cancer cell lines (OVCAR-4 and OVCAR-8) and normal human ovarian surface epithelia (OSE) cells using Sulforhodamine B assay. Isolation of the bioactive compounds was carried out through bioassay-guided fractionation. Further purification was carried out using HPLC and the isolated natural products were characterized using GC-MS, LC-MS and NMR techniques. The roles of the identified bioactive compounds in apoptosis were evaluated using Caspase 3/7, Caspase 8, Caspase 9 and Annexin V assays. The extract inhibited ovarian cancer cell growth with IC_{50} values (10.7±0.6 and 19.2±0.5 µg/ml) against OVCAR-8 and OVCAR-4 cell lines respectively. The two bioactive compounds were identified as diterpenoids: 16*α*-hydroxy-cleroda-3,13(14)*Z*-dien-15,16-olide (1) and 16-oxocleroda-3,13(14)*E*-dien-15-oic acid (2). 1 showed IC_{50} values of 4.4±0.2 and 5.7±0.3 µM, while 2 was less potent with IC_{50} of 11.8±0.5 and 16.6±2.8 µM against OVCAR-8 and OVCAR-4 cells respectively. Compound 1 and 2 were less cytotoxic to OSE cell line (IC_{50} 12.1±0.1 and 22.8±0.7µM) respectively. Both compounds induced apoptosis in OVCAR-8 and OVCAR-4 cells by increasing caspase 3/7 activities at a concentration dependent manner. 1 further increased caspase 9 activities in OVCAR-4 and OVCAR-8 cells, and increased caspase 8 in OVCAR-8 cells. 1 slightly increased early and late apoptosis in OVCAR-8 cells when compared with control. Compound 1 was identified as the most active compound with anti-ovarian cancer activities in *Justicia insularis*.

PS2-B-064

Ethnomedicinal survey of plants used in the treatment of malaria in Southern Nigeria

Macdonald Idu[1], Pass Iyamah[2]

[1]Department of Plant Biology and Biotechnology, University of Benin, Benin City, Benin City, Nigeria, [2]Department of Biology, College of Education, PMB 1251 Warri, Delta State, Nigeria, Warri, Nigeria

Malaria is one of the most severe public health problems worldwide. It is a leading cause of deaths in many developing countries, where young children and pregnant women are the groups most affected. Spread of multidrug-resistant strains of *Plasmodium* and the adverse side effects of the existing anti-malarial drugs have necessitated the search for novel, well tolerated and more efficient antimalarial drugs. This ethnomedicinal study surveyed the different types of medicinal plants used for the treatment of malaria in Southern Nigeria with the intent of identifying plants traditionally used in the treatment of malaria across geopolitical boundaries. Data were collected from 79 respondents composed of 50 traditional herb-sellers and 29 herbal practitioners using a semi-structured questionnaire. Data was analyzed using frequency and percentages. Of the 79 respondents interviewed, 24% were males while 76% were females. A total of 156 species belonging to 60 families were reported being used to treat malaria in the study area. Fabaceae was the most represented family having fourteen (14) plant species. Of the plants identified during the survey, *Azadirachta indica* was the species with the highest relative frequency of citation (RFC – 1.0). The dominant plant parts used in the preparation of remedies were leaves (50.50%) and Decoction was the main method of preparation. Analysis of regional plant occurrence revealed that South-Western Nigeria represented the region with the highest plant occurrence (60.7%) followed by South–South (24%) and South–East (15.3%). Regional occurrence of plants used in the treatment of malaria in Southern Nigeria is reported here for the first time. This study has documented a great diversity of plants used in the treatment of malaria in Southern Nigeria. Further studies may unveil future promising phytomedicines in the treatment of malaria. Conservation of these plant species is recommended to ensure their continuous availability for future use.

PS2-B-065

Synthesis and evaluation of novel hybrid antioxidant peptides

Athina Lykoura[1], Nicky Panousi[1], Virginia Dimaki[1], Nikolaos Assimomytis[2], Fotini Lamari[1], Vassiliki Magafa[1]

University of Patras, Patras, Greece, [2]University of West Attica, Athens, Greece

Dairy proteins, sea food, cereals are great sources of bioactive peptides. They can be obtained through enzymatic hydrolysis and microbial fermentation and present various qualities, i.e. antimicrobial, antioxidant, antiflammatory, antihypertensive etc.
Antioxidant peptides contribute to human health by counteracting Reactive Oxygen Species (ROS). ROS are generated in human body through normal metabolic processes and participate in important cell functions. However, when their production becomes uncontrollable and the balance is lost, oxidative stress appears. Oxidative stress is considered a major cause of many diseases such as cancer, cardiovascular diseases, diabetes etc.
In the present study, a peptide derived from rice bran protein [1], with known antioxidant activity, was synthetically produced and examined. The aim of the study was the improvement of its antioxidant properties by introducing modifications in the N-terminus, which include replacements of amino acids by natural products (cinnamic acid, caffeic acid, carnosine) or addition of those to the natural peptide. Peptide synthesis was performed with Fmoc/But solid phase methodology [2]. RP-HPLC was used to confirm their purification. Identification was performed by ESI-MS and the antioxidant activity was evaluated with FRAP and DPPH assays. FRAP and DPPH methods are correlated up to a point. Hybrid peptides with aromatic substances in the N-terminus show strong antioxidant activity in both methods. The addition of caffeic acid, improves further the antioxidant properties of the analogues. On the other hand, the addition of carnosine shows higher antioxidant activity in the FRAP method, while in DPPH shows reduced action, compared to the natural peptide.

References:

[1] L Wattanasiritham, Ch Theerakulkait, S Wickramasekara, C S Maier, J F Stevens. Food Chem 2016; 192: 156–162.
[2] B G Fields, L R Noble. Int J Pept Protein Res 1990; 35: 161–214.

PS2-B-066

Assessment of tyrosinase inhibitory effects and antioxidant properties of *Glaucium corniculatum* (L.) Rud. subsp. *refractum* (Nab.) Cullen and *Glaucium Ieiocarpum* Boiss. growing in Turkey

Fatma Ayaz[1], Yavuz Bağcı[2], Nuraniye Eruygur[1], Esra Maltaş[3], Hamide Filiz Ayyıldız[4], Cengizhan Ceylan[5]

[1]Department of Pharmacognosy, Faculty of Pharmacy, Selcuk University, Konya, Turkey, [2]Department of Pharmaceutical Botany, Faculty of Pharmacy, Selcuk University, Konya, Turkey, [3]Department of Biochemistry, Faculty of Pharmacy, Selcuk University, Turkey, [4]Department of Analytical Chemistry, Faculty of Pharmacy, Konya, Turkey, [5]Department of Clinical Pharmacy, Faculty of Pharmacy, Selcuk University, Konya, Turkey

The genus *Glaucium* Mill. (Papaveraceae), as known "boynuzlu gelincik" (horned poppy), is widely distributed in temperate and subtropical regions of the Northern hemisphere. The plants is traditionally used as hypnotic, sedative and in the treatment of cough in Turkey and has medicinal uses such as antibiotic, cardioprotective, antidiabetic and anti-cancer [1]. In the present study, we aimed to assess *in vitro* antioxidant activity and determine the content of total phenolic compounds (TPC) and flavonoids (TF) as well as investigate on tyrosinase inhibition of aerial parts of *Glaucium corniculatum* (L.) Rud. subsp. *refractum* (Nab.) Cullen and *Glaucium Ieiocarpum* Boiss. The ethanol (70%) and water extracts of the plants were proceeded for the antioxidant activities using DPPH and ABTS methods, and their content of TPC and TF were determined spectrophotometrically. According to our results, the highest content of total phenolic compounds and flavonoids in the ethanol (70%) extract of *G. leiocarpum* with 31.47 ± 3.24 mg gallic acid equivalent per gram of extract and 45.31 ± 4.00 mg quercetin equivalent per gram of extract, respectively. Among the samples, the highest DPPH and ABTS free radical scavenging activities were displayed in the ethanol (70%) extract of *G. leiocarpum* with IC_{50} values of 1261.19 ± 1.68 µg/mL and 80.17 ± 5.95 µg/mL, respectively. In addition, the ethanol (70%) extract of *G. corniculatum* subsp. *refractum* showed the highest tyrosinase inhibition with IC_{50} value 425.20 ± 6.42 µg/mL. As a conclusion, further studies on these plants need to be to identify their chemical constituents responsible for these biological activities.

Keywords: Tyrosinase inhibition, antioxidant activity, *Glaucium corniculatum* subsp. *refractum*, *Glaucium leiocarpum*

References:

[1] Baytop T. Therapy with Medicinal Plants in Turkey (Past and Present). Publication no 3255. Istanbul University. Istanbul 1984: 189.

PS2-B-067

In vitro biological effects of hydroxytyrosol on human hepatoma HepG2 cells.

Georgia Gogou[1,2], Andriana Kavallari[2], Maria Lypiridou[2], Haralambos Stamatis[1], Constantinos Pantos[2]

[1]Laboratory of Biotechnology, Department of Biological Applications and Technologies, University of Ioannina, Ioannina, Greece, [2]Laboratory of Pharmacology, School of Medicine, National and Kapodistrian University of Athens, Athens, Greece

Over recent years, several studies have related olive oil ingestion to a low incidence of several diseases including cancer. One of the major polyphenols present in virgin olive oil, hydroxytyrosol (HT) shows a variety of pharmacological activities such as antioxidant properties, anticancer and anti-inflammatory activities and beneficial effects on the cardiovascular system [1]. This study aimed to investigate HT effects on HepG2 cell proliferation and migration capacity [2]. We tested different concentrations of HT (80μM, 100μM, 200μM). Cell viability was estimated with Trypan blue exclusive test and cell migration was evaluated with two different methods, Wound Healing Assay (WHA) and Ring Cell Migration Assay (RCMA). For WHA, HepG2 cells were seeded in well-plates and cell monolayer was scraped to create a "scratch". Then, concentrations of HT were added. Pictures were taken every 12 hours, until 72 hours, to evaluate the extent of cell migration. Similarly, for RCMA we used cloning rings to create gaps in cell monolayer and we evaluated the results as in WHA. Our study showed that 200μM of HT induced cell death from the first 12 hours of treatment while lower concentrations induced cell death after 24 hours. Moreover, HT in concentrations of 80μM and 100μM seemed to inhibit cell migration as determined by WHA and RCMA. Overall, our results show that HT on 200μM has cytotoxic effects and on 80μM and 100μM inhibits cell migration.

Keywords: Hydroxytyrosol, HepG2 cell line, cell migration assays

References:

[1] Hu T *et al*. J Agric Food Chem 2014; 62(7): 1449–55.
[2] Tutino V *et al*. Anticancer Re, 2012; 32(12): 5371-7.

PS2-B-068

Anticancer effects of extracts from aerial part of *Glinus oppositifolious* (L.). Aug. DC.

Alexandru Vasincu[1], Veronica Bild[1], Christiana M Neophytou[2], Christiana Charalambous[3], Ioana-Mirela Vasincu[4], Bogdan-Gabriel Şlencu[5], Andreas Ioannou Constantinou[2], Anca Miron[6]

[1]Discipline of Pharmacodynamics and Clinical Pharmacy, University of Medicine and Pharmacy "Grigore T. Popa" Iasi, Romania, [2]Department of Biological Sciences,University Of Cyprus, Nicosia, Cyprus, [3]University of Nicosia Medical School, Nicosia, Cyprus, [4]Discipline of Pharmaceutical Chemistry, University of Medicine and Pharmacy "Grigore T. Popa" Iasi, Romania, [5]Discipline of Environmental and Food Chemistry, University of Medicine and Pharmacy "Grigore T. Popa" Iasi, Romania, [6]Discipline of Pharmacognosy, University of Medicine and Pharmacy "Grigore T. Popa" Iasi, Romania

Glinus oppositifolius (L.) Aug. DC. is a plant traditionally used in Mali in the treatment of skin disease, intestinal pain, diarrhea, joint pain, inflammation, fever, open wounds, malaria, urinary infections and liver dysfunction. These diseases are related to the immune response. Our objective is to determine if extracts obtained from the aerial part of this plant present any anti-cancer properties.

Material and methods: We prepared two extracts from the aerial part of the plant using ethanol and water as solvents. The anti-cancer potential of both extracts was initially evaluated using the MTT method to determine the viability of mammary MCF-7, alveolar A549, prostate PC-3 and colorectal CACO2 adenocarcinoma cell lines. The cell cycle distribution was analyzed by flow cytometry. Intracellular production of reactive oxygen species (ROS) was measured using the DCFH-DA assay.

Results: Our results showed that the aqueous extract significantly inhibited the viability of CACO2 cells (IC_{50} = 97.69 ± 2.03 µg/mL). The cell cycle analysis results demonstrated that the aqueous extract remarkably caused an accumulation of cell population in the sub-G1 (apoptotic) phase from 8.35 ± 3.85% in the vehicle control group to 55.7 ± 4.03% in the aqueous extract treated group. The aqueous extract also reduced CACO2 cell viability and induced an increase of intracellular H_2O_2 production that varied in a time-dependent manner.

Conclusions: Anti-tumor screening of these extracts obtained from *Glinus oppositifolius* revealed that the aqueous extract presented an inhibitory effect on the viability of CACO2 adenocarcinoma cells. The cellular mechanisms involved in this inhibitory activity will be investigated in the future.

Keywords: *Glinus oppositifolius*, MTT, cell cycle.

PS2-B-069

Acaricidal activity of constituents from *Gleditsia japonica* against *Dermanyssus gallinae*

Jin ah Kim, Yun-Hyeok Choi, Ji Eun Lee, Yeon Woo Jeong, Changon Seo, Jin Gwan Kwon, Jin Kyu Kim, Seong Su Hong, Wonsik Jeong, Chun Whan Choi

Gyeonggi Bio-center, Suwon-si, Yongtong-gu, South Korea

The aim of this study was to discover for acaricidal activity of constituents from natural source against *Dermanyssus gallinae*. The acaricidal activities of thirty-five ethanol extracts of native to Korea against *D. gallinae* were examined using the direct contact application method. Based on laboratory tests, an acaricidal activity extracts of *Gleditsia japonica* (Leguminosae) fruit was determined because of its potent activity. *G. japonica* grows widely in Korea, China and other countries, However, there are few studies on chemical constituents and biological activities of the *G. japonica*. This fruit (Gleditsiae Fructus) has long been used in traditional Chinese medicine (TCM) and as a nourishing food in Korea and China for the treatment of inflammatory, and fungal diseases. We investigated acaricidal activity using ethanol extracts and its fractions from G. Fructus to evaluate the usefulness of its extract as a functional biomaterial. Among the tested fractions, *n*-Hexane and EtOAc fraction demonstrated a significant inhibition of the acaricidal activity. Results were compared with those of the currently used animal drug such as Wagubang. The *n*-Hexane and EtOAc fraction of the EtOH extract of G. Fructus was subjected to successive column chromatography, six compounds were isolated. The compounds were identified by their spectral data such as MS, 1D and 2D NMR These results indicate that the G. Fructus-derived materials descried as poultry red mites-control agents could be useful for managing field populations of *D. gallinae*.

PS2-B-070

Biomonitored chemical study of extracts from *Pouteria ramiflora* for *in vitro* cytotoxic activity in human prostate cancer cells

Sarah Cardoso[1,2], Matheus Andrade[1], Vinícius Cunha[1], João Victor Gomes[2], Cristian Silva[2], Mayra Leão[2], Christopher Fagg[2], Eliete Guerra[3], Francisco Neves[1], Luiz Simeoni[1], Dâmaris Silveira[2], Adriana Lofrano-Porto[1,4]

[1]Molecular Pharmacology Laboratory, Health Sciences Faculty, University of Brasilia, Asa Norte, 70904-970, Brasilia, Brazil, [2]Nature Product Laboratory, Health Sciences Faculty, University of Brasilia, Asa Norte, 70904-970, Brasilia, Brazil, [3]Laboratory of Oral Histopathology, Health Sciences Faculty, University of Brasilia, Asa Norte, 70904-970, Brasilia, Brazil, [4]Gonadal and Adrenal Diseases Clinics, University Hospital of Brasilia, University of Brasilia, Asa Norte, 70904-970, Brasilia, Brazil

Introduction: Therapeutic alternatives to advanced prostate cancer are still limited.1 The Brazilian biodiversity represents a huge source of potential discovery of new drugs derived from plants.2 We aimed to conduct a biomonitoring chemical study of plant extracts from *Pouteria ramiflora* previously shown to be cytotoxic in androgen-sensitive (LNCaP) human prostate cancer cell lines. Methods: The crude extract of *P. ramiflora* branches and stem was pre-fractionated through sintered funnel. The pre-fraction hexane: ethyl acetate 1:1 was fractionated by column chromatography into 649 fractions, grouped according to the profile in thin layer chromatography into 46 samples. Among those, two samples were selected, based on their abundance and cytotoxicity by MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide), the samples were then purified, by solvent, flash and preparative chromatography. The compounds isolated were then identified by Nuclear Magnetic Resonance (NMR) spectroscopy. Results: The hexanic extract of *P. ramiflora* showed a dose-dependent cytotoxic effect in LNCaP, with a high selectivity index (5.5 after 48h of treatment), as compared to a non-cancerous human cell lines keratinocytes (HaCat). Among the 46 samples obtained from this extract, fraction "8" was purified and the compound epifriedelanol was isolated, however, it did not demonstrate cytotoxic activity. Fraction "4" was purified and the compound beta amyrin acetate was initially isolated and a mixture of beta amyrin acetate and lupeyl acetate were also identified. Those latter compounds, however, were not soluble to provide a preparation for *in vitro* cytotoxicity testing. Conclusion: Our biomonitored analysis of *Pouteria* sp. extracts has provided the purification of a number of cytotoxic fractions, and triterpens were isolated. The triterpens have been shown to exert cytotoxic effects on different types of cancer, including prostate adenocarcinoma, at low activity towards normal cells.

PS2-B-071

Antifungal activity of *Sapindus saponaria* L.

Cristian Aldemar Gasca Silva[1], Marline Dassoler[1], Guilherme Brand[2], Christopher W Fagg[3], Pérola O. Magalhães[1], Yris M Fonseca-Bazzo[1], Sueli Maria Gomes[4], Dâmaris Silveira[1]

[1]University Of Brasilia, Brasilia, Brazil, [2]Instituto de Química, University of Brasilia, CEP 70910-900, Brasília, Brazil, [3]Department of Pharmacy, University of Brasilia, CEP 70919-970, Campus Ceilândia, Brasília, Brazil, [4]Department of Botany, University of Brasília, CEP 70910970, Brasília, Brazil

The fruits of *Sapindus saponaria* L. (Sapindaceae) possess mono- and bi-desmosidic saponins able to defend plants against fungal attacks [2]. The objective of our research was to investigate the potential of the ethanolic extract (EE) and semi-purified fractions of *S. saponaria* on the inhibition of *Colletotrichum musae* (226/12I). The fruits were collected in the University of Brasília, Brasília DF (Brazil) and the pericarp was separated and extracted by maceration using hexane and ethanol. The EE was submitted to partition with hexane and ethyl acetate. Hydroalcoholic fraction (FROH) was extracted with *n*-butanol saturated with water. The butanolic phase was partitioned with NaOH 1%, furnishing Bu-sF, that was subject to preparative TLC to afford Bu-sF(D). The antifungal activity was measured by disk diffusion method. Bu-sF(D) was analyzed by CG-MS/MS and ^{1}H and ^{13}C NMR. EE and FROH were active against *C. musae* in concentrations upper to 500 and 250 µg/mL respectively, showing zones of inhibition (ZI) of 12,24 ± 0,5 mm and 10,53 ± 0,42 mm respectively. Bu-sF and and Bu-sF(D) showed IZ (mm) of 12,71 ± 0,50 and 10,68 ± 0,30 at 125 µg/mL respectively. The saponin 3-*O*-(*β*-D-xylopyranosyl)-(1→3)-*α*-L-rhamnopyranosyl-(1→2)-*α*-L-arabinopyranosyl hederagenin and the acyclic sesquiterpene oligoglycoside were identified in Bu-sF(D). The extracts and semi-purified fractions of *S. saponaria* showed antifungal effect. These findings are significant because of their potential use as a phytotherapeutic agent in Anthracnose management.

Acknowledgments: The authors thank FAP-DF, CNPq, CAPES and DPG-UnB.

References:

[1] Damke E, Tsuzuki J, Chassot F, Cortez DA, Ferreira IC, Mesquita CS, da-Silva VR, Svidzinski TI, Consolaro ME. BMC Complementary Altern Med 2013; 13 (1): 196.

[2] Murgu M, Santos LF, Souza GD, Daolio C, Schneider B, Ferreira AG, Rodrigues-Filho EJ. Braz. Chem. Soc. 2008; 19 (5): 831–835.

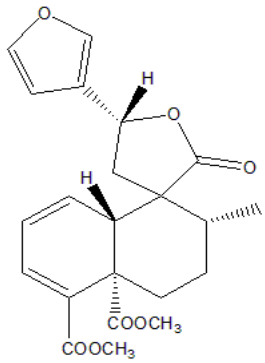
PS2-B-072

Ent-clerodanes and other constituents from the bark of *Croton oligandrus* (Euphorbiaceae) and evaluation of their cytotoxicity

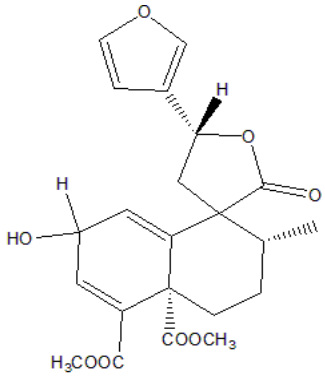
Stephanie T Guetchueng[1], Lutfun Nahar[1], Kenneth J Ritchie[1], Fyaz M D Ismael[1], Nicola M Dempster[1], Andrew R Evans[1], Satyajit D Sarker[1]

[1]Medicinal Chemistry and Natural Products Research Group, School of Pharmacy and Biomolecular Sciences, Liverpool John Moores University, Faculty of Science, James Parsons Building, Byrom Street, Liverpool L3 3AF, United Kingdom

Clerodanes are a group of bicyclic diterpenes widespread in the family Euphorbiaceae [1]. They are known for their wide range of biological activities including anticancer, anti-ulcer, anti-inflammatory and antifeedant among others [1]. RP-HPLC-DAD analysis of the stem bark dichloromethane extract of *Croton oligandrus*, a small medicinal aromatic tree of the Euphorbiaceae found in Central Africa mainly in Cameroon and Gabon [2], led to the isolation of five new clerodane diterpenes named 12-epi-megalocarpoidolide D and crotonolins A-F along with the known compounds 12-epi-crotocorylifuran, 7-*β*-hydroxydehydroabietic acid, 7-Oxodehydroabietic acid, crotonzambefuran A, trans-ferulic acid, 3-*β*-*O*-acetylaleuritolic acid and lupeol. The structures of the isolates were elucidated by spectroscopic means including 1D and 2D NMR, MS, IR, UV, and [*α*]D and also by comparison with the literature data. All the isolated compounds were evaluated for their cytotoxicity against three cancer cell lines (A549, MCF7 and PC3) and a non-cancer cell line (PNT2) using the MTT assay. Preliminary results revealed that the isolated compounds have a moderate level of cytotoxic effect on all the cell lines screened in the range of 80-125 µM.



Crotonzambefuran A



Crotonolin E

Acknowledgements: The Commonwealth Scholarship Commission in the UK is thanked for a 3 years PhD scholarship grant to STG.

Keywords: ent-clerodanes, *Croton oligandrus*, diterpenes, cytotoxicy, medicinal plant

References:

[1] Li R, Morris-Natschke SL, Lee K-H. Nat Prod Rep 2016; 33: 1166–1226

[2] Betti L J, Yongo D O, Mbomio OD, Iponga MD, and A. Ngoye A. Eur J Med Plants, 2013; 3: 174–205

PS2-B-073

Potential anti-hyperglycemic effects and chemical characterization of *Senecio clivicolus*

Immacolata Faraone[1], Flavio Prinzo[1], Dara Kirke[2], Lucia Chiummiento[1], Eloy Fernandez[3], Dilip K Rai[2], Luigi Milella[1]

[1]Department of Science, University of Basilicata, Potenza, Italia, [2]Department of Food BioSciences,Teagasc Food Research Centre, Dublin, Ireland, [3]Faculty of Tropical AgriSciences, Czech University of Life Sciences Prague, Prague, Czech Republic

The interest on *Senecio* genus has been increasing during the last years [1, 2]. For these reasons, the aim of this study was to investigate the potential antidiabetic activity and the phytochemical profile of the madeperennial shrub belonging to the Asteraceae family. The dried aerial parts of *S. clivicolus* were extracted with 96% ethanol. Then, the liquid/liquid extraction using an increasing solvent polarity was used to fractionate the ethanol extract.
The ethanol extract and the *n*-hexane, chloroform, ethyl acetate, *n*-butanol and water fractions were analyzed for their hypoglycaemic potential by measuring of *α*-amylase and *α*-glucosidase inhibition effects [3].
Besides the ethyl acetate fraction acted as inhibitors of *α*-amylase and *α*-glucosidase enzymes, and was subjected to characterization and quantitation of its chemical composition using LC-MS/MS analysis through accurate mass measurements, MS/MS fragmentation patterns and retention times of standards [4].
In the ethyl acetate fraction, of which 19 were tentative identified as of chlorogenic acid origin. These compounds were distributed in three major categories: phenolic derivatives of benzoic acid, cinnamic acid derivatives and flavonoids, known antioxidant compounds. It was demonstrated that chlorogenic acid and derivatives can perform a crucial role in glucose metabolism regulation and thus help to treat the diabetes, and obesity as well [5].
To the best of our knowledge, this is the first report about the evaluation of antidiabetic activity and phytochemical profile of *S. clivicolus*, underlying the importance of this specie as a source of health-promoting phytochemicals.

Keywords: *Senecio clivicolus*, phenols, *α*-amylase, *α*-glucosidase, anti-hyperglycemic.

References:
[1] Ajiboye B *et al*. J Adv Med Life Sci 2014; 1(1):1–5
[2] Parra C *et al*, Nat Prod Res, 2018; 32(6):719–722
[3] Armentano MF *et al*., Biomed Res Int, 2015; 2015
[4] Rico D. *et al*., J Funct Foods 2018; 46:185–194.
[5] Naveed M. *et al*. Biomed Pharmacother 2018; 97:67–74

PS2-B-074

Studies on flavonoids in *Schisandra rubriflora* male and female plant raw material and in their *in vitro* cultures

Agnieszka Szopa[1], Marta Klimek-Szczykutowicz[1], Paweł Kubica[1], Angelika Warzecha[1], Sebastian Granica[2], Halina Ekiert[1]

[1]Chair and Department of Pharmaceutical Botany, Jagiellonian University, Collegium Medicum, 30-688 Cracow, Poland, [2]Department of Pharmacognosy and Molecular Basis and Phytotherapy, Medical University of Warsaw, 02-097 Warsaw, Poland

Schisandra chinensis is an East-Asian medicinal plant species which fruits have been known in Europe from TCM as an hepatoprotective, antitumour and adaptogenic raw material [1]. Additionally, antioxidant properties of shoot and leaf extract were documented. Under our previous research in plant material as well as in *in vitro* cultures of this species we estimated one of the known groups of antioxidant substances – flavonoids [2].
Our current studies deal with flavonoid biosynthetic potential of parent plant material and of *in vitro* cultures of less known Chinese endemic *Schisandra* species – *Schisandra rubriflora.*
The aim of the study was the chromatographic qualitative and quantitative analysis of flavonoids in: fruit, leaf and shoot extracts of male (♂) and female (♀) specimens collected in different vegetation periods in Poland. The estimations were performed also in microshoot culture extracts (♂,♀ lines) growing on agar MS [3] medium with 1 mg/l 6-benzylaminopurine and 1 mg/l indole-3-butyric acid.
In the all analyzed extracts the presence of twenty flavonoids were confirmed by UHPLC-DAD-MS [4]. The quantification of nine of them: naringin, hyperoside, rutoside, isoquercetin, quercitrin, trifolin, quercetin, kaempferol and isorhamnetin, was performed by HPLC-DAD [5].
The maximal total contents of estimated compounds were equal (mg/100g DW): 352.61 in fruits, 2278.35♀ and 2916.04♂ in leaves, and 2819.18♀ and 1877.46♂ in shoots.
The total content of flavonoid in microshoot extracts were equal ♀-397.26 and ♂-439.26 (mg/100 g DW).
Our results documented for the first time, that both, *S. rubriflora* soil-grown plants and microshoots cultured *in vitro*, are a rich source of flavonoids of high antioxidant properties.

Acknowledgements: Funded by National Science Centre, Poland, 2016/23/D/NZ7/01316.

References:

[1] Szopa *et al*. Phytochem Rev 2017; 16: 195–218.
[2] Szopa *et al*. Phytochem.Lett. 2017; 20: 462–469.
[3] Murashige and Skoog, Physiol Plant 1962; 15: 473–497.
[4] Granica *et al*. Food Chem. 2017; 221: 1851–1859.
[5] Ellnain-Wojtaszek and Zgorka. J Liq Chromat Rel Technol 1999; 22: 1457–471.

PS2-B-075

Verbascoside neuroprotective potential through enzyme inhibition and radical scavenging activity

Nuria Acero[1], Inmaculada Zafra[2], Dolores Muñoz Mingarro[3], Carmen Martón Cordero[4].

[1]Pharmaceutical and Health Science Department. Pharmacy Faculty. San Pablo-CEU University, Madrid, Spain, [2]Department of Physical Chemistry. Pharmacy Faculty. Seville University, Seville, Spain, [3]Chemistry and Biochemistry Department. Pharmacy Faculty. San Pablo-CEU University., Madrid, Spain, [4]Department of Pharmacology, Faculty of Pharmacy. Seville University, Seville, Spain.

Neurodegenerative disorders characterized by the loss of neurons within central nervous system have rose their prevalence during the last decades. With the increase in life expectancy, and in consequence, with the aging of the general population, these disorders, as Parkinson´s or Alzheimer´s disease, have become a public health problem around the world (1). Plant derived phenolic compounds had shown capacity to inhibit enzymes involved in these diseases' development. In addition, the antioxidant activity of these compounds allows them to prevent oxidative stress responsible for neurotoxicity (2). Verbascoside is a phenolic compound widely distributed in plants belonging to the Lamiales order. Verbascoside had shown a range of pharmacological activities, such as antioxidant, anti-infammatory or hepatoprotective ability (3).

In this work, verbascoside neuroprotective activity has been studied. In this sense, its *in vitro* effect over tyrosinase, MAO-A and acetylcholinesterase has been analyzed. Verbascoside capacity to prevent oxidative stress in neuroblastoma cellular culture (SH-SY5Y) was also assessed. Results showed that verbascoside is a potential therapeutic tool to prevent or treat neurodegenerative disorders. This compound inhibited both acetylcholinesterase and MAO-A. Verbascoisde was also able to reduce intracellular ROS levels after H_2O_2 induced oxidative stress. These results pointed out that verbascoside could act as a chemopreventive agent, which enhance neurons survival.

Acknowledgements: Authors express their gratitude to Fundación Universitaria San Pablo-CEU and Banco de Santander for the financial support (PPC 20/2015)

Keywords: Verbascoside, neuroprotection, ROS, acetylcholinesterase, MAO-A

References:

[1] Yacoubian, T.A. 2017. Neurodegenerative Disorders. In: Drug Discovery Approaches for the Treatment of Neurodegenerative Disorders, Adeboye Adejare (ed). Academic Press. Elsevier.
[2] Hardy J, Selkoe DJ. Science. 2002; 297(5580): 353–356.
[3] Gay NH, Phopin K, Suwanjang W, Songtawee N, Ruankham W, Wongchitrat P, Prachayasittikul S, Prachayasittikul V. Neurochem. Res. 2018; 43(3): 619–636.

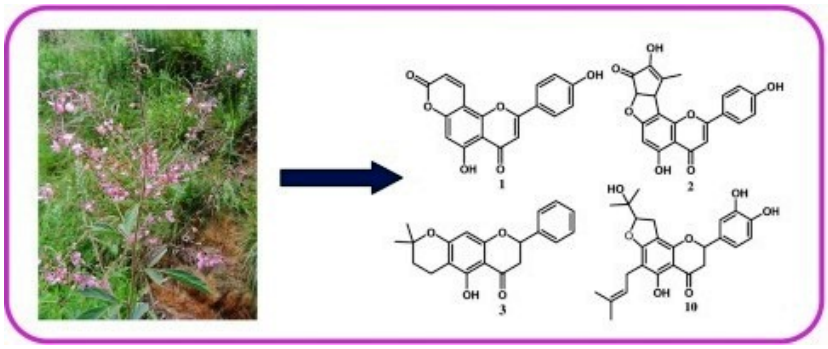
PS2-B-076

Insulin secretory, antibacterial and antiproliferative activity of flavonoidal isolates from *Pseudarthria hookeri* Wight & Arn.

Joseph Tchamgoue[1], Simeon Kouam F[1], Jean Paul Dzoyem[2], Muhammad H Rahman[3], Udo Bakowsky[4], Muhammad Iqbal Choudhary[3]

[1]Department of Chemistry, Higher Teacher Training College, University of Yaounde I, Yaounde, Cameroon, [2]Department of Biochemistry, Faculty of Science, University of Dschang, Dschang, Cameroon, [3]International Center for Chemical and Biological Sciences, University of Karachi, 75270, Pakistan, Karachi, Pakistan, [4]Department of Pharmaceutics and Biopharmaceutics, Philipps University of Marburg, Robert-KochStr.4, D-35037 Marburg, Marburg, Germany

Diabetes is projected to become one of the world's main disablers and killers within the next 25 years and is undoubtedly one of the most challenging health problems of the 21st century. Insulin deficiency is the prime basis of all diabetic manifestation and the conditions associated with diabetes promote the development of pancreatic cancer. In addition, microbial infections are reported to be more frequent and/or serious in patients with diabetes mellitus. Flavonoids are known to encompass antidiabetic, antimicrobial and anticancer properties and could thus be of pivotal significance for the treatment of diabetes and associated complications. In light of this, we carried out the chemical investigation of *Pseudarthria hookeri* in search of bioactive flavonoids which could be further used as leads for the development of new drugs. Our research led to five new and fifteen known flavonoids which were assessed for their insulin secretory, antibacterial and antiproliferative activity. Of the tested compounds, 6-prenyl-3'-methoxyeriodictyol and dorsmanin F were shown to possess potent insulin secretion activity which were 1.6- and 1.8-fold higher than that of the standard, tolbutamide. In addition, pseudarflavone A and 6-prenylpinocembrin showed the highest antibacterial activity with minimum inhibitory concentration values ranging from 16 to 32 and 8 to 64 µg/mL, respectively. Finally, all the tested compounds showed inhibitory activity to the tumor cell lines in a variable extent with IC_{50} values varying from 3.59 to 92.32 µg/mL. Pseudarflavone A and 6-prenylpinocembrin were the most active towards human leukemia cells Jurkat with the lowest IC_{50} values of 3.59 and 5.59 µg/mL, respectively. Interestingly, the tested compounds showed no toxic effect on MIN-6 and 3T3 cell lines up-to 400 µM, suggesting their safety profile. Further galenic formulation from the flavonoid-rich extract of this plant might be useful for the treatment of diabetes and its associated diseases



PS2-B-077

Stability of phenolic compounds and antioxidant capacity of carob pulp products subjected to simulated *in vitro* digestion

Vlasios Goulas, Andriani Hadjisolomou

Cyprus University of Technology, Limassol, Cyprus

Carob product compounds is a dynamic research area as they have shown to protect against many chronic diseases such as diabetes symptoms, hyperlipidemia and also reduces the risk of cancer. These disease-preventing effects have been mainly attributed to the phenolic fraction of carob pulp, but, a little information is available for the stability of phenolics during digestion. The objective of this work was to study the impact of gastrointestinal digestion on the stability of phenolics and antioxidant potency in carob products. Thus, the pure phenolics and carob pulp products are subjected to simulated digestion. The digestion protocol reproduced three steps of the digestive physiology process, including a simulated oral, gastric and small intestinal digestive step according to previous study.
Results demonstrated that the phenolic composition and antioxidant potency of carob pulp products are strongly influenced by digestion. In general, the phenolic acids had enhanced resistance under gastrointestinal conditions, whereas flavonols were the most unstable among compounds studied. Our findings also highlight a great impact of food matrix on the stability of phenolics. More specific, the carob phenolics released from food matrix or bound to the food constituents. Furthermore, the intestinal digestion caused the major losses of phenolics, while the gastric step had mostly positive effect since the acidic conditions may release phenolic compounds from food matrix. In addition, a noteworthy reduction of antioxidant activity (20-50%) was found after digestion as it measured by DPPH and FRAP assays. In conclusion, significant amounts of phenolic antioxidants in carob products remain after gastrointestinal digestion. In a next step, in vivo assays should be performed to evaluate the real impact of carob phenolics towards oxidants status in human body after gastrointestinal digestion.

Keywords: phenolic acids, flavonoids, carob powder, carob syrup, carob fibre, *Ceratonia siliqua* L

PS2-B-078

Natural phthalides as mono and dimer in *Kelussia Odoratissima Mozaff.*: insight from chemistry to biological activity

Mahdi Ayyari[1,2], Mohammad Rezaei[2], Safiollah Raeissi[3], Maxime Le Bot[4], Aniseh Alimohammadpour[1], Shahram Eisa-Beygi[5], Sara Pahlavan[2], Gilda D'urso[6], Mahya Hosseini[2], Mehrnaz Namiri[2], Alexander D Crawford[7], Dimitri Bréard[4], Yaser Tahamtani[2], Seyed Hamzeh Hossieni Kahnouj[8], Ingrid Freuze[4], Sonia Piacente[6], Pascal Richomme[4], Hossein Baharvand[2,9]

[1]Department of Horticultural Science, Tarbiat Modares University, Tehran, Iran, [2]Royan Biodiscovery Initiative, Department of Stem Cells and Developmental Biology, Royan Institute, Tehran, Iran, [3] FTSS Co., Incubator Center of Chaharmahal & Bakhtiari Science and Technology Park, Sharekord, Iran, [4]SONAS, SFR4207 QUASAV, University of Angers, Beaucouzé, France, [5]Department of Radiology, Medical College of Wisconsin, 53226, Milwaukee, USA , [6]Dipartimento di Scienze Farmaceutiche, Università degli Studi di Salerno, Via Ponte Don Melillo, 84084, Fisciano, Italy, [7]Department of Basic Sciences and Aquatic Medicine, Faculty of Veterinary Medicine, Norwegian University of Life Sciences (NMBU), Oslo, Norway , [8]Department of Biology, Faculty of Science, Jiroft University, Jiroft, Iran, [9]Department of Developmental Biology, University of Science and Culture, Tehran, Iran

Phthalildes are a relatively small group of natural products found in several higher plants and even in the fungal genera. They occur in monomeric and dimeric forms, exhibiting diverse biological activity profiles. Plants with these kinds of metabolites are common in different traditional medicines around the world. *Kelussia odoratissima Mozaff.* (Apiaceae), known as “Kelows”, is an endemic Iranian plant and a rich source of phthalides. It grows in the central Zagros Mountains at altitudes of 2500 m and above. There is a plethora of traditional usage along with biological and pharmacological activities ascribed to this species, including treatment for cardiovascular diseases, rheumatism, ulcers, indigestion, hypertension and inflammation. Kelows is also popular as a wild culinary herb used in soups, salads, pickles and yogurt. In this study, the metabolite profiling of kelussia essential oils and different extracts has been carried out using GC/MS and LC/MS (LC-ESI/LTQOrbitrap/MS and LC-PDA-ESI-MSn). The major components of this plant are phthalides, found as monomers and dimers. However, stability, isolation and identification of these natural compounds are cumbersome. Several biological activities, including anti-angiogenesis (in zebrafish and HUVECs), antiepileptic potential (in zebrafish) and antioxidant properties showed that kelussia can be a promising plant for further study as natural supplement or drug. The popularity, ethnobotanical background, extinction risks, regeneration and domestication of the Kelussia will also be explained [1-3].

PS2-B-079

Traditional herbal remedy revisited - the example of bioactive compounds from carnivorous plants combined with silver nanoparticles used to combat *Staphylococcus aureus*

Marta Krychowiak[1], Anna Kawiak[1], Magdalena Narajczyk[2], Agnieszka Borowik[3], Eng. Aleksandra Krolicka[1]

[1]Department of Biotechnology, Intercollegiate Faculty of Biotechnology, University of Gdansk and Medical University of Gdansk, Gdansk, Poland, [2]Laboratory of Electron Microscopy, Faculty of Biology, University of Gdansk and Medical University of Gdansk, Gdansk, Polska, [3]Department of Molecular and Cellular Biology, Intercollegiate Faculty of Biotechnology, University of Gdansk and Medical University of Gdansk, Gdansk, Polska

Carnivorous plants (e. g. *Drosera* spp. and *Dionaea muscipula*) are plants that evolved to trap and consume insects, or even small animals. Tissues of plants from Droseraceae family have been also used for centuries in traditional medicine. Versatile secondary metabolites are responsible for biological activity of infusions derived from these carnivorous plants. In the era of antibiotic resistance, we employed synergistic approach to combat *Staphylococcus aureus* with extract from *Drosera binata* and silver nanoparticles (AgNPs) [1]. These two simultaneously used components showed high bactericidal activity towards different strains of *S. aureus*, susceptible as well as resistant to antibiotics. What is more, we identified naphthoquinones (NQs) present in the tissues of *D. binata* as the most potent secondary metabolites with synergistic potential in combination with AgNPs. Results obtained in this study revealed the significance of synergistic interaction of NQs and AgNPs – not only did it improve the antimicrobial potential but it also helped to fine-tune doses of these two agents. Moreover, synergy between tested nanoparticles and secondary metabolites allowed to overcome the problem of cytotoxicity of NQs towards eukaryotic cells. During further analyses we explored the potential of AgNPs combined with the most prevalent NQs in tissues of carnivorous plants and identified plumbagin, ramentaceone and 3-chloroplumbagin as the most potent. We also determined the mechanism by which the AgNPs enhance the anti-staphylococcal activity of naphthoquinones and confirmed the relevance of silver ions, the membrane damage and the direct interaction of nanoparticles and NQs. The synergistic strategy employed in our study allowed to explore the antibacterial potential of plant-originated compounds and to develop a multi-target antimicrobial approach.

