Conference Book

20th International Congress of the International Society for Ethnopharmacology

Virtual Congress

18-20 April 2021

www.ethnopharmacology2021.org

ISE

Local Organiser

ARISTOTLE UNIVERSITY OF THESSALONIKI
FACULTY OF HEALTH SCIENCES
School of Pharmacy

Co-organisers

Hellenic Ethnopharmacology Society

HELLENIC REPUBLIC
National and Kapodistrian University of Athens

UNIVERSITY OF PATRAS, DEPARTMENT OF PHARMACY

Under the Auspices

HELLENIC REPUBLIC
MINISTRY OF INTERIOR
(MACEDONIA-THRACE)

20th International Congress of the International Society for Ethnopharmacology

Virtual Congress

18-20 April 2021

ISE

www.ethnopharmacology2021.org

WELCOME LETTER

Dear friends and colleagues,

On behalf of the International Society for Ethnopharmacology (ISE) it is a special privilege and a pleasure to welcome you to the 20th International Congress of the International Society for Ethnopharmacology (ISE), which will be held from April 18th – 20th, 2021 on a Virtual Congress Platform.

Greece, that was to host the physical Congress, is one of the most influential cultures in European and Mediterranean area. Through the emblematic figures of Hippocrates, Dioscorides and later on, with the Byzantine physicians, Greece has deeply influenced the European Traditional Medicine and has left a legacy which is traced down today in modern Pharmacopoeias. A common feature of this rich heritage of traditional medicine, which covers three millennia, is the wealth of the Greek Flora, endowed with more than 6500 plant species and still under exploration.

ISE International Virtual Congress 2021 brings together renowned scientists from all over the world and we hope you will enjoy your active participation in this exciting event. The congress aims to increase our knowledge on traditional medicinal products the influence of herbal medicinal products in the Pharmacopoeias and the role of natural products in drug discovery, nutrition and applied chemistry. The scientific program of the main conference includes plenary lectures and invited short lectures, contributed short lectures and posters.

On behalf of the Universities of Thessaloniki, Athens and Patras and the Hellenic Society of Ethnopharmacology,

We welcome you to the ISE 2021 International Virtual Congress and wish you a fruitful meeting!

Karioti Anastasia

Assistant Professor
Department of Pharmacognosy-Pharmacology
Aristotle University of Thessaloniki, Greece

Lazari Diamanto

Associate Professor
Department of Pharmacognosy-Pharmacology
Aristotle University of Thessaloniki, Greece

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ORGANIZING COMMITTEE

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University of London, United Kingdom

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Università di Cagliari, Italy

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Jadavpur University, India

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Aristotle University of Thessaloniki

Konstantinos Xanthopoulos
Aristotle University of Thessaloniki

SCIENTIFIC COMMITTEE

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Aristotle University of Thessaloniki

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Karl-Franzens-Universität Graz, Austria

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Vasileios Roussis
National and Kapodistrian University of Athens

Olga Tzakou
National and Kapodistrian University of Athens

Caroline Weckerle
University of Zurich, Switzerland

Zhao Zhongzhen
Hong Kong Baptist University, China

KEYNOTE SPEAKERS

Prof. **Anna Rita Bilia,** University of Florence, Italy

Prof. **Alessandra Braca,** University of Pisa, Italy of America

Assoc. Prof. **Angela Calderon,** Auburn University, United States

Assist. Prof. **Ilias Giannenas,** Aristotle University of Thessaloniki, Greece

Prof. **Chris Hogstrand,** King's College London, United Kingdom

Dr. **Eleni Maloupa,** Hellenic Agricultural Organization – Demeter-ELGO, Greece

Prof. **Helen Skaltsa,** National and Kapodistrian University of Athens, Greece

Prof. **Hermann Stuppner,** University of Innsbruck, Austria

Prof. **Alvaro Viljoen,** Tshwane University of Technology, Pretoria, South Africa

Prof. **Merlin Willcox,** University of Southampton, United Kingdom

Prof. **Günter Vollmer,** Technische Universität Dresden, Germany

INVITED SPEAKERS

Assoc. Prof. **Nektarios Aligiannis,** National and Kapodistrian University of Athens, Greece

Prof. **Rudolf Bauer,** University of Graz, Switzerland

Prof. **Mariangela Caroprese,** University of Foggia, Italy

Prof. **Ioanna Chinou,** National and Kapodistrian University of Athens, Greece

Prof. **Raymond Coopaer,** The Hong Kong Polytechnic University

Prof. **Javier Echeverría,** University of Santiago, Chile

Prof. **J.N. (Kobus) Eloff,** University of Pretoria, South Africa

Prof. **Dimitris Fatouros,** Aristotle University of Thessaloniki, Greece

Prof. **Emerson Ferreira Queiroz,** University of Geneva, Switzerland

Dr. **Bertrand Graz,** University of Geneva and Antenna Foundation, Switzerland

Prof. **Namrita Lall,** University of Pretoria, South Africa

Assoc. Prof. **Marco Leonti,** University of Cagliari, Italy

Dr. **Chia-Hsin Lin,** China Medical University

Prof. **Pulok K. Mukherjee,** Jadavpur University, Kolkata, India

Prof. **Stergios Pirintsos,** University of Crete, Greece

Prof. **Hung – Rong Yen,** China Medical University

THEMATIC TOPICS

> Application of hyphenated techniques in quality assurance of ethnopharmacological sources
> Aromatic and medicinal plants in animal health: ethnopharmacological approach
> Aromatic and medicinal plants of the Mediterranean basin as food and dietary supplements
> Conservation and sustainable use of herbal resources
> Ethnobotanical approach to drug discovery: Theory, methodology and limitations
> Formulation of Natural products and medicinal plants: from traditional to modern phytotherapeutics
> Greek ethnopharmacology
> Pharmacological and clinical studies of medicinal plants & natural products
> Traditional use of plants and herbal pharmacopoeias: the contribution of the Mediterranean flora

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Graphics design, DTP: Kyriakos Zagliveris
IT Coordinator: George Kanakaris

SHORT PROGRAM

Agenda time zone: East European Summer Time

Sunday 18 April

10:00-11.00	Opening of the Congress and Welcome Ceremony
11:00-11:45	Plenary lecture
11:45-12:15	Break
12:15-13:50	Plenary lectures: Pharmacological and clinical studies of medicinal plants & natural products
13:50-14:30	Lunch
14:30-15:30	Poster session 1
15:30-18:30	Parallel Sessions

Breakout 1		Breakout 2	
15:30-16:45	ISE-GA African workshop	15:30-16:45	Greek Ethnopharmacology
16:45-17:15	Break	16:45-17:15	Break
17:15-19:00	ISE-GA African workshop	17:15-18:30	Greek Ethnopharmacology

Monday 19 April 2021

10:00-12:00	Plenary lectures - Short lectures: Applications of Hyphenated Techniques in quality assurance of ethnopharmacological sources.
12:00-12:30	Break
12:30-14:00	Plenary lectures - Short lectures: Pharmacological and clinical studies of medicinal plants & natural products
14:00-15:00	Virtual Guided Thessaloniki Tour
15:00-16:00	Poster session 2
16:00-18:45	Parallel Sessions

Breakout 1		Breakout 2	
16:00-18:00	Aromatic and medicinal plants in animal health: ethopharmacological approach	16:00-16:45	Conservation and sustainable use of herbal resources

		16:45 - 17:45	Formulation of natural products and medicinal plants: from traditional to modern phytotherapeutics
		17:45-18:00	Break
		18:00-18:45	Ethnobotanical approach to drug discovery

Tuesday 20 April 2021

10:00-11:30	Parallel Sessions		
Breakout 1		Breakout 2	
10:00-11:30	Ethnobotanical approach to drug discovery	09:30-10:30	Traditional use of plants and herbal pharmacopoeias
		10:30-11:30	Pharmacological and clinical studies of medicinal plants &natural products
11:30-12:00	Break		
12:00-13:00	Journal of Ethnopharmacology Workshop		
13:00-14:00	Lunch		
14:00-15:00	Poster session 3		
15:00-17:00	Aromatic and medicinal plants of the Mediterranean basin as food and dietary supplements		
17:00-18:00	Closing Ceremony		

DETAILED PROGRAM

Agenda time zone: East European Summer Time

Sunday 18 April

10:00 - 11:00	**Opening of the Congress and Welcome Ceremony**
11:00 - 11:45	**Plenary lecture**
	From Hippocrates to the modern Phytotherapy. Prof. H. Skaltsa *National & Kapodistrian University of Athens, School of Pharmacy, Department of Pharmacognosy & Chemistry of Natural Products, Greece*
11:45 - 12:15	**Break**
12:15 - 13:50	**Plenary lectures: Pharmacological and clinical studies of medicinal plants & natural products** Moderator: Prof. Rudolf Bauer
	Designing clinical trials of herbal medicines as alternatives to antimicrobials. M. Willcox, C. Simpson, M. Logue, XY. Hu, A. Flower, B. Stuart, P. Little, M. Moore *University of Southampton, United Kingdom*
	Study of the mechanism of antitubercular and antiplasmodial activities of plants and natural products using mass spectrometry-based assays. Assoc. Prof. A.I. Calderón *Department of Drug Discovery and Development, Harrison School of Pharmacy, Auburn University, AL, USA*
	Traditional Chinese Medicine for Cancer Patients in Taiwan. Prof. H.-R. Yen *School of Chinese Medicine, College of Chinese Medicine, China Medical University, Taichung, Taiwan* *Chinese Medicine Research Center, China Medical University, Taichung, Taiwan* *Department of Chinese Medicine, China Medical University Hospital, Taichung, Taiwan*
	Ethnopharmacological investigations of Vietnamese medicinal plants used for the treatment of inflammation and cancer. H. Thi Tran, T. Thanh Nguyen, N. Kretschmer, E.-M. Pferschy-Wenzig, R. Bauer *Institute of Pharmaceutical Sciences, Department of Pharmacognosy, University of Graz, Austria*
13:50 - 14:30	**Lunch**

14:30 15:30	**Poster session 1** **Aromatic and medicinal plants of the Mediterranean basin as food and dietary supplements.** **Ethnobotanical approach to drug discovery: Theory, methodology and limitations** **Greek ethnopharmacology.** **Traditional use of plants and herbal pharmacopoeias: the contribution of the Mediterranean flora.** **Pharmacological and clinical studies of medicinal plants & natural products.**	
15:30 19:00	**Parallel Sessions**	
15:30 19:00	**ISE-GA African workshop** **Moderator: Dr. Cica Vissienon, African Research Network (on medicinal plants and natural product research) – Mission, Opportunities, Registration**	**15:30 18:30** **Greek Ethnopharmacology** **Moderator: Prof. Helen Skaltsa**
	Reverse pharmacology and traditional medicine Dr. B. Graz *University of Geneva and Antenna Foundation, Switzerland*	**Expanding the scope of Greek Ethnopharmacology - eurasian medicinal plants on the Mediterranean.** A. Touwaide[1], E. Appetiti[2] [1] *Division of Humanities, UCLA, Los Angeles, USA* [2] *Institute for the Preservation of Medical Traditions, Washington, DC, USA*
	Multidirectional pharma-toxicological investigations on *Harpagophytum procumbens* DC. ex Meisn.: focus on water extract activity. G. Orlando *Department of Pharmacy, University "G. d'Annunzio" of Chieti-Pescara, Italy*	**Herbal remedies in an Alexandrian manuscript of late antiquity.** M. Marselos *Department of Pharmacology, Medical Faculty, School of Health Sciences, University of Ioannina, Greece*
	The antifibrotic potential and pharmacokinetic assessment of ***Euclea natalensis*** A.DC. as a hepatoprotective agent. A.M. Reid, N. Lall *Plant and Soil Sciences, University of Pretoria, South Africa*	**Galen's formulas in his book on antidotes.** C. Grivas[1], E. Appetiti[2], A. Touwaide[3] [1] *Akadimia of Ancient Greek and Traditional Chinese Medicine, Athens, Greece* [2] *Institute for the Preservation of Medical Traditions, Washington, DC, USA* [3] *Division of Humanities, UCLA, Los Angeles, USA*

The holistic anti-ageing potential of *Persicaria senegalensis* (Meisn.) Soják ethanolic and fermented extracts.

B. D. Payne[1], C.-H. Lin[2], N. Lall[1,2]

[1] *Medicinal Plant Science, Department of Plant and Soil Sciences, University of Pretoria, South Africa*
[2] *School of Natural Resources, University of Missouri, Columbia, USA*

16:45-17:15	**Break**	16:45-17:15	**Break**
	Innovative strategies for the efficient isolation of natural products and discovery of potential new leads. E.F. Queiroz, JL. Wolfender *Institut des Sciences Pharmaceutiques de Suisse Occidentale (ISPSO), Section des Sciences Pharmaceutiques, Université de Genève, Switzerland*		**Exploration of Traditional Herbal Knowledge in the South Balkan and East Mediterranean region for the discovery of bioactive natural products.** N. Aligiannis *Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Greece*
	Anthelmintic screening of five *Palmatilobae* species used in Traditional Medicine in Katanga Province (DR Congo). M.G. Nzuzi[1,2,3], N.W. Muyumba[2,4], M.L. Gauthier[2], S.K. Panda[5], W. Luyten[5], A. Nachtergael[2], S. Vandenput[3], S.A. Bakari[6], E.V. Okombe[1], P. Duez[2] [1] *Faculty of Veterinary Medicine, Unit of Pharmacology and Therapeutic, University of Lubumbashi (UNILU), DR Congo* [2] *Unit of Therapeutic Chemistry and Pharmacognosy, University of Mons (UMONS), Belgium* [3] *Bibliothèque des Sciences de la vie, University of Liège (ULiège), Belgium* [4] *Department of Chemistry-physics, High college training teachers of Lubumbashi, DR Congo* [5] *Animal Physiology and Neurobiology Section, Department of Biology, KU Leuven, Belgium* [6] *Laboratory of Pharmacognosy, Faculty of Pharmacy, University of Lubumbashi (UNILU), DR Congo*		***Origanum dictamnus*: phytochemical analyzes and relaxing effect on rabbit intestine.** C. Paloukopoulou[1], S. Govari[1], A. Soulioti[1], G. Tasi[1], T. Psarra[2], M. Koutsoviti-Papadopoulou[2], A. Karioti[1] [1] *School of Pharmacy, Aristotle University of Thessaloniki, Greece* [2] *School of Veterinary Medicine, Aristotle University of Thessaloniki, Greece*

Bio-guided fractionation and in vitro anti-inflammatory potential of Entada Africana from Benin used in the management of pain related-conditions.