Acknowledgements: This work was supported by the grant from the National Science Centre in Poland (PRELUDIUM 10 grant no. 2015/19/N/NZ7/02802).

References:

[1] Krychowiak M *et al.* Plos One 2014; 9(12): e115727.

PS2-B-080

Biological properties of *Tasmannia lanceolata* CO_2 extract

Marina Humbert, Emmanuelle Gaillard

Robertet Group, France

Tasmannia lanceolata L. (Fam Winteraceae) is a wild plant native to northern Tasmania, already known for its applications in flavours and perfumes. Polygodial has been identified as its primary active compound, and is also responsible for its peppery taste. For the first time, we have investigated the cosmetic properties of a CO_2 extract of *Tasmannia lanceolata* leaves [Tasmanol]. Before studying the biological activities of the extract, the safety has been validated through a series of toxicological tests. Then, a first *in vitro* study on primary human fibroblasts has been performed to evaluate Tasmanol effects on inflammation, collagen I synthesis and wound healing. A significant increase in collagen 1 was observed on human fibroblasts after 7 days as well as a significant increase in cell migration after 24h on a wound healing model. These preliminary results suggest a potential positive role on stretch marks. Stretch marks are thought to occur when the skin stretches faster than the fibroblast cells can produce collagen. At the molecular level, disruption of dermal connective tissue, including collagen fibrils and elastin fibers, likely decrease strength and elasticity of the skin. A second work using a human skin model issued from abdominoplasty has been done to investigate the role of Tasmannia on stretch marks. The activity of Tasmanol on specific stretch mark biological targets such as TGFß, Pro-collagen I, Collagen III and Elastin has been assessed and will be discussed.

PS2-B-081

Response of *Bradyrhizobium japonicum* nodule variables to cucurbitacin-containing phytonematicides in cowpea (*Vigna unguiculata*) on N-deficient soil

Kola M. E. [1,2*], Mashela P. W. [1], Lukhele-Olorunju P. [2]

[1]*University of Limpopo, Green Biotechnologies Research Centre of Excellence, Private Bag X1106, 0727, Sovenga, South Africa*
[2]*University of Mpumalanga, Department of Research, 1200, Mbombela, South Africa*

**(e-mail: Elizabeth.kola@ump.ac.za)*

The Curve-fitting Allelochemical Response Data (CARD) had been adopted to develop the non-phytotoxic concentration of phytonematicides, technically referred to as the mean concentration stimulation point (MCSP). The MCSP for cucurbitacin-containing phytonematicides on Bradyrhizobium-nodulated legume crops had not been documented in low-input agricultural farming systems. The objective of this study was to determine the MCSP values of Nemarioc-AL and Nemafric-BL phytonematicides on B. japonicum-nodulated cowpea (*Vigna unguiculata*) on Nitrogen-deficient soils. Inoculated cowpea seeds were sown in 20-cm-diameter plastic pots containing steam-pasteurised N-deficient loamy soil and Hygromix at 3:1 (v/v) ratio. Two parallel experiments comprised Nemarioc-AL and Nemafric-BL phytonematicides, each had seven treatments (0, 2, 4, 8, 16, 32 and 64%) and applied once weekly. At 56 days after the treatments the MCSP values for Nemarioc-AL and Nemafric-BL phytonematicides on cowpea were 5.4% and 6.3%, respectively, with the overall sensitivity (∑k) of one for each phytonematicide. In conclusion, the observed MCSP and ∑k values suggested that the two cucurbitacin-containing phytonematicides were suitable for Bradyrhizobium-nodulated cowpea production systems on N-deficient soils.

Keywords: Climate-smart agriculture, density-dependent growth, *Meloidogyne* species, nitrogen-fixing bacteria, sustainable agriculture, quadratic relations, phytonematicides

PS2-B-082

Actiscent®: boosting cosmetic activity with a fragrance

Anthony Pegard

Robertet, Grasse, France

Fragrances are a complex mixture between synthetic and natural ingredients.
Robertet has studied for more than 18 years the cosmetic properties of natural ingredients used by perfumers to create fragrances.
From essential oils (water distillation) to absolutes (extraction with organic solvent), our researchers have discovered that some could be true treasures for skin.
Based on these observations, active fragrances called Actiscents® have beeen formulated. They are 2-in-1 fragrances, bringing the smell to the final product, but also boosting the cosmetic activity.

PS2-B-083

Cucurbitacin A affects mobility of *Meloidogyne incognita*

Zakheleni Dube

University of Mpumalanga, Mbombela, South Africa

Registration authorities of botanical inputs in crude extract form demand that the active ingredients be isolated, identified and efficacy tests on target pests be investigated [1]. Nemarioc-AL phytonematicide, with active ingredient cucurbitacin A, consistently suppress bioactivities of root-knot (*Meloidogyne* species) nematode, with limited information on the efficacy of its purified active ingredient. The objective of this study was to investigate the effects of cucurbitacin A on mobility of *M. incognita* second-stage juveniles (J2). Approximately 450 freshly hatched J2 were exposed to pure cucurbitacin A concentration at 0.00, 0.25, 0.50, 1.00, 1.25, 1.50, 1.75, 2.00, 2.25 and 2.50 µg.mL–1 distilled water, arranged in a completely randomised design, with three replicates. Experimental units were placed in an incubator at 25 ± 2°C for 12-, 24-, 48- and 72-h. After each duration, mobile and immobile J2 were each counted using a stereomicroscope. Cucurbitacin A concentration effects were highly significant on J2 immobility at all four exposure periods, contributing 84, 99, 99 and 99% in total treatment variation of the variable at the respective periods. Relative to untreated control, J2 immobility increased with increasing concentrations in density-dependent growth (DDG) patterns. In conclusion, cucurbitacin A had significant bioactivity effects on mobility of *M. incognita* J2 and could be one of the bioactivities through which Nemarioc-AL phytonematicide suppressed nematode population densities.

Acknowledgements: The authors are grateful to the Land Bank Chair of Agriculture- University of Limpopo, Agricultural Research Council - Universities Collaboration Centre, National Research Foundation (NRF) of South Africa and the Flemish Interuniversity Council (VLIR) of Belgium for funding this study.

Keywords: Cucumin, *Cucumis myriocarpus*, leptodermin, paralysis, triterpenes

References:

[1] Organisation for Economic Co-operation and Development (OECD). (2017). guidance document on botanical active substances used in plant protection products. OECD Environment, Health and Safety Publications. Series on Pesticides No. 90.

PS2-B-084

Effects of fermented *Cucumis africanus* and *Momordica balsamina* extracts on *Pseudococcus citri*

Zakheleni Dube[1,2], Foster Mokhoelele[2]

[1]School of Biology & Environmental Sciences, University of Mpumalanga, Private Bag X11283, Mbombela, South Africa, [2]University of Limpopo, Private Bag X1106, Sovenga, South Africa

Environment-friendliness and compatibility of phyto-insecticides makes them ideal alternatives to synthetic chemical pesticides in pest management. *Cucumis africanus* and *Momordica balsamina* extracts had consistently suppressed root-knot nematodes but their bioactivities against insect pests had not been empirically confirmed. The objective of this study was to assess the bioactivities of fermented crude extracts of *C. africanus* and *M. balsamina* on *Pseudococcus citri* nymphs. *In vitro* experiments with treatments namely; 0, 3, 9, 12, 15, 18, 21, 24 and 27% phytoinsecticides were conducted with experiments laid-out in a CRD with six replications. Three independent experiments were conducted for each phyto-insecticide. *Mimusops zeyheri* leaves first dipped in respect concentrations of phyto-insecticide were placed upside down in 9-cm-diameter petri dish containing moistened filter paper. Ten *P. citri* nymphs of the same size collected from *M. zeyheri* plants by carefully placed on surface sterilised leaves of *M. zeyheri* and incubated at 25 ± 2 °C. Repellence and mortality of the plant extracts were measured after 24 and 48h, respectively. There were no significant differences ($P > 0.05$) between the three independent experiments hence the data were pooled and reanalysed. *Cucumis africanus* and *M. balsamina* extracts had no significant effect on repellence but significantly ($P \leq 0.05$) affected mortality of the pest. Mortality of *P. citri* exposed to plant extracts increased in a dose-response manner. The LC_{50}s of the two extracts were comparable at 7 and 8% for *M. balsamina* and *C. africanus* extracts, respectively, whereas *C. africanus* had approximately twice as high LC_{90} as *M. balsamina* extracts at 47 and 25%, respectively. In conclusion, extracts of *C. africanus* and *M. balsamina* had bioactive effects on *P. citri* hence they have potential for use as alternative to synthetic chemical insecticides in the management of *P. citri* on *M. zeyheri*.

Keywords: Allelochemicals, botanicals, mealybug, mortality, wild watermelon

PS2-B-085

The effect of natural hydroxynaphthoquinones on 3T3-L1 cells and adipogenic differentiation

Athanasios Arampatzis, Olga Tsave, Vassilios P Papageorgiou, Andreana N Assimopoulou

Laboratory of Organic Chemistry, School of Chemical Engineering, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece

Alkannin and Shikonin (A/S) are two enantiomeric hydroxynaphthoquinones with a broad spectrum of biological properties. These are biosynthesized in the roots of Boraginaceae plants, such as *Alkanna tinctoria, Lithospermum erythrorhizon* and others. In recent years, shikonin has been reported to possess anti-adipogenic, anti-obesity and insulin-like effects. The aim of the current study was to investigate the potential structure anti-adipogenic activity relationship (SAR) of alkannin, shikonin and derivatives.

Experiments were carried out *in vitro* using the 3T3-L1 cells. Cell viability, migration and morphology were investigated upon treatment with several hydroxynaphthoquinones. Various concentrations of the tested compounds were applied and the EC_{50} values were calculated. Cell differentiation was induced in the presence of purified shikonin, alkannin and other derivatives. 3T3-L1 pre-adipocytes were treated with either 10 ng/mL of insulin and/or selected compounds (100nM & 1μM). On the 14th day of the differentiation process, cell differentiation was examined and validated by oil red O staining. Real-Time PCR was employed in order to assess the relative expression of Peroxisome proliferator-activated receptor gamma (PPAR-γ) and Adiponectin (ADIPOQ) genes.

Adipogenic differentiation was attenuated in the presence of the naphthoquinones tested. All compounds significantly inhibited mRNA expression of the PPAR-γ gene. In the case of ADIPOQ, shikonin and alkannin reduced mRNA expression, whereas deoxyshikonin increased the levels of ADIPOQ, indicating that this compound might induce adipogenesis, compared to insulin under the current experimental conditions.

In summary, the major findings show that A/S possess anti-adipogenic properties which might also be a potential indicator of an anti-diabetic profile.

Acknowledgements: This study was supported by the project "MICROMETABOLITE" that has received funding from the European Union's Horizon 2020 research and innovation programme, under the Marie Skłodowska-Curie grant agreement No 721635. The authors are grateful to Professor Constantinos Panayiotou (School of Chemical Engineering, AUTh) for giving open access to the laboratory.

PS2-B-086

Anti-fouling potential from the LEGE culture collection (LEGE-CC)

Joana Azevedo[1,2], Jorge Antunes[1,2], Pedro Leão[1,2], Vítor Vasconcelos[1,2]

[1]CIIMAR, Porto, Portugal, [2]University of Porto, Porto, Portugal

Marine biofouling organisms, namely the hard macrofoulers barnacles and mussels, represent an economical problem worldwide, particularly to the shipping industry, due to frequent ship hulls maintenance and also increased fuel consumption. Many antifouling coatings have been formulated and are available in the market, however, these products are based on persistent and toxic compounds which may seriously harm the marine environment. Thus, it is of great importance to find alternative environmentally friendly antifouling strategies, namely secondary metabolites from cyanobacteria which have already showed some antifouling potential. In this work, 71 cyanobacterial strains from the culture collection of the LEGE – CIIMAR (https://lege.ciimar.up.pt) were cultured in an appropriate medium- Z8 medium [1] and freeze-dried biomass was extracted accordingly to Edwards *et al.* [2]. The cyanobacterial crude extracts were then used in vivo against anti-fouling mussels plantigrade larvae (*Mytilus galloprovincialis*) in order to evaluate the effect of the cyanobacterial extracts on the production of their adhesive structure. The extracts were also tested against bacteria and diatoms which are involved in marine biofilm formation and are part of the microfouling communities. The cyanobacterial strains with more bioactivity were cultured in a large scale, extracted and Vacuum Liquid Chromatography (VLC) fractionated. One strongly bioactive fraction was HPLC-PDA purified and analyzed by proton NMR and LC-MS in order to elucidate the molecule structure. This compound is being tested to fully determine its anti-fouling potential.

Keywords: Marine fouling, cyanobacteria compounds, antifouling

References:

[1] Kotai J. 1972. Instructions for the preparation of modified nutrient solution Z8 for algae. Oslo: Norwegian Institute for Water Research Blindern NIVA B-11/69.

[2] Edwards D J, Marquez B L, Nogle L M, McPhail K, Goeger D E, AnnRoberts M, Gerwick W H. Chem. Biol. 2004; 11: 817–833.

PS2-B-087

α-glucosidase inhibitory activity of isolated compounds from the endophytic fungi *Annulohypoxylon stygium* derived from the leaves of *Pandanus* simplex

Roberth Riggs Rondilla[1], Thomas Edison de la Cruz[1], Fang-Rong Chang[2], Maribel Nonato[1]

[1]The Graduate School, University of Santo Tomas, Manila, Philippines, [2]Graduate Institute of Natural Products, Kaohsiung Medical University, Kaohsiung, Taiwan

Endophytic fungi isolated from *Pandanus* simplex leaves were evaluated for its antidiabetic potential through *in vitro α*-glucosidase inhibition assay. The crude extract of *Annulohypoxylon stygium* gave the highest percentage of activity and thus selected for further purification. A mixture of unsaturated fatty acids and 8-methoxynaphthol were isolated from the crude extract which showed inhibition (IC_{50} value of 321.2 and 676.3 μg/mL, respectively). The isolation of the mentioned compounds from *A. stygium* is reported herein for the first time. The results demonstrated that endophytic fungi from *P. simplex* is a sustainable source of secondary metabolites that can be further explored as alternative chemical entities to alleviate diabetes.

PS2-B-088

Physalis angulata calyces fraction promoted mice recovery in AOM/DSS-induced colon cancer model

Luis Franco[1], Yanet Ocampo[1], David Rivera[1], Daneiva Caro[1], Johana Cuadro[1], Jenny Castro[1], Coralia Osorio[2]

[1]University of Cartagena, Cartagena, Colombia, [2]National University of Colombia, Bogota, Colombia

Introduction: *Physalis angulata* is recognized by its numerous ethnopharmacological applications in tropical and subtropical regions of world. The anti-cancer activity of *P. angulata* is its most popular traditional use, as well as one of the most studied bioactivities of this plant [1]. However, no studies are available concerning the anti-proliferative effect of calyces, an organ from *Physalis* genus recognized as a prolific source of new bioactive compounds. The aim of this study was to assess the potential of a fraction obtained from the ethanolic extract of *P. angulata* calyces to inhibit the colonic carcinogenesis in AOM/DSS-induced colon cancer model.
Methods: Calyces were collected and extracted with ethanol and fractionated. Fraction F0-2 was evaluated in the mice model of azoxymethane (AOM) and dextran sodium sulphate (DSS) induce colonic cancer.
Results: The administration of F0-2 promoted mice recovery. In agreement, necropsy showed strongly reduction of the tumor burden without affecting significantly the number of tumors. Furthermore, no toxicological effects were detected in healthy mice treated with F0-2.
Conclusion: To our knowledge, this is the first report of *P. angulata* plant using an in vivo model of CRC. Our results indicate the potential of these calyces as an alternative treatment for CRC. Further investigation is warranted for the identification of the bioactive components.

PS2-B-089

Cytotoxic activity of an enriched fraction of *Physalis angulata* calyces against two human colon cancer cell lines

Daneiva Caro[1], Yanet Ocampo[1], Johana Cuadro[1], David Rivera[1], Guillermo Schinella[2], Ruben Salas[1], Luis Franco[1]

[1]University of Cartagena, Cartagena, Colombia, [2]National University of La Plata, La Plata, Argentina

Physalis angulata L. (Solanaceae) is used in Colombian folk medicine. The anti-cancer activity is the most popular traditional use, and its effectiveness has been demonstrated against cervix, skin, lung, liver, gastric, and colorectal cancer [1]. The objective of this work was to evaluate the *in vitro* cytotoxicity activity of an enriched fraction isolate from the total ethanolic extract obtained of *P. angulata* calyces against 2 human colon cancer cell lines. *P. angulata* extract was fractionated by liquid-liquid partition to obtain the active primary fraction in dichloromethane F02 (IC_{50}=13.8 µg/mL on HT-29 cells), which was subsequently fractionated by column chromatographic to obtain an enriched fraction (EFPA, yield = 25.78%). The cytotoxicity of EFPA on a HT-29 and CaCo2 colon cancer cell lines as well as the epithelial normal cells CCD 841 CoN, was evaluated employing the MTT colorimetric method. The IC_{50} was calculated and the selectivity index (SI) was determined, as the ratio from the IC_{50} on normal cell and IC_{50} on cancer cell lines. EFPA showed a prominent increase in the cytotoxicity activity in comparation to F02 fraction with IC_{50} of 3.19 µg/mL and 15.23 µg/mL on HT-29 and CaCo2 cells, respectively. Regarding the SI, EFPA exhibit a high degree of selectivity for HT-29 cells (SI = 3.3). EFPA was subsequently fractionated by column chromatography to obtain 14 sub-fractions (EFPA1-EFPA14) with 99.56% efficiency. The cytotoxic activity observed in most of these subfractions were lower than the parental fraction, suggesting a synergy affect between different metabolites with different polarities. Only EFPA3 retained the activity of the parental fraction (IC_{50} = 3.24 µg/mL), which indicates that the most active metabolites are concentrated here. Our results indicate the potential of *P. angulata* calyces as an alternative treatment for CRC. Further investigation is warranted for the identification of the bioactive components present in EFPA3.

PS2-B-090

Cape gooseberry (*Physalis peruviana*) modulates the release of inflammatory mediators in LPS-activated RAW 264.7 macrophages

Jenny Castro, Daneiva Caro, Nely Mejia, Jaime Salgado, Daniela Amaris, Jesus Cortez, Luis Franco, Ruben Salas

University of Cartagena, Cartagena, Colombia

Chronic non-communicable diseases (NCD) like diabetes, inflammatory bowel disease, cancer, among others are the main cause of morbidity and mortality of the world population. NCD incidence is linked to modifiable risk factors, such as diet. The high consumption of fruit it has been associated with the decrease the risk of developing NCD1. *Physalis peruviana* L. is native to the South American and is one of the best-known species of this genus. It is widely used in folk medicine for the treatment of inflammatory diseases. In this work we evaluated the antioxidant potential and the effect on the production of inflammatory mediators of the ethanolic extract obtained of *P. peruviana* fruits (Cape gooseberry). The fruit pulps were lyophilized and a total extract was obtained by maceration with ethanol. Secondary metabolites were identified by preliminary phytochemical screening. The content of phenols and flavonoids was determined by the methods of Folin-Ciocalteu and aluminum trichloride, respectively. The antioxidant potential of the extract was determined using the free radical scavenging DPPH● and ABTS●+ spectrophotometric methods. The anti-inflammatory activity of the extract was evaluated determining their activity on the production of NO•, IL-6 and TNF-α, in supernatants of LPS-activated RAW264.7 macrophages. The results showed that the extract of Cape Gooseberry had a moderate content of phenols (0,53 ± 0,01 mg of gallic acid/g fruit pulps). In addition, the extract showed a scavenging effect of free radicals DPPH● and ABTS●+, in a concentration-dependent manner, with IC_{50} values of 3270 µg/mL (CI95% = 3130 – 3413) and 1178 µg/mL (CI95% = 1073 - 1285) respectively. Cape Gooseberry extract significantly reduced the levels of NO• (70.14 %), IL-6 (70.16%) and TNF-α (27.7%) at non-toxic concentrations on RAW264.7. The extract of Cape Gooseberry should continue to be explored since their consumption in the diet could provide benefits beyond basic nutrition.

PS2-B-091

Synthetic cannabinoids AM–251 and AM–1241 induce apoptosis in prostate cancer cells

Sotia Louka, Christiana Neophytou, Andreas Constantinou

University of Cyprus, Nicosia, Cyprus

Despite the many advances in prostate cancer treatment in recent years, there is still need for the discovery of novel therapeutic approaches. Synthetic cannabinoid compounds have attracted attention as potential anticancer drugs that affect different levels of cancer progression, including inhibition of proliferation, invasion and induction of apoptosis. We investigated the effects of synthetic cannabinoids AM–251 and AM–1241 in cannabinoid receptor–expressing DU145 prostate cancer cells. AM–251 is a potent inverse agonist of the CB1 receptor while AM–1241 is a selective agonist for the receptor CB2. Our aim was to examine the pro–apoptotic and anti–proliferative effects of these compounds in prostate cancer cells. We applied MTT proliferation, flow cytometry, Annexin V/Propidium Iodide and DNA fragmentation assays. Our results show that AM–251 and AM–1241 inhibit the proliferation of prostate cancer cells with an IC_{50} of 20 and 29 μM respectively. Both compounds increased DNA fragmentation at 30 μM 18–fold compared to the control. Their mechanism of action possibly involves the induction of caspase–dependent apoptosis. Pan–caspase inhibitor z.vad.fmk partially restored viability of cells treated with 30 μM AM–251, while the viability was completely restored in cells treated with AM–1241. Consistently, DNA fragmentation was completely abolished in the presence of the inhibitor. Apoptosis induction was also evident by the increase of the sub–G1 fraction (30 μM of AM–251 induced 21.2% subG1 fraction and 60 μM AM–1241 induced 20.2%). In addition, cells treated with 30 μM AM–1241 for 72 hours stained positive for Annexin V indicating early apoptosis. A late apoptotic effect was observed at 72 h, with 39.1 % and 17.8 % staining double positive for AnV/PI following treatment with 30 μM of AM–251 and AM–1241 respectively. Further studies will elucidate the mechanism of action of these compounds in prostate cancer and normal cells and will highlight their potential use in prostate cancer treatment.

PS2-B-092

In vitro anti–proliferative and apoptosis–inducing activity of *Rhodiola rosea* L. against human glioblastoma cells

Aney Marchev[1], Iliyan Manoylov[2], Gabriela Boneva[2], Silviya Bradyanova[2], Ivanka Koycheva[1], Aney Tchorbanov[2], Milen Georgiev[1]

[1]Group of Plant Cell Biotechnology and Metabolomics, The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences, 139 Ruski Blvd, 4000 Plovdiv, Bulgaria, [2]Laboratory of Experimental Immunology, The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences, 26 Georgi Bonchev Str., 1113 Sofia, Bulgaria.

Rhodiola rosea L. is the most popular plant species among family Crassulaceae acting as adaptogen and possessing anti–depressive, immune stimulating and apoptosis–inducing activities in glioma and human neuroblastoma cells. The most valuable molecules in this plant are the phenylethanoids and phenylpropanoids, found in its rhizomes [1].
The aim of the present study was to quantify and qualify the secondary metabolites in *R. rosea* rhizomes wild–grown in Bulgaria by NMR–based metabolomics and HPLC. The anti–proliferative and apoptotic activity of 70% methanolic extract of *R. rosea* rhizomes, as well as, the major molecules identified were evaluated towards human glioblastoma cell line U87 MG.
The ^{1}H NMR spectra of the rhizomes revealed that the dominant identified metabolites were salioside, rosarin, rosavin and rosin and their respective estimated amounts by HPLC were 2.67 %, 0.37%, 1.97% and 0.04%.
The level of cell proliferation (MTT assay) was evaluated on the last four hours after a three–day culture period. The most prominent activity was observed by the rhizomes extract at 100 μg/mL with approximately 30% decrease of the cells proliferation. Among the identified metabolites the most active were rosarin and salioside at 25 μg/mL with 20.30% and 16.39% inhibition of the cell proliferation. Rosin, *p*–tyrosol and rosavin had an anti–proliferative activity in the same range, but at 100 or 200 μg/mL. The analysis of the cell apoptosis showed that incubation with the extract and the pure compounds influenced in a non–dose dependent manner this process in the homogenous glioblastoma cell culture.

Acknowledgements: This study has been supported by a grant from National Science Fund of Bulgaria (contract number DM11/3).

Keywords: *Rhodiola rosea*, NMR, salioside, *p*–tyrosol, rosavins, glioblastoma, apoptosis

References:

[1] Marchev A, Dinkova–Kostova A, György Z, Mirmazloum I, Aneva I, Georgiev M. Phytochem Rev 2016; 15: 515–536.

PS2-B-094

Field assessment of a carvacrol rich essential oil as a multitask mosquito control agent

Epameinondas Evergetis[1], Romeo Bellini[2], George Balatsos[3], Marco Carrieri[2], Rodolfo Veronesi[2], Arianna Puggioli[2], Vassiliki–Nafsika Kapsaski–Kanelli[1], Serkos Haroutounian[1], Dimitrios Papachristos[3], Antonios Michaelakis[3]

[1]Department of Nutritional Physiology and Feeding, Agricultural University of Athens, Athens, Greece, [2]Centro Agricoltura Ambiente "G. Nicoli", Crevalcore, Italy, [3]Department of Entomology & Agricultural Zoology, Benaki Phytopathological Institute, Kifisia, Greece

Under the frame of the LIFE CONOPS project [1] we developed a bio–prospecting experiment aiming to investigate the potencies of 8 Mediterranean wild gathered foods essential oils (EOs) as mosquito control agents. The initial bioassay screening against *Aedes albopictus* (Asian tiger mosquito) highlighted as most potent three Lamiaceae EOs, derived from *Satureja thymbra*, *Origanum onites*, and *Thymbra spicata*. All presented as principal component the molecule of carvacrol, exhibited DEET–like repellent performance, and total larvae mortality defining the carvacrol rich EO (CREO) as a promising mosquito control agent [2]. Second phase experimentation involved the dose–response definition and eco–toxicity delineation of CREO [3]. As CREO source in this phase was selected *Origanum vulgare* ssp. *hirtum*, a taxon of wide expansion and with numerous cultivation varieties, while as standard was used pure carvacrol. The findings indicated an *Ae. albopictus* larvae LC90 of 58,75 mg/L, a minimum *Ae. albopictus* adult's repellent concentration of 0,2 μL/cm2, and LC90 of 12,80 mg/L against *Macrocyclops albidus* (non–target organism). Third phase experimentation was focused on CREO formulation and its performance in field conditions. CREO source was a commercial cultivar of *O. vulgare* ssp. *hirtum*, which was evaluated as an emulsion for spatial repellent and larvicidal activity, and per se as larvicidal agent. The emulsified CREO made possible the large area coverage that proved it as a potent spatial repellent; its maximum efficacy 86% was recorded in day 1, and gradually declined in the following two days (81%, 69%). Both emulsified and crude CREO proved to be efficient larvicidal agents as they suppressed the mosquito population for two weeks, with crude CREO to outperform slightly the emulsified in terms of endurance.

References:

[1] LIFE12 ENV/GR/000466 (www.conops.gr).
[2] Parasitol Res 2018; 117: 1953–1964.
[3] Integrated Pest Management and Pest Control 26: 613–637, InTech Publisher ISBN 2012; 978–953–307–926–4.

PS2-B-095

Evaluation of *Stevia rebaudiana* extracts as potential active ingredients for natural cosmetics

Katarzyna Gaweł–Bęben[1], Wirginia Kukuła–Koch[2], Marcelina Strzępek[1], Uliana Hoian[1], Paweł Osika[1], Kazimierz Głowniak[1,2]

[1]Department of Cosmetology, University of Information Technology and Management in Rzeszów, 2 Sucharskiego Str., 35–225 Rzeszów, Poland, [2]Chair and Department of Pharmacognosy with Medical Plant Unit, Medical University of Lublin, 1 Chodźki Str., 20–093 Lublin, Poland

Stevia rebaudiana Bertoni (Asteraceae) is a perennial plant native to South America, used mainly as a zero–calorie natural sweetener. The sweet taste of stevia depends on the high content of ent–kaurane–type diterpene glycosides, known as steviol glycosides. In the leaves of stevia, the most abundant are stevioside, rebaudiosides A, C, D, E and F, dulcoside A and steviolbioside [1]. In addition to steviol glycoside, stevia is also a good source of phenolic compounds including many flavonoids and tannins [2]. Apart from its sweetener capacity, several studies demonstrated health beneficial activities of extracts and purified compounds from stevia, including anti–diabetic, antihypertensive [3] antioxidant [4], anti–inflammatory and anticancer [5] activities.

Some properties of stevia indicate that this plant might be also a valuable source of active ingredients for skin care products. Cosmetics containing stevia are already available on the South–American and Indian markets. In this study we aimed to evaluate the biological activity of aqueous, aqueous–ethanolic (1:1, v/v) and ethanolic extracts from the leaves of *Stevia rebaudiana* as cosmetic ingredients. We analyzed the total content of phenolics and flavonoids in these extracts as well as their antioxidant activity and compared with extracts from other plants, commonly used in cosmetic formulations: *Calendula officinalis*, *Viola tricolor*, *Betula pendula*. The quali–quantitative LC–MS analysis of phenolic compounds in the extracts was performed. We have also tested stevia extracts for their ability to absorb UV radiation, inhibit tyrosinase activity and influence the survival and proliferation of normal human keratinocytes *in vitro*.

Keywords: *Stevia rebaudiana*, cosmetics, phenolics, skin

References:

[1] Ceunen S. *et al.* J Nat prod 2013; 28;76(6):1201–28.
[2] Lemus–Mondaca R. *et al.* Food Chem. 2012;132(3):1121–1132.
[3] Carrera–Lanestosa A. *et al.* J Med Food. 2017;20(10):933–943.
[4] Gaweł–Bęben K. *et al.* Molecules. 2015; 20(4):5468–86.
[5] Ruiz–Ruiz JC. *et al.* Crit Rev Food Sci Nutr 2017;57(12):2680–2690.

PS2-B-096

Magnetic nano–formulations for drug delivery applications of natural polyphenol–metal complexes against cancer

Eleftherios Halevas[1,2], Maria Pelecanou[2], Marina Sagnou[2], Barbara Mavroidi[2], Anastasia Pantazaki[3], Antonios Hatzidimitriou[3], George Katsipis[3], Theodore Lialiaris[4], Dimitrios Chronopoulos[4], Athanasios Salifoglou[5], George Litsardakis[1]

[1]Department of Electrical & Computer Engineering, Aristotle University, Thessaloniki, Greece, [2]Institute of Biosciences & Applications, National Centre for Scientific Research "Demokritos", Athens, Greece, [3]Department of Chemistry, Aristotle University, Thessaloniki, Greece, [4]Department of Medicine, Democritus University of Thrace, Alexandroupolis, Greece, [5]Department of Chemical Engineering, Aristotle University, Thessaloniki, Greece

Flavonoids and curcuminoids constitute well–studied classes of natural polyphenols with excellent antioxidant, antitumor, anti–inflammatory, anti–acidogenic, neuro–, cardio–, and radio–protective properties [1–5]. Despite, however, their health benefits, their therapeutic efficacy relies on the improvement of the pharmacokinetic profile, since their limited water solubility, bioavailability, and permeability render them unstable and sensitive to exogenous factors such as light, pH, and temperature [6–8]. To that end, metal ion complexation and/or encapsulation in nanocarriers, suitable for drug delivery applications, improves their bio–utilization by increasing solubilization, thereby altering the absorption pathways and preventing metabolic degradation. Magnetic liposomes and dendrimers constitute advanced targeted drug delivery systems, which due to their diversified properties promote activity commensurate to the desired therapeutic efficacy. Herein, novel hybrid natural polyphenol–metal complexes were synthesized, and encapsulated into the magnetic nanocarriers. Physicochemical, structural and magnetic characterization of the synthetic compounds and their magnetic carriers was performed by several techniques. In addition, the DNA–binding and anticancer potential of the novel complexes, free and encapsulated, were also investigated. The novel hybrid materials exhibit enhanced solubility and bioavailability, low toxicity, and significant anti–cancer and DNA–binding activity, advantageously associated with magnetic targeting.

Keywords: Flavonoids, curcumin, magnetic carriers, liposomes, dendrimers, drug delivery

References:

[1] Jayaprakasha GK *et al.* Food Chem 2006; 98(4): 720–724.
[2] Amalraj A *et al.* J. Tradit Complement Med 2017, 7(2), 205–233.
[3] Ross JA *et al.* Annu Rev Nutr 2002; 22: 19–34.
[4] Yao LH *et al.* Plant Foods Hum Nutr 2004; 59(3): 113–122.
[5] Heim KE *et al.* J Nutr Biochem 2002; 13(10): 572–584.
[6] Manach C *et al.* Am J Clin Nutr 2005; 81: 230–242.
[7] Stahl W *et al.* Mol Aspects Med 2002; 23(1–3), 39–100.
[8] Tsolaki MN *et al.* In Bentham eBooks series: Front Anti–Infective Drug Disc 2017, 6, 1–153.

PS2-B-097

Valorization of citrus juicing industries by–products

Eleni Myrtsi, Epameinondas Evergetis, Grigor Kheiranov, Sofia Koulocheri, Serkos Haroutounian

Agricultural University of Athens, Athens, Greece

Citrus juicing industries generate considerably large amounts of residues, since only a 50% of the fresh fruit's mass is transformed into juice [1]. These by–products, consisted of peel, pulp and seeds are of limited economic interest, although their content in bioactive natural products is indicative of their potential for utilization as biorefinery raw materials. Among the numerous by–products obtained during the juice–making process, only the Cold Pressed Essential Oils (CPEOs) currently have found wide applications by food, beverage, cosmetic and pharmaceutical industries, mainly as flavouring and fragrance agents due to their characteristic aroma profile [2]. Citrus juicing CPEOs consist of two fractions, a major volatile and a minor non–volatile, both containing a sum of more than 200 compounds [3]. The volatile fraction, consisting mainly by D–limonene, is well studied as compared to the minor, non–volatile residue, which is composed by hydrocarbons, sterols, fatty acids, waxes, carotenoids, coumarins, psoralens and flavonoids [4].
Endeavour herein refers to the exploitation of the non–volatile fractions as a rich source of carotenoids and their utilization as pigments and/or bioactive extracts. Specifically, the CPEOs of four different *Citrus* species –namely orange (*C. sinensis*), lemon (*C. limon*), grapefruit (*C. paradisii*) and mandarin (*C. reticulate*)– were investigated in order to determine their efficacy as carotenoids source under various conditions (technique, temperature, pressure). Results concerning the respective yields, the carotenoids content of these extracts (determined with LC/MS–MS) and the bioactivities evaluation data will be presented in detail.

Acknowledgements: We acknowledge support of this work by the project "Research Infrastructure for Waste Valorization and Sustainable Management of Resources, INVALOR" (MIS 5002495) which is implemented under the Action "Reinforcement of the Research and Innovation Infrastructure", funded by the Operational Programme "Competitiveness, Entrepreneurship and Innovation" (NSRF 2014–2020) and co–financed by Greece and the European Union (European Regional Development Fund).

PS2-B-098

Exploitation of plant derived essential oils as potent means for the control of *Sitophilus oryzae*

Apostolia Tzimopoulou[1], Dimitrios Papachristos[2], Antonios Michaelakis[2], Epameinondas Evergetis[1], Serkos Haroutounian[1]

[1]Agricultural University of Athens, Athens, Greece, [2]Benaki Phytopathological Institute, Athens, Greece

Insects constitute a major threat for agricultural products and commodities. The quality of these products is degraded either through insects feeding and/or contamination by proteins found in insects' bodies, eggs, feces and secretions, all accused to initiate various allergic reactions in humans [1,2]. Among 1,660 insects identified to affect the agricultural products and commodities, *Sitophilus oryzae* L. (Coleoptera: Curculionidae) –known as the rice weevil– has found to attack various stored crops such as wheat, rice and maize. The annual loss of these products, occurred during the storage, processing and transport, accounts the 10% of their global production, emerging the necessity of developing potent means for the efficient control of this insect.

To date, several synthetic insecticides and fumigants are used for the control of *S. oryzae*, all displaying the serious disadvantage of resistance development and affording harmful residues that contaminate foods and environment [3]. Recently, the utilization of natural products preparations for the control of the stored–product insects, provided promising alternatives to currently used traditional synthetic pesticides [4,5]. In this context, we were isolated a broad variety of Essential Oils (EOs) from Greek biodiversity and evaluated their insecticide properties against *S. oryzae*. Respective results highlighted as more potent those retrieved from *Foeniculum vulgare* and *Ruta graveolens* species, both displaying 70–80% insecticide ability for concentrations ranging from 1.3 to 1.5 μM, indicative of their strong capability to act as potent natural biocides. Data concerning the chemical compositions and the insecticide properties of all EOs investigated will be presented in detail.

Keywords: Essential Oil, insecticide, *Foeniculum vulgare, Ruta graveolens, Sitophilus oryzae*

PS2-B-099

Smell the Roses: Rosaceae family plants extracts as potent natural antiviral agents against influenza

Anna Apostolou[1], Olympia Vangelatou[1], Sandra Liekens[2], Serkos Haroutounian[1]

[1]Agricultural University of Athens, Athens, Greece, [2]Rega Institute for Medical Research, Leuven, Belgium

Influenza viruses are vastly infectious. They attack the respiratory system of living organisms to initiate an evolving process leading to influenza illnesses that bounce among various species often triggering epidemics (or pandemics) in humans [1]. Influenza is highly transmittable and zoonotic, inducing symptoms in various systems and displaying high potency for rapid crossing the international borders [2]. During the last 150 years, various strains of influenza virus (eg Spanish flu, Asian flu, Hong Kong flu, Bird flu, and Swine flu) were responsible for high morbidity and mortality for both human and animals [3]. Annually, flu is responsible for approximately 226,000 hospitalizations costing more than $5 billion, while in case of influenza epidemic the related costs surpass $12 billion along with millions of lost work hours [4].
Although there are several antiviral agents, such as oseltamivir and zanamivir, approved for the treatment of influenza's illnesses, there is an ongoing research interest for the development of plant origin antiviral agents. In this respect, during the 1981-2010 period, the 80% of 46 entities registered as antiviral agents are synthetic derivatives or analogues of botanical natural products [5]. Work herein refers to the investigation of extracts of twenty-three plants of Greek biodiversity belonging to Rosaceae family. The antiviral efficacy of the extracts was evaluated against four common viruses known affect the respiratory system (H1N1, H3N2, influenza B and Human Parainfluenza Virus-HPIV). The respective results highlighted the methanolic extracts of sixteen plants as very active, displaying IC_{50} values ranging from 0.8 to 4 μg/mL, activity comparable to those of antiviral agents currently in use. Data concerning the chemical compositions and the antiviral properties of all extracts investigated will be presented in detail.

PS2-B-100

Exploitation of Rosaceae Family plants extracts as potent natural antiviral agents

Anna Apostolou[1], Sandra Liekens[2], Sofia Koulocheri[1], Serkos Haroutounian[1]

[1]Agricultural University of Athens, Athens, Greece, [2]Rega Institute for Medicinal Chemistry, Leuven, Belgium.

Viruses constitute a serious menace for all organisms. They invade in living cells of blood, respiratory system, liver etc., causing a broad variety of infectious diseases ranging from common flu to very severe illnesses such as HIV/AIDS and Ebola. Thus, there is a global emerge towards the discovery–development of potent antiviral agents. Among various drug discovery endeavors, the screening of natural products is of particular interest, since their enormous structural diversity provides high screening hit rates against diverse panels of viruses allowing the discovery of novel structural leads.
Work herein refer to the antiviral potency exploitation of twenty–three extracts, sampled from Rosaceae family plants of Greek biodiversity. This botanical family plants are known for long–time as active ingredients of diverse ethnic preparations because of their established therapeutic properties [1] against several diseases–disorders such as diarrhea, diabetes mellitus, cancer, microbial and viral infections [2]. In this context, the antiviral efficacies of the extracts were evaluated against a broad panel of viruses, namely *Herpes Simplex Virus* (type-1KOS, type–1TK KOS ACV, type-2 G), *Vaccinia Virus*, *Vesicular Stomatitis Virus*, *Adeno Virus–2*, *Reovirus–1, Sinbis Virus, Coxsackie Virus B4, Punta Toro Virus*, indicating that the extracts of *Pyracantha coccinea* and *Crataegus orientalis* exhibit significant activities against the investigated strains of *Herpes Simplex Virus*, with IC_{50} values of 4 μg/mL. Data concerning the chemical compositions and the antiviral properties of all extracts investigated will be presented in detail.

Keywords: Rosaceae family, plant extracts, antiviral activity

References:

[1] Tomczyk M. Latte KP.J Ethnopharmacol 2009; 122: 1840294.
[2] Hao D, Gu XJ, Xiao Gen XP. Chem Bio Omics 2015.

PS2-B-101

PlantUp – Upgrading the plant capital | Aristotle University of Thessaloniki (AUTh) Core Node

Andreana Assimopoulou[1], Vassilios Papageorgiou[1], Stella Kokkini[2], Maria Tsimidou[3], Assoc. Diamanto Lazari[4], Dimitrios Vlachos[5], Dimitrios Christofilos[6]

[1]Organic Chemistry Laboratory, School of Chemical Engineering, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [2]Laboratory of Systematic Botany and Phytogeography, School of Biology, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [3]Laboratory of Food Chemistry and Technology, School of Chemistry, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [4]Laboratory of Pharmacognosy, School of Pharmacy, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [5]Laboratory of Statistics and Quantitative Analysis Methods, Division of Industrial Management, Department of Mechanical Engineering, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece, [6]Laboratory of Physics, School of Chemical Engineering, Aristotle University of Thessaloniki, 54124, Thessaloniki, Greece

PlantUp is a Research Infrastructure (RI) that belongs to the National Roadmap elaborated by GSRT in 2014, in alignment with the objectives of the NSRTDI (2014–2020). It is implemented under the Action "Reinforcement of the Research and Innovation Infrastructure", funded by the Operational Programme "Competitiveness, Entrepreneurship and Innovation" and co–financed by Greece and the European Union (European Regional Development Fund). It comprises 5 core nodes and aspires to establish new roadmaps for securing biodiversity and crop production, focusing on systematic recording, preservation, protection and exploitation of the wealth and heritage of the Hellenic plant biodiversity, while assuring consumers' health and environmental protection.

AUTh team focuses on the systematic classification and identification of Hellenic plants, extraction and isolation of secondary metabolites of pharmaceutical, cosmeceutical, or nutritional interest. It consists of a multidisciplinary group of scientists from different areas: quality control of raw materials and final products, standardization of Natural Product (NP) extracts and final products, "green" technologies, development of high added–value products from NPs, process scale–up, feasibility and sustainability studies and supply chain management studies.

AUTh core node aims to establish an ESFRI–relation between PlantUp and two pan–European RIs approved, boosting the role of the Hellenic hub: (i) Prof. M. Tsimidou and Assoc. Prof. A. Assimopoulou participate in "METROFOOD" and (ii) Dr. D. Koureas is head of the coordination team in "DiSSCo".

The goal of PlantUp RI is to provide a holistic approach for the sustainable valorization of the Hellenic plant biodiversity via the creation of high–added value products.

PlantUp AUTh partners

Name	Position	School/Department
A. Assimopoulou	Assoc. Professor	Chemical Engineering
V. Papageorgiou	Emeritus Professor	Chemical Engineering
D. Christofilos	Assoc. Professor	Chemical Engineering
K. Kontogiannopoulos	Post-Doc	Chemical Engineering
E. Kampasakali	Post-Doc	Chemical Engineering
A. Nakas	PhD Candidate	Chemical Engineering
A. Koletti	PhD Candidate	Chemical Engineering
S. Kokkini	Professor	Biology
D. Koureas	Post-Doc	Biology
D. Mertzanidis	PhD Candidate	Biology
M. Tsimidou	Professor	Chemistry
M. Papapostolou	PhD Candidate	Chemistry
D. Lazari	Assoc. Professor	Pharmacy
O. Tsiftsoglou	Post-Doc	Pharmacy
D. Vlachos	Professor	Mechanical Engineering

PlantUp AUTh Workflow

Acknowledgements: This research has been supported by the project "Upgrading the Plant Capital (PlantUp)" (MIS 5002803) which is implemented under the Action "Reinforcement of the Research and Innovation Infrastructure", funded by the Operational Programme "Competitiveness, Entrepreneurship and Innovation" (NSRF 2014–2020) and co–financed by Greece and the European Union (European Regional Development Fund).

PS2-B-102

The roles of triterpenes and phytosterols in Talang Mamaks' traditional medicines of Indonesia

Hilwan Yuda Teruna[1], Adel Zamri[1], Jasril Karim[1], Titania T. Nugroho[1], Yum Eryanti[1], Muhamad Almurdani[1], Kamal Rullah[2]

[1]Department of Chemistry, Faculty Mathemathics and Natural Sciences, Universitas Riau, Pekanbaru 28293, Indonesia, [2]Department of Pharmacy, STIFAR Riau, Pekanbaru 28293, Indonesia, [3]Nanotechnology & Catalyst Research Centre, Institute of Postgraduate Studies, University of Malaya, 50603 Kuala Lumpur, Malaysia

Traditional usages and secondary metabolites from ten medicinal plants of Talang Mamak tribe have been examined. The tribe is one of indigenous people left in Sumatra, Indonesia. They still use plants as a major source of medicines. The ten medicinal plants, *Flacourtia rukam, Lithocarpus bancanus, Helminthostachys zeylanica, Dyera costulata, Tabernaemontana macrocarpa, Macaranga bancana, Macaranga gigantea, Anisophyllea disticha, Coleus scutellaroides* and *Litsea elliptica* showed significant quantities of triterpenes and/or phytosterols. The most frequent usage of the plant was as an antiinflamatory medicine. The isolated triterpenes and phytosterol were *β*–sitosterol, stigmasterol, daucosterol, *β*–amyrin, *β*–amyrin acetate. These compounds were proven to possess antiinflammatory agents or as adjuvants to other secondary metabolites contained in the plants. The usages of the plants and their secondary metabolites will be discussed further.

PS2-B-103

Antifungal alkaloids from the aerial parts of *Glaucium corniculatum* subsp. *refractum* against *Candida albicans*

Mohsen Bagheri[1,2], Samad Nejad Ebrahimi[1], Ali Nazarirad[1], Atousa Aliahmad[1], Emerson F. Queiroz[2], Pierre–Marie Allard[2], Laurence Marcourt[2], Abdulelah Alfattani[2], Jean–Luc Wolfender[2]

[1] Medicinal Plants and Drugs Research Institute, Shahid Beheshti University, G.C., Evin, Tehran, Iran,
[2]School of Pharmaceutical Sciences, University of Geneva, University of Lausanne, CMU– Rue Michel Servet 1, 1211 Geneva 11, Switzerland

Introduction

Emerge of resistance of *Candida* species to current antifungal drugs including itraconazole (ITC) as well as the appearance of adverse consequence of certain anti–fungal agents are universal crisis. Therefore, it is crucial to investigate for more promising and less–toxic ideal novel antifungal agents that would overcome these drawbacks. Plant–derived natural products or their derivatives may offer potential leads which could act on these pathogenic fungi. Accordingly, in this study we focused on discovery of new antifungal compounds from Iranian plants.

Methods

Screening was achieved by using the dot–blot direct bioautography method, which is very convenient and effective. In preliminary screening experiment, TLC Bioautography, the most active was a methanolic extract from aerial parts of plant at 100 µg/spot. The fractionated active extract showed significant inhibition against *C. albicans* at 25 µg/spot. The activity of extract was localized by different chromatography chromatography techniques and TLC bioautography assay.

Results

Around 68 extracts from Papaveraceae, Lamiaceae, Boraginaceae, Apiaceae and Asteraceae screened against *C. albicans* ATCC10231. The methanolic extract of *Glaucium corniculatum* showed high activity. The large–scale isolation and purification of active compounds in alkaloid rich fraction by combination revers and normal phase chromatography resulted on identification of several alkaloids. Their structures were established by using different spectroscopic methods, including 1D (^{1}H NMR) and 2D–NMR (COSY, HSQC and HMBC) and HRMS.

Keywords: *Candida albicans,* TLC Bioautography, *Glaucium corniculatum*

PS2-B-104

The potential of macroalgae to be used as a functional ingredient in meat products

Ana Rita Circuncisão[1], Catarina Marçal[1], Carla Monteiro[2], Artur Silva[1], Susana M. Cardoso[1]

[1]QOPNA & Department of Chemistry, University of Aveiro, Aveiro, Portugal, [2]Irmãos Monteiro, S.A., Aveiro, Portugal

Meat products are known as a rich source of valuable nutrients, although they are also associated with the prevalence of some diseases (1). Hence, the development of meat–based functional products through the incorporation of heathier ingredients such as macroalgae may improve their health benefits beyond the basic nutritional value (2,3). The present work studied the impact of the use of macroalgae as an ingredient in meat sausages, aiming to evaluate their nutritional composition and phytochemical contents.

Two fortified sausages (S1 and S2), formulated with distinct percentages of *Ulva rigida, Fucus vesiculosus* and *Palmaria palmata* were analysed regarding their contents of protein, fat, ash, fiber, carbohydrates and fatty acids profile, and compared to control ones (C) (4,5). Moreover, levels of pigments and phenolic compounds, as well as their antioxidant ability were measured.

The major effect caused by the fortification of sausages with macroalgae was observed for their ash levels (7.0–7.4% DW), which were higher than the control (5.6% DW). This tendency was also observed for their mineral profile, particularly in I, K, Mg and Fe. Further, lower Na/K ratio was found in the fortified sausages, while minor impacts were observed for the remaining nutrients.

Besides, although no significant differences were observed for the phenolic contents and antioxidant activity of the fortified sausages with respect to the controls, the latter showed superior pigment levels, which included chlorophyll a, b, pheophytin a and lutein, fucoxanthin, probably due to the pigment composition of incorporated macroalgae. The bioaccessibility and bioavailability of the incremented compounds are presently being evaluated, either in raw and cooked sausages.

Acknowledgements: The authors acknowledge to SHARP project (POCI–01–0247–FEDER–003419), co–financed by FEDER through COMPETE2020, under the PT2020 program. Thanks are also due to FCT/MEC for the financial support to the research unit QOPNA (FCT UID/QUI/00062/2013), through national founds and where applicable co–financed by FEDER, within PT2020 Partnership Agreement. Susana Cardoso thanks the research contract under the project AgroForWealth (CENTRO–01–0145–FEDER–000001), funded by Centro2020, through FEDER and PT2020.

References:

[1] Cofrades S, Benedí J, Garcimartin A, Sánchez–Muniz FJ, Jimenez–Colmenero F. Food Research International. 2017;99:1084–1094.
[2] Pádua M, Cavaco AM, Aubert S, Bligny R, Casimiro A. Physiologia Plantarum. 2010;138:301–311.
[3] Gupta S, Abu–Ghannam N. Innovative Food Science & Emerging Technologies 2011;12:600–609.
[4] AOAC. Official Methods of Analysis. 17th ed. Maryland, USA: Association of Official Analytical Chemistry; 2000.
[5] O'Fallon JV, Busboom JR, Nelson ML, Gaskins CT. J Animal Science. 2007; 85: 1511–1521.

PS2-B-105

Impact of *Fucus vesiculosus* aqueous extracts as ingredients in a vegetarian "alheira"

Catarina Marçal[1], Ana Rita Circuncisão[1], Carla Monteiro[2], Artur Silva[1], Susana Cardoso[1]

[1]QOPNA & Department of Chemistry, University of Aveiro, Aveiro, Portugal, [2]Irmãos Monteiro, S.A., Aveiro, Portugal

Macroalgae are a rich and balanced source of bioactive nutrients and phytochemicals with different health–promoting benefits (1,2). Although their direct consumption is still very insipient in Europe, efforts have been made mainly by food industries to introduce macroalgae as a functional ingredient in several food products. Hence, the present work aimed to evaluate the potential of *Fucus vesiculosus* aqueous extracts as a food ingredient in vegetarian products. The aqueous extracts were obtained at room temperature (RT) and at 90ºC (HT), and further characterized for their nutritional composition (3), as well as for antioxidant and ability of inhibition of key enzymes with impact on diabetes and obesity (α–amylase (4), α–glucosidase (5) and pancreatic lipase (6)). Both extracts exhibited high contents of ashes (28.8–29.4% DW) and of soluble fibers (17.2–24.5% DW), while sugars and proteins were less representative (7.5–13.8 e 6.6% DW, respectively). Moreover, the two extracts showed similar amounts of total phenolic compounds (1.5–1.7 EAG/100g extract) and exhibited close antioxidant abilities (0.6–0.8 EAA/100g extract and 2.4–2.7 EBHA/100g extract in DPPH and FRAP assays, respectively). Yet, the RT extract showed higher ability to inhibit α–glucosidase when compared to HT, revealing an IC50 of 0.54 and 2.31 mg/mL, respectively.The RT and HT *F. vesiculosus* extracts were used as ingredients in a vegetarian Portuguese bread sausage called "alheira" at 0.4% (m/m fresh weight). Then, the fortified products were evaluated in terms of sensorial analysis, colour, nutritional composition and levels of lipid peroxidation. The results showed that the new formulated vegetarian "alheira" products were in general well accepted by the panellists, while colour changes, namely a decrease of the L* parameter, was observed in comparison to the control. In opposition, the incorporation of *F. vesiculosus* extracts in the "alheira" had no impact at the chemical level of the vegetarian products.

Acknowledgements: The authors acknowledge to SHARP project (POCI–01–0247–FEDER–003419), co–financed by FEDER through COMPETE2020, under the PT2020 program. Thanks are also due to FCT/MEC for the financial support to the research unit QOPNA (FCT UID/QUI/00062/2013), through national founds and where applicable co–financed by FEDER, within PT2020 Partnership Agreement. Susana Cardoso thanks the research contract under the project AgroForWealth (CENTRO–01–0145–FEDER–000001), funded by Centro2020, through FEDER and PT2020

References:

[1] Lordan S, Ross RP, Stanton C. Marine Drugs 2011; 9: 1056–1100.
[2] Paiva L, Lima E, Patarra RF, Neto AI, Baptista J. Food Chemistry 2014;164:128–135.
[3] AOAC. Official Methods of Analysis. 17th ed. Maryland, USA: Association of Official Analytical Chemistry; 2000.
[4] Wickramaratne MN, Punchihewa JC, Wickramaratne DBM. BMC Complement Altern Med 2016; 16: 466.
[5] Sarkar D, Orwat J, Hurburt T, Woods F, Pitts JA, Shetty K. Scientia Horticulturae 2016; 212: 193–202.
[6] Panteghini M, Bonora R, Pagani F. Annals of Clinical Biochemistry 2001; 38: 365–370.

PS2-B-106

Nutritional and phytochemical content of hemp seeds (*Cannabis sativa* L.) from Finola cultivar grown in Greece

Maria Irakli, Eleni Tsaliki, Apostolos Kalivas, Eirini Sarrou

[1]Hellenic Agricultural Organization - Demeter - Institute Of Plant Breeding & Genetic Resources , Po Box 60411 Thermi Thessaloniki, Greece

Cannabis sativa L. has been an important source of food, fiber, oil and medicine as well as a psychoactive drug since ancient times. Nowadays, there is a growing interest of the commercial products made with edible hempseeds recognized as a legitimate source of medicaments, cosmeceuticals and nutraceuticals. The objective of this study was to investigate the seed nutritional, phytochemical composition and antioxidant activity of Finola hemp cultivar grown in Greece at three different locations. The protein, oil, ash and dietary fiber as well as fatty acid composition, tocopherols, pigments, total phenolics and flavonoids were quantified. Antioxidant capacities were estimated by DPPH, ABTS and FRAP assays. Greece-grown Finola seeds were rich in oil and protein concentrations with average values of 30% and 25%, respectively. The hemp seed oil is mainly composed of unsaturated fatty acids and the dominant fatty acids were linoleic acid and a-linolenic acid. γ-tocopherol was the main tocols isomer in hemp seeds followed by δ-tocopherol. Lutein followed by zeaxanthin were the predominant carotenoids, whereas α-chlorophyll was the major constituent in hemp seed extract. The effectiveness of ultrasonic extraction of phenolics and flavonoids from hemp seed cakes was also investigated using response surface methodology. Optimal extraction conditions were determined to be a solvent composition of 80% aqueous methanol, sample-to-solvent ratio of 1:100, ultrasonic time of 30 min and ultrasonic temperature of 65 °C. Under these conditions, 660 mg GAE/100 g of phenolics and 360 mg CE/100 g of flavonoids were obtained. The highest values for DPPH, ABTS and FRAP assays were also obtained. Cannabisin A, *N*-trans-caffeoyltyramine and cannabisin B were the major lignanamides identified in hemp seed extracts. Minor phenolics were naringenin, quercetin, neo-chlorogenic, protocatechuic, chlorogenic, caffeic and ferulic acids. This study suggests that the seed of Greece-grown hemp could be a functional component for balanced health product.

PS2-B-107

Variation of phenolic compounds in Greek oregano (*O. vulgare* L. subsp. *hirtum*) accessions

Eirini Sarrou, Paschalina Chatzopoulou, Maria Irakli

Hellenic Agricultural Organization – Demeter – Institute of Plant Breeding & Genetic Resources, Po Box 60411 Thermi Thessaloniki, Greece

The genus *Origanum* comprises 41 taxa and most of them have a very local distribution around Mediterranean. The economically important O. vulgare is the most widely distributed Origanum species. The subspecies *O. vulgare* L. subsp. *hirtum* (Link) Ietswaart, known as Greek Oregano, is rich in essential oil and main bioactive compounds and monoterpene phenols carvacrol (CAR) and thymol (THY). There is a growing interest nowadays on Oregano phytochemicals, other than the essential oils, which exhibit significant bioactivity. Several compounds have been reported in many Oregano species, while rosmarinic acid (RMA) is the most abundant. The phytochemical composition of plants varies even among the same species, depending on genetic factors, the environmental conditions at the growing sites, the developmental stage etc. Oregano is known for its genetic and phenotypic variability, and the detection of chemotypes rich in bioactive compounds is of major importance, for breeding purpose. In the present study we assess the chemo– diversity of cultivated populations of Greek oregano, in terms of the total phenolic content (TPC) and main phenolics identified in methanol extract as well as their antioxidant activity measured by ABTS radical scavenging activity. TPC ranged from 33.6 to 58.2 mg GAE/g dw presenting strong relationship with ABTS values. HPLC analysis showed that the presence of CAR, RMA and THY that contributed on average 42, 23 and 19% on the total identified phenolics, respectively. Specifically, RMA ranged from 9.0 to 38.7 mg/g, while CAR and THY changed between 17.1 to 56.8 mg/g and 2.2 to 26.1 mg/g in oregano accessions tested, respectively. Other compounds detected in significant amounts were eriodyctiol, naringenin, apigenin, apigenin–7–*O*–glucoside and luteolin. As a result, oregano chemotypes, rich in phenolic compounds, with high antioxidant potential, may be selected, for high added value oregano products.

PS2-B-108

Identification of piperine based P-gp inhibitors with decreased cross reactivity with CYP3A4

Safiulla Basha Syed[1], Hemant Arya[1], Hsing-Pang Hsieh[2], Mohane Coumar Selvaraj[1]

[1]Centre For Bioinformatics, School of Life Sciences, Pondicherry University, Pondicherry, India, [2]Institute of Biotechnology and Pharmaceutical Research, National Health Research Institutes, 35 Keyan Road, , Zhunan, Miaoli County, 350, Taiwan

P-glycoprotein (P-gp)/MDR-1 plays a major role in the development of multidrug resistance (MDR) by pumping the chemotherapeutic drugs out of the cancer cells and reducing their efficacy. Large numbers of P-gp inhibitors were tested in the clinical trials; unfortunately, none entered clinical use due to toxicity issues. Many of the P-gp inhibitors tested in the clinical trials have been reported to have cross-reactivity with CYP450 drug metabolizing enzymes, resulting in unpredictable pharmacokinetics and toxicity.

Previously, we reported two piperine based analogs Pip1 and Pip2 modulating the P-gp function. However, both analogs Pip1 and Pip2 inhibited CYP3A4 enzyme activity in in vitro more than that of piperine, suggesting that they may also interfere with chemotherapeutic drug metabolism. Therefore, in this study two more piperine analogs (Pip1-1 and Pip1-2) were designed based on the lead Pip1, *in silico* molecular docking and dynamic simulation studies were carried out. Later, the analogs were synthesized and *in vitro* tested in KBChR 8-5 and SW480-VCR MDR cell lines. The *in silico* results showed strong affinity of both Pip1-1 and Pip1-2 for P-gp. The vincristine resistance reversal activity for Pip1-1 and Pip1-2 at 16 µM concentration in KBChR 8-5 was found to be 42-fold and 59-fold and in SW480-VCR 8-fold and 13-fold, respectively. Both the analogs exerted less in vitro CYP3A4 inhibitory activity (54% and 50%, respectively) compared to Piperine (84%) and verapamil (72%). Moreover, the P-gp Glo assay results showed that both the analogs could act as competitive inhibitors of P-gp.

Acknowledgements: SBS thanks University Grants Commission (UGC) Government of India, for JRF fellowship (20/12/2015(ii)EU-V, Roll No. 316388) and DBT-IPLS for providing fellowship and the lab facility.

Keywords: P-glycoprotein, multidrug resistance, chemotherapy, piperine analogs and CYP3A4

PS2-B-109

Isolation and characterization of terpenoids from the root extracts of *Euphorbia grandicomita*

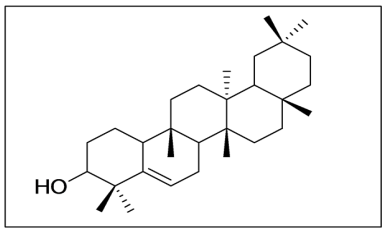
Douglas Magozwi[1], Douglas M. Kemboi[1], Jacqueline Tembu[1], Xolani Peter[2], Moses Langat[3]

[1]Department of Chemistry, Tshwane University of Technology, Private Bag X680, Pretoria, [2]Department of Biosciences, Council of Scientific Industrial Reserach (CSIR), Box 395, Pretoria, 001, [3]Department of Chemistry, FEPS, University of Surrey, Guildford, Surrey, GU27XH, UK

Plants of the Euphorbiaceae family have been the subject of many investigations because of their biologically active constituents that can be used as lead or scaffolds in drug discovery. As a result, an increasing attention has been paid to Euphorbia species because of their diverse terpenoids structural compounds such as oleanane, phorbol esters and taraxane, many of which possess pharmacological activities such as antiviral, antitumor and cytotoxic [1]. The research focused on isolation and characterization of secondary metabolites from *Euphorbia grandicomita*, a species which has not been adequately studied.

The dried, ground aerial and roots plant materials were successively extracted using dichloromethane and ethanol. The crude extracts (14.3g) were fractionated using column chromatography and the fractions were monitored using TLC. Repeated column chromatography of dichloromethane root extracts afforded 11mg, pentacyclic triterpenoid commonly known as *β*-glutinol (3-beta-hydroxy-5-glutinene) of oleanane subclass. The structure was elucidated and identified based on Mass Spectroscopy (GC-MS), Infrared Spectroscopy (IR), 1D and 2D Nuclear Magnetic Resonance (NMR) data, determination of melting point and by comparison with literature values.

The results give a scientific validity and credence to the ethnomedicinal use of this plant in folk medicine.



Acknowledgements: Supervisors, Nation Research Foundation (NRF-TWAS), Tshwane University of Technology

Keywords: triterpenoids, Euphorbiaceae, *Euphorbia grandicomita*.

References:

[1] Sulyok E, Vasas A, RedeI D, Forgo P, Kele Z, Pinke G, Hohmann J. Tetrahedron 2011; 67: 7289–7293.

PS2-B-110

Chemical characterization, biological assessment and molecular docking studies of essential oil of *Ocimum viride* for potential antimicrobial and anticancer activities

Monica Sangral, Madhulika Bhagat

University of Jammu, Jammu, India

Ocimum viride (family: Lamiaceae) is a medicinally important aromatic plant that grows widely in north western Himalayan range of Indian subcontinent. Essentials oils (EOs) and purified aromatic compounds derived from plants of genus *Ocimum* have long been used in traditional system of medicine to treat various chronic disorders. In this study we made an attempt to assess the chemical composition of essential oil (EO) obtained from *Ocimum viride* for potential antimicrobial and anticancer properties. Gas chromatography–Mass spectrometry (GC-MS) analysis revealed that EOs of aerial parts (leaves) of *Ocimum viride* contain high amounts of oxygenated monoterpenes, thymol and gamma terpinene. Notably, thymol (~50%) and *γ*-terpinene (~18%) were identified as the most abundant components of the oil. EOs showed most prominent antibacterial effect against *Bacillus subtilis* and *in silico* molecular docking analyses of antibacterial action against bacterial cell wall of Bacillus subtilis showed interaction of thymol with Sec A protein of *Bacillus subtilis* (binding energy of -15 kcal/mol) with active site Lys284, Trp275, Leu269, Arg19, Glu277, pro270. While, in vitro cytotoxic effect of EO against six human cancer cell lines showed maximum effect with IC_{50} value of ~0.034 ± 0.001µL/ mL against HT-29 colon cancer cell line. DNA fragmentation analysis and cell cycle analysis revealed that EO inhibits the growth of HT-29 colon cancer cells probably through induction of unrepairable DNA damage and subsequent cell death. Taken together, our results indicate that EO possesses potent antimicrobial and anticancer properties, and may find applications in bacterial growth inhibition and cancer therapeutics.

PS2-B-111

Effects of the *Thumbergia laurifolia* Lindl. extract on gastric ulceration in rats

Linda Chularojanamontri

Division of Pharmacology, Department of Preclinical Science, Faculty of Medicine, Thammasat University, Rungsit Campus, Thailand, Thailand

Background: From the previous study in animals, *Thumbergia laurifolia* has been previously reported to possess anti-inflammatory effect in mice. Its mechanism of action was shown might be similar to non-steroidal anti-inflammatory drugs – drugs that often, which commonly cause gastrointestinal side effects.
Objective: To study the anti-ulcerogenic activity of *T. laurifolia* extract in animals.
Materials and Methods: Male Sprague Dawley rats were used. Anti-ulcerogenic activity of *T. laurifolia* extract was tested by using four different *in vivo* models. These gastric-ulcer-inducing models induced including those the gastric ulcer was induced by HCl/Ethanol/hydrochloride administration OH, restraint water immersion stress, indomethacin administration and as well as pylorus ligation model.
Results: *T. laurifolia* extract at the doses of 100, 200 and 400 mg/kg was found to decrease the formation of gastric ulcers formation induced by EtOH/HCl administration /EtOH and restraint water immersion stress. Moreover, the extract at the doses of 200 and 400 mg/kg decreased the gastric ulcer formation induced by indomethacin. Regarding to the mechanism of anti-ulcerogenic action, the extract at the dose of 400 mg/kg decreased the gastric secretion rate and as evidenced by the reduction of the total acidity of gastric juice in the pylorus ligation model.
Conclusion: The findings of this study indicate that the *T. laurifolia* extract possesses anti-ulcerogenic activity in ratanimals.

Keywords: *Thumbergia laurifolia* extract, anti-ulcerogenic activity

PS2-B-112

Strategies for the sustainable production of novel high-value terpenoids

Norbert Mehlmer, Wolfgang Mischko, Thomas Brück

Werner Siemens-Chair of Synthetic Biotechnology, Department of Chemistry, Technical University of Munich, Garching bei München, Germany

Terpenoids are a large and diverse class of naturally occurring organic compounds found in a vast number of organisms. As secondary metabolites, they have evolved manifold functions including signaling, biotic and abiotic defense mechanisms. Especially biotic defense strategies make these compounds attractive, since vulnerable structures are conserved in biological systems and can be used for pharmaceutical targets. Even though the first steps in the synthesis are well characterized (MEP and DXP pathways), the following synthesis steps, including cyclisation and modification, are still poorly understood. For that reason, finding novel terpene syntheses and modifying enzymes are key issues. Since most bio-based insecticides are of plant origin, their production competes with agricultural food production. The sustainable, biomass-based production of insecticides has not yet been addressed. We have tackled this issue by primarily converting agricultural residues, such as wheat bran, to an efficient fermentation base medium. We have developed an enzyme system for the almost quantitative conversion (80% w/w) of milling residues to a high sugar containing fermentation medium. We have engineered an *E. coli* system harboring a synthetic DXP pathway to fermentatively generate an insecticidal, diterpenoid natural product. The diterpene Cyclooctatin from *Streptomyces melanosporofaciens* is a strong lysophospholipase inhibitor acting as anti-inflammatory agent. In a reverse genetics approach using structure guided simulation, we were able to establish an efficient whole-cell synthesis and oxidation system for the production of Cyclooctatin in *E. coli* yielding 50 times more than the original host organism. Using iterative process engineering in conjunction with continuous strain improvements, we aim to design a continuous process, which provides for economical, bio-based production of novel bioactive compounds.

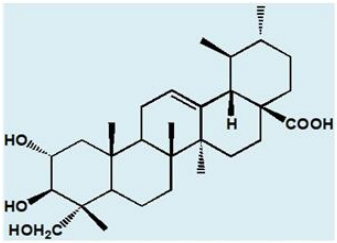
PS2-B-113

Phytochemical and pharmacological investigation on medicinal plants of different origin

Syed Imran Hassan[1], Majekodunmi Fatope[1], Sabira Begum[2], Essam Ads[3], Saravanan Rajendrasozhan[3]

[1]Department of Chemistry, Sultan Qaboos University, Muscat, Oman, [2]International Center for Biological and Chemical Sciences, University of Karachi, Karachi, Pakistan, [3]Department of Chemistry, University of Hail, Hail, Kingdom of Saudi Arabia

In virtue of the importance of medicinal plants we carried out investigations on three different medicinal plants *Psidium guajava, Pergularia tomentosa* and *Ziziphus spina christi*.
Psidium guajava (Myrtaceae) has been traditionally used in digestive disorderness, and considered to be spasmolytic. Its bark is used as an astringent, flowers are used to treat bronchitis whereas, fruits are laxative and good against bleeding gum [2,3]. Phytochemical investigation on alcoholic extract of leaves results in isolation of several new and known triterpenoids. Structures of these compounds were determined by extensive spectroscopic techniques. Pharmacological studies showed that the triterpenoid Asiatic acid (1) has dose dependent (10–500 mg/ml) spasmolytic activity (Figure 1). The ethanolic extract showed significant spasmolytic, antioxidant and antibacterial activities [4-9].
Pergularia tomentosa (Apocynaceae) is a perennial twining herb with much milky latex. Preparations from its root and shoot are used to treat skin diseases and other ailments. It is often classified as a toxic plant that causes spasm and gastroenteritis. *P. tomentosa* showed antitumor, molluscicidal, and hypoglycemic properties. Different cardenolides, taraxasterol-type triterpenes, and alkaloids have been isolated from the root and leaf which are responsible for the cytotoxic, antitumor, and antimicrobial properties. The studies on the ethyl acetate extract of this plant yielded ursane and lupane type of triterpenoids. The extract also inhibited *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Escherichia coli* at 250 µg /mL [10].
Ziziphus (Rhamaceae) is another genus which is traditionally used against various human ailments. Many species of zizphus showed sedative-hypnotic, antipyretic and analgesic properties. In Saudi Arabian folk medicine, the leaves of *Ziziphus spina christi* are used to heal wounds, treat skin diseases, some inflammatory conditions, sores, against ringworm, fever, gonorrhea, sex diseases and ulcers. The bark of the *Ziziphus spina christi* was collected from Al-Ha'il, Saudi Arabia, and its ethyl acetate extract showed prominent antimicrobial activity and cytotoxicity [11,12].



Dose (µg/ml)	Effect (% Relaxation)
10	13.1 ± 4.9
30	40.4 ± 0.5
100	53.8 ± 4.5
300	71.1 ± 3.3
500	84.5 ± 3.0

Fig. 1. Asiatic acid and its spasmolytic activity.
Values shown represent means ± SEM of 4 determinations, with EC_{50} = 80.0 ± 7.8 µg/ml

PS2-B-114

Tissue specific effects of sheep/goat whey protein on antioxidant enzymes in rats

Efthalia Kerasioti[1], Maria Gasdrogka[1], Ioannis Taitzoglou[2], Demetrios Kouretas[1]

[1]BIOCHEMISTRY-BIOTECHNOLOGY/ University of Thessaly/LARISSA, Greece, [2]Department of Veterinary, Aristotle University of Thessaloniki, Thessaloniki, Greece

Milk components have been recognized as functional foods with many health benefits [1]. Whey is the liquid waste of dairy industry and is also characterized as a functional food [2]. It is the liquid that remains during cheese manufacturing after casein removal. Greek whey is largely derived from sheep/goat milk compared to the whey from European countries where milk mainly derives from cows. In the present study, it was investigated the effect of sheep/goat whey protein administration on protein and gene expression of catalase (CAT), superoxide dismutase-1 (SOD-1) and glutamate cysteine ligase (GCL) by the method of Western blot and Real-time PCR, respectively, in rats' brain, small intestine, liver and lung. The findings of the present study have shown tissue-specific effects. In brain tissue, the protein and gene expression of GCL were increased while the protein and gene expression of SOD-1 were decreased. In small intestine, protein and gene expression of CAT and SOD-1 were decreased while protein and gene expression of GCL were increased. In liver tissue, protein and gene expression of CAT were decreased. In lung tissue, protein and gene expression of CAT and SOD-1 were decreased while protein and gene expression of GCL were increased. The determination of the molecular mechanism through which sheep/goat whey protein exerts its antioxidant action could lead to nutritional intervention strategies to prevent and assist in the treatment of oxidative stress-associated diseases.

Acknowledgments: The present study was supported by IKY Fellowships funded by the action "Enhancement of post-doctoral researchers'' from the resources of the European Program '' Development of Human resources, Education and Life-Long Learning'' co-funded by the European Social Fund.

Keywords: whey protein, protein expression, gene expression

References:

[1] Gill HS *et al*. Royal Society of Chemistry Press 2000; 248: 82–90.
[2] Marshall K. Altern Med Rev 2004; 9(2): 136–156.

PS2-B-115

Effects of feed supplemented with olive oil mill wastewater on detoxification enzymes in lamb tissues

Sotiria Makri, Sofia Raftopoulou, Demetrios Kouretas

University of Thessaly-Department of Biochemistry and Biotechnology-Larissa, Larissa, Greece

Olive Oil Mill Wastewater (OMW) is a by-product that is produced mainly from the water using during the various stages of oil production [1]. Its polyphenolic content exhibit important biological activities that can prevent animal welfare from oxidative stress associated diseases [2]. For this purpose, 24 lambs received breast milk until the postnatal period, then they were divided into two groups and received control and OMW feed for 55 days. Tissue samples were collected at 42 & 70 days post birth in order to determine antioxidant activity of GST, SOD enzymes and protein expression of γ-GCS. Results showed that GST activity was significantly increased in liver and spleen and decreased in heart at 42 experimental day and increased in heart at 70 day. Protein concentration of γ-GCS was decreased both at 42 and 70 days in liver, increased in heart only at 42 days and not affected in spleen. SOD activity was not affected in any tissue at sampling times. Feed supplemented with OMW improve lambs' redox status, which may improve their wellbeing and productivity, while the exploitation of these by-products may reduce the environmental pollution in areas with olive oil industries.

Acknowledgements: The present study was implemented by the State Scholarship Foundation funded by the «Scholarship Program for postgraduated studies of second cycle" from the resources of the Operational Program "Human Resources Development, Education and Lifelong Learning" 2014-2020 with co-funded from The European Social Fund (ESF) and the Greek State.

Keywords: lambs, olive oil wastewater, antioxidant enzymes

References:

[1] Makri S *et al*. In Vivo, 2018; 32(2): 291–302.
[2] Kerasioti E *et al*. Toxicol Reports, 2017; 4: 364–372.

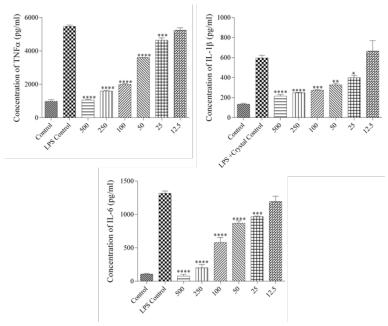
PS2-B-116

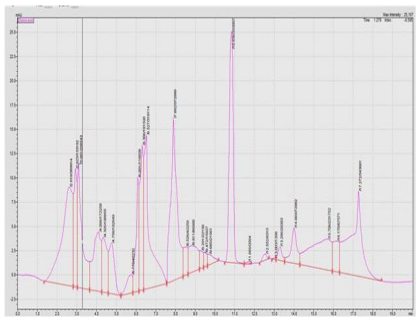
LC-MS fingerprinting and chemical analysis of TNF-α, IL-6 and IL-1β inhibitory culture broth extract of *Pseudomonas* species

Kirti Hira, Pragya P Pal, A Sajeli Begum

Department of Pharmacy, Bits-pilani, Hyderabad, India

Chronic Inflammation is a direct impact of the imbalance in the level of pro-inflammatory cytokines like TNF-α and Interleukins like IL-1β and IL-6. Serious degenerative disorders like renal injury, alzheimers, heart diseases, inflammatory bowel syndrome, atherosclerosis, etc., can occur due to sustained chronic inflammation. For the discovery and development of potential anti-inflammatory drugs, microorganisms are recognized as potential source.
Pseudomonas species ABS-36 strain used for the study was isolated from rhizospheric soil. This was cultivated in bulk at 30°C for 7 days using King's broth media. The broth was then extracted using ethyl acetate and the extract was evaporated and made free from solvent to obtain culture broth extract. ELISA studies were carried out to determine the pro-inflammatory cytokine inhibition efficacy of culture broth extract. Pretreated RAW 264.7 cell lines were stimulated using lipopolysaccharide (LPS) and TNF-α and IL-6 inhibitory potential was determined using ELISA kit. IL-1β inhibitory efficacy was determined by stimulating RAW 264.7 cells with LPS and crystals of calcium oxalate. The extract exhibited inhibition effect with IC50 values of 73.66, 95.49 and 132.09 µg/ ml against IL-6, TNF-α and IL-1β, respectively (Figure 1).
Chemical fingerprinting of culture broth extract was performed using LC-PDA-ESI-MS technique (Schimadzu LCMS- 8040) following gradient method. Mobile phases used acetonitrile and Milli Q Water having flow rate 0.3ml/min using Shim Pack, XR-ODS, 100 X 3mm (Table 1). It revealed the presence of compounds having MW ranging between 59-740 daltons (Figure 2). Further, the extract was subjected for column chromatography to isolate the major compounds, which yielded four compounds. The pure compounds were characterized as proline based cyclic dipeptides based on NMR spectroscopic analysis.





Time (min)	Mobile Phase (A%)	Mobile Phase (B%)	Profile
0.00-0.01	60	40	Isocratic
0.01-7.00	60→10	40→90	Linear ramp to 90%B
7.00-9.00	10	90	Isocratic
9.00-12.00	10→95	90→05	Linear ramp to 95%A
12.00-15.00	96→60	05→40	Linear ramp to 40%B
15.01	Stop		

Keywords: *Pseudomonas aeruginosa*, cyclic dipeptides, pro-inflammatory cytokines

PS2-B-117

Coffee improves rat redox status by increasing GSH biosynthesis

Alexandros Alexandros[1], Vasiliki Soursou[1], Anthi-Styliani Makiou[1], Fotis Tekos[1], Ioannis Taitzoglou[2], Dimitrios Kouretas[1]

[1]University of Thessaly, Larissa, Greece, [2]Aristotle University of Thessaloniki, Thessaloniki, Greece

Coffee is an extremely popular beverage throughout the world due to its stimulating effect. In addition, many beneficial health effects have been attributed to coffee, however these often come from observational studies. In the current study, a lightly roasted coffee extract from *Coffea arabica* beans was administered for two weeks in the water of rats to examine its effect on tissue and blood redox status, at a dose that corresponds to a moderate human daily consumption of 3-4 cups. According to the results, coffee had beneficial effects in all eleven tissues tested, mainly by increasing reduced glutathione (GSH) levels, an important endogenous antioxidant tripeptide. Interestingly, coffee bioactive compounds showed the ability to pass through the blood brain barrier, as the brain redox status was significantly improved. In addition, protein and lipid oxidation was reduced in several tissues in the coffee group. The observed increase in GSH was attributed to increased levels of the rate-limiting enzyme in its biosynthesis pathway, namely γ-glutamylcysteine ligase, as demonstrated by both Real-time PCR and Western blot analysis. Overall, moderate coffee consumption showed beneficial short term effects in all tissues by stimulating the endogenous antioxidant mechanisms.

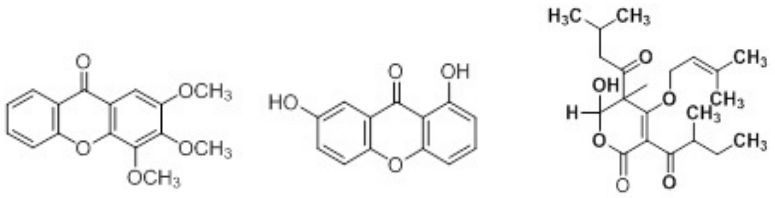
PS2-B-118

Vasodilating activity of *Hypericum revolutum* through nitric oxide (NO) synthase-dependent pathway

Noha Timraz[1], Hossam M. Abdallah[1,2], Hany A. El-Bassossy[3,4], Sabrin R. Ibrahim[5], Ali M. El-halawany[1,2], Ibrahim A. Shehata[1,2]

[1]Department of Natural Products, Faculty of Pharmacy, King Abdulaziz University, JEDDAH, Saudi Arabia, [2]Department of Pharmacognosy, Faculty of Pharmacy, Cairo University, Cairo 11562, Egypt, [3]Department of Pharmacology and Toxicology, Faculty of Pharmacy, King Abdulaziz University, Jeddah 21589, Saudi Arabia, [4]Department of Pharmacology, Faculty of Pharmacy, Zagazig University, Zagazig 44519, Egypt, [5]Department of Pharmacognosy and Pharmaceutical Chemistry, College of Pharmacy, Taibah University, Al Madinah Al Munawwarah 30078 , Saudi Arabia

Hypertension is a chronic disease which is considered one of the leading causes of many cardiovascular disorders. In spite of the significant developments in the antihypertensive therapy, serious side effects still exist. Drugs from herbal origin are prescribed widely in the treatment of different aliments because of their effectiveness, fewer side effects and relatively low cost. Here, the possible vasodilating activity of *Hypericum revolutum Vahl* (Guttiferae) was fully investigated using the isolated artery technique. Cumulative addition of *H. revolutum* total extract (1-10 µg/ml) led to a concentration-dependent relaxation of phenylephrine-precontracted aortae. Bio-guided fractionation revealed that the chloroform fraction is responsible for this effect. The observed vasodilation is endothelial nitric oxide (NO) synthase-dependent as it was blocked by endothelial denudation or *N*ω-nitro-L-arginine methyl ester hydrochloride (L-NAME), the NO synthase inhibitor. Chemical investigation of chloroform fraction of *H. revolutum* gave rise to, one new dihydropyranone derivative (3) in addition to two known xanthones namely, 2,3,4-trimethoxy xanthone (1) and 1,7-dihydroxy xanthone (2). The isolated compounds (at concentrations 1, 3, and 10 µM) showed vasodilating activity; where compound 2 showed the highest potency. Endothelial-denudation or L-NAME blocked the vasodilating activity of the two compounds as well. In conclusion, *H. revolutum* produced two vasodilating compounds that act through the endothelial nitric oxide (NO) synthase-dependent pathway.



1 2 3

Acknowledgements: This project was funded by King Abdulaziz City for Science and Technology number (No. P-S 1928-37).