M. Codo-Toafode[1,4], V. Spiegler[2], C. Vissiennon[3,6], V. Ahyi[1], A. Hensel[2], K. Fester[4,5]

[1] University of IRGIB Africa, Inter-Regional University of Industrial Engineering, Biotechnologies and Applied Sciences, Cotonou, Benin
[2] University of Münster, Institute for Pharmaceutical Biology and Phytochemistry (IPBP), Germany
[3] University of Leipzig, Institute for Medical Physics and Biophysics, Germany
[4] University of Leipzig, Institute of Pharmacy, Germany
[5] University of Applied Sciences Zittau/Görlitz, Faculty of Natural and Environmental Sciences, Germany
[6] REPHA GmbH Biologische Arzneimittel, Langenhagen, Germany

In the course of thyme: the use of *Thymus* spp. in European traditional medicine & the example of *Thymus thracicus*.

T.K. Papagrigoriou[1], O.St. Tsiftsoglou[1], D.I. Hajipavlou-Litina[2], D.M. Lazari[1]

[1] Aristotle University of Thessaloniki, Faculty of Health Sciences, School of Pharmacy, Laboratory of Pharmacognosy, Greece
[2] Aristotle University of Thessaloniki, Faculty of Health Sciences, School of Pharmacy, Department of Pharmaceutical Chemistry, Greece

A natural choice for antimicrobial protection using an indigenous South African plant for acne vulgaris.

I.A. Lambrechts, N. Lall

Plant and Soil Sciences, University of Pretoria, South Africa

Discovering herbal teas/mountain teas in Kozani-Western Macedonia-Greece, an ethnopharmacological journey.

E. Lampretsa

Earth's Pharmacy, Kozani, Greece

The Regulatory aspects and challenges for commercialisation of the natural Products.

Prof. N. Lall

Plant and Soil Sciences, University of Pretoria, South Africa

Ethnopharmacological study of the medicinal plants used in the folk medicine in Drama, Nothern Greece.

S. Tsiotsiou-Papaemmanouil[1], D. Lazari[1], D. Hadjipavlou-Litina[2], T. Dardavesis[3]

[1] Laboratory of Pharmacognosy, School of Pharmacy, Faculty of Health Sciences, Aristotle University of Thessaloniki, Greece
[2] Laboratory of Pharmaceutical Chemistry, School of Pharmacy, Faculty of Health Sciences, Aristotle University of Thessaloniki, Greece
[3] Laboratory of Hygiene, Medical School, Faculty of Health Sciences, Aristotle University of Thessaloniki, Greece

Monday 19 April 2021

10:00 - 12:00 **Plenary lectures - Short lectures: Applications of Hyphenated Techniques in quality assurance of ethnopharmacological sources.**

Moderator: Assist. Prof. Maria Halabalaki

Modern analytical approaches for Natural Products Analysis.

Prof. H. Stuppner

University of Innsbruck, Austria

The application of classic and modern pharmacognosy in the study of African traditional medicines.

A.M. Viljoen, S. Combrinck, I. Vermaak, M. Sandasi

Department of Pharmaceutical Sciences and SAMRC Herbal Drugs Research Unit, Faculty of Science, Tshwane University of Technology, Pretoria, South Africa

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TCM needs TQM: New applications using NMR for standardizing botanical mixtures.

Prof. R. Cooper

The Hong Kong Polytechnic University, Hong Kong SAR China

Qualitative and quantitative characterization of chemical profiles in three different parts of *Poria cocos*.

L. Zhu, J. Xu , X. Dong, Z. Zhao

School of Chinese Medicine, Hong Kong Baptist University, Kowloon, Hong Kong Special Administrative Region, China

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PEGASUS: An analytical chemometrics platform for the discovery of bioactive natural compounds.

A. Cheilari[1], A. Vontzalidou[1], Vl. Boka[1], D. Benaki[2], N. Aligiannis[1]

[1] *Department of Pharmacognosy and Natural Products Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Greece*

[2] *Department of Pharmaceutical Chemistry, Faculty of Pharmacy, National and Kapodistrian University of Athens, Greece*

12:00 - 12:30 **Break**

12:30 - 14:00 **Plenary lectures - Short lectures: Pharmacological and clinical studies of medicinal plants & natural products**

Moderator: Prof. Günter Vollmer

Functional consequences of stimulation of multiple biological targets by botanical extracts and their bioactive compounds – hops as an example.

Prof. G. Vollmer

Faculty of Biology, Technische Universität Dresden, Germany

Risk assessment of chemicals that occur as mixtures: additivity and interactions.

Prof. C. Hogstrand

King's College London, School of Life Course Sciences, London, UK

***Artocarpus tonkinensis,* a remedy from Vietnamese ethnopharmacology, protects mice against collagen-induced arthritis and decreases Th17 cell function.**

S. Adorisio[1], I. Muscari[2], T.T. Thuy[3], D.V. Delfino[1,2]

[1] *Foligno Nursing School, Italy*
[2] *Secion of Pharmacology, Department of Medicine, University of Perugia, Italy*
[3] *Institute of Chemistry, Vietnam Academy of Science and Technology, Hanoi, Vietnam*

Investigation on role of Indigenous medicinal herb of North East India for the management of dysregulated glutathione biosynthesis associated with metabolic syndrome.

B. Deka[1,2], P. Manna[1], J.C Borah[1], N.C Talukdar[1,2]

[1] *Biochemistry and Drug Discovery Lab, Institute of Advanced Study in Science and Technology (IASST) PaschimBoragaon, Guwahati-35, India*
[2] *Cotton University, Panbazar, Guwahati-01, India*

14:00 - 15:00 **Virtual Guided Thessaloniki Tour**

15:00 - 16:00 **Poster session 2**

Application of hyphenated techniques in quality assurance of ethnopharmacological sources.

Aromatic and medicinal plants in animal health: ethnopharmacological approach.

Conservation and sustainable use of herbal resources.

Formulation of Natural products and medicinal plants: from traditional to modern phytotherapeutics.

Miscellaneous.

Pharmacological and clinical studies of medicinal plants & natural products.

16:00 - 18:45 **Parallel Sessions**

16:00 - 18:00 **Aromatic and medicinal plants in animal health: ethopharmacological approach**

Moderator: Assist. Prof. Ilias Giannenas

Challenges and success in using plants to control internal parasites in animals

Prof. Dr. J. N. Eloff

University of Pretoria, South Africa

16:00 - 16:45 **Conservation and sustainable use of herbal resources**

Moderator: Assoc. Prof. Diamanto Lazari

Safeguarding threatened species and promoting the use of plant resources in research and development.

Dr. E. Maloupa

Hellenic Agricultural Organization-DEMETER, Institute of Plant Breeding and Genetic Resources, Laboratory of Protection and Evaluation of Native and Floricultural Species, Thessaloniki, Greece.

How can we manage dairy animal health and welfare?

Prof. M. Caroprese

University of Foggia, Italy

Understanding local knowledge as integral factor in designing strategies for sustainably conserving and using herbal resources.

Y.M. Scherrer[1], M. Heinrich[2]

[1] Sustainability Research Group, University of Basel, Switzerland
[2] Research Group 'Pharmacognosy and Phytotherapy', UCL School of Pharmacy, London, UK

The use of aromatic plants, extracts and essential oils as feed additives in animal nutrition.

Assist. Prof. I. Giannenas

EBVS® European Specialist in Poultry Veterinary Science
Laboratory of Nutrition, Faculty of Veterinary Medicine, School of Health Sciences, Aristotle University of Thessaloniki, Greece

16:45
17:45

Formulation of natural products and medicinal plants: from traditional to modern phytotherapeutics

Moderator: Assoc. Prof. Diamanto Lazari

Current status of ethnoveterinary surveys conducted in South Africa.

L.J. McGaw[1], I.M. Famuyide[1], E.T. Khunoana[1], A.O. Aremu[2]

[1]Phytomedicine Programme, Department of Paraclinical Sciences, University of Pretoria, South Africa
[2]Indigenous Knowledge Systems (IKS) Centre, Faculty of Natural and Agricultural Sciences, North West University, Mmabatho, South Africa

Evaluation of sesquiterpenes as permeation enhancers for transdermal delivery

Prof. D. Fatouros

School of Pharmacy, Aristotle University of Thessaloniki, Greece

Of Elephants and Men: From animal observation to veterinary and human pharmacopeia in Laos.

JM. Dubost

Museum National d'Histoire Naturelle-UMR 7206, Paris, France
UMR 152 Pharmadev, Université de Toulouse, IRD, UPS, France

Natural products loaded in nanocarriers to cross biological barriers.

Prof. AR. Bilia

Department of Chemistry "Ugo Schiff", Florence, Italy

Effect of dietary aromatic plants and essential oils in combination with tributyrin on broiler chicken meat composition and oxidative stability.

E. Bonos[1], E. Sidiropoulou[2], I. Giannenas[2], A. Karamoutsios[1], O. Tsiftsoglou[3], D. Lazari[3], E. Christaki[2], I. Skoufos[1], A. Tzora[1]

[1] *Department of Agriculture, School of Agriculture, University of Ioannina, Kostakioi, Arta, Greece.*
[2] *Laboratory of Nutrition, School of Veterinary Medicine, Faculty of Health Sciences, Aristotle University of Thessaloniki, Greece.*
[3] *Laboratory of Pharmacognosy, School of Pharmacy, Faculty of Health Sciences, Aristotle University of Thessaloniki, Greece.*

17:45 - 18:00 **Break**

18:00 - 18:45 **Ethnobotanical approach to drug discovery**

Moderator: Prof. Michael Heinrich

Archaeoethnopharmacological studies in South America: back to the roots of ethnopharmacology?

Dr. J. Echeverría

Departamento de Ciencias el Ambiente Facultad de Química y Biología, Universidad de Santiago de Chile

Plants used as ethnomedicine for rheumatic disorders by Williche people.

Dr. R. Saaverdra-Rodríguez

Departamento de Ciencias Básicas, Universidad Santo Tomás, Osorno, Chile

Tuesday 20 April 2021

10:00 - 11:30	Parallel Sessions		
10:00 - 11:30	**Ethnobotanical approach to drug discovery** Moderator: Assoc. Prof. Marco Leonti	09:30 - 10:30	**Traditional use of plants and herbal pharmacopoeias** Moderator: Prof. Pulok K. Mukherjee
	Recommended standards for conducting and reporting ethnopharmacological field studies. M. Leonti *Department of Biomedical Sciences, University of Cagliari, Italy*		**Pharmacopoeias on herbal medicine in India.** Prof. P.K. Mukherjee *Professor, Dept. of Pharmaceutical Technology, Jadavpur University, India*
	Traditional Chinese Medicine: A Global Exchange. E.R. Qi, E. Brand, M. Zhou, Z. Zhao *School of Chinese Medicine, Hong Kong Baptist University, Kowloon Tong, Hong Kong SA*		**Traditional Herbal Resources and Herbal Pharmacopoeia: the contribution of the Taiwanese flora** Dr. Chia-Hsin Lin *China Medical University, Taiwan*
		10:30 - 11:30	**Pharmacological and clinical studies of medicinal plants & natural products**
	Nagoya protocol and the need for new forms of collaboration. F. Scotti[1], M. Berger[2], A. Hesketh[3], M. Hitziger[4], M. Heinrich[1] [1] *UCL School of Pharmacy, London, UK* [2] *Centro de Estudios en Salud, Universidad del Valle de Guatemala* [3] *Indigena Biodiversity Limited, Gerrards Cross, UK* [4] *CITES Secretariat, Palais des Nations, Genève 10, Switzerland*		**Efficacy of honey-based ophthalmic formulation in patients with corneal ulcer: A randomized clinical trial.** M. Nejabat, K. Soltanzadeh, M. Yasemi, S. Daneshamouz, AR Akbarizadeh, M. Heydari *Shiraz University of Medical Sciences, Iran*

Mitigating climate change in Valmalenco (so, Italy): how can Ethnobotany contribute?

M. Bottoni[1,2], C. Giuliani[1,2], L. Colombo[1,2], F. Milani[1,2], K. Nallio[1,2], P.S. Colombo[1,2], P. Bruschi[3], G. Fico[1,2]

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[3] *Department of AgriFood Production and Environmental Sciences, University of Florence, Italy*

Wound-healing activity of essential oils from five *Hypericum* spp. growing wild in Greece.

M.E. Grafakou[1], A. Diamanti[1], E. Simirioti[2], A. Terezaki[2], C. Barda[1], I. Sfiniadakis[3], M. Rallis[2], H. Skaltsa[1]

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Ethnopediatric knowledge among mothers living in Southern Romania.

M. Petran, M. Gilca

Department of Functional Sciences 1- Biochemistry, Carol Davila University of Medicine and Pharmacy, Bucharest, Romania

In vitro assessment of anti-influenza activity and neuraminidase inhibition of *Cistus creticus* L. subsp. *eriocephalus* (Viv.) Greuter & Burdet grown in sea coast area of South Tuscany, Italy.