Keywords: *Hypericum revolutum*, nitric oxide, xanthones, vasodilatation

PS2-B-119

Expanding the scope of biosynthetic cooperativity between polyketide megasynthases through genome mining

Michio Sato[1], Yi Tang[2], Kenji Watanabe[1]

[1]University of Shizuoka, Shizuoka, Japan, [2]University of California, Los Angeles, Los Angeles, USA

Fungal polyketides comprise a structurally complex and diverse family of natural products, and show a wide range of biological activities. Much of the structural complexity is synthesized by the iterative polyketide synthases (IPKS) that generate the polyketide scaffold. Standalone IPKSs are capable of assembling entire polyketides, such as aromatic compounds by nonreducing PKSs (NRPKS), or more reduced products by highly reducing PKSs (HRPKSs). Nature can generate significantly more structural diversity by cooperatively pairing IPKSs of different functions in the same biosynthetic pathway. As can be seen in the biosynthesis of resorcylic acid lactones (RALs) in fungi, the cooperatively assemble of sequential iterative PKSs makes RALs structure more complicated. The cooperative IPKSs are frequently colocalized in the same biosynthetic gene cluster in the fungal producer, a genetic feature that can be utilized in the genome mining of new natural products.

Through BLAST search, we found a gene cluster in which PKS and PKS-nonribosomal peptide synthetase (NRPS) hybrid enzyme are tandemly arranged in *Penicillium oxalicum*. To date, all PKS-NRPSs are found to function alone and no biosynthetic cooperativity has been observed. Since this gene cluster was transcriptionally silent, we overexpressed a Gal4-like transcription factor to activate the cluster in *P. oxalicum*.

Metabolic analysis of the activated strain using LC/MS revealed the production of twelve new compounds, oxaleimides which contain maleimide or succinimide moiety, and the chemical structures of the compounds were determined by spectroscopic analyses. Gene disruption and stable isotope incorporation studies provided the biosynthetic pathway of these compounds.

We demonstrate here the formation of the maleimide moiety requires the collaborative function of two megasynthetases. Such collaboration expands the biosynthetic capabilities of fungal PKSs as well the chemical space occupied by polyketide natural products.

PS2-B-120

The mechanism of action of zingerone in the pacemaker potentials of interstitial cells of Cajal isolated from murine small intestine

Jeong Nam Kim[1], Hyun Jung Kim[1], Iksung Kim[1], Yun Tai Kim[2], Byung Joo Kim[1]

[1]Pusan National University School of Korean Medicine, South Korea, [2]Korea Food Research Institute, Research group of innovative special food, South Korea

Background/Aims: Zingerone, a major component found in ginger root, is clinically effective for the treatment of various diseases. Interstitial cells of Cajal (ICCs) are the pacemaker cells responsible for slow waves in the gastrointestinal (GI) tract. We investigated the effects of zingerone on the pacemaker potentials of ICCs to assess its mechanisms of action and its potential as a treatment for GI tract motility disorder.
Methods: We isolated ICCs from small intestines, and the whole-cell patch-clamp configuration was used to record the pacemaker potentials in cultured ICCs.
Results: Under the current clamping mode, zingerone inhibited pacemaker potentials of ICCs concentration-dependently. These effects were blocked not by capsazepine, a transient receptor potential vanilloid 1 (TRPV1) channel blocker, but by glibenclamide, a specific ATP-sensitive K^+ channel blocker. Pretreatment with SQ-22536 (an adenylate cyclase inhibitor), LY294002 (a phosphoinositide 3-kinase inhibitor), and calphostin C (a protein kinase C (PKC) inhibitor) did not block the effects of zingerone on the pacemaker potentials relative to treatment with zingerone alone. However, zingerone-induced pacemaker potential inhibition was blocked by 1*H* [1,2,4] oxadiazolo [4,3 *a*] quinoxalin 1 one (ODQ; a guanylate cyclase inhibitor), KT5823 (*a* protein kinase G (PKG) inhibitor), and L-NAME (a non-selective nitric oxide synthase (NOS) inhibitor). In addition, zingerone stimulated cyclic guanosine monophosphate (cGMP) production in ICCs. Finally, pretreatment with PD98059 (a p42/44 mitogen-activated protein kinase (MAPK) inhibitor), SB203580 (a p38 MAPK inhibitor), and SP600125 (c Jun N terminal kinases (JNK) specific inhibitor) blocked the zingerone-induced pacemaker potential inhibition.
Conclusion: These results suggest that zingerone concentration-dependently inhibits pacemaker potentials of ICCs via NO/cGMP-dependent ATP-sensitive K^+ channels through MAPK-dependent pathways. Taken together, this study shows that zingerone may have the potential for development as a GI regulation agent.

PS2-B-121

On the relevance of DPPH, ABTS and FRAP assays for the evaluation of antioxidant properties of natural extracts

Radu Claudiu Fierascu[1,2], Alina Ortan[1], Irina Fierascu[1,2], Cristina Elena Dinu-Pirvu[1,3], Ioana Catalina Fierascu[3,4]

[1]University of Agronomic Sciences and Veterinary Medicine of Bucharest, Bucharest, Romania, [2]National Institute for Research & Development in Chemistry and Petrochemistry – ICECHIM Bucharest, Bucharest, Romania, [3]University of Medicine and Pharmacy "Carol Davila", Bucharest, Romania, [4]Hofigal Export Import S.A., Bucharest, Romania

The consumer appetite for natural, "clean label" products shifted the attention of both scientific community and industry towards the development of food products and supplements with a very consistent natural component. Application of natural extracts and essential oils opens the possibility of developing products with longer shelf-life and multiple health benefits. One of the most studied property of natural extracts is represented by their antioxidant potential. Among the assays currently performed for the evaluation of antioxidant activity, the assays involving scavenging of free radicals, such as 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2′-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid (ABTS) and the ferric reducing/antioxidant power (FRAP) assays are the most encountered in the literature data [1, 2]. In spite of their wide use, the selected antioxidant assays present several shortcomings, the main concern being related to their relevance for living systems, as both DPPH and ABTS radicals are not naturally occurring in living organisms, while the FRAP assay is performed at "unnatural" pH levels [3]. The present study appeared due to the need to use the antioxidant assays in good knowledge regarding their application and relevance for living systems. The paper reviews the scientific literature published in the last decade, presenting the mechanisms behind each antioxidant assay and the relevance of the obtained results, by comparison with the results obtained using cell lines or *in vivo* assays. Although the shortcomings of those assays should not be overlooked, their application as a primary evaluation tool is often encouraged, due to the simple equipment and procedure employed.

Acknowledgements: The authors gratefully acknowledge the support obtained through the project SusMAPWaste, SMIS 104323, Contract No. 89/09.09.2016, from the Operational Program Competitiveness 2014-2020, project co-financed from the European Regional Development Fund.

PS2-B-122

Application of gamma radiation for the development of "green" products

Irina Fierascu[1], Radu Claudiu Fierascu[1], Eduard Marius Lungulescu[2], Nicoleta Nicula[2], Raluca Somoghi[1], Sorin Marius Avramescu[3], Camelia Ungureanu[4], Anca Nicoleta Sutan[5], Liliana Cristina Soare[5]

[1]National Institute For Research & Development in Chemistry and Petrochemistry – Icechim Bucharest, 202 Spl. Independentei, Bucharest, Romania, [2]National Institute for R&D in Electrical Engineering ICPE – CA, 313 Spl. Unirii, Bucharest, Romania, [3]Research Center for Environmental Protection and Waste Management, University of Bucharest, Bucharest, Romania, [4]Faculty of Applied Chemistry and Material Science, University Politehnica of Bucharest, Bucharest, Romania, [5]Department of Natural Sciences, University of Pitesti, Pitesti, Romania

The development of natural, safer and cleaner products represents one of the main goals of green chemistry. The present paper discusses the application of gamma radiation for the development of "green" products, focusing on two main directions: synthesis of silver nanoparticles - NP (especially on the differences in terms of biological activities between radiochemical synthesized nanoparticles and those phytosynthesized using hydroalcoholic extracts obtained from *Asplenium scolopendrium* L. leaves) and the effect of irradiation on the biological properties of *A. scolopendrium* extracts. The phytosynthesis of silver NP was previously demonstrated by our group using alcoholic extracts obtained from leaves and rhizomes of *A. scolopendrium* [1]. Radiochemical treatment (synthesis of nanoparticles and irradiation of natural extracts) was achieved by exposure to gamma radiation in air at room temperature at different irradiation doses. The irradiation dose rate was about 1.1 kGy/h. The obtained NPs were characterized using XRD, TEM and DLS [2], while the extract composition was evaluated using HPLC [3]. The evaluated biological properties are: the antioxidant potential, antimicrobial properties (against bacteria, yeast and mold - lines and isolates) and cytogenetic effects (using the *Allium cepa* assay). The obtained NPs had dimensions between 2 and 40 nm (the diameter being influenced by the irradiation dose for radiochemical synthesized NP and by the extract used in the case of phytosynthesized NPs), possessing enhanced antioxidant and antimicrobial properties, as well as a mitoinhibitory effect. The irradiation of the natural extracts revealed a very interesting dose-related effect on the evaluated biological properties.

Acknowledgements: This work was supported by a grant of the Romanian National Authority for Scientific Research and Innovation, CNCS/CCCDI – UEFISCDI, project BIOHORTINOV, project code PN-III-P1-1.2-PCCDI-2017-0332, project number 6 PCCDI/2018, within PNCDI III and by Ministry of Research and Innovation, through PN 18240102-35N/2018.

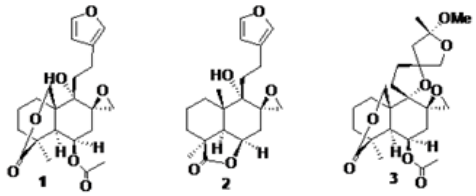
PS2-B-123

Labdane diterpenoids from *Leonotis ocymifolia* with potential anticancer activity

Jane Ncongwane[1], Comfort Nkambule[1], Vuyelwa Tembu[1], Gerda Fouche[2], Nyeleti Vukea[3], Adrienne Edkins[3], Jo-Anne de la Mare[3]

[1]Tshwane University of Technology, Pretoria, South Africa, [2]University of Pretoria, Pretoria, South Africa, [3]Rhodes University, Grahamstown, South Africa

Cancer is a worldwide problem and its burden has shifted to less developed countries which now accounts for 57% new cancer cases and 65% cancer deaths [1]. Plants have a long history of use in the treatment of diseases and natural products are a great source of vital clinically approved anticancer agents [2]. South Africa has a rich reservoir of an untapped plant biodiversity of over 20000 indigenous plants from which compounds with potential anticancer activity could be discovered [3]. In this study, *Leonotis ocymifolia* was investigated for compounds with potential cytotoxic activity. Labdane diterpenoids (Fig.1) were isolated from leaves of *Leonotis ocymifolia* through column chromatography. The structures were characterized using UPLC-MS, FTIR, optical rotation and NMR techniques as nepetaefuran (1), leonotinin (2), methoxynepetaefolin (3). Methoxynepetaefolin was found to exhibit cytotoxic activity at EC_{50} of 69.40± 1.3025 µM against triple negative breast (HCC70) cancer cell line. Additional testing of other isolated compounds is ongoing including an expansion of the array of target cancer cell lines.



Keywords: labdane diterpenoids, *Leonotis ocymifolia*, cytotoxicity

References:

[1] Torre LA, Bray F, Siegel RL, Ferlay J, Lortet-tieulent J, Jemal A. Ca-Cancer J Clin 2015; 65: 87–108.
[2] Cragg GM, Newman DJ. Biochim Biophys Acta 2013; 1830: 3670–3695.
[3] Cherry M. PLoS Biol 2005; 3: 145.

PS2-B-124

In vitro antitumor evaluation of different extracts of medicinal plants

Majed Bawazeer[1], Bathini Thissera[1], Mallique Qader[1,2], Mostafa Rateb E.[1], Mohammed Yaseen[1]

[1]University of the West of Scotland, Paisley, United Kingdom, [2]National Institute of Fundamental Studies, Kandy, Sri Lanka

Cancer is a leading cause of death worldwide. According to WHO reports, more than 8.8 million deaths were recorded in 2015. Resistivity on chemotherapeutic drugs has threatened the achievements of modern medicine. The extracts of the plant sources are rich in phytochemicals, and they are proven to have promising bioactivities. Hence, searching for new natural inhibitors has been felt paramount to find a broader range and novel products for the treatment of cancers with minimal side effects. In the current study, *Arctostaphylos uva-ursi* (leaves), *Commiphora myrrha* (resin), *Nigella sativa* (seeds), *Peganum harmala* (leaves) and *Saussurea costus* (roots) were collected from Saudi Arabia. The dried plant samples were macerated and extracted using methanol. The resulting crude extracts were tested for their anti-proliferative activity against NCI H295R (adrenal gland) and HeLa (cervix) cancer cell lines using MTT assay. The total crude extracts of all the tested samples showed more than 80% cytotoxic activity while, *A. uva-ursi* showed 50%, cytotoxic activity at 50 □g/mL conc. Further, total methanolic extracts were fractionated sequentially into hexane, dichloromethane and ethyl acetate. The resulting fractions were tested for cytotoxic activity. All the fractions of the *C. myrrha* and *N. sativa* showed good cytotoxic activity for both cervix and adrenal gland cell lines. Only the hexane fraction of *A. uva-ursi* showed good cytotoxic activity for both cell lines, while dichloromethane fraction was moderately active for cervix HeLa cell line only. Both the hexane and dichloromethane fractions of *P. harmala* and *S. costus* showed good cytotoxic activity for both cell lines. To identify the phytochemicals present in the active extracts of these plants, chemical profiling using GC-MS and LC-HR/ESI/MS are underway.

Acknowledgements: Royal Embassy of Saudi Arabia Cultural Bureau, UK is gratefully acknowledged.

PS2-B-125

Assessment of the antioxidant and antimutagenic profile from a total polyphenolic olive oil extract and its polyphenols separately

Paraskevi Kouka[1], Fotios Tekos[1], Alexios-Leandros Skaltsounis[2], Demetrios Kouretas[1]

[1]Biochemistry and Biotechnology, University of Thessaly, Larissa, Greece, [2]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, University of Athens, Athens, Greece

The olive tree is one of the most important fruit trees in Mediterranean countries and virgin olive oil possesses both health and nutritional aspects, drawing great scientific interest. It is noteworthy that all studies so far, demonstrate the beneficial effect of olive oil components, as individual substances and not as a whole, despite an increasing amount of evidence that is the complex mixture of nutritional polyphenols, more than each compound separate, which can synergistically act towards a health result [1,2]. Our aim was to firstly examine the antioxidant and antimutagenic properties of a total polyphenolic extra virgin olive oil (TPF), and secondly its polyphenols (Hydroxytyrosol (HT), Tyrosol (T), Oleocanthal (OLEO), Oleacin (OLEA)) separately, in order to assess their contribution in the total olive oil activity. For this purpose their antioxidant and antimutagenic potential were evaluated using the ABTS●+ and the plasmid relaxation assays, respectively. According to the results OLEO and OLEA contributed more readily to the TPF activity, thus exhibiting IC_{50} at lower concentrations compared to other polyphenols, in both assays.

Acknowledgements: The study was funded by the Hellenic General Secretariat for Research and Technology (GSRT) and the Hellenic Foundation for Research and Innovation (HFRI) (grant number 5547).

Keywords: olive oil, polyphenols, antioxidant activity, antimutagenic activity

References:

[1] Kouka P, Priftis A, Stagos D, *et al*. Int J Mol Med 2017; 40: 703–712.
[2] Kouka P, Chatzieffraimidi GA, Raftis G, *et al*. Phytomedicine 2018; 47: 135–142.

PS2-B-126

Anti-inflammatory and antiproliferative compounds from *Sphaeranthus africanus*

Huyen Thi Tran[1], Xuehong Gao[1], Nadine Kretschmer[1], Eva-Maria Pferschy-Wenzig[1], Olaf Kunert[2], Loi Huynh[3], Rudolf Bauer[1]

[1]Institute of Pharmaceutical Sciences, Department of Pharmacognosy, University of Graz, Universitaetsplatz 4, 8010 Graz, Austria, Graz, Austria, [2]Institute of Pharmaceutical Sciences, Department of Pharmaceutical Chemistry, University of Graz, Universitaetsplatz 1, 8010 Graz, Austria, Graz, Austria, [3]Department of Botany and Pharmacognosy, Hong Bang International University, Ho Chi Minh City, Vietnam, Ho Chi Minh, Vietnam

Sphaeranthus africanus L. has been used in traditional Vietnamese medicine to treat sore throat, and to relieve pain and swelling [1]. Previously, we isolated five carvotacetones (1-5) from this plant that displayed cytotoxicity against several cancer cell lines [2]. However, anti-inflammatory activity of this plant had not been investigated yet.

The objective of this study was to investigate the anti-inflammatory activity of all constituents (1-9), and to evaluate the anti-proliferative activity of the newly isolated compounds (6-9).

In addition to the previously isolated compounds (1-5), we could identify two isomeric carvotacetones (6-7), asperglaucide (8), and chrysoplenol D (9).

All compounds isolated from *S. africanus* (1-9, Figure 1) have been evaluated for *in-vitro* inhibitory activity on COX-1 and COX-2 enzymes. The results showed that the previously isolated compounds 1 and 2 possess potent selective cyclooxygenase-2 inhibitory effect with IC_{50} values of 3.6 and 0.5 μM, respectively. Apart from 1 and 2, none of the other compounds exhibited anti-inflammatory activities against COX-1 and COX-2.

Anti-proliferative activity was evaluated by XTT viability assay with four cancer cell lines, namely CCRF-CEM, MDA-MB-231, HCT-116, and U-251 cells. XTT assay revealed that within the newly isolated compounds, the two isomeric carvotacetone derivatives (6-7) exhibited considerable anti-proliferative activity with IC_{50} values ranging from 1.23 to 8 μM.

In conclusion, this is the first-time that the isomeric compounds (6,7) were separately isolated and their antiproliferative activity was determined. Selective *in-vitro* COX-2 inhibitory activity exhibited by some of the major constituents of *S. africanus* supports the traditional medical application of this plant for the treatment of inflammation-related disorders.

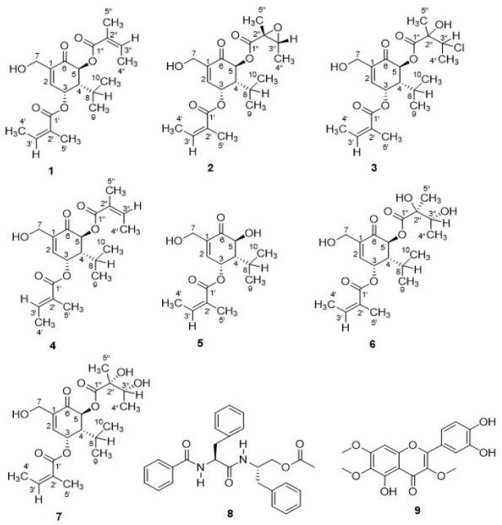


Figure 1. Structures of **1-9**

References:

[1] Chi VV. Từ điển cây thuốc Việt nam (Dictionary of medicinal plants in Vietnam), Y hoc 2014.
[2] Tran HT, Pferschy-Wenzig EM, Kretschmer N, Kunert O, Huynh L, Bauer R. J Nat Prod 2018; 81(8):1829–1834.

PS2-B-127

Dual enzymes inhibitor of tiger milk mushroom (*Lignosus rhinocerus*) extracts on HIV-1 protease and reverse transcriptase

Chanin Sillapachaiyaporn[1], Siriporn Chuchawankul[2]

[1]Program in Clinical Biochemistry and Molecular Medicine, Department of Clinical Chemistry, Faculty of Allied Health Sciences, Chulalongkorn University, Bangkok, Thailand, [2]Department of Transfusion Medicine and Clinical Microbiology, Faculty of Allied Health Sciences, Chulalongkorn University, Bangkok, Thailand

Known as a medicinal mushroom with anti-inflammatory, antioxidant and anti-proliferative activities, *Lignosus rhinocerus* (LR) also showed antiviral activity on human papilloma virus and dengue virus type-2. Since protease (PR) and reverse transcriptase (RT) of human immunodeficiency virus type-1 (HIV-1) are crucial enzymes for its replication, they are targets of anti-retroviral drug development. Herein, we determined PR and RT inhibitory activity of three LR crude extracts: crude hexane (LRH), crude ethanol (LRE) and crude water (LRW). *In vitro* screening assays showed an inhibition of either PR or RT activities by the extracts. Non-toxic concentrations of the extracts based on MTS assay using T lymphocytes (MOLT-4) were used. All extracts at certain concentrations significantly inhibited HIV-induced syncytial formation in agreement with the effect of inhibitors, darunavir (DRV) and nevirapine (NVP). Moreover, all extracts exhibited significant inhibition on PR activity by reducing p24 production with percentages of inhibition ranged from 30 to 50. Meanwhile, only LRH at 0.09 mg/ml blocked RT activity at the last step of reverse transcription showing significant decrease in full-length DNA (flDNA) levels. Furthermore, gas chromatography-mass spectrometry (GC-MS) and liquid chromatography-mass spectrometry (LC-MS) revealed fatty acids, peptides and terpenoids as major groups of phytochemical constituents in the extracts. Computation simulation assay by AutoDock 4.0 presented that some compounds strongly bound to both enzyme targets. Especially, thunbergol which is found in LRH showed binding energy with PR and RT in line with original inhibitors of these enzymes. In conclusion, LRH showed not only PR but also RT inhibitory activities in in vitro non-cell and infected cell based assays as well as *in silico* approach. These results suggested that LR could be a potential source of compounds to inhibit HIV-1 replication. Furthermore, the identified compounds could be beneficial data to develop novel PR and RT drugs.

PS2-B-128

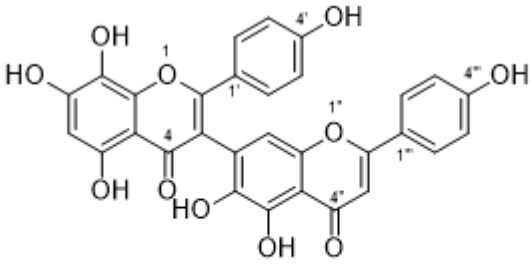
Isolation of a new bi-flavonoid from *Rhus leptodictya* leaves extract

Tshifhiwa Matamela

Vaal University of Technology, Johannesburg, South Africa

Rhus belongs to a genus that consists of 250 species and is commonly known as sumac or sumach. These plants are found to be individual species of flowering plants in the family Anacardiceae [2]. All sumac have a milky or resinous sap [3]. Some Anacardiceae are well known poisonous plants viz., rhustoxicodendron also kmown as poison Ivy while others have been used as herbal medicines. The leaves of *Rhus leptodictya* were collected, dried and gried to powder and stored at room temperature. The biological assays were done for screening of antibacterial and antioxidant activity [1]. Powdered plant material (200 g) was exhaustively extracted by shaking for 12 hours with a labotech shaker with 2 litres of acetone for the separation of compounds. Investigation of the leaves of *Rhus leptodictya* has led to the isolation of a new biflavonoid 5,5',6',7,8-pentahydroxy-2,2'-bis(4-hydroxyphenyl)-4H-4H'-[3,7'-bichromene]-4,4'-dione. The chemical structure was fully established using ^{1}H NMR, ^{13}C NMR, DEPT, HR-MS, IR and COSY spectroscopic techniques. The bioactivity of the crude extract shows antioxidation potential.

Keywords: isolation, bioflavonoids, bioactive compound, 5,5',6',7,8-pentahydroxy-2,2'-bis(4-hydroxyphenyl)-4H-4H'-[3,7'-bichromene]-4,4'-dione



5,5',6',7,8-pentahydroxy-2,2'-bis(4-hydroxyphenyl)-4*H*,4'*H*-[3,7'-bichromene]-4,4'-dione

References:

[1] Eloff JN. J Ethnopharmacol 1998; 60: 1–8.
[2] Mithcell JD. The poisonous Anacardiaceae genera of the world. Advance in Economic Botany 1990; pp. 103–129.
[3] Oh SH, Haw CR, Lee MH. Contact Dermatitis 2003; 48(5): 251–254.

PS2-B-129

M. charantia: short review on its phytochemicals and potential therapeutic use

Alina Ortan[1], Daniela Ionescu[1,2], Simona Spinu[1,3]

[1]University of Agronomic Sciences and Veterinary Medicine, Bucharest, Romania, [2]Hofigal Export Import S.A, Bucharest, Romania, [3]Faculty of Physics, Doctoral School of Biophysics and Medical Physics, Bucharest, Romania

Plants are rich sources for all classes of chemical compounds and at the same time have many pharmacological properties that are widely used in therapeutics due to high efficacy and low toxicity. The present paper reviews the phytochemical profile and the potential use of *Momordica charantia* L. for various diseases treatment, based on the available literature.
Momordica charantia L., commonly known as bitter melon, bitter gourd; bitter squash or balsam-pear, is a herbaceous vine from the Cucurbitaceae family., widespread in tropical and subtropical regions of Africa, Asia, Australia, South America and the Caribbean. In traditional medicine, *M. charantia* has been used for microbial infections, wound healing, inflammation, hypertension, intestinal disorders.
Phytochemical profile. *M. charantia* contains up to 228 known phytochemicals, dispersed in all plant organs [1]. Plant is rich in alkaloids, flavonoids, steroidal glycosides, saponins and tannins [2]. The fruit, the most widely used part of the plant, contains glycosides, saponins, alkaloids, triterpenes, proteins, steroids and phenolic compounds that contribute to its use in phytotherapy; the characteristic bitter taste is attributed to momordicosides K and L, and momordicines I and II [3].
Pharmacology. *M. charantia* fruits are widely used as functional food for the prevention and treatment of diabetes and associated complications. Recent studies demonstrated that bioactive compounds responsible for antidiabetic activity are charantine (saponin), momordicin (alkaloid) and p-insulin [4]. Although research is directed mainly to its antidiabetic activity, antioxidant, anti-carcinogenic and anti-inflammatory effects have been reported [3].
The extensive use of the plant in traditional medicine as well as scientific research conducted in recent years qualify *M charantia* as a useful tool for the modern treatment of many diseases.

Acknowledgements: The authors gratefully acknowledge the support obtained through the project SusMAPWaste, SMIS104323, Contract No. 89/09.09.2016, from the Operational Program Competitiveness 2014-2020, project cofinanced from the European Regional Development Fund.

PS2-B-130

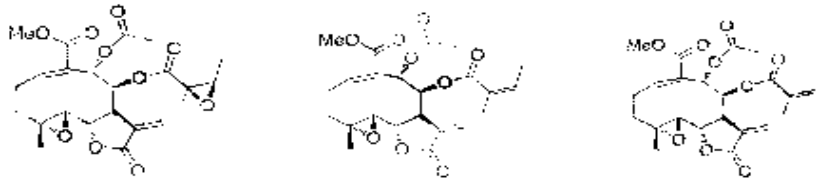
Anti-proliferative activity of melampolide-type sesquiterpene lactones from *Enhydra Fluctuans* on cancer cell lines

Thanh Triet Nguyen[1,3], Nadine Kretschmer[1], Eva-Maria Pferschy-Wenzig[1], Olaf Kunert[2], Rudolf Bauer[1]

[1]Department of Pharmacognosy, University of Graz, Graz, Austria, [2]Department of Pharmaceutical chemistry, University of Graz, Graz, Austria, [3]Department of Traditional pharmacy, University of Medicine and Pharmacy at Ho Chi Minh city, Ho Chi Minh, Vietnam

In Vietnam, the whole plant of *Enhydra fluctuans Lour*. is used as a tonic for the nervous system and for the treatment of bile secretory disorders, haemorrhage, for digestive disorders, and skin protection [1]. Melampolide-type sesquiterpene lactones are well-known to be the most abundant constituents in *Enhydra fluctuans*. Melampolide derivatives have been reported previously from several genera, such as *Melampodium*, *Smallanthus*, *Tetragonotheca*, *Sigesbeckia*, *Schkuhria* and *Acanthospermum* [2]. In this study, the cytotoxicity-guided isolation led to the identification of three sesquiterpene lactones from the aerial parts of *Enhydra fluctuans*: enhydrin, fluctuanin and fluctuadin. The structures of the compounds were elucidated by various spectroscopic methods, such as UV, LC-MS, NMR, and comparison with the literature data [1-3]. The compounds were evaluated for their anti-proliferative activity in four human cancer cell lines: CCRF-CEM, HCT-116, MDA-MB-231 and U251 cells, in comparison with normal lung fibroblast MRC-5 cells. All compounds showed significant cytotoxicity on the tested cell lines with IC_{50} values ranging from 0.18 µM to 17.34 µM. Finally, enhydrin was chosen for testing the apoptotic mechanism. There were no considerable alterations to be found in the activation of caspases - 3/7. The results suggest that the cytotoxic activity of *Enhydra fluctuans* is derived from these sesquiterpene lactones which may become lead compounds for anti-cancer drugs.

Supplementary data: Structures of three sesquiterpene lactones



enhydrin **fluctuanin** **fluctuadin**

Acknowledgments: I am thankful to Department of Pharmacognosy, University of Graz for supporting chemicals and facilities during period of thesis performance.

References:

[1] Phạm Hoàng Hộ, Cây cỏ Việt Nam, Nhà xuất bản Trẻ 1999; 1.
[2] Bohlmann F, Ahmed M, Robinson H, King RM. Phytochemistry 1982; 21(7): 1675–1678.
[3] Ali E, Ghosh Dastidar PP, Pakrashi SC, Durham LJ, Duffield AM. Tetrahedron 1972; 28: 2285 –2298.

PS2-B-131

Secondary metabolites from the leaves and seeds of *Centaurea vlachorum* (Asteraceae) and their biological activity

Entela Çeliku[1,2], Olga Tsiftsoglou[2], Erjon Mamoci[3], Sokol Abazi[4], Lulezim Shuka[5], Dimitra Hadjipavlou-Litina[6], Diamanto Lazari[2]

[1]Department of Chemistry, Faculty of Biotechnology and Food, Agricultural University of Tirana, Pajsi Vodica St., 1029 , Tirana, Albania, [2]Laboratory of Pharmacognosy, School of Pharmacy, Faculty of Health Sciences, Aristotle University of Thessaloniki, Thessaloniki, Greece, [3]Department of Food Sciences and Biotechnology, Faculty of Biotechnology and Food, Agricultural University of Tirana, Pajsi Vodica St., 1029, Tirana, Albania, [4]Faculty of Engineering, Canadian Institute of Technology, Zayed Centre, Tirana , Albania, [5]Department of Biology, Faculty of Natural Sciences, University of Tirana, Bld. ZOG I, 1001, Tirana, Albania, [6]Department of Pharmaceutical Chemistry, School of Pharmacy, Faculty of Health Sciences, Aristotle University of Thessaloniki, Thessaloniki, Greece

In the course of our investigation on bioactive natural products, we have investigated the chemical constituents and the biological activity of the leaves and seeds of *Centaurea vlachorum* Hartvig, collected on early July 2011 in Albania, in district of Dibra, Mt Kunora e Lures. Phytochemical investigation of the aerial parts and seeds of *Centaurea vlachorum* led to the isolation of eleven compounds including five sesquiterpene lactones, cynaropicrin (1), repdiolide (2), chlororepdiolide (3), chlorojanerin (4) and epi-cebelin J (5), two flavonoids, luteolin (6) and nepetin (7), two indole alkaloids *N*-(*p*-coumaroyl) serotonin (8) and moschamine (9) and two dibenzylbutyrolactone lignans matairesinol (10) and arctiin (11). The pure compounds were isolated using chromatographic methods (VLC, CC, TLC, HPLC) and the structures were elucidated by spectroscopic methods UV-Vis, 1D and 2D NMR. Moreover, the isolated compounds (lignans and indole alkaloids) were tested for their free radical scavenging activity using the following *in vitro* assays: (i) interaction with the free stable radical of DPPH (1,1-diphenyl-2-picrylhydrazyl), (ii) inhibition of linoleic acid peroxidation with the dihydrochloric acid of 2,2-Azobis2-amidinopropane (AAPH) and their inhibitory activity towards soybean lipoxygenase was evaluated, using linoleic acid as substrate. Finally, all the isolated compounds were tested using disc diffusion method against three bacteria such as *Escherichia coli* ATCC 25922, *Salmonella typhimurium* ATCC 14028, *Staphylococcus aureus* ATCC 6538 and yeast *Candida albicans* ATCC 10231.

Matairesinol (10) is presenting the highest interaction with the stable radical DPPH, followed by moschamine (9) and *N*-(*p*-coumaroyl) serotonin (8). *N*-(*p*-coumaroyl) serotonin (8) and arctiin (11) showed high anti-lipid peroxidation activity ranged from 82.3 to 81.6% compared to the reference compound trolox (88.0%). The tested compounds (10) and (9) significantly inhibit soybean lipoxygenase (~80%) whereas compounds (11) and (8) did not exhibit any inhibition. Finally, none of the tested compounds showed antimicrobial activity at the tested dose (1mg/mL).

PS2-B-132

Antioxidant properties and safety use of goutweed

Karolina Jakubczyk, Katarzyna Janda, Katarzyna Watychowicz, Justyna Kałdyńska, Daniel Styburski, Karolina Dec

Pomeranian Medical University in Szczecin, Department of Biochemistry and Human Nutrition , Szczecin, Poland

Goutweed (*Aegopodium podagraria* L.) is a plant, widely grown in Europe and Asia. In folk medicine, mainly the leaves of goutweed were used to treat gout, inflammatory states in kidneys and bladder and to facilitate wound healing. Despite the presence of many pro-healthy compounds, e.g. polyacetylenes (falkarinol and falkaridiol), essential oils from the group of mono- and sesquiterpenes, polyphenols, e.g. phenolic acids and flavonoids, coumarin compounds, there is not much research on the properties of this plant. The aim of the study was to investigate the antioxidant properties of ethanolic and aqueous extracts of goutweed. The antioxidant effect of extracts from flowers, leaves, seeds and rhizomes of the plant was tested on the macrophages. The effects of extracts on the parameters of cytotoxicity (proliferation, apoptosis) were investigated to verify the safeness. The extracts protect cells against oxidative stress and had a positive impact on cell proliferation. Spectrophotometric methods demonstrated the high antioxidant capacity of extracts of flowers and seeds. *In vitro* studies on macrophages show that the same extracts decrease antioxidant potential and enzyme activity, which may inhibit free radical reaction by antioxidants contained in extract. Rhizomes and leaf extract initiated stress, increased enzyme activity and antioxidative potential as a defense mechanism. Extracts of seeds of goutweed added to the food or supplements can reduce oxidative stress, which may be used in the treatment and prevention of many diseases.

Keywords: goutweed, *Aegopodium podagraria* L., oxidative stress, antioxidant

References:

[1] Jakubczyk K, Kwiatkowski P, Sienkiewicz M, Janda K. Post Fitoter 2018; 1: 3–9.
[2] Valyova M, Tashev A, Stoyanov S, Yordanova S, Ganeva Y. Journal of Chemical Technology and Metallurgy 2016; 51(3): 271–274.

PS2-B-133

Lifespan extending and oxidative stress resistant properties of *Anacardium occidentale* L. leaf extract in *Caenorhabditis elegans*

Chatrawee Duangjan[1], Tewin Tencomnao[2], Micael Wink[3]

[1]Program in Clinical Biochemistry and Molecular Medicine, Department of Clinical Chemistry, Faculty of Allied Health Sciences, Chulalongkorn University, Bangkok, Thailand, [2]Department of Clinical Chemistry, Faculty of Allied Health Sciences, Chulalongkorn University, Bangkok, Thailand, [3]Institute of Pharmacy and Molecular Biotechnology, Heidelberg University, Heidelberg, Germany

Anacardium occidentale contain a number of polyphenol compounds and exhibit antioxidant activities. However, a possible anti-aging potential of this plant extracts have not been explored. The objective of this study is to investigate the lifespan-extending effect and oxidative stress resistant properties of AO extraction in *C. elegans* model. AO extraction enhanced oxidative stress resistance by increasing the survival rate of nematodes under oxidative stress, attenuating intracellular ROS level, increasing expression of stress-response gene such as SOD-3 and GST-4 while decreasing hsp-16.2 gene expression via DAF-16/FOXO and SKN-1/Nrf-2 signaling pathway. Moreover AO extraction has beneficial effect on anti-aging and longevity by improving pharyngeal pumping function, attenuating auto fluorescent pigment and increasing life span extension. This research is a first time to study the effect of *A. occidentale* leaf extracts on anti-aging potential in *C. elegans* model and the result of study may serve as potential agents that slow aging.

PS2-B-134

Goutweed infusions as a source of antioxidants

Katarzyna Janda, Karolina Jakubczyk, Kamila Dębia, Katarzyna Watychowicz, Justyna Kałduńska, Daniel Styburski, Karolina Dec

Department of Biochemistry and Human Nutrition, Pomeranian Medical University in Szczecin, Szczecin, Poland

Goutweed (*Aegopodium podagraria* L.) is a perennial that belongs to the carrot family (Apiaceae). Its luscious leaves are added to soups, like cabbage soup, cold soup, beetroot soup or salads. It is a common practice to marinate, pickle or dry this plant and to use it in dishes due to its unique aroma. It is a valuable medicinal plant given its anti-inflammatory, antimicrobial and anticancer potential. Antioxidants of plant origin are characterized by great biological activity, including ability to neutralize oxidative stress. Bioactive components extensively present in plants, exhibit potential health benefits connected to antioxidant, anticancer, anti-atherosclerotic, antimutagenic and antiangiogenic activities. Antioxidants are not synthesized by human body, therefore its supply with food is a preventive treatment, especially in case of defense from free radicals. *In vitro* studies show, that bioactive components of herbs and spices may inhibit biochemical pathways that regulate cell division, proliferation and detoxification, as well as anti-inflammatory immunological response. This indicates that food of plant origin, because of its antioxidant activity, is an important factor in prevention of lifestyle diseases. In the presented work we compared antioxidant potential of goutweed leaves, flowers, seeds and rhizomes that was prepared in different temperatures.

Research material was leaves, flowers, seeds and rhizomes of goutweed. Leaves was collected during spring, before the appearance of flower shoots; flowers were collected at the beginning and in the middle of flowering period, in a ripening cycle, while seeds were collected right before shoveling of flowers; rhizomes were collected before first frosts. After purification plant material were frozen in -20°C, and then it was freeze dried in Alpha 1-2 LD plus machine (pressure 0,735 mmHg, temperature –20°C). After drying material was homogenized and used to prepare infusions. Infusions were prepared using 0,5g of lyophilized leaves, flowers, seeds, rhizomes. It was then inundated with 100 cm^3 of distilled water (25°C, 70°C, 80°C and 90°C). After shaking in room temperature infusions were percolated. Antioxidant properties of infusions were indicated by spectrophotometric method using synthetic free radical DPPH (1,1-diphenyl-2-picrylhydrazyl, Sigma). Absorbance was measured at 518 nm wavelength. The highest antioxidant power was observed in flower infusions and the lowest in rhizome infusions. Independently of the type of material the lowest activity was measured in infusion prepared in 25°C.

Keywords: goutweed, *Aegopodium podagraria* L., infusions, antioxidant activity

References:

[1] Orav A, Viitak A, Vaher M. Procedia Chem 2010; 2: 152–160
[2] Kunstman P, Wojcińska M, Popławska P.Post Fitoter 2012; 4: 244–249.
[3] Yoo KM, Lee CH, Lee H, Moon B, Lee CY. Food Chem 2008; 106: 929–936
[4] Jakubczyk K, Kwiatkowski P, Sienkiewicz M, Janda K. 2018; 1: 3-9.
[5] Valyova M, Tashev A, Stoyanov S, Yordanova S, Ganeva Y. J Chem Technol Metall 2016; 51(3): 271–274.

PS2-B-135

Dietary quercetin impacts the concentration of tau-fluvalinate in honey bees (*Apis mellifera*)

Hamidreza Ardalani, Nanna Hjort Vidkjær, Per Kryger Inge S. Fomsgaard

Department of Agroecology, Faculty of Science and Technology, Aarhus University, Forsøgsvej 1, 4200 Slagelse, Denmark, Slagelse, Denmark

Honey bees are important pollinators and subject to numerous stressors such as reduced flower diversity, new parasites, and chronical exposure to agrochemicals [1]. We have limited knowledge of metabolisation pathways and synergistic effects of xenobiotics in honey bees. Quercetin is one of the most abundant phytochemicals in plants and therefore, honey bees ingest quercetin consistently. Tau-fluvalinate, a pyrethroid acaricide, is used for control of Varroa mites in bee colonies as well as a frequent insecticide in agriculture. In this study, we investigated the effect of dietary quercetin on the metabolisation rate of tau-fluvalinate in honey bees. Honey bees (n=240) were divided in two groups and fed with either quercetin and sucrose candy or only sucrose candy for 72 h. Hereafter, they were contact-exposed to 0.9 µg/bee tau-fluvalinate. After 24 h, the bees were anesthetized with CO_2 and sacrificed by freezing. The bees were extracted with a validated QuEChERS method (recovery percentage 86.3±2.8). Finally, the concentration of tau-fluvalinate was determined by LC-MSMS. The mean concentration of tau-fluvalinate was 0.17±0.02 and 0.22±0.03 µg/bee in bees that were fed with and without quercetin, respectively (significantly different; P=0.03). The results of this study demonstrate that intake of quercetin leads to a reduction in the concentration of tau-fluvalinate in honey bees 24 hours after contact-exposure. Johnson *et al*. [2], showed that honey and pollen feeding upregulates the bees P450 detoxification system and dietary quercetin reduced the toxicity of tau-fluvalinate. Thus, we hypothesized that dietary quercetin may lead to lower concentrations of tau-fluvalinate by increasing the metabolisation rate of the acaricide. Our results support this and upon confirmation, quercetin-rich plants may be exploited in future bee-keeping.

Keywords: *Apis mellifera*, pesticide, quercetin, tau-fluvalinate.

References:

[1] Goulson D, Nicholls E, Botias C, Rotheray EL. Science 2015; 347: 1255957
[2] Johnson RM, Mao W, Pollock HS, Niu G, Schuler MA, Berenbaum MR. PLoS One 2012; 7: e31051.

PS2-B-136

Antioxidant and lifespan extension effects of *Caesalpinia mimosoides* extracts in nematode *Caenorhabditis elegans*

Panthakarn Rangsinth[1], Anchalee Prasansuklab[1], Michael Wink[2], Tewin Tencomnao[3]

[1]Program in Clinical Biochemistry and Molecular Medicine, Department of Clinical Chemistry, Faculty of Allied Health Sciences, Chulalongkorn University,Bangkok, Thailand, [2]Institute of Pharmacy and Molecular Biotechnology (IPMB), Heidelberg University, Heidelberg, Thailand, [3]Department of Clinical Chemistry, Faculty of Allied Health Sciences, Chulalongkorn University, Bangkok, Thailand

Caesalpinia mimosoides is a native plant of Southeast Asia, as well as in northern and north-eastern parts of Thailand. This plant has several active compounds including gallic acid and quercetin that has been reported to exhibit antioxidant activity *in vitro*. However, *in vivo* experiment, still need further investigation. *Caenorhabditis elegans*, a small free-living soil nematode, plays an established role in several medical researches. The aim of this research is to study the antioxidant activity of *C. mimosoides* leaf extracts in *C. elegans* and its underlying mechanisms. The nematodes were treated with *C. mimosoides* extracts in various concentrations. To investigate the protective effects of the extract against oxidative stress, wild-type N2 were used to determine survival rate under oxidative stress and intracellular ROS. To study underlying mechanisms, the mutant strains with GFP reporter gene including TJ356, CF1553, EU1 and LD4 were used to study DAF-16, SOD-3, SKN-1 and GST-4 gene, respectively. The extracts were able to enhance stress resistance and to ameliorate intracellular ROS accumulation after treatment. The antioxidant effects were mediated through DAF-16/FOXO pathway as well as SOD-3 expression, whereas SKN-1 and GST-4 cannot detect any alteration. To detect any toxicity of the extracts, brood size and body length of the worms were not detected any sign of the toxic. To further investigate lifespan extension, *C. mimosoides* extracts were also observed to prolong lifespan of the worms. These findings establish the evidence of antioxidant activity of *C. mimosoides* extract *in vivo* that can be used for development as alternative drug against oxidative stress in the future.

Keywords: *Caesalpinia mimosoides*, Caenorhabditis elegans, antioxidant, lifespan

PS2-B-138

The use of innate microflora of honey in fermented milk drink

Hosam Baksamawi[1], Izlem Haktanir[1], Dimitrios Kafetzopoulos[2], Georgios Kafetzopoulos[2], Kostas Gkatzionis[1]

[1]University of Birmingham, Birmingham, United Kingdom, [2]University of the Aegean, Myrina, Lemnos, Greece

Honey is a natural food with properties and composition depending on its geographical and floral origin, seasonality, environmental factors and treatment during processing. Moreover, honey is a natural source of probiotic bacteria beneficial to human health. This study aimed to assess the effect of honey microflora on milk fermentation. Thus, honeys of various floral origins were compared (two monofloral Manuka, multiflora Italian and Greek organic honey, 'raw' and after thermal treatment). The honey samples were utilized for inoculation of pasteurized whole milk (100 ˚C, 5 minutes followed by cooling to 42 ˚C) in order to produce a fermented milk drink. Lactic acid production, acidity, pH, microbial counts of lactic acid bacteria, yeasts a total aerobic count (MRS, RBCA, PCA respectively) were monitored during three days of fermentation (24, 48 and 72 hours). The results showed a significant decrease ($p < 0.05$) in the pH value in accordance to increase of acidity of the honey samples. The microbial counts showed a significant increase ($p < 0.05$) in the lactic acid bacteria counts during fermentation; however, not all the honey samples presented the same microbial growth pattern. Yeasts growth was evidenced in all samples except Manuka. The results of this study indicate that honey samples containing lactic acid bacteria could enhance lactic acid production in milk fermentation.

Keywords: Honey, probiotic bacteria, fermented milk, lactic acid bacteria

PS2-B-139

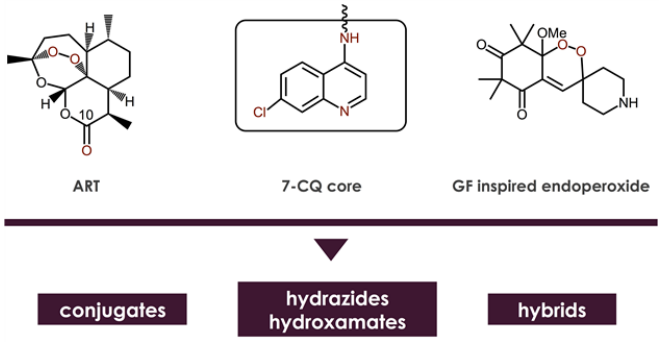
Synthesis of novel artemisinin dimers and hybrids with other antimalarial agents

Dionissia Pepe[1], Dimitra Toumpa[1], Christophe Menendez[2], Christiane André-Barrès[2], Christina Koumpoura[1], Michel Baltas[2], Constantinos Athanassopoulos[1]

[1]*Organic Chemistry Laboratory, Department of Chemistry, University of Patras,, GR 265 04, Patra, Greece,* [2]*Laboratoire SPCMIB (CNRS UMR 5068, Université de Toulouse,, 118 Route de Narbonne, 31062 Toulouse, France*

Malaria is a mosquito-borne infectious disease caused by parasitic protozoans of *Plasmodium* genus. Despite the fact that there is a wide variety of drugs used in clinical practice, malaria still has high mortality rates due to the development of resistant *Plasmodium falciparum* (Pf) strains resulting from excessive use of antimalarial drugs. For more than 40 years, chloroquine (CQ) [1], which bears the pharmacophore core of 7-chloroquinoline, was one of the most widely used antimalarial drugs. In 2004, Artemisinin (ART) [2] was introduced into clinical practice and currently it is the drug of choice against most *Plasmodium* strains, especially the CQ-resistant ones. Furthermore, Growth Factors (GFs) [3] which bear an endo-peroxidic bond like ART, have recently been attractive targets for the development of novel synthetic analogues which present interesting antimalarial properties. This work presents synthetic protocols for a library, which contains potential novel antimalarial agents, combining structural elements from ART [4], CQ and GFs, aiming to improve the pharmacological profile of the parent molecules. This is envisaged in ART/7-CQ/GFs hybrids and conjugates. In addition, the synthesis of hydroxamate and hydrazide analogues of both endoperoxides (ART and GF inspired ones) is a very interesting functional modification since the combination of two pharmacophores in these analogues would hopefully improve their antimalarial and/or antitumor activities.

Keywords: artemisinin, chloroquine, growth factors, endoperoxides, malaria, antimalarials, polyamines, hydroxamics, hydrazides, conjugates, hybrids



References:

[1] Wellems TE, Plowe C V. J Infect Dis 2001; 184: 770–776
[2] Hastings IM, Ward SA. J Infect Dis 2005; 192: 1303–1305
[3] Ruiz J, Azema J, Payrastre C, Baltas M, Tuccio B, Vial H, Andre-Barres C. Curr Top Med Chem 2014; 14: 1668–1683
[4] Magoulas GE, Tsigkou T, Skondra L, Lamprou M, Tsoukala P, Kokkinogouli V, Pantazaka E, Papaioannou D, Athanassopoulos CM, Papadimitriou E. Bioorg Med Chem 2017; 25: 3756–3767.

PS2-B-140

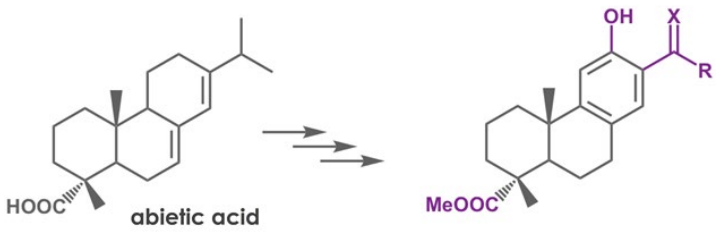
Abietane diterpenes - high value natural precursors for the development of new antimicrobial agents

Antonia Antoniou[1], Macarena Funes Chabán[2], Catherine Karagianni[1], Claudia Solá[3], Despina Palla[1], Myrsini-Irene Tachliabouri[1], Maria C. Carpinella[2], Constantinos Athanassopoulos[1]

[1]Synthetic Organic Chemistry Laboratory, Department of Chemistry, University of Patras, 265 04 Patra, Greece, [2]Research Institute of Natural Resources and Sustainability José Sánchez Labrador S.J. (IRNASUS-CONICET), School of Chemistry, Catholic University of Córdoba, Córdoba, Argentina, [3]CIBICI-CONICET. National University of Córdoba, Córdoba, Argentina

Bacterial infections are dramatically increasing every day for diverse reasons, mainly due to the development of resistance to conventional antibiotics. Methicillin-resistant *Staphylococcus aureus* (MRSA) is one of the major pathogen being responsible for 60-89% of nosocomial infections.
The urgent need to develop new antibiotics to deal with ever more resistant bacteria is therefore of great interest to researchers and pharmaceutical industries.
Plants have played a dominant role in the development of sophisticated traditional medicine systems. The chemodiversity in nature offers a valuable source of new drugs, new drug leads, and new chemical entities (NCEs).
Abietanes are a family of naturally occurring diterpenoids with a highly diverse repertoire of bioactivities. They have generated significant interest for the medicinal and pharmacological communities since they exhibit anticancer, antiviral, antifungal and antibacterial activity [1]. Regarding the latter, abietane derivatives are valuable in the battle against the antimicrobial resistance and imminent antibiotic shortage.
In the present work, we designed and synthesized a suitable for structure-activity relationship studies (SARS) library of abietane analogues, using abietic acid as the natural precursor [2]. Biological evaluation of these products against different *S. aureus* strains, methicillin-susceptible *S. aureus* (MSSA), CA-MRSA, HA-MRSA and LA-MRSA), revealed some promising compounds one of them being the most potent with MIC values ranging from 3.9 to 15. Moreover, interesting results were obtained regarding the structural elements being responsible for the improvement in activity.
In conclusion, the synthesized compounds proved to be potential candidates for effective agents to tackle infections caused by pathogenic bacteria, especially by resistant phenotypes.

Keywords: Diterpenes, abietic acid, SARS, Antimicrobial, multidrug resistance, MRSA



References

[1] Gonzalez MA. Nat Prod Rep 2015; 32: 684–704.
[2] Antoniou A, Chatzopoulou M, Bantzi M, Athanassopoulos CM, Giannis A, Pitsinos EN. Med Chem Comm 2016; 7: 2328–2331

PS2-B-141

Investigation of the *in vivo* oral acute toxicity and genotoxicity of Chios mastic gum in male Wistar rats

Eirini-Christina Psarou, Katerina Kyriakopoulou, Aikaterini Termentzi, Pelagia Anastasiadou, Marios Meidanis, Kyriaki Machera

Benaki Phytopathological Institute, Greece

Chios mastic gum (CMG), the resin of *Pistacia lentiscus* L. var*. chia*, is a product of Protected Designation of Origin (PDO) and of major financial importance in Greece. Mastic presents increasing interest in the global nutraceutical and cosmeceutical market, as scientific evidence about its biological effects continuously grows [1]. Although it presents promising therapeutic properties including antibacterial, hypolipidemic, antiatheromatic activity and activity against gastrointestinal disorders, little is known concerning its potential toxicity, especially after extended or high dose consumption [2,3].

In this study the acute toxicity and genotoxicity of CMG were investigated. CMG was orally administered to rats at 2000mg/kg b.w. according to the OECD 423 guideline. The bioavailability of the resin was ensured after the unambiguous detection of its characteristic triterpenic acids in plasma by UHPLC-HRMS/MS. No signs of toxicity, mortality or adverse effects in terms of body weight changes and gross organ pathology were observed. Histopathological analysis of the liver, was also carried out. Ongoing HRMS-based metabolomics analysis in plasma, urine and liver is expected to produce valuable information regarding the metabolic pathways possibly triggered or suppressed after oral administration of CMG.

The genotoxic potential of CMG was investigated *in vivo* using the Mammalian Erythocyte Micronucleus Test according to OECD 474 guideline. CMG was orally administered to rats at 2000mg/kg b.w. for three consecutive days. The administered doses were well tolerated by the rats and no signs of toxicity were observed. The characteristic mastic triterpenic acids were unambiguously detected in bone marrow, one and two hours after administration. The analysis of the results as regards the frequency of micronucleated polychromatic erythrocytes is still in progress aiming to bridge the scientific information gap in relation to the genotoxicity of CMG, not studied before.

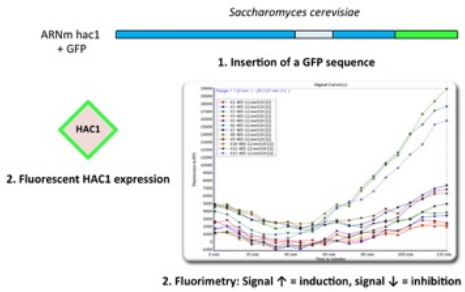
PS2-B-142

Xanthone derivatives from NPs library as potential UPR inhibitors for alternative crop protection: Molecular modelling and biological activity.

G. Viault[1], D. Bréard[1], CP Dinh[1], N. Blon[1], AM Le Ray[1], N. Bataillé-Simoneau[2], T. Guillemette[2], P. Simoneau[2], P. Richomme[1]

[1]University of Bretagne Loire, University of Angers/SFR 4207 QUASAV, EA 921 SONAS, Campus du végétal, 42 rue Georges Morel 49070 Beaucouzé, France, Angers, France, [2]University of Bretagne Loire, University of Angers/SFR 4207 QUASAV, UMR 1345 IRHS, Campus du végétal, 42 rue Georges Morel 49070 Beaucouzé, France, Angers, France

"Alternaria Leaf Spot", a common disease of crucifers caused by the fungal pathogen *Alternaria brassicicola*, affects different vegetables such as cabbage or Brussels sprout. Indole phytoalexins (camalexin and brassinin) play, in planta, a key role in crop protection against this necrotrophic agent. However, fungi mutants quickly become phytoalexin-resistant when activating signalling pathways like the Unfolded Protein Response (UPR) [1]. Indeed, it has already been shown that UPR deficient and avirulent mutants of *A. brassicicola* appear as hypersensitive to camalexin and brassinin [2,3]. However very few UPR inhibitors such as the synthetic STF-083010 [4] are known today. A preliminary *in silico* docking study pointed to potential interactions of xanthones with IRE1 – the sole reported sensor in fungi – murin cristallised protein (PDB4PL3). Therefore, we have developed an original screening assay, detecting the production of a HAC1 fluorescence-induced protein, a transcriptional activator issued from IRE1 activation, in *Saccharomyces cerevisiae* cultures (Figure 1). Different natural products from an in-house library were tested whereas RT-PCR was used to validate (or not) the observed effects. Xanthones (dibenzo gamma-pyrone) isolated from endemic *Calophyllum* or *Garcinia* species appeared as promising inhibitors of the UPR pathway whereas good correlations between Gold® scores and biological results were observed. We envisage now the design and semisynthesis of new bioactive derivatives in order to bypass the mechanism of defense of phytopathogens exploiting this signalling pathway, with the aim to restore the natural defences of cultivated plants.



Keywords: Crop protection, UPR pathway, inhibition assay

References :

[1] Joubert A, Simoneau P, Campion C, Bataillé-Simoneau N, Iacomi-Vasilescu B, Poupard P, François JM, Georgeault S, Sellier E, Guillemette T Mol Microbiol 2011; 79: 1305–1324
[2] Guillemette T, Calmes B, Simoneau P. Virulence 2014; 5: 357–364
[3] Chakraborty R, Baek JH, Young Bae E, Kim WY, Lee SY, Gab Kim M. 2016

[4] Papandreou I, Denko NC, Olson M, Van Melckebeke H, Lust S, Tam A, Solow-Cordero DE, Bouley DM, Offner F, Niwa M, Koong AC.. Blood 2011; 117: 1311–1314.

PS2-B-143

Escalating low-dose Δ^9-THC treatment during adolescence affects motor function and dopaminergic activity in adult male rats

Foteini Delis[1], Nafsika Poulia[1], Charalampos Brakatselos[1], Nikolaos Kokras[2,3], Christina Dalla[2], Katerina Antoniou[1]

[1]Department of Pharmacology, University of Ioannina, Ioannina, Greece, [2]Department of Pharmacology, National and Kapodistrian University of Athens, Athens, Greece, [3]First Department of Psychiatry, Eginition Hospital, Athens, Greece

Introduction: Delta-9-tetrahydrocannabinol (Δ^9-THC) is the main psychoactive component of cannabis. Cannabis use in adolescence induces long-term changes in behavioural parameters and in the activity of brain neurotransmitter systems in adult rats.
Aim: Evaluation of specific behavioural, neurochemical, and neurobiological indices associated with the dopaminergic system, after adolescent chronic Δ^9-THC administration.
Methods: From postnatal day (PND) 35 to PND45 adolescent male rats received escalating low-dose Δ^9-THC treatment twice daily (0.3 mg/kg PND35–37; 1 mg/kg PND38–41; 3 mg/kg PND42–45) or vehicle.
During adulthood: a) open-field motor activity was recorded, b) dopaminergic activity was assessed in the prefrontal cortex (PFC), hippocampus (HIPP), dorsal striatum (DS), and nucleus accumbens (NAC), by determining tissue dopamine and dopamine metabolite levels with High Performance Liquid Chromatography and c) dopamine transporter (DAT) protein levels were determined in specific brain regions with Western blot.
Results: a) Δ^9-THC-treated rats showed increased spontaneous locomotor activity in the open-field. b) Δ^9-THC-treated rats had lower dopamine turnover rate in the PFC and higher dopamine turnover rate in the HIPP, compared with vehicle. Furthermore, lower striatal dopamine levels and higher DOPAC levels were measured in the nucleus accumbens of Δ^9-THC-treated rats, compared with vehicle. c) No effects on DAT protein levels were found in both striatal and cortical regions.
Conclusions: Adolescent low-dose Δ^9-THC administration induced psychomotor stimulation in adult rats, accompanied by region-specific alterations in dopaminergic activity, without long-term effects on total DAT protein levels. The present findings provide novel information on the consequences of Δ^9-THC treatment during adolescence, while essentially contribute to preclinical/clinical studies focusing on the effects of adolescent Δ9-THC use on the dopaminergic system later in adulthood.

PS2-B-144

Escalating low-dose Δ^9-THC treatment during adolescence affects cognitive functions and neuroplasticity markers in adult rats

Nafsika Poulia[1], Foteini Delis[1], Charalampos Brakatselos[1], Alexia Polissidis[2], Katerina Antoniou[1]

[1]Department of Pharmacology, University of Ioannina, Ioannina, Greece, [2]Laboratory of Neurodegenerative Diseases, Biomedical Research Foundation Academy of Athens, Athens, Greece

Introduction: Preclinical studies suggest that during adolescence the main psychoactive component of cannabis, delta-9-tetrahydrocannabinol (Δ^9-THC), can trigger long-term behavioural and neurobiological alterations in adulthood.
Aim: Evaluation of behavioural and neurobiological profile of adult rats following chronic adolescent Δ^9-THC exposure.
Methods: From postnatal day (PND) 35 to PND45 adolescent male rats received escalating low-dose Δ^9-THC treatment twice daily (0.3 mg/kg PND 35–37; 1 mg/kg PND 38–41; 3 mg/kg PND 42–45) or vehicle.
On PND 75: a) open-field motor activity was recorded b) short-term spatial memory was assessed using the Object Location Task and the Morris Water Maze test c) higher-order cognitive functions were examined using the Attentional Set Shifting test and d) specific neuroplasticity-related molecular markers were evaluated in the hippocampus (HIPP) and prefrontal cortex (PFC) of adult rats.
Results: a) Δ^9-THC treatment led to increased spontaneous motor activity and affected the pattern of behavioural motor habituation in the open field test, compared with vehicle. b) Δ^9-THC exposure affected adult short-term spatial recognition memory and learning as deduced by decreased discrimination ability in the Object Location Task and impaired spatial memory and learning in the Morris Water Maze test. c) Δ^9-THC treatment did not affect the execution of the higher-order cognitive task. d) Δ^9-THC treatment led to reduced BDNF protein levels in the HIPP and the PFC, compared with vehicle.
Conclusions: Adolescent low-dose Δ^9-THC administration induced psychomotor stimulation accompanied by changes in the pattern of behavioral habituation, cognitive impairments, and region-specific changes in protein levels related to neuroplasticity markers.
Based on these findings and our previous results, we propose that low-dose Δ^9-THC during adolescence induces psychomotor and cognitive impairments paralleled with region-specific neurobiological alterations

PS2-B-145

Ficusnotins, diarylbutanoids from the Philippine endemic plant *Ficus nota* (Blanco) Merr.

Mylene Uy[1], Felmer Latayada[2], Shinji Ohta[3]

[1]Premier Research Institute of Science and Mathematics, Mindanao State University-Iligan Institute of Technology, Iligan City, Philippines, [2]Department of Natural Sciences, Caraga State University, Butuan City, Philippines, [3]Graduate School of Biosphere Science, Hiroshima University, Higashi-Hiroshima, Japan

Six diarylbutanoids, designated as ficusnotins A–F (1-6), with a rare carbon skeleton consisting of two aromatic rings separated by an unbranched C4-chain have been isolated from the leaves of the Philippine endemic plant *Ficus nota*. The structures were determined on the basis of spectroscopic data as well as X-ray crystallographic analysis. The isolated compounds were evaluated for their antibacterial activity against *Bacillus subtilis*. Ficusnotins A–F (1-6) exhibited modest antibacterial activity against *B. subtilis* with IC_{50} values of 97, 155, 578, 152, 116, and 54 μM, respectively.

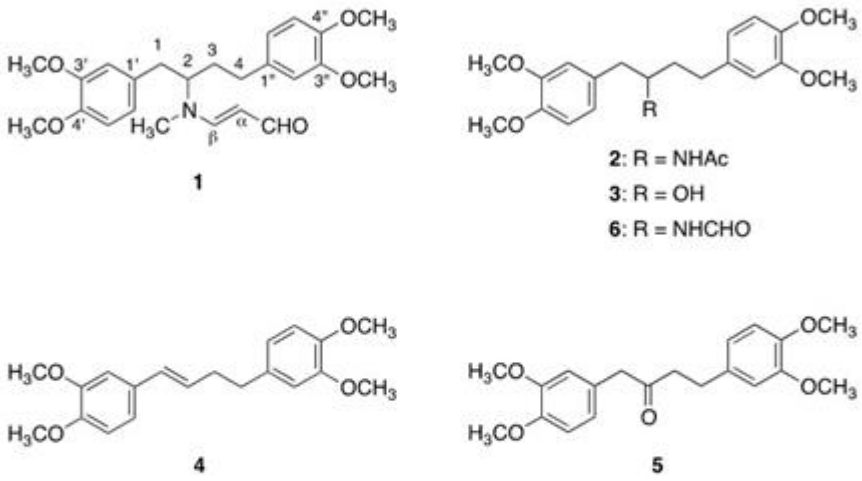


Fig. 1. Structures of Ficusnotins A-F from *F. nota.*

PS2-B-146

The impact of chronic low-dose Δ⁹-THC administration during adolescence on psychotic-like behaviour of adult rats

Charalampos Brakatselos[1], Nafsika Poulia[1], Foteini Delis[1], Alexia Polissidis[2], Katerina Antoniou[1]

[1]Department Of Pharmacology, University Of Ioannina, Ioannina, Greece, [2]Laboratory of Neurodegenerative Diseases, Biomedical Research Foundation Academy of Athens, Athens, Greece

Introduction: Clinical observations and preclinical studies suggest that adolescent exposure to delta-⁹-tetrahydrocannabinol (Δ⁹-THC), the main psychoactive component of cannabis, is associated with the emergence of central nervous system pathologies in adulthood, which may also involve the exacerbation of sensitivity to other drugs of abuse.
Aim: To determine if chronic adolescent low-dose Δ⁹-THC administration affects psychosis-associated behavioral parameters of adult rats.
Methods: From post-natal day (PND) 35 to PND 45 adolescent male rats received escalating low-dose Δ⁹-THC treatment twice daily (0.3 mg/kg PND 35–37; 1 mg/kg PND 38–41; 3 mg/kg PND 42–45) or vehicle.
During adulthood (PND>75): A) Evaluation of adolescent Δ⁹-THC treatment on adult motor response in the open field, after an acute d-amphetamine dose (1 mg/kg), which acts as an indirect dopaminergic agonist, or an acute ketamine dose (25 mg/kg), which acts as a non-competitive NMDA receptor antagonist. B) Evaluation of sensorimotor gating, which is severely disrupted in schizophrenia, using the pre-pulse inhibition (PPI) test.
Results: Adolescent Δ⁹-THC treatment: A) dampened the motor effects of d-amphetamine and abolished those of ketamine during adulthood, as deduced by horizontal and vertical activity evaluation in the open field. B) Did not influence the filtering of sensorimotor information of adult rats, as evaluated in the PPI behavioural paradigm.
Conclusions: Adolescent low-dose Δ⁹-THC administration leads to specific changes in motor response to d-amphetamine and ketamine, but not in a way that imitates hypersensitivity to these drugs as observed in specific psychopathologies. Moreover, the absence of significant changes in the PPI test, during adulthood, suggests that adolescent Δ⁹-THC-induced long term behavioural changes do not correspond to aspects of positive psychotic-like symptomatology, as modeled by the PPI experimental procedure.

PS2-B-147

On the dual role of natural compounds in crop protection

Valeria Terzi[1], Caterina Morcia[1], Roberta Ghizzoni[1], Nesrine Salhi[2]

[1]Research Centre for Genomics and Bioinformatics (CREA-GB), Council for Agricultural Research and Economics, Fiorenzuola d'Arda 29017, Italy, [2]Facultè des Sciences de la Nature et de la Vie, Université Kasdi Merbah Ouargla, Ouargla 30 000, Algérie

In the frame of the Integrated Pest and Disease Management-IPDM, renewed interest is now focused on the antimicrobial potentialities of medicinal plant extracts, as fungicidal agents for crop protection [1,2]. In our work, we have evaluated the antifungal properties of different plant extracts derived from a panel of aromatic and medicinal plants. Moreover, the potential role of natural compounds as priming agents has been explored. More in detail, priming is an adaptive strategy triggered by different stimuli that enhance the defensive capacity of plants against environmental stresses. The evaluation of the priming role of natural molecules has been done in an experimental system in which barley plants, infected with *Blumeria graminis* (or not), were treated (or not) with sub-lethal concentrations of Tea Tree Oil, TTO, derived from the Australian tree *Melaleuca alternifolia* - alone or in mixture with eugenol and thymol. A whole transcriptome characterization of the barley plants and the physiological evaluation of the impact and priming properties of TTO and mixtures were done. The results obtained suggest that these natural compounds can have a double role in crop protection, thanks to their direct antimicrobial activity and to their capacity to stimulate plant response against pathogen attack.

Acknowledgements: This work has been supported by the Italian-Algerian collaborative project "Molecole naturali per una cerealicoltura sostenibile", Ministero degli Affari Esteri e della Cooperazione Internazionale, Direzione Generale per la Promozione del Sistema Paese.

References:

[1] Morcia C, Tumino G, Terzi V (2013). M.R. Abyaneh and M. Rai eds, 14: 401-427. ISBN 978-3-642-38075-4

[2] Morcia C, Mehani M, Salhi N, Nazari L, Khelil A, Bara A, Ghizzoni R, Tumino G, Terzi V (2015). Méndez-Vilas eds., pp.193-198. ISBN: 978-84-942134-6-5

PS2-B-148

Phytochemical profiling using LC–PDA–MS and determination of the antioxidant activity of date palm (*Phoenix dactylifera L.*) seed extracts (Kentichi cultivar) from Tunisia

Sahar Ben Youssef[1], Olfa Taktak[1], Kenn Foubert[2], Luc Pieters[2], Noureddine Allouche[1]

[1]Laboratory of organic of chemistry, Natural Substances Team (LR17E508), Faculty of Sciences of Sfax, University of Sfax, BP 1171. 3000, Sfax, Tunisia, [2]Natural Products & Food Research and Analysis (NatuRA), Department of Pharmaceutical Sciences, University of Antwerp, Universiteitsplein 1 2610 , Antwerp, Belgium

Date palm (*Phoenix dactylifera L.*) is the most economically important plant in the Middle East due to its nutritionally value. The Kentichi cultivar is rich in phenolic compounds. In the current study, the profile of flavonoid glycosides, procyanidins and saponins of date seed extracts of the Kentichi cultivar was determined using liquid chromatography with online photo diode array and mass spectrometric detection (LC–PDA–MS). The purification of the methanolic extract was performed by column chromatography using Sephadex LH-20 as a stationary phase. All the fractions were analyzed by PDA-MS. The good separation of fractions containing saponins allowed identifying a series of triterpenoid saponins and tetracyclic saponins. The methanolic extract showed a powerful antioxidant activity with IC_{50} = 30.03 µg/ml, which was comparable to that of BHT (IC50= 12.80 µg/ml). This potent activity was associated with a high total phenolic (330.01±4.56 mg GAE/g DW) and total flavonoid (15.23±0.67 mg QE/ g DW) levels in the methanolic extract. Consequently, date palm (*Phoenix dactylifera L.*) seeds, Kentichi cultivar, can be considered as excellent sources of bioactive compounds.

PS2-B-149

The cytotoxicity and phytochemical characterization of *Ipomoea pileata* Roxb. extracts

Piotr Gorski[1], Idowu Eniafe Fadayomi[1], Fidelia I Uche[1], Elisabete Pires[2], James McClaugh[2], Wen-Wu Li[1]

[1]Institute for Science and Technology in Medicine, Keele University, Stoke-on-trent, United Kingdom, [2]Chemical Research Laboratory University of Oxford, United Kingdom, Oxford, United Kingdom

Discovering new therapeutic agents to treat cancer is especially evident in the instance of ovarian cancer as there is a high prevalence of chemotherapy resistance. DNA methylation inhibition is a new potential area, as high levels of methylation play a key role in the altered expression of genes associated with cancer formation, progression and metastasis. *Ipomoea pileata* Roxb. [(synonym *Involucrata* P. Beauv.) family Convolvulaceae] a related species of the sweet potato (*I. batatas*) is a neglected and hardly studied member of the *Ipomoea* genus. Members of this genus have been previously used in traditional medicine to treat asthma, diabetes and nausea. This study was performed to elucidate the bioactive compounds found in *I. pileata* from Nigeria and helps discover phytochemicals which can be used in cancer treatment. Cytotoxicity testing determined the *I. pileata* leaves extract to be more active than the stem extract in the high grade serous ovarian OVCAR-8 cancer cells (IC_{50} values: 36.8 ± 6.5 µg/mL vs. 80.4 ± 18.3 µg/mL respectively) and in immortalized normal human ovarian epithelial (HOE) cells (33.8 ± 6.3 µg/mL vs. 253 ± 34.3 µg/mL respectively). However, the selectivity index (SI) showed that the stem extract was more cytotoxic specifically to cancer cells than leave extracts (3.2 vs 0.9 respectively). Further sub-fractionation of the leaves extract was performed, and the cytotoxicity and the SI of the fractions determined helping narrow down the bioactive compounds. The *I. pileata* extracts were also analysed using analytical HPLC, GC-MS and LC-MS/MS methods to determine the compounds present. This study has shown the presence of many compounds including 3-*O*-caffeoylquinic acid, 1,3-*O*-dicaffeoylquinic acid, 3,5-*O*-dicaffeoylquinic acid, ellagic acid and quercetin in *I. pileata* leaves and stem extracts for the first time, suggesting further work on elucidating their mechanisms of action and further potential uses as anti-cancer agents.

PS2-B-150

Role of plant polyphenols in intervening aging and associated events

Kanti Bhooshan Pandey

Csir-central Salt & Marine Chemicals Research Institute, Bhavnagar, India

Aging is a progressive functional inability with time leading to mortality. Advancing age invites several chronic complications including decline in immunity, memory, and hyperglycemia with increased susceptibility to disease. The biological mechanism(s) that underlie aging involve damage of vital cellular molecules followed by their accumulation that leads to altered cellular physiology. In our studies we have provided experimental evidence that many polyphenols such as resveratrol, catechins and quercetin found largely in variety of fruits and vegetables, possess significant potential in limiting the deleterious events of aging and promoting health. The upregulation of housekeeping compensatory mechanisms, restoration of redox homeostasis, modulation of cellular transporters are some of the key mechanisms in addition to modulation at gene level that establish these secondary metabolites as promising agents in combating the anticipated burden of aging and related manifestations. The information on absorption, metabolism, and the consequent bioavailability are still under investigation. However, despite the areas linking consumption of polyphenols and purported health benefits, the studies till date provide enough evidence to claim amelioration of age associated cellular impairments. Since polyphenols are naturally present in many fruits and vegetables, a diet rich in polyphenols may provide protection against age-associated complications.

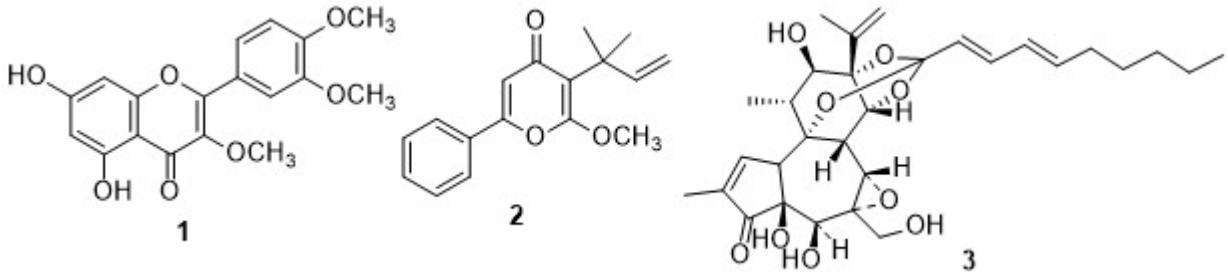
PS2-B-151

Antiplasmodial, antimicrobial and antiviral compounds from South African plants

Fanie Van Heerden, Nasir Tajuddeen, Wonder Mpapane, Jude Chukwujekwu

University of Kwazulu-Natal, Pietermaritzburg, South Africa

Natural products have always been a good source of antimalarial, antimicrobial and antiviral compounds. Herewith we want to report on our phytochemical investigation of South African plants which has led to the identification of several compounds with antiplasmodial, antibacterial and antiviral activity. The leaves of the fever tree, *Vachellia xanthophloea* (Fabaceae), led to the isolation of several quercetin derivatives of which the dimethyl and trimethyl derivatives have antiplasmodial activity. The trimethyl derivative (1) has an IC50 of 4.5 µg/mL. An investigation of *Hypericum roeperanum* (Hypericaceae) led to the isolation of hyperenone A (2) with antiviral properties. From the roots of *Gnidia splendens* (Thymelaceae), three diterpenes with anti-HIV activity were isolated, of which compound 3 shows the highest activity. In the presentation the role of ethnobotany in the selection of plants, the structures of the active compounds and structure-activity relationships will be discussed.



PS2-B-152

Kushenol E, a Prenylated Flavonoid, Attenuated Autophagy Maturation and Impaired Lysosome Localization through VCP/p97 Inhibition

Min Cheol Kwon, Sung-Kyun Ko, Jong Seog Ahn

Anticancer Agent Research Center, Korea Research Institute of Bioscience and Biotechnology (KRIBB), 30 Yeongudanjiro, Ochang, Cheongju, Korea

The root of *Sophora flavescens* (SF) has been used as a traditional herbal medicine to treat skin disease, gynecological diseases and cancer. Prenylated-flavonoids of SF have a variety of biological activity, but their activities are not fully investigated as anti-cancer activity. Autophagy, an evolutionarily conserved mechanism, is a catabolic lysosomal degradation pathway essential for cellular homeostasis and cell survival. In the course of screening for autophagy modulating compounds, we found kushenol E (KE), a prenylated-flavonoid from *S. flavescens*, suppressed tumor survival by regulating autophagy maturation and lysosome positioning. Dose- and time-dependent experiments of KE resulted in increased LC3B puncta and p62 accumulation. Indeed, immunostaining results of LAMP-1, a lysosomal marker, revealed that KE disrupts lysosome localization into peripheral site of cells. In target identification experiment using thermal shift assay (TSA), VCP/p97, an AAA-ATPase, was detected as the target of KE. VCP is known as a transporter of autophagosome and waste-cargo of proteasome. siVCP or overexpression of VCP dominant-negative (DN) form suppressed autophagy maturation like KE and DBeQ, a VCP inhibitor, by blocking lysosomal translocation. So, we suggest that KE is a potent anti-cancer agent by modulating autophagy and lysosome positioning as a VCP/p97 inhibitor.

Acknowledgements: The authors acknowledge financial support by grants International Joint Research Project (ASIA-16-011) of the NST (National Research Council of Science & Technology) and KRIBB Research Initiative Program from the funded by the Ministry of Science ICT (MSIT) of the Republic of Korea.

Keywords: *Sophora flavescens*, prenylated-flavonoid, kushenol E, autophagy induction, VCP inhibition

PS2-B-153

Natural Products with Antioxidant and Anticancer Potentials from Philippine's Biodiversity

Mylene Uy

Premier Research Institute of Science and Mathematics, Mindanao State University-Iligan Institute of Technology, Iligan City, Philippines

The Philippines is considered one of the megadiverse countries in the world. Its rich terrestrial and marine ecosystems, particularly in the Mindanao region have remained under-tapped and under-explored. The Philippines has about 1,500 medicinal plants from more than 13,500 plant species of which more than 3,500 are considered indigenous. Meanwhile, the Philippines, with its long coastal lines, has drawn on its marine capital only to a small extent. Only a few marine organisms collected from various parts of the Philippines have been documented and investigated in terms of their potential as source of bioactive secondary metabolites, particularly anticancer compounds. This paper presents the preliminary results of our studies on some Philippine medicinal plants and marine sponges with emphasis on their antioxidant and antiproliferative activities against various cancer cells lines.

PS2-B-154

Punicalagin quantification by CZE in extracts of *Combretum aculeatum* used traditionally for TB treatment

Assane E. Diop

University Of Geneva - Analytical Sciences, Geneva, Switzerland

Tuberculosis (TB) is an infectious bacterial disease caused by *Mycobacterium tuberculosis*, which most commonly affects the lungs. In the last years the number of resistant TB cases dramatically increased around the world [1]. In the search of novel lead compound or medicine an ethnopharmacological survey combined with a guided host-pathogen assay has highlighted an aqueous extract of *Combretum aculeatum* [2]. This extract showed significant antimycobacterial activity in an intracellular and extracellular model of infection with *M. marinum* [3]. Ellagitannins such as Punicalagin (PNG) and Punicacortein D were isolated and linked to the biological activity. Punicalagin has α- and β-anomers, which are mainly separated by HPLC while they are co-migrating in one peak by CZE. Capillary zone electrophoresis is the most common electrophoretic technique used in herbal medicines quality control. Internal standards approach was used for a better reproducibility on mobility and integral data [4].
Assessments of different methods were done in this work and the conditions below were used for the quantification: Fused silica capillary 75cm (65cm to detector), 75µm ID, running buffer Phosphate 25mM at pH 7.4, hydrodynamic injection of samples, 15kV as analysis constant voltage and the detection by UV absorbance at 280nm. A standard addition method was developed to avoid matrix effect and this method was applied on different batch of plants and different type of extracts. Concentrations of PNG were calculated using the ratio of peak areas between PNG and diclofenac as internal standard. The primary application for qualitative and quantitative analysis to extracts permits us to evaluate the content of PNG related to extraction duration, origins of herbs and harvesting period. This assessment in a limited number of samples should be repeated on a larger sample amount for a validation procedure.

Keywords: *Combretum aculeatum*, Punicalagin, Tuberculosis (TB), Capillary zone electrophoresis (CZE)

PS2-B-155

Exploring the Traditional Medicine of Atacama: an inestimable source of bioactive compounds.

M. Sergio Ortiz[1], Marylin Lecsö-Bornet[2], Christine Bonnal[3], Sandrine Houze[3], Ali Tahrioui[4], Emeline Bouffartigues[4], Sylvie Chevalier[4], Aikaterini Gioti[5], Roxane Tenta[5], Naveen Kumar[6], Thorsten Rose[6], Bernd Fiebich[6], Konstantina Stathopoulou[7], Maria Makropoulou[7], Nektarios Aligiannis[7], Sylvie Michel[1], Raphaël Grougnet[1], Sabrina Boutefnouchet[1]

[1]Laboratoire Pharmacognosie, Université Paris Descartes, Paris, France, [2]Laboratoire Ecosystème Intestinal, Probiotiques, Antibiotiques, Université Paris Descartes, Paris, France, [3]Laboratoire de Parasitologie-Mycologie, Hôpital Bichat Claude Bernard, Paris, France, [4]Laboratory of Microbiology Signals and Microenvironment, Normandy University, University of Rouen, Evreux, France, [5]Department of Nutrition & Dietetics, Harokopio University, Athens, Greece, [6]VIVACELL Biotechnology GmbH, Denzlingen, Germany, [7]Department of Pharmacognosy and Natural Products Chemistry, University of Athens, Athens, Greece

In northern Chile, culture of Atacameños called "Tradicion del desierto" is present. Few modern ethnobotanical studies related to Atacameñas communities are available with about 200 plant species for therapeutic uses reported [1]. In previous ethnobotanical surveys in this region, treatment of respiratory, gastro-intestinal and urinary disorders as well as pain and inflammation were among the most cited traditional indications. A collaborative work has been engaged within the Atacama Community of Taira (Ollagüe, Chile), in order to validate the use of their traditional medicinal plants. A collection of 18 plants has been selected for ethnopharmacological study.

Extractive task was envisaged using standardised ASE (Assisted solvent extraction) with EtOAc and MeOH. Extracts were screened for antibacterial, antifungal, antioxidant, anti-inflammatory and cytotoxic activities using a bio-guided approach.

Junellia serephioides, *Aloysia deserticola* and *Krameria lappacea*, traditionally used as anti-infectious revealed the presence of highly promising compounds. Bioguided fractionation allows us to isolate and identify triterpenoids from *J. serephioides* and *A. deserticola* and neolignans from *K. lappacea*. (+)-Conocarpan (1) presented an impressive MIC values varying from 1 to 5 µg/mL against various Gram-positive bacterial strains. In addition, regarding the pathogenicity of the Gram-negative *Pseudomonas aeruginosa PAO1*, compound 1 was able to inhibit the biofilm formation and at the same time, the pyocyanin production, which is a marker of the production of virulence factors of this bacteria.

Regarding anti-inflammatory activity, the apolar extracts of *Fabiana* species (*F. denudata* and *F. squamata*) and from *Artemisia copa* showed anti-inflammatory activity, inhibiting the production of mIL-6 and mTNFα in RAW264.1 in the range of 40-94 % at 50 µg/mL. EtOAc extract from *F. denudata* even inhibit 70% of mTNFα production at 1 µg/mL.