M. Biagi[1], G. Baini[1], E. Miraldi[1], C. Terrosi[2], M.G. Cusi[2]

[1] *University of Siena, Department of Physical Sciences, Earth and Environment, Italy*
[2] *University of Siena, Microbiology Unit, Department of Medical Biotechnologies, Italy*

Antiprion effects of naturally occurring compounds.

A. Karsanidou[1], E. Kanata[1], A. Karioti[2], N. Bekas[3], A. Sachinidis[1], A.K. Kanellis[4], D. Dafou[3], K. Xanthopoulos[1], T. Sklaviadis[1]

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[4] Group of Biotechnology of Pharmaceutical Plants, Laboratory of Pharmacognosy, School of Pharmacy, Aristotle University of Thessaloniki, Greece

11:30-12:00	**Break**
12:00-13:00	**Journal of Ethnopharmacology Workshop** Mrs. Anne Pordon, Prof. Alvaro Viljoen
13:00-14:00	**Lunch**
14:00-15:00	**Poster session 3** **Pharmacological and clinical studies of medicinal plants & natural products.**
15:00-17:00	**Aromatic and medicinal plants of the Mediterranean basin as food and dietary supplements** Moderator: Assist. Prof. Anastasia Karioti

Chemical profile and nutraceutical features of some Mediterranean food plants.

Prof. A. Braca

Department of Pharmacy, University of Pisa, Italy

Evaluating Greek honey-bees' products (Honey, Propolis, Royal Jelly) through centuries, as vehicle of high value chemical and pharmacological potential.

Prof. I. Chinou

Laboratory of Pharmacognosy and Chemistry of Natural Products, Dept. of Pharmacy, National and Kapodistrian University of Athens

***Pistacia lentiscus* resin (Chios mastic gum): chemistry and biology.**

Assist. Prof. M. Halabalaki

Laboratory of Pharmacognosy and Natural Products Chemistry, Department of Pharmacy, University of Athens, Greece

A case of pharmaceutical development based on the therapeutic potential of the essential oil combination of three Cretan herbs (Cretan IAMA®).

Prof. S.A. Pirintsos

Department of Biology, University of Crete, Greece
Botanical Garden, University of Crete, Greece

Human gastrointestinal and colonic metabolisation study of olive secoiridoids using an in vitro continuous flow dialysis system.

M.E. Sakavitsi[1], A. Breynaert[2], A. Angelis[1], L. Pieters[2], N. Hermans[2], S. Mitakou[1], M. Halabalaki[1]

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17:00 - 18:00 **Closing Ceremony**

Keynote speeches

20th International Congress of the International Society for Ethnopharmacology

KS01. From Hippocrates to the modern Phytotherapy.

H. Skaltsa

National & Kapodistrian University of Athens, School of Pharmacy, Department of Pharmacognosy & Chemistry of Natural Products, Greece

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Plants have formed the basis of sophisticated traditional medicine systems that have been in existence for thousands of years, and their uses by many cultures have been extensively documented.

The most extensive source for the study of ancient pharmacology is *Corpus Hippocraticum*, which contains almost 1500 recipes. Early on, Hippocrates was celebrated as the founder of the pharmacological discipline and his name served as an anchor for the preservation of the pharmacological material. In antiquity, Pharmacology and Botany were intimately linked. Theophrastus's *Enquiry into Plants* (4th c. BC) includes much information about pharmacologically active plants, while Dioscorides's *Materia Medica* (1st c. AD), also contains some excellent botanical information. Later on, Galen's *Art of Medicine* was one the most well-known medical handbooks in medieval and Renaissance times with a tremendous impact on the science of medicine.

During the medieval period, the medical manuals were the official pharmacopoeias of their era and had a great impact on scientists in Europe over time, until the 17th century. Among the byzantine physicians, Nikolaos Myrepsos (13th c.) wrote in Greek the largest known collection of pharmaceutical recipes entitled *Dynameron* with 2667 recipes based mainly on medicinal plants, most of them inherited from classic ancient Greek and Hellenistic periods. The most important point in Byzantine medicine is the compilation of oriental practices and herbal products with those inherited from classic antiquity and its propagation to medieval occident, as many recipes have been copied in later medieval Pharmacopoeias of the Europe. Byzantine medicine is an important bridge for the passage from the ancient Greek medicine into the medieval medicine. Nowadays, it is advantageous to compare the plants quoted in medical manuscripts of the past according to the current scientific knowledge. Actually, among these plants, more than 50 plants are listed in herbal medicines by the European Medicine Agency.

KS02. Designing clinical trials of herbal medicines as alternatives to antimicrobials.

M. Willcox, C. Simpson, M. Logue, X.-Y. Hu, A. Flower, B. Stuart, P. Little, M. Moore

University of Southampton, United Kingdom

Antimicrobial resistance (AMR) is a growing challenge and is already causing 700,000 deaths per year; by 2050 this is predicted to rise to 10 million deaths per year. In many cases antimicrobials are used for minor infections which are self-limiting and in which they provide little objective benefit. Herbal medicines are a possible alternative which may provide symptom relief while avoiding unnecessary use of antimicrobials, thus reducing the risk of resistance. We have conducted several clinical trials of herbal medicines for the treatment of malaria, urinary tract infections, and acute respiratory infections, using a variety of designs: randomised controlled trials (RCTs) of herbal medicine in addition to or instead of a conventional antimicrobial; a factorial design; feasibility randomised controlled trials using individualised or standardised herbal medicine; and nested qualitative studies. The presentation will summarise these designs and lessons learned.

It has been feasible to recruit and retain patients to RCTs of herbal medicines for a variety of infectious diseases in primary care both in Africa and in the UK. It is particularly important to ensure that the dose, preparation and duration of treatment are optimised prior to launching a RCT and that good follow-up mechanisms are in place to maximise retention. Choice of primary outcome measure can be controversial. Many patients and practitioners are willing to consider herbal alternatives for treatment of uncomplicated infections, and these may reduce use of conventional antimicrobials, thus reducing the risk of resistance. High-quality evidence from RCTs is needed to inform the choice of the most effective treatments and to provide trustworthy advice to patients and practitioners.

KS03. Study of the mechanism of antitubercular and antiplasmodial activities of plants and natural products using mass spectrometry-based assays.

A.I. Calderón

Department of Drug Discovery and Development, Harrison School of Pharmacy, Auburn University, Alabama, USA

Strategies for developing new drugs to treat diseases include the phenotypic and target-based screening of libraries of natural products and extracts to identify novel bioactive chemical entities. Mass spectrometry (MS) and hyphenated techniques such as liquid chromatography-mass spectrometry (LC-MS) have become primary tools in these endeavors. In the last thirteen years, my research group has been developing and applying methods based on qualitative and quantitative high-resolution mass spectrometry to a wide variety of assays for profiling the biological activity of natural products. We have developed MS-based bioassays to study the ligand-target binding and profile the mechanism of inhibition. The bioassays were designed using affinity selection mass spectrometry and the MS detector to determine the reaction product of the targeted enzyme reaction. The assays identify natural products, which can selectively target 1) *Plasmodium falciparum* thioredoxin reductase (*Pf*TrxR), 2) *Plasmodium falciparum* glutathione reductase (*Pf*GR) enzymes, and 3) *Mycobacterium tuberculosis* shikimate kinase (*Mt*SK). We have successfully profile *Mycobacterium tuberculosis* shikimate kinase inhibitors such as manzamines and ilimaquinone from marine sponges, natural products, and plants (*Alpinia galanga* (L.) Willd. (Zingiberaceae) and *Santalum album L (Santalaceae)*. Oleamide from *Guatteria recurvisepala* R.E. Fr. (Annonaceae), and other plant-derived natural products, were identified as *Plasmodium falciparum* thioredoxin reductase (*Pf*TrxR) inhibitors. These MS-based assays are valuable tools to speed up the bioactive molecule discovery in complex mixtures of natural products.

KS04. Modern analytical approaches for Natural Products Analysis.

H. Stuppner

University of Innsbruck, Austria

The wide chemical diversity of natural products has challenged analysts all over the world and has been a driving force for the development of innovative technologies since decades. Among the different analytical separation systems available, a predominance of high and ultra-high performance liquid chromatography (HPLC&UHPLC) and gas chromatography (GC) can be observed in natural product analysis. Although GC still is unmet in its separation capacity, its application is however limited by the need to vaporize samples prior to analysis. Electrophoretic separation techniques such as capillary electrophoresis (CE) or capillary electrochromatography (CEC) are alternatives to HPLC or GC, but its routine use is hampered by Its limited robustness and sometimes difficult manageability. One of the most recent techniques is supercritical fluid chromatography (SFC) which is considered as green technology. Unlike initial SFC approaches, using neat supercritical carbon dioxide, modern SFC technology employs mixtures of CO2 and organic solvents (mostly alcohols) as mobile phase which increases the solvent strength significantly. Consequently, the technique could largely expand its suitability and develop to a valuable alternative in modern natural product analysis. In contrast to GC, which in natural product research relies mostly on the unspecific flame ionization detector (FID) to monitor analytes if a mass spectrometry hyphenation (GC-MS or GC-MS/MS) is not realized, analyte detection in HPLC, CE/CEC and SFC can be facilitated in many ways. Besides mass spectrometry, mainly diode array detectors (DAD) and ELSD detectors are used to monitor analytes. Beside a short overview on common analytical techniques described above this presentation will mainly focus an SFC and its applicability for the analysis of natural products.

KS05. The application of classic and modern pharmacognosy in the study of African traditional medicines.

A.M. Viljoen, S. Combrinck, I. Vermaak, M. Sandasi

Department of Pharmaceutical Sciences and SAMRC Herbal Drugs Research Unit, Faculty of Science, Tshwane University of Technology, Pretoria, South Africa

Southern Africa harbours a unique flora comprising of over 24 000 species of flowering plants. Woven within this tapestry of botanical diversity is the traditional use of indigenous plants as ethnomedicines. Developing official monographs, establishing a national repository of botanical standards, and producing validated analytical methods is a fundamentally important prerequisite to encourage research and commercialisation of Africa's medicinal flora. The unfortunate underrepresentation of pharmacognosy in the curricula of many pharmacy schools has left a void of expertise, which has hampered the development of a comprehensive herbal Pharmacopoeia. A further challenge involves the inherent complexity of medicinal plants, exacerbated by extensive chemotypic variation. Chemical fingerprinting is a crucial component in characterising plant material and requires a dedicated approach to develop analytical methods for the profiling of complex herbal extracts. Funding from the National Research Foundation (NRF/DST SARChI Initiative) and the South African Medical Research Council has catalysed initiatives at the Tshwane University of Technology to develop herbal monographs that aid in the identification and quality control of important South African herbal medicines. Selected examples will be presented to illustrate the daunting workflow, which includes extensive sampling, the development of analytical methods to profile volatile and non-volatile compounds using GC-MS and LC-MS, HPTLC, vibrational spectroscopy, as well as the use of preparative chromatography to isolate biomarkers. The powerful tandem application of analytical chemistry and chemometric modelling will be highlighted. Developing comprehensive species monographs requires a multidisciplinary collaborative effort which will inevitably contribute to the safety, efficacy and quality of African Traditional Medicines and commercial herbal formulations.

KS06. Functional consequences of stimulation of multiple biological targets by botanical extracts and their bioactive compounds – hops as an example.

G. Vollmer

Faculty of Biology, Technische Universität Dresden, Germany

The multiple biological targets of hops (humulus lupulus L.) and its bioactive constituents have recently been reviewed [1]. Spent hops, among other constituents contains the chalcone xanthohumol (XH) as the major prenylated flavonoid. However, less abundant but biologically important prenylated flavonoids contained in the extract in addition are isoxanthohumol (IX), 8-prenylnaringenin (8-PN), and 6-prenylnaringenin (6-PN). Importantly XH undergoes phase I metabolism to form IX, 6-PN and particularly 8-PN, which is the most potent phytoestrogen known to date. As would be expected from a potent phytoestrogen, 8-PN triggers numerous estrogen receptor-initiated responses among them growth and proliferation of experimental uterine and mammary gland models. The latter responses are associated with an increased risk for cancer of these hormone dependent organs. For 8-PN contained in hops extracts, these activities are not detectable, although at least in vivo a rapid metabolism of XH to relatively high levels of 8-PN occurs. In vitro data suggest that in the case of the extracts 6-PN attenuates the estrogen receptor dependent activities of 8-PN through activation of the aryl hydrocarbon receptor (AhR) pathway. In summary, constituents of hops extracts stimulate multiple pathways. These activated pathways interact and result in modulation of functional outcomes. For hops, the activation of the AhR pathway attenuated unfavorable and estrogen receptor dependent proliferative events induced in mammary gland and uterus by 8-PN. In conclusion, these results are relevant for prevention strategies of tumorigenesis in hormone dependent organs.

[1] J.L. Bolton, T.L. Dunlap, D.A. Hajirahimkhan, O.C. Mbachu, S.-N. Chen, L. Chadwick, D. Nikolic, R.B. van Breemen, G.F. Pauli, B.M. Dietz (2019) The Multiple Biological Targets of Hops and Bioactive Compounds, Chem. Res. Toxicol. 32: 222-233.

KS07. Risk assessment of chemicals that occur as mixtures: additivity and interactions.

C. Hogstrand

King's College London, School of Life Course Sciences, London, United Kingdom

Chemical risk assessment has traditionally been carried out for single chemicals in isolation. However, chemicals can have combined effects and it is therefore reason to assess effects caused by chemical mixtures. Risk assessment of combined exposure to multiple chemicals presents numerous challenges to researchers, risk assessors and risk managers, particularly because of the complexity of the problem formulation, the vast numbers of chemicals that potentially could be involved, and the amount of data needed to characterize hazard of and exposure to chemical mixtures. It is therefore a priority area to develop methodologies for combined exposure to multiple chemicals. Like for single chemicals, frameworks to assess risk of chemical mixtures consists of problem formulation, exposure assessment, hazard assessment, and risk characterisation, and can be applied for both the whole mixture and the combined effect of components of the mixture. The default assumption is usually that chemicals act in an additive fashion, but in some cases synergism or antagonism exist. For component-based approaches chemicals are grouped into assessment groups based on the same or similar modes of action and a common critical effect.