At last but not least, methanolic extract of *Chuquiraga atacamensis* presented high antiproliferative activity against A549, H1299, HCT116 and RT4 cancer cell lines (CC50: 6-0.6 µg/mL).

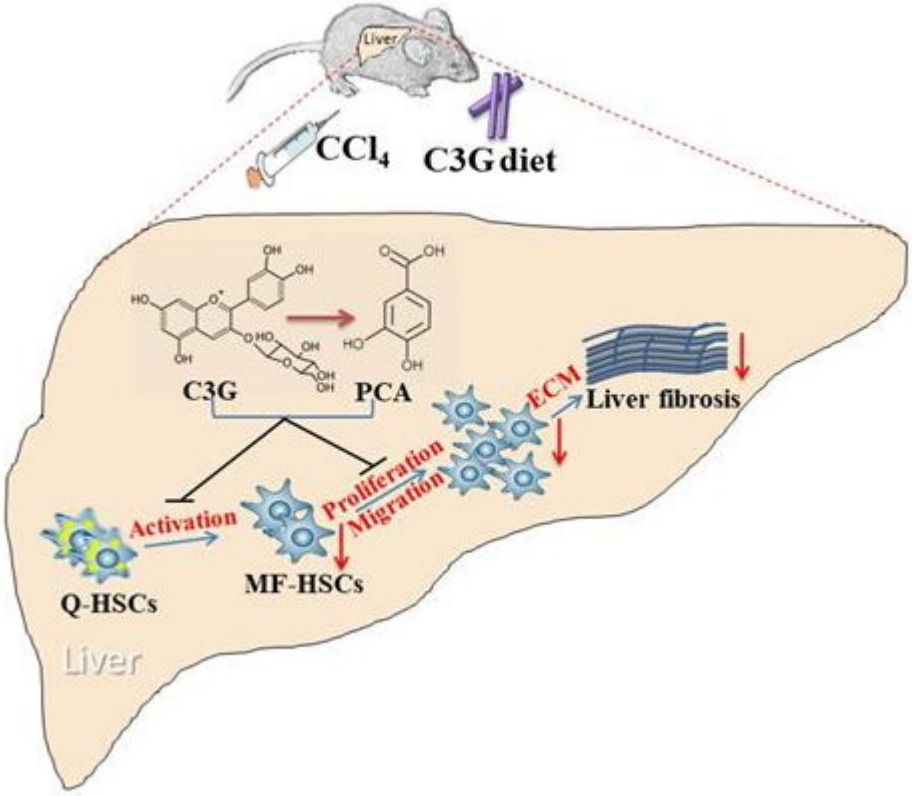
PS2-B-156

Cyanidin-3-*O*-*β*-glucoside combined with its metabolite protocatechuic acid attenuated the activation of mouse primary hepatic stellate cells *in vivo* and *in vitro*

Xinwei Jiang

Jinan University, Guangzhou, China

Previous studies indicated that cyanidin-3-*O*-*β*-glucoside (C3G) as a classical anthocyanin exerted an anti-fibrotic effect in the liver, but its bioavailability was quite low. This study was undertaken to explore the restraining effect of C3G and its metabolite protocatechuic acid (PCA) on the activation of hepatic stellate cells (HSCs). Our data demonstrated that the treatment of a carbon tetrachloride-treated mice model with C3G inhibited liver fibrosis and HSC activation. *In vitro*, both C3G and PCA preserved the lipid droplets and retinol in primary HSCs, and additionally inhibited the mRNA expression of α-smooth muscle actin and collagen I, but elevated the level of matrix metalloproteinase-2 and liver X receptors. Only PCA suppressed the levels of tumor necrosis factor alpha (TNF-α) and interleukin-6 (IL-6) secreted from HSCs significantly. In addition, C3G and PCA inhibited the proliferation and migration of HSCs. In conclusion, PCA mainly explained the *in vivo* inhibiting effect of C3G on HSC activation and liver fibrosis.



PS2-B-157

Evaluation of antioxidant and anti-inflammatory potential of various extracts from different parts of *Tanacetum cilicicum* (Boiss.) Grierson

Ali Şen[1], Aybeniz Yıldırım[1,2], Ahmet Dogan[3], Leyla Bitis[1]

[1]Department of Pharmacognosy, Faculty of Pharmacy, Marmara University, Istanbul, Turkey, [2]Department of Pharmacognosy, Faculty of Pharmacy, Adıyaman University, Istanbul, Turkey, [3]Department of Pharmaceutical Botany, Faculty of Pharmacy, Marmara University, Istanbul, Turkey

The aim of this study was to comparatively investigate the antioxidant and anti-inflammatory activities of various extracts from different parts of *Tanacetum cilicicum* (Boiss.) Grierson. In total 15 extracts, ethanol extracts and their n-hexane, chloroform, ethyl acetate, and aqueous ethanol fractions from the capitula, leaves, and stem of *T. cilicicum* were obtained using maceration and liquid-liquid extraction methods, respectively. The antioxidant activity of the extracts, was tested by DPPH, ABTS and FRAP methods and the results were expressed as IC50 values (50% inhibitory concentration) and mg of trolox equivalent per g dry extract. The total phenolic and flavonoid contents were determined by Folin-Ciocalteu method and AlCl3, and the results were expressed as mg of gallic acid and quercetin equivalents per g dry extract, respectively. Anti-inflammatory activity of extracts was evaluated against 5-lipoxygenase enzyme (5-LOX)[1]. Among all the extracts analyzed, ethyl acetate fraction of ethanol extract from the capitula of *T. cilicicum* (TCCEA) showed the highest DPPH and ABTS radical scavenging activity, with IC50 values of 22.44 and 30.86 µg/mL, respectively. TCCEA also possessed higher activity compared to other extracts in the ferric-reducing antioxidant power (FRAP) assay (42.02 mg TE/g extract). TCCEA and ethyl acetate fraction of ethanol extract from the leaves of *T. cilicicum* (TCLEA) were found to have the highest total phenolic and flavonoid contents. Also, TCCEA exhibited the best 5-LOX inhibitor activity with IC50 value of 9.44 µg/mL. These results indicate that the TCCEA possesses potent antioxidant and anti-inflammatory properties.

Keywords: *Tanacetum cilicicum*; antioxidant activity; anti-inflammatory activity.

References:

[1] Sen A . P. Mill. Marmara Pharmacy Journal, 2018; 22(2): 328–333.

PS2-B-158

A study on the antioxidant activity, total phenolic and flavonoid content of *Calepina irregularis (Asso)* Thell. (Brassicaceae)

Sevda Deniz Dalğın[1], Ali Şen[1], Gizem Bulut[2], Leyla Bitis[1]

[1]Department of Pharmacognosy, Faculty of Pharmacy, Marmara University, Istanbul, Turkey, [2]Department of Pharmaceutical Botany, Faculty of Pharmacy, Marmara University, Istanbul, Turkey

In this study, it was aimed to evaluate the antioxidant activity, total phenolic and flavonoid compound amounts of ethanol extract obtained from *Calepina irregularis* (Asso) Thell. (Brassicaceae) which are traditionally used for food purposes and also medical values around Diyarbakır/Silvan district of Turkey. Aerial parts of the plant were collected during the consumption time from Diyarbakır/Silvan and ethanol extracts were obtained by maceration. The antioxidant activity of the ethanol extract was determined by DPPH and ABTS radicals scavenging, metal chelating capacity and reducing power methods. Total phenolic and flavonoid contents were investigated by Folin Ciocalteu and $AlCl_3$ methods, respectively. *Calepina irregularis* ethanol extract showed a good antioxidant activity (IC_{50}: DPPH: 560.3, ABTS: 180.20 and metal chelating: 19.4 µg/mL, reducing power: 19.77 mg TE/g extract). Total phenol and flavonoids contents were found to be 46.50 mg gallic acid and 8.26 mg quercetin equivalents, respectively. Antioxidant activity and amounts of total phenolics and flavonoids of *C. irregularis* were investigated for the first time. According to these results, *C. irregularis* ethanol extract has been found to have good radical scavenging, reducing power and metal chelating activities. Therefore, the plant can potentially be used as a natural antioxidant source in food, cosmetic and pharmaceutical industries. However, there is a need for cytotoxic studies on the extract. In addition, a study can be conducted on the identification of the active compounds responsible for the antioxidant activity [1].

Acknowledgements: This work was supported by the Research Fund of the University of Marmara, Project No SAG-C-YLP-131217-0663.

Keywords: *Calepina irregularis*, antioxidant activity, total phenolic contents, total flavonoid contents

References:

[1] Dalğın SD, Bitis L (2018). An Investigation of Antioxidant Activities of some Wild Edible Medicinal Plant Species in Silvan District of Diyarbakır Province, Master Thesis, Marmara University, Institute of Health Sciences, İstanbul.

PS2-B-159

Evaluation of anti-inflammatory activities of some medicinal plants

Leyla Bitiş, Ali Şen

Department of Pharmacognosy, Faculty of Pharmacy, Marmara University, Istanbul, Turkey

Medicinal plants have been used by people for a long time to treat different inflammatory conditions and, especially, skin inflammations. The aim of this study was to evaluate the anti-inflammatory activities of ethanol extracts obtained from *Eryngium campestre* L. var. virens (Link) Weins (EC), *Lathyrus cicera* L. (LC), *Mentha longifolia* subsp. *thphoides* var. thphoides (Brig. Harley) (ML), *Ranunculus muricatus* L. (RM), *Rumex acetocella* L. (RA). Shade-dried ground plants were macerated with EtOH, using an ultrasonic bath. The solvent was evaporated using a rotary evaporator at 40 °C. *In vitro* anti-inflammatory activity of the obtained ethanol extracts was measured by lipoxygenase inhibition assay [1,2]. ML extract at a concentration of 20 µg/mL showed highest anti-lipoxygenase activity with inhibition rate of 46.45%, followed by RM (32.63%), RA (6.59%), EC (4.34%) and LC (3.03%) extracts, respectively. ML extract showed strong anti-lipoxygenase activity with an IC_{50} value of 21.36 µg/mL. These results indicate that *Mentha longifolia* subsp. *thphoides* var. thphoides has significant anti-inflammatory activity.

Keywords: Medicinal plants, ethanol extract, anti-inflammatory activity

References:

[1] Sen A. Marmara Pharmacy Journal, 2018; 22(2): 328–333.
[2] Dalğın SD, Bitis L (2018). An Investigation of Antioxidant Activities of some Wild Edible Medicinal Plant Species in Silvan District of Diyarbakır Province, Master Thesis, Marmara University, Institute of Health Sciences, İstanbul.

PS2-B-160

Insecticidal potential of aqueous extracts from *Dittrichia viscosa*, *Cistus villosus*, *Rosmarinus officinalis* and *Lavandula stoechas* from the Tlemcen area (Algeria)

Ahmed Taibi, Choaib Lokbani, Mohammed Zerriouh

Department of Agronomy, faculty SNV/STU, University of Tlemcen, Tlemcen, Algeria

In the search for new strategies for insect management in agricultural systems, a great interest is to use the plant extracts to replace or integrate insect control. The objective of this research is the formulation of a biological pesticide to treat bean aphids (*Aphis fabae*) by aqueous extracts based on plant species harvested in the Tlemcen area *Lavandula stoechas*, *Rosmarinus officinalis*, *Cistus villosus* and *Dittrichia viscosa*. These extracts are used either singly or in combination by contact on the aphid to estimate the mortality rate. For this purpose and according to laboratory analyses noted that the aqueous extract of *Cistus* and *Dittrichia* simple or in combination give important results and allowed us to say that the biopesticide based on both plants are effective against black aphid of the bean.

PS2-B-161

Insecticidal effects of some *Sinapis* aqueous extracts on the control of *Aphis fabae* and *Myzus percisae*

Ahmed Taibi, Amel Bechta

Department of Agronomy, faculty SNV/STU, University of Tlemcen, Tlemcen, Algeria

The present research is conducted under laboratory condition; it has encountered the study of the insecticidal effect of aqueous extracts at the base of 2 mustard plants (*Sinapis arvensis* and *Sinapis alba*). The aqueous extracts are tested by contact and spray on aphids (*Aphis fabae* and *Myzus persicae*). This insecticide on *Aphis* makes aqueous extracts of *Sinapis alba* records a significant mortality in the first observation, it increases with time to reach up to 93.3% by contact and 100% by spray after 72 hours. *Sinapis alba* has a mortality rate of *Myzus persicae* which increases with time, reaching 83.3% by contact and 86.7% by spray after 72 hours. The chi-2 test show a significant difference between the aqueous extracts of the plants on the corrected after 72 h of the two species of aphids. There is an effectiveness of the aqueous extract of mustard plants separated from the mixture. We noted an action in favor of *Aphis fabae* (up to 100%) compared to *Myzus persicae*.

Keywords: Sinapis arvensis, Sinapis alba, Aphis fabae, Myzus persicae, aqueous extracts, Tlemcen.

PS2-B-162

Activation of silent natural product biosynthetic pathways in fungi using epigenetic modifiers

Mohammed Aldholmi[1,2], A Ganesan[1]

[1]School of Pharmacy, University of East Anglia, Norwich Research Park, Norwich NR4 7TJ, United Kingdom, [2]Natural Products and Alternative Medicine, College of Clinical Pharmacy, Imam Abdulrahman Bin Faisal University (University of Dammam), Dammam 31441, Saudi Arabia

Since most genes responsible for the production of secondary metabolites are silent under laboratory conditions, there is great interest in activating these gene clusters. The epigenetic processes including the modifications of DNA and amino acids on histones have been revealed to be extensively used by organisms to regulate the expression of genes involved in natural product biosynthesis. Hence, it is possible to influence these processes during microbial fermentation by the addition of chemical epigenetic modulators. Five Fungal strains were cultivated in the presence of HDAC inhibitors (e.g. SAHA) or DNMT inhibitors (e.g. 5-Azacytidine). Morphological changes were monitored for seven days before extraction with Ethyl acetate or Methanol. The extracts were run on HPLC and LC-MS, and the metabolic profiles were compared using chromatogram overlay and Volcano plots. The overlayered chromatograms and volcano plots presented significant differences between the cultures treated with epigenetic modifiers and control cultures, in addition to differences in morphology and colour. The extract from one of the fungal cultures contained two secondary metabolites significantly induced with 5-Azacytidine and slightly induced with SAHA. Interestingly, the induction of these metabolites was doubled when the culture was treated with both 5-Azacytidine and SAHA suggesting a synergistic effect of these modifiers on secondary metabolism. MS analysis of the first metabolite provided molecular ions in both positive and negative mode corresponding to M+H (331.1950) and M-H (329.1840), respectively. The second metabolite was only ionizable in the negative mode providing a molecular ion with a mass of 681.3420. Databases were searched using the accurate mass and UV λmax , but no natural products were found matching both accurate mass and UV profile. Therefore, large-scale fermentation is in progress to isolate the compounds and elucidate their structure. Subsequently, the pure compounds will be tested for antifungal, antibacterial and cytotoxic activity.

Acknowledgements: I would like to thank Imam Abdulrahman Bin Faisal University for financial support to do my PhD in the UK.

Keywords: Natural products, epigenetic, secondary metabolites, fungi

References:

[1] Felsenfeld G, Groudine M. Nature 2003; 421(6921): 448–53.
[2] Cichewicz RH. Nat Prod Rep 2010; 27(1): 11–22.
[3] Williams RB, Henrikson JC, Hoover AR, Lee AE, Cichewicz RH. Org Biomol Chem 2008; 6(11): 1895–7.
[4] Wang X, Sena Filho JG, Hoover AR, King JB, Ellis TK, Powell DR, *et al*. J Nat Prod. 2010; 73(5): 942–8.
[5] Yang X-L, Huang L, Ruan X-L. J Asian Nat Prod Res 2014; 16(4): 412–7.
[6] González-Menéndez V, Pérez-Bonilla M, Pérez-Victoria I, Martín J, Muñoz F, Reyes F, *et al*. Molecules 2016; 21(2).

PS2-B-163

The *Crocus sativus* compounds *trans*-crocin 4 and *trans*-crocetin modulate the amyloidogenic pathway and tau misprocessing in Alzheimer disease neuronal cell culture models

Ioanna Chalatsa[1], Demetrios A. Arvanitis[2], Nikolaos Stavros Koulakiotis[3], Athina Giagini[1], Alexios Leandros Skaltsounis[4], Zeta Papadopoulou-Daifoti[5], Anthony Tsarbopoulos[3,5], Despina Sanoudou[1,2]

[1]Clinical Genomics and Pharmacogenomics Unit, 4th Department of Internal Medicine, Medical School, National and Kapodistrian University of Athens, Athens, Greece, [2]Molecular Biology Division, Center for Basic Research, Foundation for Biomedical Research of the Academy of Athens, Athens, Greece, [3]GAIA Research Center, Bioanalytical Department, The Goulandris Natural History Museum, Athens, Greece, [4]Department of Pharmacognosy and Natural Product Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [5]Department of Pharmacology, Medical School, National and Kapodistrian University of Athens, Athens, Greece

Crocus sativus L., also known as saffron, has been shown to have a therapeutic potential for different illnesses including central nervous system diseases. Since Alzheimer's Disease (AD) is the most common form of dementia in the elderly, with a current estimate of 47 million patients worldwide [1], we studied the molecular effects of the *Crocus sativus* natural compounds *trans*-crocin 4 and *trans*-crocetin on the key pathways of AD development, namely amyloidogenesis and tau phosphorylation. Two well-established AD neuronal cell models were used namely, differentiated SH-SY5Y cells overexpressing APP [2] and differentiated PC12 cells overexpressing hyperphosphorylated tau [3]. Biologically relevant concentrations of *trans*-crocin 4 and *trans*-crocetin, ranging from 0.1 μM to 1 mM, applied for 24h or 72h, were well tolerated by differentiated wild type SH-SY5Y and PC12 cells. Treatment of the differentiated SH-SY5Y-APP with *trans*-crocin 4 significantly decreased BACE1 and APP-C99, suggesting the suppression of the amyloidogenic pathway. Meanwhile it markedly decreased PSEN complexes, the catalytic components of *γ*-secretases that generate toxic beta-amyloid peptides. Similarly, *trans*-crocetin treatment led to a significant reduction of BACE1 and PSEN complexes. Studies of the neuronally differentiated PC12-htau cells, showed that both compounds were effective in suppressing the active forms of GSK3β and ERK1/2 kinases, as well as significantly reducing total tau and tau phosphorylation on the pThr231 and pSer199/Ser202 epitopes. In summary, the present findings demonstrate that treatment with *trans*-crocin 4 or *trans*-crocetin can reverse a series of protein changes observed in APP misprocessing and tau hyperphosphorylation, supporting their preventive and possible therapeutic potential against AD.

PS2-B-164

Sideritis scardica and *Cichorium spinosum* reduce amyloidogenesis and tau aggregates in Alzheimer's disease models

Demetrios A. Arvanitis[1], Ioanna Chalatsa2, Eleni V. Mikropoulou[3], Athina Giagini[2], Nektarios Aligiannis[3], Maria Halabalaki[3], Zeta Papadopoulou-Daifoti[4], Anthony Tsarbopoulos[4,5], Alexios Leandros Skaltsounis[3], Despina Sanoudou[1,2]

[1]Biomedical Research Foundation Academy Of Athens, Athens, Greece, [2]Clinical Genomics and Pharmacogenomics Unit, 4th Department of Internal Medicine, Medical School, National and Kapodistrian University of Athens, Athens, Greece, [3]Department of Pharmacognosy and Natural Product Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [4]Department of Pharmacology, Medical School, National and Kapodistrian University of Athens, Athens, Greece, [5]GAIA Research Center, Bioanalytical Department, The Goulandris Natural History Museum, Athens, Greece

Key components of the Mediterranean diet, the decoction of *Sideritis scardica* leaves, commonly known as "mountain tea", and the wild edible greens (chórta) of Crete, *Cichorium spinosum*, an endemic Mediterranean plant also known as "stamnagkathi", represent traditional remedies for thousands of years [1,2]. Herein, we demonstrate that treatment of cell line models of Alzheimer's disease (AD) with *S. scardica* or *C. spinosum* extracts modulates APP misprocessing and tau hyperphosphorylation. In specific, we initially analyzed the extracts' composition using LC-HRMS methods. The extracts were then applied to SH-SY5Y and PC12 neuronal-like cells to determine the maximal tolerated concentration over time. The optimal concentrations were studied for their effects on the key molecules of the amyloidogenic and tau misprocessing pathways, in the SH-SY5Y-APP and PC12-htau cells, respectively. We found that both the *S. scardica* and *C. spinosum* extracts are likely to promote APP processing through the alpha, non-amyloidogenic pathway, while they both significantly decrease total tau, the activation of the GSK3β, ERK1 and/or ERK2 kinases of tau, as well as tau hyperphosphorylation. Collectively, our data suggest that *S. scardica* and *C. spinosum* extracts could have protective effects in different AD models and may be important supplements in an AD preventive or curative diet.

PS2-B-165

Screening of plants extracts from Reunion Island as biopreservatives for cosmetic applications

Laure-Anne Peyrat, Gwenn Atheaux, Nicolas Gaboriaud-Kolar

Bioval Ocean Indien, Montpellier, France

Located in the Indian Ocean, Reunion Island is considered as a hot spot of biodiversity. With a large diversity of natural environments, this island presents a high level of endism with more than 850 native plants species including 232 endemic plants. The Reunion flora is notably characterized with the large presence of epiphytic plants such as Orchidaceae or Bromeliaceae. Since 2010, this Island and its biodiversity are listed on the UNESCO World Heritage. Locally, around 200 plants are used for medical applications and 19 have been recently registered in the French Pharmacopoeia.1 Nowadays, the public awareness about more natural ingredients in food or cosmetics has increased. Moreover, the recent reports pointing out health-threatening chemical preservatives (formaldehyde, parabens…) have deeply impacted these market sectors. Therefore, active research is led to find innovative and natural alternatives. Among them, some natural extracts or natural products are now commercialized such as grapefruit seed extract, naticide (PlantaservQ®), benzoic acid or radish root ferment. However, these products have a weak long-term efficacy and are then more appropriate for home-made cosmetics. Bioval Ocean Indien is specialized in the use of green extraction techniques applied to the valorization of plants and by-products from Reunion Island. In order to find new sources of biopreservatives, we undertook a study including 60 plants and agricultural by-products. A total of 87 original extracts have been produced using solely green solvent (ethanol). The extracts were tested for their antioxidant, antiradical and antibacterial activities. Among them, 16 displayed similar or better activities than the commercialized natural products. Interestingly, some of them combined both antibacterial and antiradical/antioxidant properties highlighting them as interesting candidates for further investigations (other green solvents, process, biological evaluation…).

PS2-B-166

Quantitative determination of the prodrug of the naturally occurring antibiotic colistin in formulations

Ioanna Dagla[1], Evangelia Karkoula[2,3], Eirini Baira[1], Anthony Tsarbopoulos[2,3], Evagelos Gikas [1,3]

[1]Faculty of Pharmacy, School of Health Sciences, National And Kapodistrian University Of Athens, Panepistiomiopolis, Zografou, 157 71, Athens, Greece, [2]Medical School, National and Kapodistrian University of Athens, 115 27, Athens, Greece, [3]GAIA Research Center – The Goulandris Natural History Museum, 145 62, Kifissia, Greece

Colistin is a lipopeptide antibiotic, produced by *Bacillus polymyxa* var. *Colistinus* [1]. Colistin A and B are its main constituents, which are differentiated to the length of the lipid acid side chain [2]. Colistin is used mainly in the form of prodrug - colistimethate sodium (CMS), and it is widely used as a last resort antibiotic for severe infections caused by resistant gram-negative bacteria. CMS is formed by a sulphomethylation reaction and the final prodrug is an extremely complex mixture of molecules, containing a varying degree of substitutions in the five amino moieties of colistin [3]. Due to its complexity, no chemical quality control methods for the quantitative assessment of the various forms of drug in formulations have been developed. Until now, the quality control methods are based on microbiological methods raising concerns about toxicity and bioavailability of colistin [4]. A new UPLC-QTOF quantitative method has been developed for the determination of colistin in commercial prodrugs based on its mass spectral profiles, using selected ion peaks as quality markers. The method is validated according to the ICH procedures for the final prodrugs.

Keywords: colistin, colistimethate sodium, QTOF, quantitation

References:

[1] Suzuki T, Fujikawa K. J Biochem 1964; 56: 182–189.
[2] Govaerts C, Rozenski J *et al.*, Rapid Commun Mass Spectrom 2002; 16: 823–833.
[3] McMillan FH, Pattison IC, J Pharm Sci 1969; 58: 730–737.
[4] Sivanesan S, Roberts K *et al.*, J Nat Prod 2017; 80: 225–229.

PS2-B-167

Ethnopharmacological study of plants from the greek flora for the discovery of cytotoxic agents and inhibitors of acetylcholinesterase, hyaluronidase and phospholipase

Rozalia Michalea[1], Vasiliki Dedousi, Panagiotis Polychronopoulos[1], Konstantina Stathopoulou[1], Dimitra Benaki[2], Emmanuel Mikros[2], Nektarios Aligiannis[1]

[1]Department of Pharmacognosy & Natural Products Chemistry, Faculty of Pharmacy, University of Athens, Athens, Greece, [2] Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Athens, Athens, Greece

The contribution of ethnopharmacology in the discovery of new drugs is important and has led to findings such as galanthamine and artemisinin. Two texts of two Greek physicians–grammarians, Nicander of Colophon (2nd century BC) and Nikolaos Ieropais (17th century) were the source of this study. The ancient poem of the first writer, Theriaca, the translation of which was accomplished in 2014, is describing venomous creatures, the causes of their envenomation and several herbs used as antidotes [1]. The second work consists of different chapters depending on the pathological situation such as malignancies, where the suitable therapeutic plants are presented. The exhaustive study of them led to the interpretation of 217 medicinal plants and two lists of plant species with anticancer and anti-venom properties were developed, having some species in common. Thirty one of them were collected and extracted with different solvents. Three hydrolytic enzymes, acetylcholinesterase, hyaluronidase and phospholipase, involved in snake envenomation, and three cancer cell lines and a normal one constituted the pharmacological targets. After chemical profiling and biological evaluation of crude extracts, the hydroalcoholic extracts of the roots and aerial parts of *Paeonia parnassica* (Paeoniaceae), *Aristolochia hirta* (Aristolochiaceae), the aerial parts of *Teucrium divaricatum* ssp. *divaricatum* (Lamiaceae), *Adiantum capillus-veneris* (Pteridaceae) and *Anchusa undulata* ssp. *hybrida* (Boraginaceae) were selected for phytochemical analysis. They were fractionated through FCPC and the fractions were evaluated for their content and therapeutic potency. A bioactivity-guided-NMR dereplication method was performed for three extracts in order to detect their bioactive metabolites prior to any isolation. Finally the purification, identification and biological assessment of 50 compounds from the seven extracts were accomplished. They belong to several chemical categories e.g. phenolics, flavonoids, iridoids, terpenoids etc. and some of them such as pentagalloyl glucose, caffeoyl-malic acid, teucrioside exhibited considerable inhibitory activities of the enzymes.

PS2-B-168

Chemical Constituents from the Stem Bark of *Pentaclethra macrophylla* Benth (Fabaceae)

Augustine Ahmadu[1], Chinaka Nnennaya[2], Sunday Garba[2], Nikolaos Tsfantakis[3], Nikolas Fokaliakis[3]

[1]Kaduna State University, Kaduna, Nigeria, [2]National Kapodistrian University of Athens, Athens, Greece, [3]National Kapodistrian University of Athens, Athens, Greece, [4]University of Portharcourt, Portharcourt, Nigeria, [5]University of Portharcourt, Portharcourt, Nigeria

Plants have successfully served as source of lead discovery in drug development; there is the need to look into our ethnomedicinal plants for the purpose of identifying bioactive antibacterial agents to combat the growing antibacterial resistance to current drugs. The present study investigated the antibacterial activity of the extract and soluble fractions the *Pentaclethra macrophylla* used in Nigerian ethnomedicine to treat infectious diseases, diarrhea and inflammation, against some selected pathogens and the isolation and structure elucidation of three compounds using different chromatographic and spectroscopic techniques. The antibacterial activity was evaluated on the crude hydroalcoholic extract, ethylacetate and n-butanol fractions against five pathogenic bacteria using the agar diffusion assay method. The ethylacetate soluble fraction at 10 mg/ml showed a good activity against *B.subtilis* and *S.aureus* with inhibition zone diameter of 16.0 and 13.5 mm compared with Levofloxacin (1.5 µg/ml) having diameter of 21.1 and 16.6 mm respectively. The crude hydroalcoholic and n-butanol fraction proved to be less active against *B.subtilis*, with zones of inhibition of 8±2.89 and 9.5±1.78mm. None of the extract or fractions showed activity against *K.pneumoniae*. The active ethylacetate and n-butanol fractions were subjected to column chromatography and subsequent purification over sephadex LH-20 afforded methyl gallate (I), Bergenin (II) and 11-*O*-galloylbergenin (III). The structures of these compounds were elucidated using NMR and MS and are reported in this plant for the first time.

Keywords: *Pentaclethra macrophylla*, antibacterial, Methyl gallate, Bergenin and 11-O-galloylbergenin.

References:

[1] Bhat ZA. Journ of Pharm Res 2012; 5(5): 2457–2459.
[2] Calixto JB. Braz Journ of Med Biol Res 2000; 2: 179–189.
[3] Iwu IC, Uchegbu R. Int Journ of Emerging Knowl 2014; 1(10): 39–44.
[4] Katiyar C, Gupta A, Kanjilal S, Katiyar S. Int quaterly Journ of Res in Ayurveda 2012; 33(1): 10.

PS2-B-169

Dendropanax morbifera extract ameliorates thioacetamide-induced hepatic fibrosis via TGF-β1/Smads pathways

Kyeong Seok Kim, Hae Ri Kim, Jong Seung Im, In Su Kim, Hyung Sik Kim

School of Pharmacy, Sungkyunkwan University, Suwon, South Korea

The aim of this study was to assess whether aquatic extract of *Dendropanax morbifera* (DM) ameliorates thioacetamide (TAA)-induced hepatic fibrosis. Hepatic fibrosis was induced by an intraperitoneal (i.p.) injection (60 mg/kg, twice per week) of TAA for 6 weeks. After the TAA injections, DM (60 mg/kg/day) or silymarin (50 mg/kg/day) was administered daily for 6 weeks by oral gavage. Hepatic damage was evaluated by measuring markers of hepatic function such as aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), and gamma-glutamyl transferase (rGTP) in the serum. The levels of transforming growth factor β1 (TGF-β1), 4-hydroxyproline, oxidative stress, and α-smooth muscle actin (α-SMA) expression were also evaluated as markers of hepatic stellate cell (HSC) activation. DM and silymarin markedly attenuated TAA-induced liver injury, as indicated by a significant reduction in serum activities of AST, ALT, ALP, and r-GTP. DM administration markedly decreased TAA-induced apoptosis of hepatocytes. DM and silymarin significantly ameliorated the TAA-mediated reduction in total glutathione (GSH) concentration, diminished superoxide dismutase (SOD) and catalase (CAT) activity, and reduced hepatic malondialdehyde (MDA) concentration. TAA enhanced protein expression of α-SMA, and administration of DM significantly reduced expression of type I collagen and vimentin. DM and silymarin markedly inhibited expression of TGF-β1 and p-Smad2/3 in rats with TAA-induced fibrosis. The protective effects of DM on liver fibrosis were clearly demonstrated in 4-hydroxyproline concentration and histopathological examination in the liver. Our results revealed that DM prevented hepatic fibrosis by inhibiting TAA-mediated oxidative stress through the induction of antioxidant responses and the inhibition of the TGF-β1/Smads signaling pathways.

PS2-B-170

Afrocyclamine A, a triterpenoid saponin, induces apoptosis and autophagic cell death via the PI3k/Akt/mTOR pathway in human prostate cancer cells

Sachan Richa, Ji Yeon Son, Hae Ri Kim, Hyung Sik Kim, In Su Kim

School of Pharmacy, Sungkyunkwan University, South Korea

Afrocyclamin A (AnumN-19), an oleanane-type triterpene saponin, was isolated from *Androsace umbellate*, a vegetable also used as a traditional herbal medicine. The aim of this study was to investigate the anticancer activity of AnumN-19 on human prostate cancer cells. Cytotoxicity, cell cycle distribution, apoptosis, and autophagic cell death were measured following exposure to AnumN-19. The *in vivo* antitumor activity of AnumN-19 was assessed in a xenograft model. The protein expression levels of pAKT, mTOR, Bax, Bcl-2, caspase-3, and caspase-9 were detected using western blot analysis. AnumN-19 increased cytotoxicity, caused changes in cell morphology and induce sub-G0/G1 phase arrest which indicating an increase in the number of apoptotic cells in DU145 cells. AnumN-19 robustly induced autophagy in DU145 cells, as demonstrated by the conversion of LC3B-I to LC3B-II, & the formation of autophagic vacuoles as revealed by western blot analysis and fluorescence staining respectively. AnumN-19 also inhibited the phosphorylation of PI3K, AKT, and mTOR, suggesting their role in AnumN-19 induced cell death. In addition, AnumN-19 also inhibited cell migration and invasion in a concentration and time-dependent manner. In an in vivo xenograft model AnumN-19 inhibited the growth of DU145 cells. This study shows that AnumN-19 exerts its anticancer activity through the PI3K/AKT/mTOR pathway leading to cell death. A strong foundation for further research will be needed to establish a theoretical basis the design and development of new anticancer drugs.

PS2-B-171

The beneficial effect of natural compounds in the treatment of post-menopausal osteoporosis.

A. Vontzalidou[1,2], M. Makropoulou[1,2], A. Meligova[2], D.J. Mitsiou[2], S. Mitakou[1], M.N. Alexis[2], N. Aligiannis[1]

[1]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece, [2]Institute of Biology, Medicinal Chemistry and Biotechnology, National Hellenic Research Foundation, Athens, Greece

Osteoporosis is a menopause-associated degenerative disease characterized by progressive bone loss affecting millions of women worldwide. Treatments include hormone replacement therapy (HRT) and selective estrogen receptor modulators (SERMs). However, nowadays traditional herbal medicines and plant derived supplements are increasingly used as a "natural" alternative to drugs to prevent post-menopausal disorders, including osteoporosis [1]. Our aim was the discovery of novel osteoprotective natural agents deriving from Greek flora, exploiting information from traditional medicine sources, ancient Greek physicians and current literature, concerning the beneficial effects of plants in bone health [2]. To this direction, 78 plant species were selected, 144 extracts were prepared and evaluated using well-validated cellular models of menopause-related diseases. Preliminary phytochemical and biological screening revealed *Iris* sp., *Rhamnus* sp., *Lupinus albus*, *Psoralea bituminosa*, *Ceratonia siliqua*, *Hippocrepis comosa* and *Cytisus villosus* between the most interesting extracts. Consequently, their major constituents were isolated and their structure was elucidated by NMR spectroscopy. In total, 84 compounds from various chemical categories were identified, whereas 8 were characterized as new natural products. All isolated compounds were in vitro evaluated using a cell–based screening concerning: a) the differentiation of MC3T3-E1 cells to osteoblasts b) the inhibition of differentiation of RAW264.7 macrophages to osteoclasts c) their estrogenic properties using MCF-7 and Ishikawa cell lines. According to the combined results phenolic acids (vanillic acid, gallic acid), benzophenones (iriflophenone) terpenes (β-amyrin, lupeol, stigmastenone) coumestans (coumestrol, 2-α,α-dimethylallyl-coumestrol, isotrifoliol) coumarin, iridal and isoflavone derivatives were considered as the most promising agents for potentially preventing and treating post-menopausal osteoporosis.

Acknowledgments This study has been carried out with the financial support Thalis 2011 programme "SERMENCO MIS 375617". This Programme is co-funded by the European Development Fund and National Resources

References:

[1] Evid Based Complement Alternat Med. 2013; Article ID 356260, 11 pages
[2] Am J Chin Med. 2012; 40(6): 1289–1305.

PS2-B-172

Exacerbative effect of *Paullinia pinnata* methanol leaf extract on ethylene glycol monomethyl ether-induced testicular dysfunction in male Wistar rats

Oluwatoyin Adeyemo-salami

Department of Biochemistry, College of Medicine, University of Ibadan, Oyo Stae, Nigeria, Ibadan, Nigeria

Paullinia pinnata (PP) is a medicinal plant whose parts are used for medicinal purposes traditionally in the treatment of various diseases including malaria, diarrhea and to help infertility which is currently a scourge globally. The aim of this study is to validate the traditional use of *P.pinnata* leaves in the treatment of infertility using ethylene glycol mono methyl ether (EGME)-induced reproductive dysfunction model.

Sixty adult male Wistar rats were weight-matched into six groups of ten animals each. All administrations were done orally daily for twenty-one consecutive days as follows: Group I (control) – distilled water; Group II – 1.5 ml/kg body weight of 10% dimethyl sulfoxide (vehicle); Group III- EGME only (200 mg/kg); Group IV- PP only (200 mg/kg); Group V- EGME+PP at 100 mg/kg body weight and Group VI- EGME+PP at 200 mg/kg body weight. On day 22, blood was collected for the analysis of the reproductive hormones. The animals were euthanized, dissected, and the testes, epididymes, seminal vesicles, prostate gland and brain were excised and weighed. The brain, testes and epididymes were processed and used for spermatozoa analysis, antioxidant and anti-inflammatory assays, and histological examination appropriately.

The plasma concentrations of the reproductive hormones including luteinizing hormone were significantly increased in the co-administered groups while the plasma testosterone concentration was decreased. Similarly, the spermatozoa parameters including testicular spermatozoa number were reduced in the EGME only and co-administered groups. Antioxidant parameters including catalase and glutathione-*S*-transferase were affected in the epididymis, testes and brain in the EGME only and co-administered groups. The inflammatory markers were also elevated. The results were complemented by the histological observations.

Paullinia pinnata leaves lack chemopreventive potential against Ethylene glycol mono methyl ether- induced gonadotoxicity rather, it exacerbates the deleterious effects.

Keywords: *Paullinia pinnata*, reproductive hormones, ethylene glycol mono methyl ether

PS2-B-173

Investigation of potential cytotoxicity and genotoxicity of olive secondary metabolites and an olive oil polyphenols extract

Agathi Charistou[1], Anastasia Spyropoulou[1], Effrosyni Katsanou[1], Apostolis Angelis[2], Dimitris Michailidis[2], Maria Halabalaki[2], Alexios-Leandros Skaltsounis[2], Kyriaki Machera[1]

[1]Benaki Phytopathological Institute, Athens, Greece, [2]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, University of Athens, Athens, Greece

Olive oil is the basis of the Mediterranean diet and a valuable resource of health-beneficial compounds. Amongst them, the polar components which constitute the total phenolic fraction (TPC), as oleocanthal, oleacein and maslinic acid, are considered as key olive secondary metabolites dew to their numerous biological activities. In this study, the potential cytotoxicity and genotoxicity induced by these three compounds and the TPC, to a human cell line (hepatocellular carcinoma, HepG2 cells) have been investigated.

TPC was recovered from extra virgin olive oil (EVOO), procured in Northern Greece (Mount Athos), using a recently development liquid-liquid extraction method. High purity oleocanthal, oleacein and maslinic acid were recovered from the treatment of the above extract in a two steps separation process.

Cytotoxicity was first determined by using the MTT assay. Significant cytotoxicity was induced at the highest concentration tested, i.e. 10-4 M for the individual compounds and 100 mg/L for the TPC. The genotoxic and cytotoxic potential of the three compounds and the TPC was further investigated by using the In-Cell-Western (ICW) assay using the Odyssey CLx Infrared system. Oleacein and maslinic acid, at the highest concentration tested (10-5 M), exhibited a significant 1.35-fold and 4.5-fold induction, respectively, of γH2AX in comparison to control (DMSO), while the respective induction exhibited by oleocanthal was 1.1-fold. The tested TPC exhibited a 1.6-fold induction of γH2AX in comparison to control (DMSO) at the highest concentration tested (50 mg/L). It should be noted that the tested molecules exhibit high biological activity and the concentrations tested in the presented assays are thousands of times higher than the respective daily human intake through olive oil consumption.

Additional experiments are being carried out to investigate further the genotoxic and beneficial potential of all tested olive secondary metabolites and the specific polyphenol extract.

PS2-B-174

Identification of additional anti-microbials and anti-oxidants in South African *Hypoxis hemerocallidea* Lam (African potato) extracts

Kokoette Bassey[1], Noel-David Nogbou[2], Andrew Musyoki[2]

[1] Pharmaceutical Sciences Unit, School of Pharmacy, Sefako Makgatho Health Sciences University. Ga-Rankuwa. P. O. Box 60 Medunsa, 0204, Pretoria, South Africa, [2] Department of Microbiological pathology, School of Medicine, Sefako Makgatho Health Sciences University. Ga-Rankuwa. P. O. Box 60 Medunsa 0204., Pretoria, South Africa

Hypoxis hemerocallidea, commonly known as African potato or "Inkomfe" is one of only 14 plant species indigenous to South Africa that is highly commercialized hitherto. Interest in the species stirred from its reputation as a cure for prostate hypertrophy and the patented anti-cancer activity of the extract. It is also use to treat other, human and animal diseases that include urinary, venereal and bladder disorders, diabetes, skin conditions and in the management of HIV/AIDS. However, the biological activity of this plant drug is allied only to rooperol, an aglycon from its major prodrug hypoxoside and/or *β*-sitosterol(in). We herein report on the identification of other active molecules present in *Hypoxis hemerocallidea* extracts. Two isolated standards (galpinoside and hemerocalloside) indicated in vitro qualitative free radical scavenging properties against DPPH when analyzed with the extract by TLC using $CHCl_3$:MeOH: H_2O (v/v/v) and vitamin C as the positive control. Orcinal glycoside and colchicoside were identified as active anti-oxidants in the methanol extract from their Rf values of 0.70 and 0.28 ($CHCl_3$:MeOH: H_2O (v/v/v) respectively. In addition, some of the molecules indicated microbial inhibition for some pathogens including *A. baumannii*, *E. coli*, *K. pneumonia*, *P. aeruginosa*, *E. faecium* and *E. fecalis*.