KS08. The use of aromatic plants, extracts and essential oils as feed additives in animal nutrition.

I. Giannenas

EBVS® European Specialist in Poultry Veterinary Science
Laboratory of Nutrition, Faculty of Veterinary Medicine, School of Health Sciences, Aristotle University of Thessaloniki, Greece

Animal production globally depends on feed efficiency to sustain growth and profitability. Sustainability of poultry and pig industries should be further regarded as a crucial factor to match worldwide increased food demands and reservation of feed sources. Similarly egg and milk production need support to sustain health and profitability. Several feed additives have been used in animal nutrition in order to increase sustainability of the Agrofood sector, in both organic and conventional livestock. The aim of the current report is to provide a comprehensive description on the use of aromatic plants, extracts and essential oils as feed additives alone or in combination with feed additives of different categories in our most recent research studies. Combinations of phytogenics with synbiotics and acidifiers have been investigated in terms of efficacious replacement of antimicrobial growth promoters in poultry, pig and dairy animals. These feed additives may further support animals to overcome long-standing health problems including bacterial challenges, as well as promote welfare especially for layer hens. This report will finally explore the potential of the aforementioned feed additives to tackle with environmental problems. Examples include use of local feed additives together with enzymes, which can enhance the use of local feedstuffs, especially in cases where high fibre content is the main constraint. Emphasis will be placed on the consequences of using local feed sources versus imported feedstuffs on global warming potential, primary energy use, nutrient excretion and the feed additive influence on lessening the pollution from animal operations. *In vitro* testing of antioxidant and antimicrobial bioactive herbal compounds and novel tools, such as life cycle assessment and genomic microbiota analysis, are necessary to verify effectiveness of aromatic plants, extracts and essential oils as feed additives in supporting livestock health, performance and sustainability. Further, practical examples and solutions for the production of broiler, turkey, pork meat or eggs with functional properties and minimal environmental impact are provided. The sustainability of meat, egg or milk industry is based on health, environmental protection and steady farmer income.

KS09. Safeguarding threatened species and promoting the use of plant resources in research and development.

E. Maloupa

Hellenic Agricultural Organization-DEMETER, Institute of Plant Breeding and Genetic Resources, Laboratory of Protection and Evaluation of Native and Floricultural Species, 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.

Throughout history, botanic gardens have continuously responded to pressing environmental changes. Their collections of living plants provide an essential resource for scientific research, conservation and public engagement.

Botanic gardens attract many of the diverse audiences which Responsible Research and Innovation (RRI) seeks to engage aiming to align the outcomes of scientific and technological advances with the values and needs of the society.

Plant diversity is disappearing as a direct impact of the way humankind uses the natural resources. Our flora is fundamentally important to human life as a source of food, shelter and medicine amongst many other things. Wild plants offer a wealth of services and goods of essential livelihood value.

Since its establishment, the BBGK has been focused on the conservation and the sustainable use of native plants of Greece and the Balkans, leaving aside the exotic and ornamental plants. The Balkan Botanic Garden of Kroussia (BBGK) was founded on 19 May 2001. It is member of the Botanic Gardens Conservation International (BGCI). 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 70km from Thessaloniki, 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) with almost 20% with 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 as flavorings and seasonings, for the preservation and storage of various foods and help to maintain their organoleptic properties. Plants that are often used are: oregano (*Origanum vulgare* ssp. *hirtum*), sage (*Salvia officinalis*), thyme (*Thyumus* spp.) and rosemary (*Rosmarinus officinalis*). Through this presentation we are going to identify successful models and draw lessons from sustainable management and commercialization of unique native flora and its products.

KS10. Natural products loaded in nanocarriers to cross biological barriers.

AR. Bilia

Department of Chemistry "Ugo Schiff", Florence, Italy

Natural products (NPs) are fascinating molecules not exclusively for their exciting structure variability but also for their ability to interact with diverse targets. In spite of these advantages, in many cases, the impressive *in vitro* activity to less or not significant *in vivo* efficacy is generally due to drug poor water solubility, low lipophilicity and inappropriate molecular size resulting in reduced absorption due to difficulties to cross biological barriers. In the gastrointestinal tract, a variety of physiological and morphological barriers such as gastric pH, proteolytic enzymes, colonic microflora and mucus layer can severely affect NPs bioavailability. Skin is a further physiological barrier, which essentially consists of four layers, and the stratum corneous, the outer layer of the skin (nonviable epidermis) represents the rate-controlling barrier for diffusion for almost all compounds. Small lipophilic NPs such a mono- and sesquiterpenes have high permeation properties and largely used as penetration enhancers, but they can also need of specific formulations because susceptible to volatilization and degradation, mainly by oxidation and isomerization. Other important barriers are the pulmonary and nasal mucosae where NPs bioavailability is often limited by rapid degradation and/or clearance by the mucociliary system and alveolar macrophages. Additionally, blood retinal barrier and blood-brain barrier (BBB) are among the most challenging, ensuring proper homeostasis, mainly due to very selective and restrictive bidirectional transport of endogenous and exogenous compounds. Absorption through each pathway is dependent on different physical characteristics, such as molecular weight, hydrophobicity, ionization constants, and stability of absorbing molecules as well as biological barriers. Different approaches can be used to increase barrier-crossing properties of NPs, based on chemical permeation enhancement using small lipophilic NPs, polysaccharides, dendrimers, cyclodextrins, and the design and production of appropriate drug delivery systems, in particular nanosized ones, which is the most attractive to enhance the permeation through paracellular, transcellular, carrier-mediated, and receptor-mediated transport. Some properties such as mucoadhesion and retention to the mucosa can also be used to increase the cross biological barriers to achieve optimal pharmacological action at pathological sites.

Bilia et al. (2018) Nanocarriers: A Successful Tool to Increase Solubility, Stability and Optimise Bioefficacy of Natural Constituents. Curr Med Chem. 2018 Nov 1. doi: 10.2174/0929867325666181101110050.

Bilia et al. (2017). Improving on Nature: The Role of Nanomedicine in the Development of Clinical Natural Drugs. Planta Med. 83(5):366-381. doi: 10.1055/s-0043-102949

Bilia et al. (2014). Essential oils loaded in nanosystems: a developing strategy for a successful therapeutic approach. Evid Based Complement Alternat Med. 651593. doi: 10.1155/2014/651593.

Bilia et al. (2016). Vesicles and micelles: two versatile vectors for the delivery of natural products. Journal of Drug Delivery Science and Technology, 32, 241-255.

Bilia et al. (2014). Flavonoids loaded in nanocarriers: an opportunity to increase oral bioavailability and bioefficacy. Food Nutr Sci. 5: 1212–1227

Bilia et al. (2018). Plants Extracts Loaded in Nanocarriers: an Emergent Formulating Approach. NPC, 13 1157-1160 https://doi.org/10.1177/1934578X1801300914

KS11. Chemical profile and nutraceutical features of some Mediterranean food plants.

A. Braca

Department of Pharmacy, University of Pisa, Italy

From an epidemiological standpoint, natural active principles from fruits and vegetables have been reported to protect humans from several diseases. In recent years, the scientific community has strongly emphasized the role played by diet in the prevention of chronic degenerative pathologies [1]. The mechanism of action of these dietary components is not completely understood, however, at least partially the effectiveness of these substances may be explained in terms of antioxidant and anti-inflammatory properties, since the different pathogenesis of many diseases are marked by the role played by free radicals [2].

In Italy, many spontaneous plants are used as food in folk traditions and are now being re-evaluated as healthy products with high nutritional value. In this context, our recent research is focused on chemical study of some Mediterranean food plants selected because they are simply forgotten or poorly investigated. Among them, "common dogwood" or "bloody dogwood" (*Cornus sanguinea* L., Cornaceae), "albero dei coralli" (*Elaeagnus umbellata* Thunb., Elaeagnaceae), "agretti" or "barba di frate" (*Salsola soda* L., Amaranthaceae) [3], and some old Tuscany cherry cultivars (*Prunus avium* L., Rosaceae) were selected. Plant materials were extracted and subjected to different column chromatographies to isolate and structurally characterize, through NMR and MS analyses, their main chemical components that were assayed to evaluate their beneficial effects as antioxidant or anti-inflammatory remedies. In some cases extracts were simply analysed by LC-PDA/UV-MS. Results on kaempferol 3-*O*-β-D-glucopyranosyl-(1→2)-β-D-galactopyranoside 7-*O*-α-L-rhamnopyranoside isolated from *E. umbellata* as possible local protective agent against oxidative stress and inflammatory stimuli in gingival tissue, revealed this new property for a flavonoid glycoside; while the appreciable inhibitory effectiveness of quercetin 3-*O*-glucuronopyranoside from *S. soda* on enzyme involved in diabetic complications and inflammatory process is indicative of its valuable nutraceutical potential. Extracts of cherry displayed a potential anti-angiogenic activity in the CAM e zebrafish assays.

[1] Traka, Maria H.; Mithen, Richard F. Plant science and human nutrition: challenges in assessing health-promoting properties of phytochemicals. Plant Cell 2011; 23: 2483-2497.

[2] Han, X.; Shen, T.; Lou, H. Dietary polyphenols and their biological significance. International Journal of Molecular Sciences 2007; 8: 950-988.

[3] Pignatti S. Flora d'Italia, Edagricole: Bologna, 2017.

Invited lectures

20th International Congress of the International Society for Ethnopharmacology

IL01. Traditional Chinese Medicine for Cancer Patients in Taiwan.

H.-R. Yen

School of Chinese Medicine, College of Chinese Medicine, China Medical University, Taichung, Taiwan
Chinese Medicine Research Center, China Medical University, Taichung, Taiwan
Department of Chinese Medicine, China Medical University Hospital, Taichung, Taiwan

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Many cancer patients seek complementary therapies. We investigated the adjunctive use of traditional Chinese medicine (TCM) by Taiwanese cancer patients and conducted basic and clinical studies to delineate its potential benefits. We analyzed the registry database of the National Health Insurance Research Database and included all cancer patients in Taiwan. Patients were categorized as adjunctive TCM users or non-TCM users, followed by matching with age, sex, comorbidity, conventional treatment, and index year to compare the risk of mortality and survival rate. The prescribed Chinese herbal medicine (CHM) was further investigated in the laboratory or clinical trial. Compared to non-TCM users, adjunctive TCM users were younger and more likely to be female, white-collar workers, and reside in highly urbanized areas. The average interval between cancer diagnosis and TCM consultation was 15.3 months. The most common cancer type was breast cancer in adjunctive TCM users (19.4%), and intrahepatic bile duct cancer in non-TCM users (13.6%). A total of 33.1% of adjunctive TCM users visited TCM clinics more than 9 times per year. Overall, adjunctive TCM users had a lower adjusted hazard ratio for mortality (aHR = 0.69, 95% CI: 0.68-0.70) compared with those non-TCM users. For leukemia, we conducted a laboratory investigation and clinical trial on Sheng-Yu-Tang to prove its efficacy. For nasopharyngeal carcinoma, we have also shown the prescribed CHM, Gan-Lu-Yin, is effective against tumor in vitro and in a tumor-bearing murine model. This study provides an overview of complementary TCM usage among cancer patients in Taiwan. Our big data-bench-bedside approach could be developed as a powerful tool for the investigation of ethnopharmacology.

IL02. Ethnopharmacological investigations of Vietnamese medicinal plants used for the treatment of inflammation and cancer.

H. Thi Tran, T. Thanh Nguyen, N. Kretschmer, E.-M. Pferschy-Wenzig, R. Bauer

Institute of Pharmaceutical Sciences, Department of Pharmacognosy, University of Graz, Austria

Vietnam is very rich in traditional medicinal knowledge and still 80% of the population is using it in the primary health care system. In our ongoing research on plants used for the treatment of inflammation and cancer, we have recently screened and investigated a series of Vietnamese plants with high activity.

From *Helicteres hirsuta* we isolated the new natural product 4,4'- Sulfinyl - bis - (2-(*tert*-butyl)-5-methylphenol, as well as 7-*O*-methylisoscutellarein, 3*β*-*O*-acetylbetulinic acid, and simiarenol, which were found in Sterculiaceae for the first time. 4,4'- sulfinyl - bis - (2-(*tert*-butyl)-5-methylphenol and 3*β*-*O*-acetylbetulinic acid showed cytotoxicity in CCRE-CEM and HCT-116 cancer cells [1].

From *Enhydra fluctuans*, the known sesquiterpene lactones enhydrin, fluctuanin, and fluctuadin showed significant cytotoxicity against four cancer cell lines and a normal lung fibroblast cell line.

Four sesquiterpene derivatives were also isolated from the rhizomes of *Stahlianthus involucratus*, including the two new natural products 5-hydroperoxylacorafuran and isoacotatarone C, plus the known tatarinowin B and tatarinowin C. Besides tatarinowin C, all showed cytotoxicity.

Sphaeranthus africanus has been used in traditional Vietnamese medicine to treat sore throat, and to relieve pain and swelling. A series of carvotacetones from this plant that displayed cytotoxicity against several cancer cell lines and showed anti-inflammatory effects. [2,3]. 3-angeloyloxy-5-angeloyloxy-7-hydroxycarvotacetone, 3-angeloyloxy-5-[2?,3?-epoxy-2?-methyl-butanoyloxy]-7-hydroxycarvotacetone, 3-angeloyloxy-5-[3?-chloro-2?-hydroxy-2?-methyl-butanoyloxy]-7-hydroxycarvotacetone, 3-tigloyloxy-5-angeloyloxy-7-hydroxycarvotacetone, 3-angeloyloxy-5-hydroxy-7-hydroxycarvotacetone were identified together with two diastereomeric carvotacetones (3-angeloyloxy-5-[2»*S*,3»*R*-dihydroxy-2»-methyl-butanoyloxy]-7-hydroxycarvotacetone and 3-angeloyloxy-5-[2»*R*,3»*R*-dihydroxy-2»-methyl-butanoyloxy]-7-hydroxycarvotacetone, asperglaucide, and chrysoplenol D. Some of them showed very potent anti-inflammatory and antiproliferative effects with IC_{50} values in the nM and low μM range.