PS2-B-175

Characterization of traditionally used medicinal plants in the treatment of HIV

Babalwa Tembeni[1], Dashnie Naidoo-Maharaj[1], Thomas Klimkait[2], Vinesh Maharaj[1]

[1]University of Pretoria, Hatfield, South Africa, [2]University of Basel, Basel, Switzerland

Traditional medicine (TM) is widely used all over the world, despite its wide spread use, there are many limitations faced by TM, such as lack of recognition by the formal mainstream primary health care sector. Scientific evidence substantiating the efficacy, administration, safety and quality control is required to formally authenticate its claimed medicinal use is required. The unavailability of natural product drugs used as anti-viral agent.

Hence, the aim of this research is to characterize four South African medicinal plants used as a mixture by a traditional healer to treat HIV infected people and identify the biological active compounds responsible for the observed anti- HIV activity and to authenticate the traditional medicine through the development of herbal medicines for treatment of HIV.

100 g of each plant material was extracted using a 1:1 v/v of dichloromethane: methanol. The obtained extracts were screened in vitro against the HIV using a cell-based anti-HIV assay (DeCIPhR) that uses clinical isolates of different HIV variants. Primary dual point screens at concentrations of 25 µg/ml and 2.5 µg/ml, served as a filter to select most active plant species for full dose investigations. Crude extracts of Plant4 and the mixture both showed good potency, inhibiting the virus at both concentrations.

Plant 4 was sequentially extracted using hexane, dichloromethane, ethyl acetate and methanol. The chemical fingerprints of the DCM, ethyl acetate and methanol extracts was investigated using UPLC-MS-qTOF. This revealed the presence of daphnane diterpenoids which are reported to have anti-HIV properties at picomolar levels.

The DCM fraction was able to inhibit the virus up to 0.1 µg/ml with an inhibition of 93 % leading to its further purification using chromatographic techniques such HPLC, LC-MS-SPE-NMR and SFC. Further purification of active fractions is still ongoing.

PS2-B-176

Halogenated metabolites from the red alga *Laurencia majuscula* collected in the Red Sea

Mohamed Tammam, Efstathia Ioannou, Vassilios Roussis

Section of Pharmacognosy and Chemistry of Natural Products, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771 Athens, Greece

The red algal genus *Laurencia* (Rhodomelaceae) comprises approximately 150 cosmopolitan species with a wide distribution throughout the world. Even though they have been the subject of intense research over the last 60 years, they are still a prolific source of new natural products in the marine environment. To date, more than 1050 secondary metabolites, mostly halogenated terpenes and acetogenins frequently displaying new carbocycles, have been reported from *Laurencia* species and mollusks feeding on them. Due to their relatively high degree of halogenation, many of these molecules either are biologically active or play an ecological role in their ecosystem, often exhibiting antibacterial, antifungal, antiviral, anti-inflammatory, antiproliferative, cytotoxic, antifouling, antifeedant, ichthyotoxic, and/or insecticidal activity.

Coral reefs are renowned for their extraordinary natural beauty, biological diversity and high productivity, representing crucial sources of income and resources through their role in tourism, fishing, building materials, coastal protection, and the discovery of new drugs and biochemicals. Among them, the coral reefs of the Red Sea are considered the best-developed reefs in the western Indian Ocean.

In the course of our investigations on the chemistry of marine organisms, specimens of the red alga *Laurencia majuscula* were collected by SCUBA diving from Hurghada in the Red Sea (Egypt). The algal tissues were extracted with mixtures of CH_2Cl_2/MeOH and the organic residue was submitted to a series of chromatographic separations, leading to the isolation of a number of secondary metabolites. The structures of the isolated compounds, among which several are new natural products, were determined on the basis of thorough analysis of 1D and 2D NMR and MS data.

Acknowledgements: The authors thank Special Account for Research Grants and National and Kapodistrian University of Athens for funding to attend the meeting.

Keywords: *Laurencia majuscula*, halogenated metabolites, isolation, structure elucidation

PS2-B-179

Purification of Chios Mastic active constituents and development of analytical methods for quality control purposes

Vasiliki K. Pachi[1,2], Ekaterini Arguropoulou[1], Efi Thoma[3], Sofia Mitakou[1], Maria Halabalaki[1], Leandros-Alexios Skaltsounis[1]

[1]Laboratory of Pharmacognosy and Natural Products Chemistry, University of Athens,Panepistimioupoli, Zografou, Greece, [2]IASIS PHARMA HELLAS S.A., Pharmaceutical & Cosmetics Industry, Kamatero, Greece, [3]PharmaGnose S.A., Chalkida, Greece

Chios Mastic gum is the resinous secretion obtained from stems and branches of *Pistacia lentiscus* var. *chia* (Anacardiaceae). In 2015, the *P. lentiscus* L., Mastic resin was recognized as a traditional medicinal product by the European Medicines Agency (EMA) with two therapeutic indications i.e. mild dyspeptic disorders & skin inflammation/healing of minor wounds [2]. However, today, there is no reliable method for quality control purposes which complicates significantly the evaluation of authenticity and quality of the resin itself as well as its products. The main reason is the absence of commercially available reference standards of the unique secondary metabolites of mastic i.e. masticadienonic (MNA) and isomasticadienonic acid (IMNA). Thus, towards this direction, the main aim of this study was the development and application of purification and analytical methods of the active constituents of Mastic gum. Based on an at-line combination of a Sephadex and Semi-Preparative HPLC methods MNA and IMNA were isolated in high purity (>95%).

Moreover, analytical methods for MNA and IMNA quantification were developed on High Performance Liquid Chromatography-Diode-Array Detector (HPLC-DAD) and High Performance Thin Layer Chromatography (HPTLC). In addition, oleanolic acid (commercially available) was also used. The ICH Q2R1 guideline was followed for the validation of the procedure and the criteria tested were recovery, accuracy, precision, specificity, robustness, limit of detection and quantification, and stability. Statistical analysis proved that the methods were repeatable and selective for the specific constituents and they were both applied for the analysis of the crude resin as well as market products.

PS2-B-180

Investigation of the absorption and colonic biotransformation of olive oil polyphenols in the GIDM-Colon, a validated *in vitro* Gastrointestinal Dialysis Model with Colon phase

Maria Eleni Sakavitsi[1], Annelies Breynaert[2], Apostolis Angelis[1], Luc Pieters[2], Maria Halabalaki[1], Nina Hermans[2]

[1]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771, Athens, Greece, [2]Research group NatuRA – Natural Products & Food Research and Analysis, Department of Pharmaceutical Sciences, University of Antwerp, Universiteitsplein 1, 2610 Antwerp, Belgium

Olive oil composes one of the principal ingredients of the Mediterranean diet. Recently, olive oil polyphenols (OOPs) attracted the interest of scientific community, due to the EFSA health claim, in which olive oil polar constituents of a specific concentration level are recognised as contributors to the protection of blood lipids from oxidative stress. Specifically, with the term OOPs, is defined the polar components of olive oil, which comprise 3-5% of it, regarding a complex mixture of several chemical classes such as phenyl alcohols, phenolic acids, secoiridoids, triterpenoids etc. Hydroxytyrosol, tyrosol and oleocanthal, oleacein along with their aglycones belonging to phenyl alcohols and secoiridoids, resp., are mostly considered as major compounds of olive oil. Besides OOPs, this study includes oleuropein, a glycosylated secoiridoid, abundant in green olives and olive leaves with significant pharmacological properties, which is also detectable in considerable amounts in olive oil as its aglycone.
Moreover, the biological effects of polyphenolic compounds in the human body are partly the consequence of their biotransformation by the colon microbiota, and the subsequent absorption of these colonic metabolites. Thus, we recently optimised and validated an *in vitro* continuous flow dialysis system, simulating the absorption from lumen to mucosa and followed by the colon phase using pooled human faecal suspensions (GIDM-Colon Model), which mimics the physiological conditions during human gastrointestinal digestion (1).
Aiming to study the gastrointestinal absorption and biotransformation of polyphenolic compounds (1) and within the framework of the international MediHealth project, the GIDM-Colon Model was applied to OOPs namely oleacein, oleocanthal, hydroxytyrosol, and tyrosol, including oleuropein, as well as the total polyphenolic extract of olive oil. During GIDM-Colon digestion, different samples were collected of both dialysate and retentate solutions, after gastric and small intestinal digestion, and at different time points of colonic digestion. Data analysis and metabolite fingerprinting were conducted using a UPLC-QTOF MS and a hybrid UPLC-IT-Orbitrap MS (2). Results of this study will provide significant insight into the bioavailability and biotransformation of OOPs in the human gut using a validated *in vitro* Gastrointestinal Dialysis Model with Colon phase.

Acknowledgements: The authors would like to thank the MediHealth project (Horizon2020-MSCA-RISE-2015-691158) for the financial support and collaboration opportunities. Also, the present work is implemented with scholarship of the General Secretariat of Research and Technology (GSRT) and the Hellenic Foundation for Research and Innovation (HFRI).

Keywords: olive oil polyphenols, *in vitro*, ADME assay, bioavailability, biotransformation

References:

[1] Breynaert A *et al*, Planta Med 2015; 81: 1075

[2] Kanakis P *et al*, Planta Med 2013; 79: 1576

PS2-B-181

Olive oil's biophenols quantitative study using NMR, LC-HRMS & MS/MS methods

Stavros Beteinakis[1], Sotirios Katsikis[1,2], Emmanouel Mikros[2], Leandros A Skaltsounis[1], Maria Halabalaki[1]

[1]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis, Zografou, 15771, Athens, Greece, [2]Department of Pharmaceutical Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis, Zografou, 15771, Athens, Greece

Olive oil, an accidental discovery of the ancient times, seems to experience an increasing superfluity of research interest and attention recently. Aside from its renowned part in the Mediterranean diet for centuries, olive oil gained major health claims by the European Food Safety Authority (EFSA) in 2011, such as anti-inflammatory, cardioprotective and antioxidative, among others [1]. Responsible for the above characterization are biophenols, molecules in significantly low, yet with great impact, concentration in extra virgin olive oil (EVOO). Olive oil's 'oily' nature and physicochemical characteristics, along with its changeable chemical composition, make it an intricate matrix that poses a challenge to any analytical technique and analyst.

To this day, only one analytical method has been suggested by the International Olive Council (IOC), and that is High Performance Liquid Chromatography (HPLC) with Diode-Array Detection (DAD). However, even that is in need of improvement and seems to be displaying several issues, as the lack of standards has led to certain compromises in the quantification process and the insufficient baseline separation of all the possible compounds in the polar fraction makes mass detection mandatory for their safe identification. Furthermore, there are flustering and contradictive results in the literature regarding actual levels of EVOO's bioactive phenols. Hence, a revision of the minimum amount of 5mg of phenols per 20g of oil (EFSA's claim) cannot be taken off the table once scientists agree upon an optimum method and great number of samples are analyzed [2].

The aim of the present study was to play a crucial role in solving the aforementioned dispute that exists among analytical methods for the quantification of EVOO's bioactive phenols that constitute the EFSA claim. To that end, methodologies, using state-of-the-art equipment, specifically Ultra-High-Performance Liquid Chromatography combined with High Resolution Mass Spectrometry and Tandem Mass Spectrometry (UHPLC-HRMS and MS/MS) and Nuclear Magnetic Resonance (NMR) spectroscopy, were initially developed and then validated. Results from both methods were compared, both with each other, as well as with the well-established method of the IOC using HPLC-DAD [3].

Acknowledgements: The present work is implemented with IKY scholarship which is funded by the action "Strengthening Human Resources Research Potential via Doctorate Research" with funds from the operational program "Human Resources Development Program, Education and Lifelong Learning, 2014-2020 with the co-funding of European Social Fund (ESF) and Hellenic government. The authors would also like to thank the European Olive-Net project (Horizon2020-MSCA-RISE-2016-734899) for the financial support and collaboration opportunities.

Keywords: Olive Oil, NMR spectroscopy, UHPLC-MS &MS/MS, Quantification, Validation

References:

[1] EFSA, Scientific Opinion. EFSA Journal, 2011; 9(4): 2033

[2] Tsimidou M Z, Boskou D. Eur J Lipid Sci Technol 2015;117: 1091–1094
[3] IOC, Determination of Biophenols in Olive Oils by HPLC, 2017, 1-8, COI/T.20/Doc No 29/Rev.1

PS2-B-182

Bio-guided isolation of volatile compounds with repellent properties against *Aedes albopictus* (Diptera: Culicidae) using CPC technology

Anastasia Liakakou[1], Apostolos Angelis[1], Nikolaos Fokialakis[1], Antonios Michaelakis[2], Dimitrios Papachristos[2], Leandros.A Skaltsounis[1]

[1]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Panepistimioupolis, Zografou, 15771, Athens, Greece, Athens, Greece, [2]Department of Entomology and Agricultural Zoology, Benaki Phytopathological Institute, 8 S. Delta Str., Athens 14561, Greece, Athens, Greece

Aedes albopictus is an invasive mosquito species, with wide spread in many countries worldwide. The protection from mosquito bites still remains the most effective way to prevent infections associated with moboviruses, such as Zika, Dengue and Chikungunya. In the effort to develop potent mosquito repellents less harmful for humans and the environment, research on natural products with strong bioactivity, such as essential oils, has been prioritized.
In the present work 6 essential oils (EOs) obtained from wood of *P. heldreichii* and *P. nigra*, needles of *P. pinea*, ripe and overripe fruits of *J. phoenicea* and fruits of *J. oxycedrus* have been prepared using Microwave-assisted hydrodistillation. Chemical composition of all EOs were determined by GC/MS analysis. Additionally, their repellent properties were evaluated against *Aedes albopictus* showing that the essential oil of *P. pinea* and *J. phoenicea* presented high activity at the dose of 0.2 μl/cm^2. Comparing the results of GC/MS analysis and biological assays we decide to further analyze the EOs of *P. pinea* and *J. phoenicea* in order to isolate the main volatile compounds and to identify those responsible for the repellent activity. The fractionation of the selected EOs took place by CPC using for the analysis the biphasic system *n*-Heptane/ACN/BuOH in ratio 1.6/1.6/0.2 (v/v/v). The results of this step were the isolation of (-) limonene and guaiol from EO of *P. pinea* and myrcene and germacrene-D from EO of *J. oxycedrus.* All isolated compounds were tested for their repellent activity at the dose of 0.2 μl/cm^2. The results of repellent bioassays, employing fractionated compounds, revealed that (-) limonene, guaiol and germacrene D presented high repellent activity, while myrcene was almost non-active.
It is worth noting that the use of CPC for bio-guided isolation of these active volatile compounds from these EOs is presented for the first time.

PS2-B-183

Mediterranean herb extracts inhibit oral microbial growth and biofilm formation of *Streptococcus mutans*

Lamprini Karygianni[1], Joachim Hickl[2], Aikaterini Argyropoulou[3], Maria-Eleni Sakavitsi[3], Maria Halabalaki[3], Ali Al-Ahmad[2], Elmar Hellwig[2], Nektarios Aligiannis[3], Alexios Leandros Skaltsounis[3]

[1]Clinic for Preventive Dentistry, Periodontology and Cariology, Center of Dental Medicine University of Zurich, Zurich, Switzerland, [2]Department of Operative Dentistry and Periodontology, Medical Center, University of Freiburg, Faculty of Medicine, University of Freiburg, Freiburg, Deutschland, [3]Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Athens, Greece

In light of the growing antibiotic resistance, the usage of plant-derived antimicrobial agents could serve as an effective alternative treatment against oral infections. The aim of this study was to investigate the antimicrobial and antibiofilm activity of Mediterranean herb extracts against representative oral microorganisms. The extraction procedures and the analysis of the obtained extracts were performed under established experimental conditions and the fingerprinting was conducted using High Performance Thin Layer Chromatography (HPTLC), UPLC-HRMS and HRMS/MS analysis. The minimum inhibitory (MIC) and bactericidal (MBC) concentrations of the methanol extracts of *Cistus creticus* ssp. *creticus*, *Cistus monspeliensis*, *Origanum vulgare, Rosmarinus officinalis, Salvia sclarea* and *Thymus longicaulis* against eight typical oral bacteria and the fungus *Candida albicans* were determined. The antibiofilm activity against *Streptococcus mutans* was also quantified using the microtiter plate test. Overall, all tested extracts inhibited effectively the screened obligate anaerobic microorganisms and in concentrations ≥0.3 mg ml-1 had moderate to high antibiofilm activity comparable to that of chlorhexidine (CHX) against *S. mutans*. In particular, *R. officinalis* (MIC: 0.08 - 5.00 mg ml-1) and *S. sclarea* (MIC: 0.08 - 2.50 mg ml-1) showed the highest antibacterial activity, while *Cistus* spp., *R. officinalis* and *S. sclarea* significantly inhibited *S. mutans* biofilm formation at 0.60, 1.25 and 2.50 mg ml-1, respectively. *Porphyromonas gingivalis* and *Parvimonas micra* were high susceptible to *O. vulgare* (MIC = 0.30 mg ml-1), whereas *T. longicaulis* eradicated all oral bacteria (MBC: 0.15 - 2.50 mg ml-1). Nevertheless, *C. albicans* showed no sensitivity to the tested extracts. In conclusion, the tested plant extracts could serve as alternative natural antibacterial and antibiofilm components against oral infections.

Figure 1

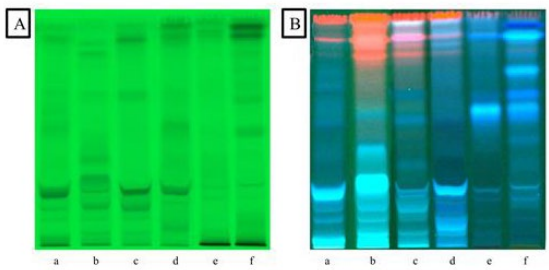
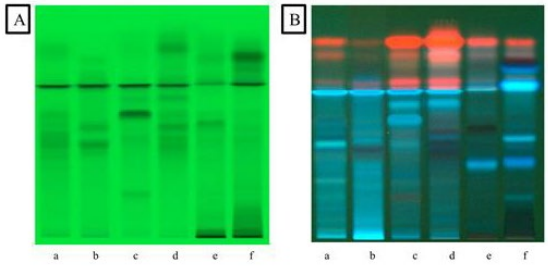
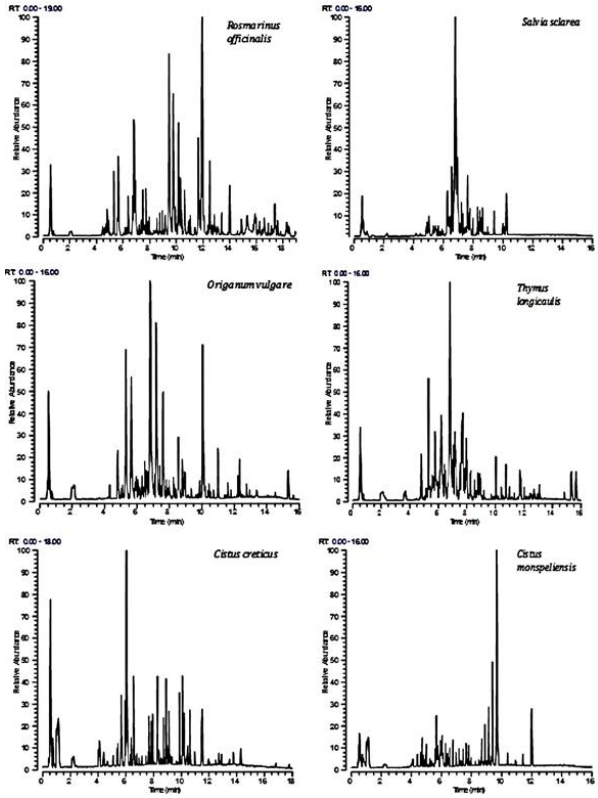
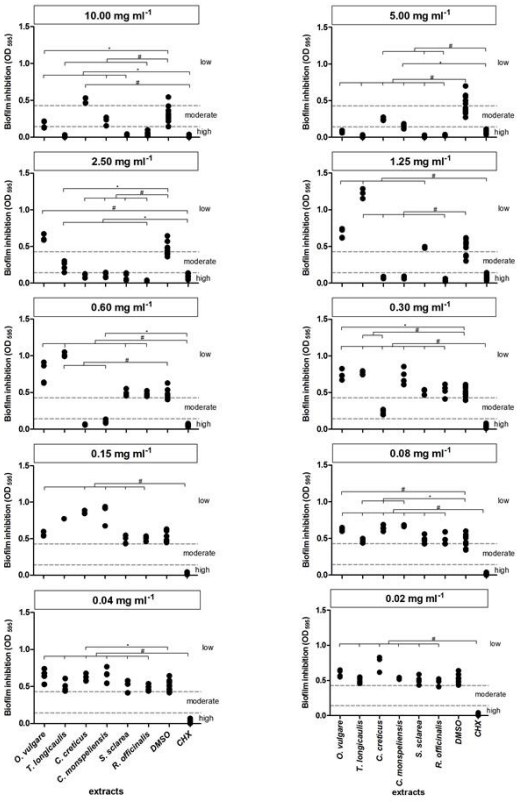


Figure 2




Rosmarinus officinalis
Salvia sclarea
Origanum vulgare
Thymus longicaulis
Cistus creticus
Cistus monspeliensis
Relative Abundance
Time (min)


10.00 mg ml-1
5.00 mg ml-1
2.50 mg ml-1
1.25 mg ml-1
0.60 mg ml-1
0.30 mg ml-1
0.15 mg ml-1
0.08 mg ml-1
0.04 mg ml-1
0.02 mg ml-1
Biofilm inhibition (OD 595)
low
moderate
high
O. vulgare
T. longicaulis
C. creticus
C. monspeliensis
S. sclarea
R. officinalis
DMSO
CHX
extracts

PS2-B-184

Indirubin analogues are promising compounds for the treatment of Leishmaniasis and Human African Trypanosomiasis

Antonia Efstathiou[1], Nicolas Gaboriaud-Kolar[2], Vassilios Myrianthopoulos[2], Emmanuel Mikros[2], Agallou Maria[1], Dimitrios Bouziotis[1], Karagouni Evdokia[1], Alexios-Leandros Skaltsounis[2], Despina Smirlis[1]

[1]Dpt of Microbiology, 127 Bas. Sofias Ave., Hellenic Pasteur Institute, 11521 Athens, Greece; [2]Dpt of Pharmacy, Panepistimiopolis Zografou, University of Athens, GR-15771 Athens, Greece;

Leishmania and *Trypanosoma brucei* are protozoan parasites that cause leishmaniases, and Human African Trypanosomiasis, respectively. These parasitic diseases are a significant cause of morbidity and mortality in developing countries. Moreover, leishmaniasis is an important public health and veterinary concern in Mediterranean countries while human cases are alarmingly increasing. Despite these alarming calls, no human vaccines are approved and the current chemotherapy is ineffective, mainly due to the emerging drug-resistance and severe toxic side effects. Consequently, there is an urgent need to discover new therapies. Indirubins, a class of bis-indole derivatives are promising candidates for targeted therapy because they are validated as inhibitors of eukaryotic protein kinases, enzymes that regulate all the essential processes of eukaryotic organisms. We have previously shown that indirubin analogues inhibit intracellular *Leishmania* growth and the essential parasite kinases CRK3 and GSK3. Herein, the antiparasitic activity of a library of 69 indirubins was evaluated against the related to *Leishmania* trypanosomatid parasite *Trypanosoma brucei.* Our results showed that 32 analogues displayed potent growth inhibition against *T. brucei* (IC50<1 μM) and satisfactory selectivity index against mammalian cells. The same compounds displayed significant inhibition against *Tb*GSK3, while there was a positive correlation between antitrypanosomal activity and *Tb*GSK3 inhibitory activity for a class of 3' bulky 6BIO analogues. Moreover, the most potent indirubin analogues (5Me6BIO and compound 11) were tested *in vivo* in a murine model of acute and chronic visceral leishmaniasis and displayed substantial reduction of the parasitic burden. In summary, the above results show promising antitrypanosomatid efficacy of GSK3 inhibitors and are anticipated to lead to drug candidates that are urgently needed for treating parasitic diseases, an arising public health concern.

Acknowledgements: We acknowledge support of this work by the project Kripis "Infectious, autoimmune and neurodegenerative diseases: study of the pathogenetic mechanisms and development of diagnostic, prognostic and therapeutic approaches" (MIS 5002486) which is implemented under the "Action for the Strategic Development on the Research and Technological Sector", funded by the Operational Programme "Competitiveness, Entrepreneurship and Innovation" (NSRF 2014-2020) and co-financed by Greece and the European Union (European Regional Development Fund). We would also like to thank for funding IKY-SIEMENS Postodoctoral scholarship of excellence/2015-2017.

PS2-B-185

Antiviral properties of the leafs extract of succulent plant *Graptopetalum paraguayense* E. Walther against sensitive and resistance Herpes Simplex virus type 2 (HSV-2) strains

Petia Genova-Kalou[1], Nadezhda Markova[2], Stefka Ivanova[1], Kamelia Yotovska[3], Venelin Enchev[2]

[1]Department of Virology, National Center of Infectious and Parasitic Diseases, 44A "Gen. Stoletov" Blvd., Sofia 1233, Bulgaria, [2]Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, "Acad. G. Bontchev" Str., bl. 9, Sofia 1113, Bulgaria, [3]Faculty of Biology, Sofia University "St. Kliment Ohridski", 8 "Dragan Tzankov" Blvd., Sofia 1164, Bulgaria

Herpes simplex virus types 1 and 2 (HSV-1 and HSV-2) are members of the Herpesviridae family and are the most common spread human pathogens. Furthermore, HSV-2 infection has been reported to be a risk factor in HIV infection. Nucleoside analogues such as the acyclovir (ACV) or its subsequent derivatives with better bioavailability such as famciclovir, valacyclovir and penciclovir remain as the mainstay for HSV infection treatment. However, with the emergence of acyclovir-resistant HSV strains particularly in immunocompromised patients, there is a need to develop an alternative antiherpetic drug.

The objective of our examination is to evaluate the anti-herpes activities of *Graptopetalum paraguayense* E. Walther (Crassulaceae) against sensitive and resistance HSV-2 strains in Vero cells by the MTT colorimetric assay. This is the first study that reported the antiviral activities of *Graptopetalum paraguayense* E. Walther. The results are present as 50% cytotoxicity (CC50) and as 50% inhibitory concentration of the viral effect (IC_{50}) for CPE by MTT assay, which give possibility to calculate the selectivity index (SI). The aqueous extract of *Graptopetalum paraguayense* E. Walther has not cytotoxic effect on Vero cells. The extract effectively inhibits HSV-2 replication in dose-dependent manner. Furthermore, the extract is more effective inhibitor of HSV-2 Bja strain (sensitive to ACV) replication in cultured cells, as their IC_{50} values are not so significantly lower than that of ACV. It was inhibited the HSV-2, Bja strain replication 87%, whereas its effect to HSV-2, strain DD (resistant of ACV) was about 75%.

The mechanism of the antiviral action of leafs extract of *Graptopetalum paraguayense* E. Walther (Crassulaceae) is not yet completely identified. Further studies are needed in order to verify which compounds could be responsible for this activity and how they exert their antiviral effects.

Acknowledgements: We acknowledge the financial support of the Bulgarian Fund for Scientific Research under Grant DH 19/16.

Keywords: Herpes Simplex virus type 2 (HSV-2), *Graptopetalum paraguayense* E. Walther, antiviral resistance, acyclovir (ACV), cytotoxicity, cell cultures

PS2-B-186

Glycyrrhiza glabra enhanced extract and Adriamycin anti-proliferative effect against PC-3 prostate cancer cells.

Katerina Gioti[1], Anastasia Papachristodoulou[1,2], Dimitra Benaki[2], Apostolos Beloukas[3,4], Argyro Vontzalidou[5], Nektarios Aligiannis[5], Alexios-Leandros Skaltsounis[5], Emmanuel Mikros[2], Roxane Tenta[1]

[1]School of Health Sciences and Education, Department of Nutrition and Dietetics, Harokopio University, Kallithea, Greece, [2]Faculty of Pharmacy, Department of Pharmaceutical Chemistry, University of Athens, Panepistimiopolis, Zografou, Greece, [3]Department of Biomedical Sciences, Athens University of Applied Sciences, Aigaleo, Greece, [4]Institute of Infection and Global Health, University of Liverpool, Liverpool, United Kingdom, [5]Faculty of Pharmacy, Department of Pharmacognosy and Natural Products Chemistry, University of Athens, Panepistiomiopolis, Zografou, Greece

Adriamycin (ADR) is a widely used chemotherapeutical drug yet with severe side effects, while GGE is an extract isolated from the plant *Glycyrrhiza glabra* (Fabaceae) tested for its cytotoxic activity in various types of cancer. PC-3 is an androgen-independent human prostate cancer cell line, widely used for the study of prostate cancer.
Previous data from our studies have demonstrated that GGE inhibited PC-3 cell proliferation in a dose-dependent manner while the co-treatment of ADR-GGE resulted in an additive inhibition of cell proliferation, compared to ADR alone. In order to elucidate the implicated mechanism of ADR-GGE action, cells were treated with ADR (25nM) or/and GGE (35 μg/mL) and apoptosis and autophagy rates were evaluated through flow cytometry. The expression levels of autophagy related genes (ULK1, AMBRA1, Beclin1, LC3A) were measured in the mRNA level with Real Time PCR, while NMR spectrometry was used to identify the implication of specific metabolites.
Our results from flow cytometry demonstrate that ADR-GGE treatment results in an enhanced apoptotic rate compared to ADR treatment alone, revealing also necrotic effects. In addition, ADR treatment induces the autophagy of PC-3 cells, yet an inhibition of autophagic flux is found after GGE and ADR-GGE treatment. The expression levels of autophagy related genes are also consistent with the induction of autophagy after ADR treatment, while GGE and ADR-GGE treatments imply alterations on ADR-induced autophagy. Cells also show a different metabolic profile according to each treatment while the levels of specific metabolites related to autophagy are altered, as ADR+GGE co-treatment increases several amino acids concentration as branched-chain amino acids (BCAAs) (namely isoleucine, valine), alanine, tyrosine, glutamic acid, as well as formic acid and o-phosphocholine, compared to ADR.
In conclusion, GGE natural extract has an additive effect on ADR cytoxicity by altering the autophagic potential of PC3 cells and triggering excessive apoptosis, thus showing promising results to be used as an adjuvant prostate cancer treatment.

Keywords: *Glycyrrhiza glabra*, ADR, apoptosis, autophagy, metabolic profile

References:

[1] Minotti G, Menna P, Salvatorelli E, Cairo G, Gianni L. Pharmacol Rev 2004; 56: 185–229.
[2] Nourazarian SM, Nourazarian A, Majidinia M, Roshaniasl E. Asian Pac J Cancer Prev 2016; 16: 8563–8566.
[3] Hsieh MJ, Lin CW, Yang SF, Chen MK, Chiou HL. Br J Pharmacol 2017; 171: 3037–3050.

PS2-B-187

Identification of Novel Natural Bioactive Compounds using in Vivo Zebrafish Phenotypic Assays

Agalou A[1], Thrapsianiotis M[2], Angelis A[2], Papakyriakou A[3], Skaltsounis AL[2], Aligiannis N[2], Beis D[1]

[1]Zebrafish Disease Model Lab, Biomedical Research Foundation Academy of Athens, Athens, Greece. [2]Department of Pharmacognosy and Natural Product Chemistry, Faculty of Pharmacy, University of Athens, Athens, Greece. [3]Institute of Biosciences and Applications, National Centre of Scientific Research "Demokritos", Athens, Greece.

Identification of new Bioactive Natural Products (BNPs) that may serve as potential drug lead compounds is a constant challenge. The current reductionist screening paradigms employ *in vitro* cell-based tests and/or one pathway or even a single protein as a drug target. In numerous cases, the compounds determined this way as hit molecules, fail to show any *in vivo* efficacy in animal models. Carrying out large-scale drug screens on mammals nowadays would be ethically and financially unjustifiable. Alternatives are offered by zebrafish embryos, which allow, in particular, *in vivo* monitoring of complex cell behavior and physiological parameters. Therefore zebrafish-based assays are gaining high popularity and wide usage in both academic and industrial drug discovery efforts.

Zebrafish is a vertebrate model extensively used in development and Human Disease modelling due to its high genetic, physiological, and pharmacological similarities with human. Zebrafish embryos develop externally and are transparent, allowing for *in vivo* non-invasive imaging. There is a plethora of transgenic and mutant lines available that mimic most human diseases. These include reporter lines for most signaling pathways related to Human Disease. In addition, it is a well established model to study organ regeneration, since zebrafish retain their capacity to regenerate most organs during their entire life span. For all these reasons, zebrafish emerges as a valuable whole animal platform for various stages of BNPs discovery efforts. These include phenotypic screenings, determinations of general and/or specific toxicity (cardiac, renal, hepatotoxicity etc) and mechanism of action studies.

One of the well established phenotype-based drug screenings regards the identification of products related to pigmentation disorders. Abnormal pigmentation has been correlated with various aesthetic and medical problems including melanoma. We use the inhibition of melanogenesis during early embryo development to identify natural compounds that block melanogenesis. We identified an extract from the Greek hawthorn *Crataegus pycnoloba* as a potent inhibitor of melanin synthesis and used activity based subfractionation to identify active subfractions and eventually three single compounds that inhibit melanogenesis. Finally, we identified a molecular mechanism that elucidates how their activity is mediated.

PS2-B-188

Aqueous ethanol extracts of red grape pomace exert potent *in vitro* anti-platelet and anti-oxidant properties

Maria Choleva, Vasiliki Boulougouri, Smaragdi Antonopoulou, Elizabeth Fragopoulou

School of Health Science and Education, Department of Nutrition and Dietetics, Harokopio University, Athens, Greece

Micro-constituents from wine exert anti-oxidant and anti-platelet properties, as reported in our previous studies [1]. Winery by-products are cheap and unexploited source of wine-like compounds [2], therefore the aim of the study is to evaluate different extraction methods in order to obtain an extract mainly with anti-platelet but also anti-oxidant actions.
Grape pomace from four red grape varieties was used and four extraction methods (Bligh-Dyer, 80% ethanol, hexane, water) were performed. Total phenolic content, extracts' ability to scavenge DPPH and to inhibit serum oxidation were determined. Extracts' ability to inhibit platelet aggregation against PAF, ADP, and TRAP was measured in human platelet rich plasma (PRP) by Light Transmission Aggregometry and IC_{50} value meaning the amount of the sample that induced 50% inhibition against each agonist was calculated.
The aqueous ethanol and Bligh-Dyer water phase extracts had the highest concentration of phenolic compounds approximately 0.4 mg gallic acid/mg winery by-products and revealed better capacity to scavenge the DPPH free radicals and to inhibit serum oxidation against copper. Regarding the platelet aggregation, more potent and dose-dependent inhibition of platelet aggregation was showed by aqueous ethanol extracts, with IC_{50} values 162.1±66.9, 181.2±82.3, 156.3±97.5 μg against PAF, ADP, TRAP respectively, followed by the extracts of the Bligh-Dyer lipoid phase with IC_{50} values 280.9±115.9, 293.2±102.7, 284.8±131.8 μg against PAF, ADP, TRAP respectively. No differences were observed concerning the grape variety.
Aqueous ethanol extracts of red grape pomace contain micro-constituents that combine potent anti-platelet and anti-oxidant actions. Their use as ingredients could lead to the production of functional foods with cardioprotective properties.

Keywords: Grape pomace, antioxidant action, platelet aggregation, ethanolic extract, PAF

References:

[1] Fragopoulou E, Demopoulos CA, Antonopoulou S. Int J Wine Res 2009; 1: 131–143.
[2] Teixeira A, Baenas N, Dominguez-Perles R, Barros A, Rosa E, Moreno DA, Garcia-Viguera C. Int J Mol Sci 2014; 15: 15638–15678.

PS2-B-189

Anti-platelet and anti-inflammatory properties of wines from Ionian Islands

Elizabeth Fragopoulou[1], Maria Choleva[1,2], Maria Detopoulou[1,2], Filio Petsini[1,2], Olga Arvaniti[2], Efi Kallinikou[2], Eleni Sakantani[2], Aggeliki Tsolou[2], Yiannis Samaras[2]

[1]Department of Nutrition-Dietetics, School of Health Science and Education, Harokopio University, [2]Department of Food Technology, TEI of Ionian Islands

Wine cardioprotective effect is thought to be attributed to its micro-constituents and during the last decade, great effort was made to isolate and identify these bioactive compounds, as well as to elucidate the mechanisms of their action [1, 2]. The observed activity could be depended on the grape variety and the vineyard region. Therefore, the scope of the study is to investigate the anti-platelet and anti-inflammatory properties of Ionian Islands wines.

For this purpose, 10 varieties of red/white wine from Ionian Islands were selected and two extraction methods were performed. In the first method, total lipids (TL) were extracted, according to the Bligh Dyer's method. In the second method, liquid/liquid extraction was performed, in order to obtain several fractions, containing different classes of phenolic compounds: FI: anthocyanins; FII: procyanidins, catechins, flavonols; FIII: phenolic acids, quercetin-3-*O*-glucuronids; FIV: the rest of the phenolic components that are not distributed to previous fractions. Extracts capacity to inhibit PAF, ADP, TRAP, arachidonic acid and collagen-induced platelet aggregation was measuring in Platelet Rich Plasma (PRP) by Light Transmission Aggregometry. Also, their ability to inhibit LPS-induced secretion of IL-1β and TNF-α was measured in human peripheral blood mononuclear cell (PBMC).

Organic fractions TL, FII and FIII revealed better biological activity compared to the aqueous ones FI and FIV. In particular, according to their anti-platelet effect TL and FII revealed the most potent effect following by FIII and according to their anti-inflammatory effect FII revealed the most potent effect following by TL and FIII. No differences were observed according to the color of wine however some varieties from both red and white wine presented better biological activity.

In conclusion, the anti-platelet and anti-inflammatory action of wine is attributed to lipid-soluble molecules and seems to be variety-depended.

Keywords: wine, inflammation, platelet aggregation, mononuclear cell

References:

[1] Fragopoulou E, Demopoulos CA, Antonopoulou S. Int J Wine Res 2009; 1: 131–143.
[2] Andriantsitohaina R, Auger C, Chataigneau T, Étienne-Selloum N, Li H, Martínez MC, Schini-Kerth VB, Laher I. Br J Nutr 2012; 108(9): 1532–49.

PS2-B-191

Development of anti-ageing natural products based on biodiversity of the Greek flora by employing environmentally friendly technologies and anti-ageing biological research

Vasiliki-Ioanna Boka[1], Aimilia Sklirou[2], Maria Angelopoulou[3], Aristidis Konstantas[4], Aikaterini Argyropoulou[1], Haris Pratsinis[3], Dimitris Bilalis[4], Eleftherios Kalpoutzakis[1], Nektarios Aligiannis[1], Dimitris Kletsas[3], Ioannis P. Trougakos[2], Marianna Ralli[5], Alexios-Leandros Skaltsounis[1]

[1]Department of Pharmacognosy & Natural Products Chemistry, Faculty of Pharmacy, University of Athens, Zografou, 15771, Athens, Greece, [2]Department of Cell Biology and Biophysics, Faculty of Biology, University of Athens, Panepistimiopolis, Zografou 15784, Athens, Greece, [3]Laboratory of Cell Proliferation and Ageing, Institute of Biosciences and Applications, National Centre of Scientific Research "Demokritos", 11855, Athens, Greece, [4]Agricultural University of Athens, Department of Crop Science Lab. Of Crop Production, Iera Odos 75, 10445, Athens, Greece, [5]Korres S.A. Natural Products, 3 Drosini & Tatoiou Street Metamorfosi, 144 52, Athens, Greece

Ageing is a complex process driven by diverse molecular pathways and biochemical events. It has been defined as the time-dependent decline of functional capacity and stress resistance and affects most of tissues and organs of the body. The aim of this study is the development of anti-ageing natural products by employing state-of-the art environmentally friendly technologies and anti-ageing biological research. Specifically, a high number of plants (600) from Greek flora was selected and extracted by using "green technologies" (SFE, ASE and MWE). The extracts were investigated for their chemical profile and for their *in vitro* antioxidant activity (DPPH• and ABTS assays). Subsequently, the most promising extracts were applied to human diploid skin fibroblast cells and their antioxidant capacity was recorded by using the DCFH-DA assay. Based on these results 25 plant extracts were selected for further investigation. Assays were mainly based on normal human cells and refer to targets known to contribute cell protection from age-related damage. Specifically, they were tested for their efficacy against UV protection, as well as for their ability to modulate the proteasome and/or the autophagy-lysosome pathways functionality. "Multi-functional" extracts that apart from exerting antioxidant activity, are also activating main cellular pathways were identified. Plants affording the most bioactive extracts were cultivated, in order to protect the Greek biodiversity, while their chemical profile and biological activity was afresh confirmed. Among others, *Rosa damascena* R. and *Sideritis scardica* L. preparations showed the ability to reduce the ROS levels (to 24% and 51% compare to the control, respectively), while at a non-toxic concentration induced the activation of the proteasome LLVY/β5 peptidase activity. Finally, the results of the aforementioned biological research revealed these extracts as promising anti-ageing agents for potential usage as cosmeceuticals.