Our findings support the traditional medical application of these plants and their potential for drug lead discovery.

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[3] Tran, H.T., Gao X.-H., Kretschmer, N., Pferschy-Wenzig, E.M., Raab, P., Pirker, T., Temml, D., Schuster, D., Kunert O, Huynh, L., Bauer, R. Anti-inflammatory and antiproliferative compounds from ***Sphaeranthus africanus*** Phytomedicine 62: 152951 (2019).

IL03. Reverse pharmacology and traditional medicine.

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In the conventional process of discovering treatments that could be widely recommended, clinical studies are conducted after several years of preclinical research. There is however, a faster track: the approach called *reverse pharmacology* (or *bedside-to-bench*). For traditional medicines, population surveys on therapeutic activities and correlation with reported clinical outcome (Retrospective Treatment Outcome -- RTO) can help select treatments with good safety profile and indices of effectiveness, hence accelerate eventual clinical validation.

The reverse pharmacology approach has four steps: After an RTO survey, the second step is a dose-response study with humans; the third step evaluates effectiveness through a clinical trial. The fourth step are laboratory studies, which in this case, and contrary to classical drug discovery, are thus post-clinical.

IL04. Innovative strategies for the efficient isolation of natural products and discovery of potential new leads.

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Natural products (NPs) have been a source of inspiration for drug development throughout history. Nature continues to represent a rich source of new molecules with interesting pharmacological properties that could be used as lead compounds for the development of new drugs. Technological development in this field is constantly evolving. New high-throughput screening strategies have been introduced for rapid and efficient evaluation the biological and chemical potential of complex matrices, such as plant extracts. Significant advances in molecular biology afford now the development of more predictable in vitro and in vivo bioassays. The combination of metabolite profiling using molecular networks and bioassays gives the possibility to distinguish between already known bioactive compounds and new molecules directly in crude natural extracts (dereplication)[1] Thus, the targeted isolation of new bioactive products or constituents presenting novel or unusual spectroscopic features can be undertaken. The structural identification of new NPs requires only very small amounts of material (sub milligram) using such approaches. On the other hand, the extensive bioactivity profile characterization of a potential lead requires milligram or tens of milligram amounts. In this case, preparative chromatographic methods for the purification of NPs at large scale are mandatory. At this level, significant improvements for the efficient targeted isolation of given NPs through chromatographic gradient transfer methods have been made [2,3]. The applications, possibilities and limitations of these latest technologies applied to the study plant and fungal metabolites will be illustrated with recent investigations performed in our laboratory.

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[3] Challal, S.; Queiroz, E. F.; Debrus, B.; Kloeti, W.; Guillarme, D.; Gupta, M. P.; Wolfender, J. L., ***Planta Med.*** 2015, 81, 1636-1643.

IL05. The Regulatory aspects and challenges for commercialisation of the Natural Products.

N. Lall

Plant and Soil Sciences, University of Pretoria, South Africa

Traditional medicine is an important foundation upon which modern medicine has been built. In these traditional practices, plants have been a crucial resource, owing to their abundance since the dawn of mankind. In modern times, synthetic drugs have become a lucrative alternative; however, plants as a resource remain the indispensable resource from which even synthetic alternatives are derived. Recently, natural medicines have shown increased popularity, not surprisingly when considering the benefits. By combining the knowledge derived from traditional medicinal practices with modern science, the possibilities for drug discovery and use of plants in the treatment of a wide array of conditions seems endless.

South Africa has a wealthy supply of plants (about 23 500 species of higher plants) together with a high degree of endemicity (36.6%) in the indigenous South African flora, of which 4000 plant taxa are ethnomedicinally used and approximately 500 species are used in traditional medicine by an estimated 70% South Africans on a regular basis. South Africa has huge potential for identifying novel compounds to treat many diseases. South African plants for various purposes such as infectious diseases, cancer, skin-hyperpigmentation problems, melasma, Periodontal diseases, and for acne problem have been scientifically investigated. Steady progress in evaluating potential medicinal plants for product development with potential in human medicine has been made.

Once the medicinal products are developed, the dynamics of various ways for benefitting the people who have been using plants for centuries, will be discussed in the talk.

IL06. Exploration of Traditional Herbal Knowledge in the South Balkan and East Mediterranean region for the discovery of bioactive natural products.

N. Aligiannis

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The geographical and ecological characteristics of the Balkan Peninsula and Mediterranean Sea, along with the historical connection among ethnic groups living in the area, gave rise to the development of a distinct flora and to the uses of common medicinal plants against various ailments. Over the past twenty years, our research group obtain evidence on the importance of collecting, assessing and exploring ethnopharmacological data for the discovery of bioactive natural compounds. As examples, the study of Nicander's "Theriaca", verified the use of several plants to treat bite symptoms from venomous animals and information obtained from semi-structured interviews for the wound healing properties of *Ficus carica* preparations were verified by *in-vivo* mouse model applications. In parallel, the establishment of a validated analytical chemometrics approach through the elaboration of contemporary chromatographic and spectroscopic techniques (FCPC, HPTLC, LC-HRMS/MS, NMR) along with sophisticated statistical algorithms for the rapid and effective identification of bioactive compounds, prior to their isolation directed us to the development of PEGASUS platform.

The aforementioned efforts led tOo join forces with Balkan countries and coordinate the European project EthnoHERBS (H2020-MSCA-RISE) for the sustainable and effective exploitation of SE European traditional knowledge and biodiversity for the development of innovative products based on natural ingredients. Within the scope of project, all ethnobotanical studies from Albania, Bosnia-Herzegovina, Bulgaria, Croatia, Cyprus, Greece, Italia, Kosovo, Montenegro, North Macedonia, Romania, Slovenia, Serbia, Turkey were assessed for archiving in a database herbal preparations used for skin ailments. The aim is to provide an overview of the ethnobotanical knowledge of medicinal plants and traditional medical practices for the treatment of skin disorders in the South Balkan and East Mediterranean region. Initially, the research was focused in Albania, Cyprus, Greece and Turkey. Literature study of ethnobotanical surveys showed that 948 taxa belonging to 417 different genera and 111 different families are used in the treatment of skin related problems. The majority of the plants belong to the families of Asteraceae (11.0%), Lamiaceae (8.5%), Rosaceae (7.1%), Plantaginaceae (4.5%) and Malvaceae (3.5%). Their usage is internal (decoction, infusion etc.), or external (compress, poultice, ointment etc.) to treat skin related ailments such as wounds. In order to relate this ethnopharmacological knowledge and trace its expansion and diversification through centuries, a comparison of findings was made with the use of the medicinal species mentioned in Dioscorides' "*De Materia Medica*" for skin disorders. This work constitutes the first comparative study performed with ethnobotanical data for skin ailments gathered in the South Balkan and East Mediterranean area. Our findings confirm the primary hypothesis that people in Albania, Cyprus, Greece and Turkey are close related in terms of traditionally using folk medicinal practices. Nevertheless, more field studies conducted, especially in remote places of these regions, can help preserve the traditional medical knowledge, aiming at the discovery of new phytotherapeutics against dermatological diseases.

IL07. TCM needs TQM: New applications using NMR for standardizing botanical mixtures.

R. Cooper

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There is a need to improve and promote the scientific inquiry on the study and use of botanicals, natural products and Traditional Chinese Medicine (TCM), in order to secure global acceptance. Although there is an enormous history of use of botanical medicines a better understanding of these preparations and formulae within the scientific and international community and a rigorous process is needed. The aim is to study select botanicals and natural products to provide scientific data used for standardization and substantiation of health claims using Total Quality Management (TQM). For example, active compounds found in the botanical extract of the Traditional Chinese Medicine, *Sambucus williamsii* Hance exhibit protective effects on trabecular bone mass and mechanical strength of cortical bone of ovariectomized rats. The main bioactive compounds were determined as lignan compounds. A novel approach was adapted to standardize and determine quantities of the lignan content in the aqueous alcoholic extract of *Sambucus williamsii* using spectroscopic methods and provide data to help meet the registration requirements. Using these data, the lignan amount in the extract was calculated. The application of this simple and reliable method can be used to estimate other compound families in complex mixtures or botanical extracts and herbal medicines.

IL08. Challenges and success in using plants to control internal parasites in animals.

JN. Eloff

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Haemonchus contortus is a nematode causing extensive losses in sheep production especially in tropical countries. Strong resistance has developed against current control agents. These agents are expensive or unavailable to poor rural farmers. Rural pastoralists have use many plant extracts to control this parasite. In contrast to external parasites where it is easy to see the efficacy of treatments, it is not easy to see the efficacy of plant extracts with internal parasites. It is possible to collect eggs of the parasite from faeces and keep it alive long enough to determine the treatment effect on egg hatching and on larval development. Because the biological assay is demanding it is not feasible to carry out random screening of large number of plants. It is also not easy to isolate the bioactive molecule(s). Using ethnoveterinary data is the only practical option. Much work has been done in this field. In some cases, the objective was to support traditional use and in other cases it was to find compounds that could be used as nematicidals. Benzimidazoles are active against fungi and nematodes. Some antifungal compounds may also be active against nematodes. It is relatively easy to screen plants for antifungal activity and isolate antifungal compounds that may be active against nematodes. The positive and negative results found with these two approaches will be discussed. The work eventually led to the isolation of agrimol G from *Leucosidea sericea* leaves with good activity against *Haemonchus contortus.* Based on electron microscopical studies it has a different mechanism of activity than ivermectin and albendazole, two commercially used nematicidals. The controversy around using ivermectin against the Corona virus will also be discussed.

Adamu M, *et al*. (2019) The phloroglucinol derivative Agrimol G isolated from *Leucosidea sericea,* induces microtubular degeneration in *Haemonchus contortus.* Experimental Parasitology 20710778.

IL09. How can we manage dairy animal health and welfare?

M. Caroprese

Department of Agriculture, Food, Natural Resources, and Engineering (DAFNE), University of Foggia, Italy

Consumers constantly require healthy food from healthy animals and are aware that milk quality has important impacts on human health. In the last few years, there is a growing concern about livestock health and welfare in relation to the spread of antimicrobial resistance caused by the misuse of antibiotics, highlighting the interest of new alternatives to antibiotics in veterinary medicine practices. As an alternative to the use of antibiotics, there is a considerable interest in the use of feed supplementation, such as oily seeds, and additives, such as compounds or mixtures, added in animal feed to improve health status, growth rate, productivity, and performance of dairy animals. Feed supplementations and additives can be used as growth promoters, antimicrobials, immunostimulators, and antioxidants, having a particular role for the livestock health, and thus satisfying the increasing consumer demand for natural foods with functional properties. On the other hands, meeting the increasing demand for animal-based food for the increasing human population is a major sustainability challenge. A possible approach could be an increase in total efficiency and the reduction of the impact of livestock by feeding livestock recycling biomass from agro-food systems unsuitable as food. The use of by-products from the agro-food system could reduce feed–food competition and, therefore, the utilization of croplands for feed production. Moreover, the use of agricultural by-products in animal feeding represents a circular economy model to guarantee the production of healthy and affordable food by reducing the livestock system impact from an environmental and societal point of view and minimizing waste outputs. The effects on the use of feed supplementation or additives, also deriving from of agro-industrial by-products, will be presented and analysed in terms of milk quality and dairy animal health and welfare.

IL10. Evaluation of sesquiterpenes as permeation enhancers for transdermal delivery.

D. Fatouros

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Chemical permeation enhancers (sulfoxides, pyrrolidones, fatty acids, alcohols and glycols, surfactants, urea and its derivatives, essential oils, terpenes and their derivatives, phospholipids) have been used extensively to promote the transport of actives across human skin. Among them active compounds derived from essential oils (terpenes) have been widely used as permeation enhancers for both hydrophilic and hydrophobic drugs. Terpenes can interact with intracellular lipids in the SC to increase the diffusion of the drugs. Beyond the relatively small monoterpenes, larger terpene molecules (sesquiterpenes) have also been evaluated as enhancers for molecules permeating human skin membranes. Recent work from our team demonstrated the effect of sesquiterpenes as chemical enhancers for the transdermal delivery of a model macromolecular compound across human skin *in vitro*.

IL11. Archaeoethnopharmacological studies in South America: back to the roots of ethnopharmacology?

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Since the term Ethnopharmacology was defined in 1967, this scientific discipline was closely linked to the study of psychoactive plants. This is demonstrated by the pioneering studies of hallucinogenic snuffs from South America (Bo Holmstedt), alkaloids from cacti collected in the Mexican desert (Jan G Bruhn), and tryptamines/beta-carbolines from ayahuasca samples collected in the Amazon (Laurent Rivier). With time, this discipline has evolved to other themes, topics, and interests, losing - to a great extent - its original essence.

Recent archaeological investigations in archaeological materials found in various archaeological sites in South America have focused their interest on the chemical and botanical study of the material evidence of the consumption of psychoactive plants by the first inhabitants of the continent, to understand and interpret the cultural complexity of these practices in prehistory.

At present, is fascinating and captivating to recognize that very distant and different disciplines such as archaeology and ethnopharmacology, have merged into common scientific interests. The most important convergence points are the context of the study material, psychoactive plants; the analytical, botanical/chemical/pharmacological approach; as well as the geographic context, South America. It is neither illogical nor unrealistic to propose the emergence of a new discipline, *Archaeoethnopharmacology*.

This conference will reveal the close relationship between the origins of ethnopharmacology and recent chemical and botanical studies of archaeological materials associated with the use of psychoactive substances in the prehistory of South America. The main plant species and their most important management practices will be reviewed.

These antecedents will seek to answer the following question: Are we facing a return to the roots of ethnopharmacology?

IL12. Recommended standards for conducting and reporting ethnopharmacological field studies.

M. Leonti

Department of Biomedical Sciences, University of Cagliari, Italy

What are the minimum methodological and conceptual requirements for an ethnopharmacological field study? How can the results of ethnopharmacological field studies be reported so that researchers with different backgrounds can draw on the results and develop new research questions and projects? And how should these field data be presented to get accepted in a scientific journal such as JEP?

We focus on conducting and reporting ethnopharmacological field studies on medicinal plants or *materia medica* and associated knowledge of a specific people or region.

We highlight the most frequent problems encountered in literature, own fieldwork and insights gained from peer-reviewing of field studies [1].

Research needs to be ethical and legal and follow local and national regulations. Primary ethnopharmacological field data need to be collected and presented in a transparent and comprehensible way. In short this includes: 1) Relevant and concise research questions, 2) Thorough literature study encompassing all available information on the study site from different disciplines, 3) Appropriate methods to answer the research questions, 4) Proper plant use documentation, unambiguously linked to voucher specimens, and 5) Qualitative and quantitative analyses of the collected data, the latter relying on use-reports as basic units.

[1] Weckerle CS, de Boer HJ, Puri RK, van Andel T, Bussmann RW, Leonti M. 2018. Recommended standards for conducting and reporting ethnopharmacological field studies. J Ethnopharmacol. 210, 125-132.

IL13. Pharmacopeias for Indian herbal drugs

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Among Indian systems of medicines, herbs/herbal formulations occupy a prominent place. The quality control of these herbal formulations is important for maintaining their safety, efficacy and export potential to other countries. Indian systems of medicine use pharmacopoeias for the quality control and standardization herbs and formulations. Various pharmacopoeias are published in India stating the quality control specification. Major among them is Indian Pharmacopoeia (IP) which contains 168 specific monographs of herbs. Herbal Products and Phytopharmaceuticals Monograph Development Subcommittee of IPC, is developing the Phytopharmaceutical Ingredient (PPI) category monograph with the support of stakeholders. AYUSH is working on promotion of Indian traditional medicine (ITM) and developing pharmacoepial specifications for drugs used in Indian medicine (IM). Ministry of AYUSH, Govt of India and Pharmacopoeia commission for Indian medicine & homoeopathy Ghaziabad publishes Ayurvedic Pharmacopoeia of India (API). API cover monographs of 645 plant derived drugs. Monographs for 191 herbal formulations are published in Ayurvedic formulary of India. Other than API, Siddha Pharmacopoeia of India part I consists of monographs of 139 plant derived drugs. It consists of a brief introduction, synonym, description (macroscopic, microscopic, powder), foreign matter, total ash, acid insoluble ash, alcohol soluble extractive, water soluble extractive, liquid chromatography assay with reference standard, thin layer chromatography for fingerprinting, chemical constituents, properties and actions, important formulations, therapeutic uses and dose. Homeopathic Pharmacopoeia of India consists of monographs of 1117 plant derived drugs. Unani Pharmacopoeia of India cover 298 herbal drugs and 150 herbal formulations. Quality standards of Indian Medicinal plants by Indian Council of Medical Research (ICMR) are a non pharmacopoeial reference guide on standards of Indian herbs. Monographs on 449 plants are included in this guide. Indian Herbal Pharmacopoeia published by Indian Drugs Manufacturers Association (IDMA) containing 40 monographs of medicinal plants. The inclusions of herbs/herbal formulations in Pharmacopoeia have helped develop quality standards of these herbal medicines. This would ensure use of quality herbs/herbal products and improve its export potential through maintaining internationally accepted standards for quality thus promoting Indian herbal drug industry.

IL14. Traditional Herbal Resources and Herbal Pharmacopoeia: the contribution of the Taiwanese flora.

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Research Center for Traditional Chinese Medicine, China Medical University Hospital
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In Taiwan, more than 30% of Taiwanese utilize Chinese medicine. However, 80% of the raw herbal materials come from abroad. Geographically, Taiwan has diverse terrains and mountains surrounded by the sea, and different climates across tropical, temperate and frigid zones. Therefore, it creates an enriched ecological environment for medicinal plants in Taiwan. Breeding more than 4000 kinds of vascular plants.

There are many indigenous peoples in Taiwan who rely on fishing, hunting, gathering and planting for their livelihoods. Therefore, natural medicinal resources have been used for many years to reduce or treat diseases, which is a valuable source for the development of pharmacy. Thanks to the assistance of the Japanese in hundreds of years ago, they began to systematically sort out the medicinal herbs in Taiwan, with more than 300 species in total. Because Taiwan is a humid and sultry environment, most of medicinal plants are the effects of clearing away heat, detoxifying, and promoting blood circulation. In Taiwan, in addition to the private use of medicinal plants, the government is also actively including native medicinal plants in the pharmacopoeia.

The first edition of the Taiwan Herbal Pharmacopoeia was published in 2004, which was revised and a third edition was published in 2018. The Taiwan Herbal Pharmacopoeia regulates the quality of Chinese medicinal materials, containing 357 items, including 329 plants, 13 animals, 6 fungi, 3 insects and 4 minerals. In addition, Taiwan's native species *Dendrobium tosaense*, *Bletilla formosana, Orthosiphon aristatus, Pteris multifda, Litsea cubeba* and *Uncaria lanosa* var. *appendiculata* are added.

The government also actively supports the development of local herb in the research of active ingredients and pharmacology. For example, the Agriculture and Food Agency and Taiwan Seed Improvement and Propagation Station have also invested in Chinese herbal medicine for many years. Some school research center uses traditional pharmacy knowledge to integrate database and literature as the basis for scientific and systematic sound quality control and application, to promote the internationalization of Taiwan's traditional Chinese medicine and the development of the traditional Chinese medicine industry.

IL15. Evaluating Greek honey-bees' products (Honey, Propolis, Royal Jelly) through centuries, as vehicle of high value chemical and pharmacological potential.

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Honey-bees (*Apis mellifera*) have played an important role through centuries. Famous Greek honey has observed an increasing interest in its use in traditional Mediterranean food.

Propolis, a resinous material by bees, derived from the Greek "pro", in front of, at the entrance to, and "polis" the city/community; a substance that is in defense of the city/hive. It has been used in folk medicine, cosmetology and in food industry for health foods, and nutrition supplements. The chemistry of propolis is highly depended on the flora of the geographic region from which it is collected. The rich flora of Greece contributes a lot to a distinguished chemical profile of proposed as "Mediterranean type of propolis", well accepted after our investigations.

Royal jelly is secreted by the hypopharyngeal gland of young worker (nurse) bees, to feed young larvae and the adult queen bee. It has been harvested in very small quantities and also is so expensive.

Some Hundreds of samples of Greek/Mediterranean bee keeping products till now, have been chemically analysed and several isolated compounds have been structurally determined by modern spectral means. Furthermore, they were tested for their antimicrobial activity against Gram (+) bacteria and fungi. Their anti-oxidative activity has been also assayed together with cytotoxicities. RJ samples, were further subjected i) in a clinical study at postmenopausal healthy women, showing a significant decrease in LDL and increase in HDL- cholesterol ii) in an *in vivo* study on Wistar male rats in Water-maze test, where RJ exerted an improvement of the spatial memory while iii) strong immunomodulatory influence. All these bioactivities, therefore, have considerable commercial appeal, ranging from the pharmaceutical and food industries up to cosmetic ones.

IL16. A case of pharmaceutical development based on the therapeutic potential of the essential oil combination of three Cretan herbs (Cretan IAMA®).

S.A. Pirintsos

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Botanical Garden, University of Crete, Greece

Observations from the island of Crete (Greece) suggest that infusions of traditional Cretan aromatic plants, well known for their ethnopharmacological use in the Eastern Mediterranean region and Near East, could be effective in the prevention and treatment of upper respiratory tract infections, including viral-induced infections. CAPeo (Cretan Aromatic Plants essential oil), a mixture of essential oils derived from *Thymbra capitata* (L.) Cav., *Salvia fruticosa* Mill. and *Origanum dictamnus* L. in olive oil, which has a remarkable antiviral activity against influenza A/H1N1 virus strains, influenza B and human rhinovirus 14 (HRV14) has been advanced by our research group and released in the market in 2015. Here, the scientific evidence which support this product, and the recently findings about its antiviral activity against SARS-CoV-2 will be presented.

Orals

20th International Congress of the International Society for Ethnopharmacology

Verdus BioHerbs.

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Verdus BioHerbs S.A. is the largest company in Greece that processes aromatic and medicinal plants from the Mediterranean and Greek herbal treasure. Our goal is to produce perfect quality essential oils, herbal extracts and dry herbs. Our competitive advantage is the use of a holistic approach that enables the monitoring of all the phases of the production from the seed sowing to the packaging of the products.

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Orals

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O01. Multidirectional pharma-toxicological investigations on *Harpagophytum procumbens DC*. ex Meisn.: focus on water extract activity.

G. Orlando

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Inflammatory bowel diseases (IBDs) are chronic, relapsing and multifactorial disorders of the colonic mucosa, which show increased and unbalanced intestinal immune response to external stimuli. Plant-derived extracts were described to possess the capability in contrasting IBDs related oxidative stress and inflammatory pathways. In the present study, we investigated the water extract of *Harpagophytum procumbens* DC. ex Meisn. in an experimental model of IBDs. Additionally, a microbiological investigation was carried out to discriminate the efficacy against bacterial and fungal strains involved in IBD. Finally, an untargeted proteomic analysis was conducted on more than 100 colon proteins involved in tissue morphology and metabolism. The extract blunted the level of selected biomarkers of oxidative stress and inflammation, including serotonin, prostaglandins, cytokines and transcription factors. Additionally, the extract inhibited the growth of *Candida albicans* and *C. tropicalis*. The extract was also able to exert a pro-homeostatic effect on the levels of multiple colon proteins, thus corroborating protective effects against the burden of inflammation and oxidative stress. On the other hand, the supraphysiological downregulation of cytoskeletal-related proteins involved in tissue morphology and antimicrobial barrier function, namely ezrin, actin, plastin-1, smoothelin and defensins, was observed, as well.

Concluding, the present multidirectional study showed protective effects of *H. procumbens* water extract in blunting the burden of oxidative stress and inflammation in LPS-stimulated colon, alongside with antimicrobial effects against pathogen fungi involved in IBD. Additionally, the fingerprint phytochemical analyses suggest the involvement of multiple active principles namely harpagoside, gallic acid, catechin, epicatechin and resveratrol in the observed pharmacological effects. Nevertheless, the supra-physiological downregulation of ezrin, actin, plastin-1, smoothelin and defensin indicates potential morphological alterations in the colon that should be taken in account in further researches. Overall, this study recommends caution in the use of botanicals enriched with *H. procumbens*.

O02. The antifibrotic potential and pharmacokinetic assessment of *Euclea natalensis* A.DC. as a hepatoprotective agent.

A.M. Reid, N. Lall

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Euclea natalensis has been used by the Tsonga, Venda and Zulu people in southern and tropical Africa for the treatment of dysentery, abdominal pain, stomach problems and chest related complaints, including tuberculosis. The pharmacodynamic properties of the ethanolic shoot extract (DER 1:10 (w:v)) from this plant were tested and validated for its antibacterial, antimycobacterial and hepatoprotectant properties. In addition, the extract showed a reduction in the Th1, and increase in the Th2 immune response. The extract was able to reduce alanine aminotransferase (ALT) in mice associated with drug-induced liver injury. In an attempt to develop the extract further, additional toxicity tests, the antifibrotic potential, as well as the absorption and metabolism of the extract, were assessed. The extract showed effective antifibrotic activity by decreasing 50% of the levels of transforming growth factor-beta (TGF-β) at a concentration of 48.46±1.21 µg/mL (IC_{50}) in rat hepatic stellate cells (HSC-T6). A lesser effect was observed on alpha-smooth muscle actin (α-SMA), where the extract showed an IC_{50} value of 2249±3.09 µg/mL). The effect of the extract on cytochrome (CYP) P450 enzyme metabolism, revealed the inhibition (IC_{50}) of CYP2A6 (68.94±1.1 µg/mL), CYP2D6 (26.44±0.89 µg/mL) and CYP3A4 (24.92±1.36 µg/mL) and the induction of CYP1A1/2. No effect was observed for CYP2B6 and CYP2C9. A metabolomics approach was used to investigate the absorption of the extract through an intact human epithelial colorectal adenocarcinoma cell (Caco2) layer. The results of this study can be used in support of the further development of this extract into an effective hepatoprotectant either as a single entity or in a polyherbal formulation.

O03. Anthelmintic screening of five *Palmatilobae* species used in Traditional Medicine in Katanga Province (DR Congo).

M.G. Nzuzi[1,2,3], N.W. Muyumba[2,4], M.L. Gauthier[2], S.K. Panda[5], W. Luyten[5], A. Nachtergael[2], S. Vandenput[3], S.A. Bakari[6], E.V. Okombe[1], P. Duez[2]

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Resistance to known anthelmintics has become a growing concern. Based on 2017–2019 ethnomedical and ethnoveterinary surveys carried out in Katanga Province, several species of shrub locally identified as *Vitex* species (Lamiaceae) are used to treat helminthiases. In this study, 5 of these species, including different morphotypes, were selected for *in vitro* screening in order to search for new anthelmintic lead compounds.

n-heptane, dichloromethane (DCM), ethyl acetate (EtOAc) and methanol (MeOH) extracts of selected plants (leaves, root barks and stem barks) were evaluated for their anthelmintic activity. The experiment was assessed using a standard motility assay on the N2 wild-type *Caenorhabditis elegans* strain.