PS2-B-192

Effects of Mastiha (*Pistacia lentiscus*) supplement on oxidative stress and on plasma free amino acid in Inflammatory Bowel Disease; A randomized, double blind, placebo-controlled trial

Efstathia Papada[1], Charalampia Amerikanou[1], Ljilja Torović[2], Nick Kalogeropoulos[1], Chara Tzavara[1], Ilias Smyrnioudis[3], Alastair Forbes[4], Andriana C. Kaliora[1]

[1]Department of Dietetics and Nutritional Science, School of Health Science and Education, Harokopio University, Athens, Greece, [2]Department of Pharmacy, Faculty of Medicine, University of Novi Sad, Novi Sad, Serbia, [3]Chios Mastic Gum Growers Association, 1 K. Monomachou St., Chios, 82100, Greece, [4]Norwich Medical School, University of East Anglia, Bob Champion Building, James Watson Road, Norwich, NR4 7UQ, United Kingdom

Background: Natural products with antioxidant properties are investigated as a non-pharmacological approach in Inflammatory Bowel Disease (IBD), since oxidative stress is present in patients with active disease.
Aim: To assess the effects of natural Mastiha (*Pistacia lentiscus*) on oxidative stress biomarkers and to determine plasma free amino acids (AA) of patients with active IBD. This was a randomized, double-blind, placebo-controlled clinical trial. A total of 60 IBD patients were randomly allocated to Mastiha (2.8g/day) or placebo for 3 months adjunctive to stable medical treatment. Oxidative stress biomarkers, namely plasma oxidized low-density lipoprotein (oxLDL), total serum oxidizability and serum uric acid were evaluated. Oxidized LDL/LDL and oxidized LDL/High Density Lipoprotein (HDL) ratios were calculated. The plasma free AA were determined with gas chromatography/mass spectrometry analysis.
Results: Levels of oxLDL ($p=0.031$) and ratios of oxLDL/HDL ($p=0.020$) and oxLDL/LDL ($p=0.005$) decreased significantly in the intervention group. The mean change between groups for oxLDL/HDL ($p=0.044$) and oxLDL/LDL ($p=0.015$) was significantly different. As regards to AA, several changes were reported. More specifically, Mastiha ameliorated a decrease in plasma free AAs seen in patients with ulcerative colitis taking placebo.
Conclusion: Favourable changes in oxidative stress biomarkers were reported in active IBD patients administered with Mastiha. Plasma free AA changes indicated possible preventive role of Mastiha in metabolic dysregulation in active ulcerative colitis.

ClinicalTrials.gov Identifier: NCT02796339

PS2-B-193

Utilizing compact mass spectrometry for detection and quantification of chemicals related to cannabis

Daniel Eikel, Simon J Prosser, Vijay Gupta and Jack D Henion

Advion Inc, 10 Brown Road, Ithaca NY 14850

The transition of cannabis from an illegal drug to a drug for medical and even recreational use raises challenging questions for regulatory agencies and analytical chemists alike. Cannabinoid content has significantly changed in the last 30 years and strains with higher tetrahydrocannabinol (THC) content are actually undesirable for medical uses. Also, pesticide use is currently poorly regulated but can pose a serious health risk to consumers. Last but not least, cannabis remains illegal at the federal level, requiring rapid and legally defensible field tests for the detection of cannabis in suspect material and surfaces of interest. Here we will show a selection of analytical techniques based on compact mass spectrometry (CMS) in combination with three different sample inlets (atmospheric solids analysis probe (ASAP®), thin-layer chromatography (TLC) as well as classical liquid chromatography (LC)) for the detection and quantification of cannabinoids and pesticides in cannabis related material and contraband.

Keywords: Cannabis, Compact Mass Spectrometry (CMS), pesticides, Atmospheric Solids Analysis Probe (ASAP), Thin Layer Chromatography (TLC)

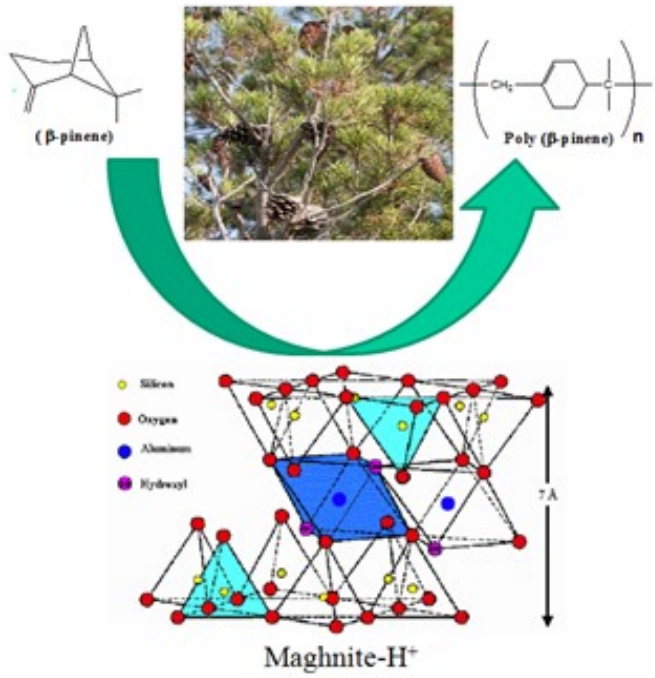
PS2-D-001

Valorisation of natural resourses: polymerization of *β*-pinene by natural montmorillonite clay

Amine Harrane[1,2], Malika Akeb[2], Mohammed Belbachir[2]

[1]University Of Abdelhamid Ibn Badis - Mostaganem, Mostaganem, Algeria, [2]University of Oran 1 Ahmed Benbella, Oran, Algeria

Environmental friendly catalytic processes of biomass valorization to produce chemicals with high added value, are an important field which are continuously attract more attentions in chemical engineering.
Biomass derived *β*-pinene issued from pin trees is an essential compound for fine chemical industry. It is the precursor of non toxic and inert poly(*β*-pinene) used as additive for rubbers, food packaging, casting industries and in the production of chewing gums. In this paper, we report an efficient and environmentally method to produce poly(*β*-pinene). We have used an algerian Montmorillonite clay as an heterogeneous non toxic catalyst to induce the polymerization of (*β*-pinene). Spectroscopic methods such as FT-IR, ^{1}H NMR, GPC chromatography and viscosimetry were used to confirm the structure of the obtained polymer. Effects of Maghnite/monomer weight-ratio, temperature and solvent on the yield of the polymerization and on the average molecular weight Mv of the resulting polymers were studied. The thermal properties (DSC) of the resulted poly(*β*-pinene) were also studied.



PS2-D-002

Utilization of lignin from a novel bio-refinery process as a co-component in 3D-printing

Lucas Lagerquist, Patrik Eklund

Johan Gadolin Process Chemistry Centre, Åbo Akademi University Biskopsgatan 8, FI-20500, Åbo, Finland

Lignin is after cellulose the second most abundant polymer in nature (up to 30% of lignocellulosic biomaterial). Its properties and possible utilization have been studied for decades. However, lignin still needs to be further explored and valorized to find novel and high-value applications. Lignocellulosic biorefineries yield enormous amounts of lignin and utilization of lignin, not only to use it as energy, is of interest for developing sustainable chemistry and a viable bioeconomy in the future.
In this work we have analyzed and characterized lignin [1] from a novel pressurized hot water extraction process [2,3] The process, developed by CH-Bioforce Oy, isolates the hemicelluloses from wood chips with hot water at oxygen-depraved conditions. After the separation of the hemicelluloses the lignin is removed by mild alkali pulping of the remaining fibers, also at oxygen-depraved conditions. This leaves both of the carbohydrate fractions as well as the lignin in both high yield and purity and as such it is of importance to determine the structure and properties of the lignin fraction for utilization. The process is currently being up-scaled to a facility being able to process up to 100 k ton biomass annually.
Based on our findings we have utilized the lignin in a PLA (poly lactic acid):lignin polymer blend. The bio-degradable PLA is a synthetic bio-polymer that has multiple uses; [4] however, its relatively high cost limits its use. To improve the compatibility with PLA the lignin was modified and the blend was used for 3-D printing.

Keywords: Lignin, natural products, structural analyzation, bio-refinery, 3-D printing

References:
[1] Lagerquist L *et al*. Ind Crops Prod 2018; 111: 306–316.
[2] Von Schoultz S. 2014; Method for Extracting Biomass. WO2014009604 A1.
[3] Von Schoultz S. 2015; Method for extracting lignin. WO 2015104460.
[4] Xu W *et al*. Carbohyd Polym 2018; 187: 51–58.

PS2-D-003

Miscibility of cellulose with synthetic polymers in ionic-liquid based solvent systems

Jiri Dybal, Nikolay Kotov, Vladimir Raus

Institute of Macromolecular Chemistry, Czech Academy of Sciences, Heyrovského nám. 2, 162 06, Prague, Czech Republic

The discovery of new materials through blending cellulose with synthetic polymers is driven by the abundancy, renewability, and good mechanical properties of cellulose and variability of synthetic, preferably biocompatible and biodegradable, polymers. However, due to its extensively hydrogen-bonded structure, cellulose is practically insoluble in water and in most organic solvents, which hinders preparation of cellulose-based polymer blends. Recently, certain ionic liquids were revealed as excellent solvents of cellulose. Cellulose-based composites were prepared from ionic liquid solutions, e.g. cellulose/polyvinyl alcohol (PVA) in 1 butyl-3-methylimidazolium chloride (bmimCl) [1]. However, cellulose solutions in bmimCl show high viscosity, complicating the polymer blending process. Addition of polar solvents has been proposed as a way to facilitate the dissolution of cellulose in bmimCl [2]. In this work, we investigated the feasibility of this strategy towards the preparation of cellulose-synthetic polymer blends. The solvent mixtures DMSO/bmimCl and DMAc/bmimCl were confirmed to be good solvents for cellulose while ensuring acceptable viscosity of the final mixtures. The role of non-covalent interactions (S=O…H-C, C=O…H-C, CH_3…Cl-) both in the solvent mixtures and in the solutions of cellulose (5 wt.%) in DMSO/bmimCl and DMAc/bmimCl were explored by spectroscopic investigations and model calculations. Dissolution of PVA and poly(*N*-vinylpyrrolidone) (PVP) in DMSO/bmimCl and DMAc/bmimCl at various concentrations was also studied. Based on the acquired data, a series of cellulose/PVA and cellulose/PVP mixtures in solutions and as polymer blends regenerated from solutions were investigated. ATR FTIR and Raman spectroscopy were used to characterize structural features and to identify key intermolecular interactions.

Acknowledgements: Financial support from the Czech Science Foundation is gratefully acknowledged (Grant No. 17-03810S).

Keywords: cellulose, polymer blends, ionic liquid, infrared spectroscopy

References:

[1] Hameed N, Xiong R, Salim NV, Guo Q. Cellulose 2013; 20: 2517.
[2] Andanson JM, Bordes E, Devémy J, Leroux F, Pádua AH, Gomes MFC. Green Chem 2014; 16: 2528.

PS2-D-004

Development of herbal sunscreen formulations from *Ophiorrhiza mungos*

Mayuri Napagoda[1], Chamika Liyanaarachchi[1], Shamila Malkanthi[1], Kaumadhi Abayawardana[1], Sudhara De Soyza[1], Sanjeeva Witharana[2], Lalith Jayasinghe[3]

[1]Department of Biochemistry, Faculty of Medicine, University of Ruhuna, Galle, Sri Lanka, [2]Faculty of Engineering, Higher Colleges of Technology, PO Box 4793, United Arab Emirates, United Arab Emirates, [3]National Institute of Fundamental Studies, Kandy 20000, Sri Lanka

The exposure to the ultraviolet (UV) component of the solar radiation is identified as a major contributing factor for conditions such as hyperpigmentation, wrinkling, erythema and inflammation [1]. To counteract this harmful UV radiation, synthetic sunscreens have been introduced to the market. However, with the disclosure of the adverse effects of synthetic sunscreen products, there is a growing demand for sunscreens of herbal origin [2]. In order to cater to this need, we have been studying the photoprotective potential of Sri Lankan medicinal plants. The work presented here focuses on the development of herbal sunscreens from *Ophiorrhiza mungos*, a plant widely utilized as a remedy for several dermatological conditions. The hydroalcoholic extract of *O. mungos* was incorporated into the aqueous cream-base at different percentages (25%, 50% and 75%) and the UV absorption measurements were obtained for each formulation to determine its UV filtering potential and subsequently the sun protection factor (SPF). To compare the efficacy of the herbal formulations, a commercial synthetic sunscreen and the aqueous cream-base were used as positive and negative controls respectively. The formulation containing 75% of the extract surpassed the other two formulations with its high SPF, photostability and broad-spectrum of UV absorption. Interestingly, the result with commercial synthetic sunscreen product was found to be inferior to our formulation. The initial SPF value of this formulation was determined as 22.9 and any significant reduction of this SPF value was not observed after its exposure to direct solar radiation for 21 days, thus, demonstrating the photostability. Furthermore, the high UV absorbance in 260-360 nm range was evident for its broad-spectrum sunscreen potential against both UV-A and UV-B radiation. Therefore, this study clearly demonstrated the suitability of *O. mungos* to be developed into a commercial herbal sunscreen and the experiments are underway to enhance its bioavailability via nanotechnology.

PS2-D-005

Biofunctional coating inspired by carob polyphenols for potential application in food packaging

Vlasios Goulas, Loukas Chadjivasiliou, Christodoulos Michael, George Botsaris

Cyprus University of Technology, Limassol, Cyprus

Polyphenols are widely distributed in plant kingdom and they are linked to diverse biological functions as antioxidant and antimicrobial properties. The carob fruit also contains significant amounts of phenolic compounds. The gallic acid is the most abundant among them; whereas catechins, flavonols and gallotannins have been also found. Recently, the formation of polyphenolic coatings via *in situ* oxidative polymerization of at alkaline pH has been described. Plant polyphenols can be polymerized onto a variety of material surfaces relevant to food packaging materials, including aluminum, glass, polyethylene terephthalate, and polypropylene. The objective of this work was to synthesize a biomimetic polyphenol coating for food packaging utilizing the phenolic fraction of carob pulp.

At first, the polyphenolic coatings were deposited in glass surfaces using different concentrations of carob extracts (2 and 4 mg mL-1) at three pH values (7, 8 and 9). Results demonstrated that the coating that is produced at pH=8 and at a concentration of 4 mg mL-1 had the most potent antioxidant and antimicrobial potential. In a next step, the coatings were applied directly on the salmon fillet (edible coating) and on the plastic container (active packaging). The peroxide and TRABs methods were used to measure the potency to inhibit lipid oxidation in salmon fillet. Furthermore, the anti-Listeria activity of coatings was also determined. Results showed a significant decrease of lipid oxidation during cold storage of salmon fillet for both coating applications. Regarding to the antimicrobial potency, one log reduction of *Listeria monocytogenes* was found after 10 days storage; no differences between edible coating and active application were observed. Overall, we describe the use of low-cost carob polyphenols as precursors for the formation of bifunctional coatings with promising application in food packaging.

Keywords: carob, coating, antioxidant potency, antimicrobial potency, active packaging, gallic acid

PS2-D-006

Thin film composite polyamide membranes embedded with Acacia gum: Properties and performance

Viktor Kochkodan, Yehia Manawi, Muataz Ali Atieh

Qatar Environment and Energy Research Institute, Doha, Qatar

Novel thin film composite (TFC) polyamide (PA) membranes blended with 0.01 – 0.2 wt. % of Acacia gum (AG), which is a natural gum collected as exudation from the stem and branches of *Vachellia* (Acacia), have been prepared using interfacial polymerization technique. The properties of the prepared membranes were evaluated using contact angle, zeta potential measurements, Raman spectroscopy, scanning electron microscopy, and surface profilometer. It was found that, the use of AG as an additive to TFC PA membranes increased the membrane's hyophilicity (by 45%), surface charge (by 16%) as well as water flux (by 1.2-fold) compared with plain PA membrane. In addition, the prepared PA/AG membranes possessed reduced surface roughness (by 63%) and improved antifouling behavior while maintaining NaCl rejection above 96%. The TFC PA/AG membranes were tested with seawater collected from the Arabian Gulf and showed higher salt rejection and lower flux decline during filtration when compared to commercial membranes (GE Osmonics and Dow SW30HR). These findings indicate that AG can be used as efficient natural additive to enhance the properties of TFC PA membranes.

Keywords: Reverse osmosis, polyamide membrane, Acacia gum, hyophilicity, surface charge, antifouling properties

PS2-D-007

Magnetically-responsive hydrogels obtained from marine plants

Tetsu Mitsumata[1,2], Mika Kawai[1,2]

[1]Niigata University, Niigata, Japan, [2]Japan Science and Technology Agency, Tokyo, Japan

Materials that physical property changes responding to a stimulus have been extensively investigated, particularly in the field of soft materials such as polymer gels, rubbers, or elastomers. Polymer gel containing magnetic particles is a stimuli-responsive gel that viscoelastic properties can be controlled by applying magnetic fields. We have fabricated magnetoelastic soft materials with various polymer matrices [1-6]. The elastic modulus of magnetic gels is enhanced by forming a chain structure of magnetic particles, similarly to magnetic fluids. Few years ago, we have succeeded to fabricate a new class of magnetoelastic gel that demonstrates drastic and reversible changes in dynamic modulus without using strong magnetic fields [7,8]. It was also succeeded to synthesize polyurethane elastomers showing drastic change in dynamic modulus by a factor of 277 [9].
In this paper, we present the magnetoelastic response and morphological properties of magnetic hydrogel consisting of a marine plant, carrageenan, that underwent wide modulation of dynamic modulus (500 times higher than off-field modulus).

References:

[1] Mitsumata T, Ikeda K, Gong JP, Osada Y, Szabo D, Zrinyi M. J Appl Phys 1999; 85: 8451.
[2] Mitsumata T, Honda A, Kanazawa H, Kawai M. J Phys Chem B 2012; 116: 12341.
[3] Mitsumata T, Ohori S, Honda A, Kawai M. Soft Matter 2013; 9: 904.
[4] Mitsumata T, Ohori S, Chiba N, Kawai M. Soft Matter 2013; 9: 10108.
[5] Nanpo J, Nagashima K, Umehara Y, Kawai M, Mitsumata T. J Phys Chem B 2016; 120: 12993.
[6] Oguro T, Endo H, Kawai M, Mitsumata T. Mater Res Express 2017; 4: 126104.
[7] Mitsumata T, Abe N. Chem Lett 2009; 38: 922.
[8] Mitsumata T, Honda A, Kanazawa H, Kawai M. J Phys Chem B 2012; 116: 12341.
[9] Mitsumata T, Ohori S. Polym Chem 2011; 2: 1063.

PS2-D-008

Electric properties for Kapton and biobased polyimide films

Mika Kawai[1,3], Tatsuo Kaneko[2,3], Tetsu Mitsumata[1,3]

[1]Niigata University, Niigata, Japan, [2]Japan Advanced Institute of Science & Technology, Nomi, Japan, [3]Japan Science and Technology Agency, Japan

The development of biobased polymers is indispensable for the establishment of a sustainable low-carbon society. A number of aliphatic biobased polymers such as polyesters; poly(lactic acid) [1], poly(hydroxyalkanote)s [2], and poly(butylenes succinate) [3], and polyamides (polyamide 11 and polyamide 66) have been developed, but their low glass transition temperature, Tg, and only a small percentage of their substitutes limited their use for various applications as superengineering plastic. We have developed so far biobased PIs from bioavailable aromatic diamines, which were photodimers of 4-aminocinnamic acid (4ACA) derived from genetically manipulated *Escherichia coli* [4-6]. These biobased PI films showed ultrahigh thermal resistance with the temperature at 10 % of mass loss, T10, values over 425 °C and no Tg values under 350 °C, which is the highest value of all biobased plastics reported thus far. This advanced thermal property can be useful for electric devices such as flexible polymeric electrodes that can be annealed at high temperature. On the application for electric devices, the electric properties such as resistivity, dielectric constant or dielectric breakdown voltage are very important. However, the electric properties for these films are not fully understood.
In this paper, we prepared biobased PI copolymer films and measured the electric resistance and dielectric constant for these films.

References:

[1] Okano K, Kondo A, Noda H. Eco Ind 2006; 11, 43.
[2] Vert M. Biomacromolecules 2005; 6, 538.
[3] Taniguchi I, Kimura Y. Biopolymers 2001 3b, 431.
[4] Kaneko T, Tran HT, Shi DJ, Akashi M. Nature Mater 2006; 5, 966.
[5] Kumar A, Tateyama S, Yasaki K, Ali MA, Takaya N, Singh R, Kaneko T. Polymer 2016; 83, 182.
[6] Suvannasara P, Tateyama S, Miyasato A, Matsumura K, Shimoda T, Ito T, Yamagata Y, Fujita T, Takaya N, Kaneko T. Macromolecules 2014; 47, 1586.

PS2-D-009

Production of an innovative dairy product using plant bioactive compounds

E-M Bata[1], P. Stathopoulos[1], L. Skaltsounis[1], Charalampos Proestos[2]

[1]Laboratory of Pharmacognosy and Natural Products Chemistry, Department of Pharmacy, University of Athens, Panepistimioupoli Zografou, 15771, Athens, Greece, [2]Laboratory of food chemistry, Chemistry Department, National and Kapodistrian University of Athens, 15771, Athens, Greece

In the Greek market, there is a tendency for purchase and consumption of dairy products with low salt concentration. The decrease of salt consumption at 3 - 4 g per day in conjunction with the increase of potassium, contributes to decrease and maintanance of pressure at normal levels. Consequently, cardiovascular diseases and brain strokes are reduced in percentage of 40% and 30%, respectively, while, death risk reduction is achieved in 20% percentage. The extract of olive tree leaves which is added, contains the antioxidant oleuropein in combination with the concentrations of necessary for the organism fatty acids (ω3, ω6, ω9) render the product thoroughly different and innovative in its category. There is no similar product with these characteristics, despite the fact that a spread with olive oil exists is the only one in the Greek market. Hence, the aim of the project was to produce a nutritional and innovative milk product that will contain bioactive compounds which will add value to final product, extend its shelf life and make possible use of health claim according to EU regulation 432/2012. The final product was a spread of cheese from organic sheep and goat's milk with olive leaf extract. Olive oil, sunflower oil, linseed oil and a small amount of flavored salt with thyme essential oil was included. The second product produced was a cheese with linseed oil, lecithin and olive oil. HPLC-DAD and UHPLC-MS was performed for the analysis of oleuropein, the bioactive compound of olive oil. Sensory evaluation of final products was also performed.

Acknowledgements: Authors would like to thank the company Family farm SA for providing milk samples.

Keywords: bioactive compounds, innovative food products, oleuropein, HPLC-DAD

References:

[1] Roidaki A, Kollia E, Panagopoulou E, Chiou A, Varzakas T, Markaki P. Current Research in Nutrition and Food Science, 138–145.
[2] Commission Regulation (EU) No 432/2012 of 16 May 2012 establishing a list of permitted health claims made on foods, other than those referring to the reduction of disease risk and to children's development and health Text with EEA relevance.
[3] Zoidou E, Magiatis P, Melliou E, Constantinou M, Haroutounian S, Skaltsounis, AL. Food Chem 2014; 158: 319–324.

PS2-D-010

Development of children's toothpaste gel with natural alternative preservatives

Spyridon Papageorgiou[1], Athanasia Varvaresou[1], Maria Giannakou[2], Irene Panderi[2], Fotini Mellou[1]

[1]Division of Aesthetics and Cosmetic Science, Department of Biomedical Sciences, Faculty of Health and Welfare Sciences, University of West Attica, Campus 1, 12243 Egaleo, Greece, [2]Faculty of Pharmacy, National and Kapodestrian University of Athens, 15771 Panepistimipolis, Greece

There is an existing research that focuses on the rational use of necessary ingredients in children's toothpaste. Necessary ingredients are those ingredients used specifically to increase the quality of children's oral care and/or contribute to the production of toothpaste. The most common detergent used in toothpaste is the anionic compound sodium lauryl sulfate (SLS), which has both antimicrobial and plaque inhibitory properties, as well. The known untoward side effects of SLS are inflammation and desquamation of oral mucosa. Preservatives are also necessary ingredients that stabilize the formulation. Due to the skepticism regarding sodium lauryl sulfate and traditional chemical synthetic preservatives, we formulated a toothpaste gel for children avoiding the use of SLS and traditional preservatives such as sodium benzoate, as well.

MATERIALS AND METHODS: We developed three fluorinated gel (1000 ppm Fluoride) formulations against dental plaque with three natural preservative systems namely, A, B and C. All formulations contain the mild surfactant Sodium Lauroyl Sarcosinate.

A: Sorbitol (30 %) and Glycerin (10%), B: Sorbitol (30 %), Glycerin (10%), Glyceryl Caprylate (0.5%), Sodium Anisate, (0.2%) and C: Sorbitol (30 %), Glycerin (10%), the alternative preservative mixture consisting of [Aqua, Glycerin, Sodium anisate, Sodium Levulinate] (2%). We performed the appropriate challenge tests according to the European Pharmacopoeia.

RESULTS: All the formulations proved to be sufficiently preserved according to criteria A and B. However, we had to be more careful regarding children's hygiene care products and therefore criterion B has to be considered "not safe" during the use by consumers. Only Formulation B fulfilled criterion A for all the tested microorganisms with *Aspergillus brasiliensis* being eliminated on day 7. Formulations A and C passed marginally the tests, (criterion B regarding *Aspergillus brasiliensis*) and therefore were not considered "safe" enough for children's use.

Acknowledgments: We thank Frezyderm S.A for sponsoring the above preservative efficacy tests.

Keywords: Children's toothpaste, natural, alternative preservatives

PS2-D-011

Electrospun micro/nanofibrous drug delivery systems for the modified release of melatonin

Stefanos Kikionis[1], Angeliki Siamidi[2], Marilena Vlachou[2], Andrew Tsotinis[3], Efstathia Ioannou[1], Vassilios Roussis[1]

[1]Section of Pharmacognosy and Chemistry of Natural Products, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771 Athens, Greece, [2]Section of Pharmaceutical Technology, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15784 Athens, Greece, [3]Section of Pharmaceutical Chemistry, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15784 Athens, Greece

The impact of nanotechnology in pharmaceutical and medical sciences is steadily increasing and nanoscale formulations such as liposomes, polymeric micelles, complexes, and nanofibers have attracted special attention during the last decade. Compared to other formulations, drug-loaded polymeric nanofibers provide great flexibility in selecting materials and drugs for various drug delivery applications. They can be easily produced from electrospinning, a versatile technique for the generation of continuous polymeric fibers from natural or synthetic polymers in the micro/nano scale under the application of a high voltage electric field.
Electrospun fibers possess high surface-to-volume ratio, small pore size, high porosity and advanced mechanical properties. Due to their high encapsulation efficiency, high loading capacity, simultaneous delivery of diverse therapies, ease of operation, and cost effectiveness they find extensive use as delivery systems for the controlled release of various active incorporated drugs. Depending on the selected polymers and the electrospinning parameters, nanofibrous systems can be easily modified for fine tuning of their release characteristics to provide rapid, immediate, delayed or modified dissolution profiles.
In the framework of our research interests towards the production of electrospun micro/nanofibrous composites, we have investigated the modified release of melatonin (MLT) from micro/nanofibrous polymeric matrices. MLT was successfully incorporated in electrospun fibers of various polymers, such as PVP, CA, PEO and HPMC, in different formulations. Scanning electron microscopy was used for the morphological characterization of the produced nanofibrous systems. The various formulations of the electrospun nanofiber mats loaded in capsules exhibited variable MLT release profiles in the gastrointestinal-like fluids, revealing that polymeric nanofibers represent a promising MLT carrier for the development of novel oral MLT delivery systems suitable for treating sleep onset and sleep maintenance problems.

Acknowledgements: This work was partially supported by the research program MARINOVA.

Keywords: Electrospinning, nanofibers, controlled release, dissolution, melatonin

PS2-I-001

Insights into the bifunctional aphidicolan–16–*ß*–ol synthase through rapid biomolecular modeling approaches

Monika Fuchs**,** Max Hirte, Thomas Brück

TUM Werner Siemens– Chair of Synthetic Biotechnology, Munich/ Garching, Germany

Diterpene synthases catalyse complex, multi–step C–C coupling reactions thereby converting the universal, aliphatic precursor geranylgeranyl diphosphate into diverse olefinic macrocycles that form the basis for the structural diversity of the diterpene natural product family. Since catalytically relevant crystal structures of diterpene synthases are scarce, homology based biomolecular modelling techniques offer an alternative route to study the enzyme's reaction mechanism. However, precise identification of catalytically relevant amino acids is challenging since these models require careful preparation and refinement techniques prior to substrate docking studies. Targeted amino acid substitutions in this protein class can initiate premature quenching of the carbocation centred reaction cascade. The structural characterization of those alternative cyclization products allows for elucidation of the cyclization reaction cascade and provides a new source for complex macrocyclic synthons. In this study, new insights into structure and function of the fungal, bifunctional Aphidicolan–16–*ß*–ol synthase were achieved using a simplified biomolecular modelling strategy. The applied refinement methodologies could rapidly generate a reliable protein–ligand complex, which provides for an accurate *in silico* identification of catalytically relevant amino acids. Guided by our modelling data, ACS mutations lead to the identification of the catalytically relevant ACS amino acid network I626, T657, Y658, A786, F789, and Y923. Our biomolecular modelling and mutational studies suggest that the ACS substrate cyclization occurs in a spatially restricted location of the enzyme's active site and that the geranylgeranyl diphosphate derived pyrophosphate moiety remains in the ACS active site thereby directing the cyclization process. This study demonstrates that a simple and rapid biomolecular modelling procedure can predict catalytically relevant amino acids.

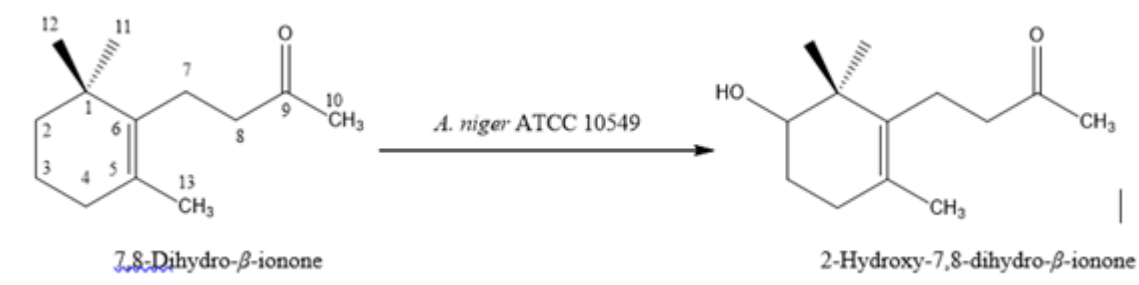
PS2-I-002

Fungal biotransformation of 7,8-dihydro-*β*-ionone

Özge Özşen Batur[1], Ismail Kiran[1], Fatih Demirci[2], K. Hüsnü Can Baser[3]

[1]Faculty of Science and Letters, Eskişehir Osmangazi University, Department of Chemistry, 26480, Eskişehir, Turkey, [2]Faculty of Pharmacy, Department of Pharmacognosy, Anadolu University, Eskişehir, Turkey, [3]Department of Pharmacognosy, Faculty of Pharmacy, Near East University, Lefkoşa, Cyprus

Flavour and fragrance compounds have a wide range of use in food, beverage, pharmaceutical industries and are consumed in a worldwide market. These compounds are mainly produced by either extraction from plant sources or by chemical syntheses. Biotechnological processes are a natural way of production, especially microbiological transformation reactions.
β-Ionone is a volatile cyclic norisoprenoid terpene found in natural products such as plant absolutes and extracts, which is a key intermediate for the synthesis of various vitamins. Ionones are used in the food and cosmetic industry due to their pleasant aroma. *β*-Ionone is found in flavour and fragrance compounds used in many daily products such as household products, pharmaceutical industries, cosmetics, shampoos, toilet soaps and other toiletries. Therefore, it has an annual production of several hundreds of tonnes.
In this present study, the objective was to obtain new derivatives from 7,8-dihydro-*β*-ionone for the biotechnological production of potential aroma compounds, through microbial biotransformation. For this purpose, experiments were conducted by using more than 15 microorganisms. Among them, biotransformation with the plant pathogenic fungus *Aspergillus niger* ATCC 10549 resulted in metabolization to the hydroxylated derivative in a 6.25% yield as shown in the scheme. The fragrant molecule was characterized by extensive chromatospectral techniques. Olfactory and biological evaluations of the metabolite were performed.



7,8-Dihydro-*β*-ionone

2-Hydroxy-7,8-dihydro-*β*-ionone

PS2-I-003

Fabrication of porous hybrid scaffolds for tissue engineering from gelatin and the marine polysaccharide ulvan

Leto-Aikaterini Tziveleka[1], Andreas Sapalidis[2], Niki Chondrogianni[3], Efstathia Ioannou[1], Vassilios Roussis[1]

[1]Section of Pharmacognosy and Chemistry of Natural Products, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, 15771 Athens, Greece, [2]Institute of Nanosciences and Nanotechnology, NCSR "Demokritos", Aghia Paraskevi 15310, Attiki, Greece, [3]Institute of Biology, Medicinal Chemistry and Biotechnology, National Hellenic Research Foundation, 48 Vassileos Constantinou Ave, 11635 Athens, Greece

Bioapplication-oriented materials have to be biocompatible, biodegradable, with chemical tuning ability and sustainability for being considered as promising materials in this field. Therefore, natural biopolymers are a valuable source of such materials. In particular, the negatively charged sulfated polysaccharides comprise a valuable category of compounds due to the plethora of bioactivities associated with them.
Ulvan is recognized as a natural marine polysaccharide of biomedical interest. Various ulvan-based biomaterials have been prepared and studied according to their structural characteristics for different biomedical applications. On the other hand, gelatin, a natural biopolymer obtained from the partial hydrolysis of collagens, has been widely used as biocompatible material due to its generally recognized safety by the US Food and Drug Administration.
Within the framework of our research towards the fabrication of biopolymer-based materials for biomedical applications, a series of novel porous hybrid structures based on chemically cross-linked ulvan and gelatin were prepared and characterized. The measured IR spectra were characteristic of the constituents and the peak intensities were analogous to the ratio of the two biopolymers used. Thermogravimetric analysis provided quantitative measurement of mass change in the prepared materials associated with dehydration, decomposition and oxidation. Additionally, their mechanical performance, water-uptake ability, weight loss upon incubation in PBS at 37 °C and biocompatibility were assessed, while their morphology was analyzed using scanning electron microscopy. The obtained results indicate the potential use of the prepared ulvan/gelatin hybrid structures as prospective scaffolds in tissue engineering.

Acknowledgements: This work was partially supported by the research program MARINOVA.

Keywords: Ulvan, gelatin, hybrid scaffolds, tissue engineering, biomaterials

PS2-I-004

Camelina sativa press cake and oil for production of vegan omega 3 and bioactive glucosinolates

Chiara Magoni[1], Francesco Saliu[2], Sofia Cavini[1], Lorenzo Guzzetti[1], Ilaria Bruni[1], Massimo Labra[1]

[1]Department of Biotechnology and Bioscience, University of Milano-Bicocca, , Italy, [2]Department of Earth and Environmental Sciences, University of Milano-Bicocca, , Italy

Camelina sativa (L.) Crantz is an oilseed crop belonging to Brassicaceae, distribute worldwide. This species grows well in poor soils and does not require great maintenance when cultivated at high density. The composition of the *C. sativa* oil makes it suitable for numerous uses such as biodiesel and feed. Among the byproducts of *C. sativa* there is press cake, which is reused in animal feed, an item in which the glucosinolate levels are limited by law. The press cake is known to be rich in glucosinolates, molecules that are hydrolyzed by myrosinases to produce different forms of isothiocyanates, some of which have anti-tumor activity. The analysis of the effects of *C. sativa* glucosinolates will allow to evaluate not only the biological effect of these phytocomplexes, but also the industrial feasibility of glucosinolates extraction from feeding products. On the other hand, our work aims to enhance the oil of *C. sativa*, that naturally occurs without eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), through a oleaginous microorganisms biotransformation and via immobilized enzymes. Such treatment allows the production of polyunsaturated fatty acids (EPA and DHA), to respond to the vegan omega 3 request. These issues are part of the Food Social Sensor Network project (Food Net), granted by Program POR FESR 2014-2020 – Accordi per la Ricerca e l'Innovazione, aimed at producing functional foods for the deficiencies of elderly healthy patients, such as the low omega 3 intake.

PS2-I-005

Artichoke waste as a source of inuline: a strategy to valorize the agricultural supply chain and its by-products

Sofia Cavini[1], Francesca Givoia[1], Lorenzo Guzzetti[1], Ilaria Bruni[1], Chiara Magoni[1], Massimo Labra[1]

[1]*Department of Biotechnology and Biosciences, University of Milano-Bicocca, Piazza della Scienza 2, 20126 Milan, Italy*

The agricultural supply chain produces a large amount of agricultural waste biomasses. Recently, this waste is more and more used as the target of valorization strategies due to its content in high value-added molecules for cosmetics and medical purposes. A class of molecules having an important effect on human health are the fructans (i.e., polysaccharides composed by monomers of fructose). One of the most important polysaccharide, is inulin. In this molecule, β-D-fructose forms a chain by β (2,1) bonds linkage, making it one of the most important prebiotic sugars used at the industrial scale. This polysaccharide is abundant within the Asteraceae family, to which many horticultural species, such as the artichoke belong. Artichoke is an important product in Italy, that is the world leading producer of this vegetable. Its production, however, is concentrated almost exclusively on the consumption of the edible flowers, which represents just the 30% of whole biomass of the plant. The rest is considered waste, and in most of the cases, left on the cultivation field or at least exploited as feed for livestock. Through this work we show that artichoke waste extracts have an inulin content reaching 70%. In fact, waste products deriving from the artichoke agricultural supply chain can be considered a potential big resource of this prebiotics molecules. After the development of reliable strategies for the extraction of inulin from these biomasses, the work focused on the definition of environmental storage variables able of influencing the inulin content and its polymerization degree. Different polymerization degrees of inulin can impact on the prebiotic properties of this polymer. Therefore, it is important to know how different storage conditions can affect extraction final yields.

PS2-I-006

Metabolically engineered *E. coli* able to produce the highly valuable hydroxytyrosol and derivatives

Emmanouil Trantas[1], Theodora Nikou[2], Maria Halabalaki[2], Leandros Skaltsounis[2], Filippos Ververidis[1]

[1]Plant Biochemistry and Biotechnology Group, Laboratory of Biological and Biotechnological Applications, Department of Agriculture, School of Agriculture, Food and Nutrition, Technological Educational Institute of Crete, P.O. 1939, GR 710 04, Heraklion, Greece, [2]Division of Pharmacognosy and Natural Product Chemistry, Department of Pharmacy, National and Kapodistrian University of Athens, Panepistimiopolis Zografou, GR-15771, Athens, Greece

Hydroxytyrosol (HT, 3,4-dihydroxyphenylethanol) is one of the most characteristic compounds of olive oil as well as olive drupes and leaves, with significant contribution to their qualitative characteristics; it was recently approved as a part of "olive oil healthy polyphenols", by EC Regulation 432/2012. Beyond the fact that HT is highly priced in the markets (highly pure form is priced at around 2860 €/gr), it has powerful antioxidant properties, antimicrobial and antiviral activity and is credited with several interesting beneficial properties for human health, such as possessing high capacity for free radical scavenging, reducing the risk of cancer, coronary heart disease and atherosclerosis, having critical effects on the formation and maintenance of bones, being used as an effective remedy in the treatment of osteoporosis symptoms, etc. However, HT biosynthetic pathway in olives is not fully decoded yet. Based on its structure, it can be presumed that HT might be derived from tyrosine which is then biotransformed to HT through sequential biosynthetic steps involving a hydroxylation, a decarboxylation, and a deamination reaction. Recent research data showed that *Escherichia coli* initially genetically engineered to overproduce the precursor molecule tyrosine and then encompassing appropriate genes encoding respective enzymes for hydroxylation, decarboxylation, and deamination reactions, was able to de-novo produce HT. Such a redirection of the carbon flow towards the production of HT directly from glucose, achieved a reasonable high HT concentration in the crude extract. Various gene combinations derived from plants or bacteria were used to form a novel, artificial biosynthetic dual pathway managing to redirect the carbon flow towards the production of HT directly from glucose. Various biosynthetic bottlenecks faced due to feaB gene function, resolved through the overexpression of a functional aldehyde reductase (ALR-K). Currently, we have achieved equimolar concentration of HT to tyrosine as precursor when overproduced straight from glucose, reaching the level of 1.76 mM (270.8 mg/L) analyzed by LC-HRMS.

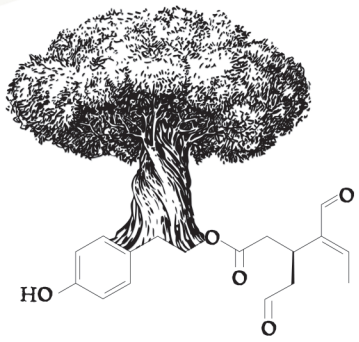
PS2-I-007

Determination of urease and catalase activities in soil samples and their incidence in the significant learning of enzymatic kinetics

Jairo Granados[1], Dolffi Rodríguez[2]

[1]Chemistry department, University of Cundinamarca, Fusagasugá, Colombia, [2]UNAD, School of basic sciences, technologies and engineering, Bogotá, Colombia

The teaching-learning of enzymatic kinetics is difficult, due to its complexity and high conceptual structure, one of these resides in the poor argumentation of students about the nature and speed of chemical reactions, due to their lack of foundation in Theories of the activated complex and collisions [1], the other, is related to the forms of spontaneous reasoning, based on functional fixation and reduction, which can act as epistemological and methodological barriers. The present study was carried out with two groups of students of agrarian sciences, enrolled in Metabolic Biochemistry (MB). A quasi-experimental test-post-test design [3] was applied, guided by a research-based metacognitive didactic strategy (MDS) and supported by laboratory practices, which focused on soil sampling, physicochemical analysis, extraction of enzymes and kinetic study of urease and catalase [3]; Results and Discussion: the major conceptual change was presented in concepts such as reaction order, activated complex theory and kinetic equation, while greater difficulty was perceived in concepts such as: activation energy, Michaellis constant and reaction mechanism; with respect to the procedural component, the students determined optimal pH, Michaelis constants, maximum speed, activation energy and kinetic equations for Urea and catalase extracted from soils coming from Sumapaz and Boyaca regions; urease exhibited better activity at a pH of 7.0 with an average maximum speed of 2.217 µmol urea / g.min and Km of 9.745 µmol / mL; Conclusions: Significant conceptual and methodological progress ($P < 0.05$) was observed in the average scores of the instruments applied, which is related to the type of strategy used, since it considered the students' previous knowledge Likewise, a very proactive attitude towards the learning of this topic was evidenced.



Professional Congress Organizer

AFEA Travel and Congress Services

AFEA S.A. Travel & Congress Services
Professional Congress Organizer
39-41 Lykavittou Str., 10672 Athens
Tel: +30 210 3668854, Fax: +30 210 3643511
website: www.iscnp30-icob10.org
e-mail: info@iscnp30-icob10.org