Anthelmintic activity was confirmed by the calculation of motility inhibition. The following extracts, tested at 200 µg extract/mL, were significantly active on this model (average ± SD; n = 3; positive control, levamisole, 50 µM: 98.7 ± 4.3 % of motility inhibition): *Oldfieldia dactylophylla* (Welw. ex Oliv.) (Picrodendraceae; locally misidentified as *V. congolensis* De Wild and T. Durand) [DCM leaves (71.0 ± 5.7 %), MeOH stem barks (90.0 ± 7.3 %) and MeOH stem roots (97.0 ± 5.9 %)] ; *V. doniana* Sweet [EtOAc leaves (64.0 ± 4.4 %)] ; *V. fischeri* Gürke [n-heptane leaves (98.0 ± 7.8 %) and MeOH leaves (96.2 ± 6,3 %)] ; *V. madiensis* subsp. Milanjiensis (Britten) F.White [DCM leaves 3-5 leaflets (65.0 ± 5.6 %) and 5 leaflets (69.0 ± 6.6 %)] and *V. mombassae* Vatke [DCM leaves 3-5 leaflets (62.0 ± 8.1 %)].

Anthelmintic screening study showed that 9 of 84 tested extracts are significantly actives on *Caenorhabditis elegans*. The motility inhibitions measured on that N2 wild-type nematode model provide support for the traditional use of these plants and call for further studies to develop compounds as possible treatments for parasitic worm infections.

O04. Bio-guided fractionation and *in vitro* anti-inflammatory potential of *Entada africana* from Benin used in the management of pain related-conditions.

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Entada africana Guill. & Perr., was reported for the management of pain in an ethnopharmacological study in Benin during treatments of fractures and sprains [1]. Additionally, the plant was used in a traditional recipe which was tested for its potential analgesic efficacy in the management of pain related-diseases [2]. The aim of the present study was to determine the anti-inflammatory effect of a hydroalcoholic extract and fractions of *E. africana* obtained with a sephadex LH 20 fractionation using a step gradient from ethanol, methanol-water, and methanol to acetone. This fractionation led to the collection of 25 combined fractions (FC) which were analyzed for their flavonoid profile on thin-layer chromatography. Then, the hydroalcoholic extract of *E. africana* (Ea) and its fractions (FC3, FC4, FC7, FC8, and FC22) were tested for their anti-inflammatory activity according to their phenolic profile. The *in vitro* anti-inflammatory assay was performed using a TNFα-stimulated human keratinocyte (HaCaT) as a skin inflammation model. The MTT assay was first performed, to determine the cytotoxicity of the extracts. FC3, FC4, FC7, FC8, and FC22 extracts did not affect the cell viability, while the hydroalcoholic extract decreased significantly the cell viability. FC7 and FC22 significantly decreased the release of IL8 with FC7 (IC_{50} = 91.74 µg/ml) and FC22 (IC_{50} = 63.47 µg/ml) respectively. For IL6 release, fractions FC3, FC4, FC8 and FC22 had a significant influence with FC3 (IC_{50} = 54.13 µg/ml), FC4 (IC_{50} = 87.55 µg/ml), FC8 (IC_{50} = 75.85 µg/ml) and FC22 (IC_{50} = 72.91 µg/ml) respectively. These results support the anti-inflammatory effect of *E. africana* and its application in the traditional Beninese medicine. Further, its fractions which demonstrated anti-inflammatory effects will be characterized in order to identify promising active compounds for the treatment of pain-related conditions.

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[2] Vissiennon, Z., Ahyi, V., Koupkaki, E., & Nieber, K., Planta Medica

O05. The holistic anti-ageing potential of *Persicaria senegalensis* (Meisn.) Soják ethanolic and fermented extracts.

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Persicaria is one of the fifty genera in the Polygonaceae family of mono- and dioecious herbs, small trees and shrubs. It is abundantly distributed throughout the Northern hemisphere but can be found throughout Africa growing along streams, rivers, pools and swamps in freshwater systems. Traditionally, it has been used for a range of skin troubles and was therefore selected for the current study. The anti-ageing potential of Persicaria senegalensis was confirmed as the ethanolic extract was found to preserve the integrity of the dermis by inhibiting elastase at a fifty percent inhibitory (IC_{50}) value of 50.59 ± 4.36 µg/mL. Additionally, the ethanolic extract was found to curb the chronic inflammation associated with ageing skin by inhibiting cyclooxygenase-2 (IC_{50} 1.97 ± 0.44 µg/mL) and 5-lipoxygenase (IC_{50} 13.3 ± 7.14 µg/mL), and scavenge superoxide (27.22 ± 4.3 µg/mL) and hydrogen peroxide (IC_{50} 71.66 ± 9.86 µg/mL). In the competitive realm of anti-ageing technologies, it is only normal that the search for more powerful actives will always be an enduring one which requires innovation and for the sake of sustainability, frugality. To investigate this, the ethanolic extract was fermented using Bifidobacterium and investigated for its elastase inhibitory potential. The fermented extract was found to have a five-fold lower IC_{50} value of 10.86 ± 2.15 µg/mL. These results support the use of the ethanolic and fermented extracts of P. senegalensis as anti-ageing actives that can be used in the development of anti-ageing technologies.

O06. A natural choice for antimicrobial protection using an indigenous South African plant for acne vulgaris.

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Acne vulgaris (AV) is a chronic inflammatory disease of the pilosebaceous follicle caused by the Gram-positive bacteria, Cutibacterium acnes. The disease affects approximately 9.4% of the world population, making it the 8th most prevalent disease worldwide. In nature, microorganisms rarely exist as planktonic microorganism suspensions, but rather as a complex biofilm. These biofilms, consisting of consortia of species, contribute to antibiotic resistance seen in AV. The Lamiaceae family are aromatic plants known for their antibacterial properties and is used by traditional healers in southern Africa for treating skin diseases. The ethanolic semi-pure fraction from the leaves of plants belonging to the Lamiaceae family was tested against C. acnes (ATCC 6919) to determine the antibacterial, prevention of biofilm formation and quorum sensing inhibition potential. In vitro studies revealed that acne-causing bacteria have a virulence effect through the increased production of the quorum sensing molecule, autoinducer-2 (AI-2). This signalling molecule contribute to the formation of biofilms and the release of virulence factors such as lipase that is associated with inflammation. The inhibition potential of the sample on biofilm formation and AI-2 release was determined. The semi-pure fraction was tested against inflammatory enzymes such as lipase, Cyclooxygenase-2 (COX-2) and matrix metalloproteinase-9 (MMP-9) which have been identified to contribute to inflammatory acne. The semi-pure fraction was confirmed to prevent biofilm formation and the release of AI-2, preventing quorum sensing. Enzymatic studies revealed that the semi-pure fraction was effective against various inflammatory enzymes associated with AV. In vivo studies confirmed that the semi-pure fraction was not irritating to the skin and formulated at 10% in a gel formulation was effective against acne after fourteen to twenty-eight days of consecutive use twice a day. These data suggest the potential of this South African plant for the treatment of acne vulgaris.

Orals

Application of hyphenated techniques in quality assurance of ethnopharmacological sources

20th International Congress of the International Society for Ethnopharmacology

O07. Qualitative and quantitative characterization of chemical profiles in three different parts of *Poria cocos*.

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Poria cocos (Schw.) Wolf has been widely used in traditional Chinese medicine for centuries. Its three medicinal parts are Poria Cutis, the epidermis or fulingpi in Chinese; White Poria, the middle part or baifuling; and Poria cum Radix Pini, the sclerotium with some part of host pine root or fushen. The hostwood in fushen is the inner part, known as fushenmu. The epidermis, middle part and middle-plus-inner part have different clinical applications, but the differences in their chemistry have not been determined. Therefore, it is of great significance to determine how the bioactive components are distributed in the three parts to provide some explanation for their differential clinical use. 13 batches of *Poria cocos* were collected; the botanical parts in each sample, namely, the epidermis, the middle part and the inner part, were manually separated. The carbohydrates (polysaccharides, oligosaccharides and monosaccharides) and secondary metabolites in the three parts were qualitatively and quantitatively characterized by high performance gel permeation chromatography coupled with charged aerosol detector (HPGPC-CAD) and ultra-performance liquid chromatography coupled with triple quadrupole mass spectrometry (UHPLC-QqQ-MS/MS), and ultra-performance liquid chromatography-quadrupole/time-of-flight mass spectrometry (UHPLC-QTOF-MS/MS), respectively. The obtained data were further processed by principal component analysis (PCA) and supervised orthogonal partial least squared discriminant analysis (OPLS-DA). The carbohydrates and secondary metabolites in the three parts varied remarkably. The epidermis contained more polysaccharides with larger molecular weight and higher amount of glucose residue than that of the middle and inner parts, and eight major bioactive triterpene acids accumulated mostly in the epidermis and inner part, with the least accumulation in the middle part. This systematic information on the differences in carbohydrates and secondary metabolites in the three botanical parts of *Poria cocos* provides a factual chemical basis not only for its distinctive therapeutic practice but also for its quality evaluation.

O08. PEGASUS: An analytical chemometrics platform for the discovery of bioactive natural compounds.

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Natural products have been a source of medicinal agents for thousands of years and the remarkable number of modern drugs that have been derived, are predominantly based on traditional medicine. However, isolation of natural compounds has always been tedious, as herbal extracts are complicated systems constituted of hundreds of chemical entities, consume time and solvent and very often the procedure ends with the re-isolation of known metabolites. Complementary, there has been a pressing need to involve approaches that accelerate the measurement of metabolite levels directly from plant extracts through the implementation of high-throughput screening (HTS) technologies and chemometrics.

The goal of PEGASUS is the establishment of a validated analytical chemometrics platform through the elaboration of contemporary chromatographic and spectroscopic techniques (FCPC, HPTLC, LC-HRMS/MS, NMR) along with sophisticated statistical algorithms for the rapid and effective identification of bioactive compounds, prior to their isolation.

The last five years, the development of an *Het*ero*C*ovariance *A*pproach (*HetCA* algorithms), has been applied at our Department for the discovery of bioactive metabolites in several biological targets such as free radicals, enzymes and cancer cell lines [1-4]. *HetCA* is a MATLAB toolbox based on Statistical Total Correlation Spectroscopy (STOCSY) and Statistical Heterospectroscopy (SHY) methodologies. Specifically, in this work is presented the establishment of *HetCA* algorithm via the study of the antioxidant capacity of a mixture consisting of known pure compounds and further more the application of *HetCA* for the discovery of antioxidant compounds from *Stachys tymphaea*, and the anti-tyrosinase activity of *Paeonia mascula* ssp *hellenica*, belonging to the Greek flora.

PEGASUS incorporates for the first time chromatographic and spectroscopic techniques and various bioactivity results along with advanced chemometrics for the rapid identification of bioactive compounds.

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Orals

Aromatic and medicinal plants in animal health ethnopharmacological approach

20th International Congress of the International Society for Ethnopharmacology

O09. Of Elephants and Men: From animal observation to veterinary and human pharmacopeia in Laos.

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In a region of Laos where domestic elephants are still kept on the outskirts of villages, we studied the interactions between the observation of elephants by their mahouts (elephant care handlers) and their own medicinal practices. This study highlights many convergences between elephant self-medication observations reported by mahouts (elephant care handler) and their own medicinal practices, as for example the root of *Harrisonia perforata* which is said to be consumed by elephants suffering from diarrhoea and also used in decoctions by some mahouts to cure this ailment in the household context. Some mahouts explain the therapeutic action of these plant items consumed by elephants by referring to certain principles derived from the semiology and etiology of Laotian traditional medicine; thus of the vegetal items of "cold" nature consumed by elephants that are supposed to counteract "hot" ailments like diarrhoea. Some mahouts combine plant items selected from elephant observation with other plants from the local pharmacopoeia in preparations intended for the care of elephants. These elements show that in this area there is a close interconnexion between ethnoveterinary practices, traditional medicine, and animal observation, providing insight into the pathways that lead to the genesis of certain medicinal practices.

O10. Current status of ethnoveterinary surveys conducted in South Africa.

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Traditional practices are often used by livestock owners in developing countries with limited access to professional veterinary services. Plant-based remedies are commonly used to cure animals suffering from internal or external parasite infestations, wounds, gastrointestinal ailments and inflammation. For such disorders, ethnoveterinary medicine (EVM) may comprise an affordable and readily available alternative or adjunct to expensive orthodox drugs, and also may play an important role in preventative medicine. Interest in the use of medicinal plants has increased markedly as there is a clear association between animal health and human health. Additionally, drug residues in meat combined with the rising occurrence of antibiotic resistance also provide motivation to search for alternatives to commercially available drugs. Indigenous communities hold valuable knowledge of plants used in traditional animal healthcare, and relatively little has been done to document and preserve this knowledge in various communities around South Africa. However, in recent years several more surveys focusing on ethnoveterinary use of plants have been conducted. In the past ten years alone, 139 plants from 50 families used against 21 animal diseases and conditions were recorded. Leaves, roots and bark remained popular plant parts used for EVM. In terms of livestock type reported, the major focus was on cattle, goats and poultry. These studies contribute valuable knowledge that may be used to support development of innovative products for use in preventing and curing certain livestock diseases. However, only four of the nine provinces in the country have been surveyed and there is much to be done to complete the ethnoveterinary medicinal plant inventory of South Africa.

The use of certain plant species for similar ailments by different ethnic groups offers an indication of efficacy and non-toxicity, generating interesting leads for prioritising research.

O11. Effect of dietary aromatic plants and essential oils in combination with tributyrin on broiler chicken meat composition and oxidative stability.

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Meat quality of broiler chicken in terms of chemical composition along with its oxidative stability is of immense interest for consumers. Various feed additives are under examination in order to improve the broiler chicken production and meat quality. This trial examined the combined dietary use of a mixture of aromatic plants and essential oils together with tributyrin, an ester of butyric acid and glycerol that could attenuate intestinal inflammation and oxidative stress. In a 37 days trial, 360 one-day-old male broiler chicks (Ross-308) were randomly allocated to 4 groups with 6 replicates (of 15 chicks), housed in floor pens with rice hulls litter. Commercial breeding, management and vaccinations procedures were employed. Group A (Control) was fed a basal diet. Group B was given the basal diet supplemented with a mixture of oregano oil (50ppm), garlic oil (5 ppm), dried ground sage (2 g/kg) and *Crithmum (1 g/kg)*; Group C the basal diet with tributyrin (1 g/kg); Group D diet with both the mixture of dried aromatic plants and essential oils and tributyrin. At slaughter, breast and thigh samples were collected for analysis. Meat chemical composition was evaluated using the FoodScan Analyser. Meat resistance to oxidation after one day of refrigerated storage was determined as malondialdehyde (MDA). Data were analysed with the ANOVA and Kruskal-Wallis procedures, using SPSS v20 software. The breast chemical analysis showed that the Group D had higher ($P<0.05$) fat percentage compared to the group A, while the other parameters were not significantly affected. Thigh chemical composition did not differ between the groups. The oxidative stability analysis showed that the breast meat of group B had lower ($P<0.05$) MDA values compared to group C, whereas the thigh meat of group B had the lower (<0.001) MDA values and the meat of group C had the higher MDA values.

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Orals

Aromatic and medicinal plants of the Mediterranean basin as food and dietary supplements

20th International Congress of the International Society for Ethnopharmacology

O12. *Pistacia lentiscus* resin (Chios mastic gum): chemistry and biology.

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Chios mastic, is the resinous secretion obtained from the wounds of the trunk and branches of *P. lentiscus* L. var. *chia*, which is endemic to the Greek island of Chios [1]. Since antiquity (500 BC), Chios Mastic has been well recorded for its medicinal and pharmaceutical properties. From 1997, Chios mastic has been identified as a product of Protected Designation of Origin (PDO) while cultivating mastic has been inscribed by UNESCO in 2014 in its Representative List of the Intangible Cultural Heritage of Humanity. In July 2015, mastic was recognized as a traditional medicinal product by the European Medicines Agency (EMA) with two therapeutic indications (mild dyspeptic disorders & skin inflammation/ healing of minor wounds) [2]. In the frame of a continuation study on *Pistacia* sp. an integrated, complementary bottom up approach has been designed. This approach includes isolation of active, marker compounds from starting material with fast and state-of-the-art techniques (CPC-UV, SFC-UV-MS); profiling and characterisation of composition via multiple analytical methods (HPTLC, HPLC-DAD, UPLC-HRMS & HRMS/MS & NMR); and validation of methods for quality control purposes. Additionally, pharmacokinetic characteristics of major mastic constituents have been determined after a human cohort and metabolomics approaches (LC-MS and NMR) have been implemented for revealing of biomarkers. The current work could be considered as an example of a complete workflow implemented in medicinal plants, from the natural entity to human organism.

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O13. Human gastrointestinal and colonic metabolisation study of olive secoiridoids using an in vitro continuous flow dialysis system.

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Olive oil biophenols such as tyrosol (T), hydroxytyrosol (HT) along with secoiridoids oleacein (OLEA), oleocanthal (OLEO) and oleuropein (OLEU) and total polyphenolic fraction (TPF) are genuinely associated with the Mediterranean diet, bearing diverse health promoting and disease preventing abilities. Additionally, a large body of evidence suggests that the potential biological effects of edible phenolic compounds in the human body are partly consequence of their biotransformation by the colon microbiota. Yet, metabolisation studies of the aforementioned compounds are limited.

Aiming towards a detailed investigation into the absorption, metabolism and microflora-dependent transformation of olive oil polar constituents, and within the framework of the international MediHealth project, the GIDM-Colon Model was applied. The above-mentioned model is an optimized, validated *in vitro* continuous flow dialysis system, simulating the absorption from lumen to mucosa; followed by the colon phase using pooled human faecal suspensions, which mimics the physiological conditions during human gastrointestinal digestion [1]. Throughout GIDM-Colon digestion, different samples were collected of dialysate solutions, after gastric and small intestinal digestion, and at four different time points of colonic digestion.

Overall, tentative identification of more than 50 metabolites was achieved by using the information obtained from statistical analysis, the different metabolism stages, chromatographic and spectrometric features and the high-resolution information from the Orbitrap mass analyzer. The metabolisation patterns seemed to differ greatly among the various compounds, and the most abundant metabolites came from compounds containing a 1,2-dihydroxybenzene group, such as the phenylalcohol HT and the secoiridoids OLEU and OLEA. Also, the results indicated that most of the metabolites studied are reaching the maximum level in the human gut metabolism up to 6 h, whereas in the samples taken 24 h after entering the colon phase, a fast elimination of the compounds was observed.

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Orals

Conservation and sustainable use of herbal resources

20th International Congress of the International Society for Ethnopharmacology

O14. Understanding local knowledge as integral factor in designing strategies for sustainably conserving and using herbal resources.

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Local communities have to be familiar with the environment sustaining them. Consequently, knowledge on natural resources and related regional ecological processes is essential in identifying adequate local development strategies also contributing to advancing sustainable development (SD). However, understanding what matters is often challenging. Next to issues of power, this is foremost due to a lack of insight by the (often highly specialized) Western actors into a) the complexity of local knowledge (LK) and b) their own biases in studying the topic stemming from their disciplinary, organizational and personal backgrounds. Thus, researching LK also encompasses a reflection on one's own contribution to shaping said 'understanding'.

In this context, this research aims at facilitating an inclusive approach by suggesting a social-science based framework approach to *understanding LK* that is applicable to a broad range of LK forms in industrializing and industrialized countries. Moreover, next to being accordant with basic sustainability principles and allowing for a critical reflection of one's own situatedness, the framework is also expected to allow for comparative studies.

Methodically, the research is based on an extended literature analysis across sociology, philosophy, anthropology, geography and the ethno-sciences. Against this backdrop, suitable theory-based generic dimensions – in total 16 – have been identified and aggregated in a conceptual-analytical framework according to Jabareen[1]/Dowding[2]/Stanley[3].

In conclusion, an interdisciplinarily-founded framework on *understanding LK* is proposed. Its purpose consists in facilitating the understanding of principally any given LK form through a theory-based minimal set of interconnected key dimensions and questions. Given its normative foundation in SD, this social-science based approach to LK provides interested parties with a differentiated way to gain comprehensive insights into local contexts as a basis for determining suitable conservation and management approaches.

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Orals

Ethnobotanical approach to drug discovery Theory, methodology and limitations

20th International Congress of the International Society for Ethnopharmacology

O15. Mitigating climate change in Valmalenco (so, Italy): how can Ethnobotany contribute?

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This ethnobotanical survey is part of the Interreg Italy-Switzerland project *B-ICE* [1], which is aimed to create a new management model for the ongoing climate change and to reach new sources for the local enhancement. The work was performed in Valmalenco (SO, Italy) and aimed at investigating the traditional uses of the local plants. The retreat of the glaciers and the snowfall reduction are the main transformations in the natural landscape [2,3,4]. The territory is no longer suitable for winter tourism and this has major consequences for the local economy. Traditional ethnobotanical uses are an important part of the rich biocultural heritage locally available; promotion of this heritage could represent an alternative strategy for the economic development of the area. Attention was paid to the common and dialectal names of the plants, their survival, their past or actual use, the preparation forms, the administration methods and the therapeutic indications. In summer 2019, 300 interviews were collected and it emerged that the used species are 150. Out of the most cited ones, *Achillea moschata* Wulfen is used for digestive purposes, as analgesic, hypotensive, for gynaecological infections caused by *Candida* spp., as anti-inflammatory and ulcers healing in the lower limbs and against intestinal swelling in bovine. *Malva sylvestris* L. is used as emollient, relaxing and purifying, in case of allergies, for acne reactions and soothing baths, *Thymus pulegioides* L., as expectorant, for sore throat and cold and *Achillea millefolium* L., for menopause symptoms, footbaths, as disinfectant for wounds on bovine paws. This knowledge will define the "cultural landscape" of the territory and will allow to select symbol species, with the purpose of realizing a high-rise Botanic Garden. This place will aim to preserve the plants and the biocultural diversity and to attract a new form of tourism based on the rediscovery of ethnobotanical traditions.

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O16. Ethnopediatric knowledge among mothers living in Southern Romania.

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Medicinal plants have a long history of ethnopediatric use in Romania. Our aim is to establish which plants are used for treating children diseases nowadays in Southern Romania. Ethnopediatric knowledge was recorded using structured interviews focused on respondent demographics (age, community of residence), local names of useful plants, part(s) used, means of preparation and administration, local folk indications of taxa. All the mothers (104) with 0 to 18-year-old children hospitalised between September 2019- January 2020 in "Grigore Alexandrescu" Children Emergency Hospital Bucharest and living in Southern Romania were enrolled in the study and interviewed. Use Value Citation Index (UV) and Informant Consensus Factor (ICF) were calculated. A total of 22 plants were found to be used to treat children diseases in Southern Romania; 68 informants are living in urban area and 36 in rural area. Most frequently used plants were mint (*Mentha spicata L.*) in 51 cases (UV=0.519) in diarrhoea – ICF=0.97, chamomille (*Matricaria chamomilla L.*) in 32 cases (UV=0.307) in colic – ICF=0.5 and skin infections – ICF=0.75, and silver linden (*Tilia tomentosa Moench*) in 19 cases (UV=0.182) in acute respiratory diseases – ICF=0.66 and anxiety and agitation - ICF=0.66. Less used plants were garlic and pumpkin (*Cucurbita pepo L.*) in intestinal worms, onion in acute respiratory diseases, heartsease (*Viola tricolor L.*) in hives, wheat bran (*Triticum aestivum L.*) in dermatitis, dill (*Anethum graveolens L.)* in colics and greater celandine (*Chelidonium majus L.*) in skin verruca and acute hepatitis. One informant reported insertion of a garlic clove in the rectum to treat oxyuriasis. In 44 cases plant treatment was associated with drugs. About 1/3 plants were recommended by physicians and 2/3 by family members. In 4 cases plant indication was collected from internet. In conclusion, medicinal plants still represent an important indication in the treatment of various pediatric diseases in Southern Romania.

O17. Nagoya protocol and the need for new forms of collaboration.

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The Convention on Biodiversity (CBD), followed by the Nagoya Protocol (NP), responded to the need to proactively engage in ethical practices when investigating the use of genetic resources across countries to overcome a long history of resource exploitation. In terms of access and benefit sharing (ABS), the CBD defined the framework while the NP proposed more practical directions as to how to resolve those problems. Implementation at national levels needs to ensure that all collaborators are treated fairly and that our scientific projects take account of each party's requests and desires.

The lack of clarity at a national level about their implementation drove some investors and collaborators away, putting countries that ratified the NP or CBD at an economic disadvantage. Examples of NP-abiding projects with lucrative commercial outcomes are rare and are often not in the public domain. In this case-study, funded by the UK's Darwin Initiative and with indigenous and government stakeholders in Guatemala we jointly develop a new paradigm on *how to* conduct a scientific project that follows the rules and fulfils all requirements prescribed by the NP. A collaboration between UCL (London), UVG (Guatemala), an industrial partner (Indigena Biodiversity Ltd.) and an indigenous group in Guatemala, analyses case-studies for patients' medicinal plants uses which potentially are also of interest for industrial development.

The outcome will be the documentation of the process (agreement/disagreements on all points, discussions, ABS terms). Tools for the application of NP will be produced and made available openly. The importance of continuous discussion and regular meeting of all stakeholders, alongside classic negotiation methods, is a core element of these transdisciplinary processes. Therefore, the inclusion of indigenous partners is an essential element of this equitable partnership.

One year into the project, we will be presenting our strategy, methods, as well as some preliminary ethnobotanical/ethnomedical data.

O18. Traditional Chinese Medicine: A Global Exchange.

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The history of medicine is not complete without considering the history of trade. Throughout time, the exchange of goods helped shape the medical field in various cultures. While physical goods are the easiest to track, the introduction of new knowledge and illnesses also shifted priorities and understanding in medicine. Traditional Chinese medicine is not exempt from the influence of global trade. For example, the Ming Dynasty scholar Li Shizhen's Compendium of Materia Medica recorded 10% of imported medicinals that were known and used in traditional Chinese medicine; some well-known imports include myrrh, frankincense, etc. However, the exchange was not one-sided. Various Chinese medicinals, such as tea and gingko biloba, have also been exported. Some of these medicinals are used for the exact same purpose in the exported country, while others found different indications. The current idea of drug discovery is geared towards conventional medicine--extracting, refining, and chemically reproducing. The limitations of traditional medicinals translating to conventional medicine is well explored with concerns from the misunderstanding of its original system to the ability to reproduce results clinically. However, with the age of information, drug discovery should also be considered on the level of traditional medicine systems. For example, whether common Western herbals can be integrated into systems such as traditional Chinese medicine. As modern globalization brings the world together, information is easier to access than ever, and modern technologies offer a different perspective on old knowledge. Moving forward, traditional Chinese medicine should consider the integration of other medicinals as it did historically, and conventional medicine should evaluate traditional medicinals within their systems as they are meant to be.

O19. Plants used as ethnomedicine for rheumatic disorders by Williche people.

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The present study assesses the state of ethno-medicinal plant knowledge by Williche people in Los Lagos Region, Southern Chile. Fieldwork was conducted to the study area from August 2015 to March 2016, supplemented by follow-up visits during 2016. Data were collected in San Juan de la Costa through collection of medicinal plants, semi- structured interviews, forest walks, participant observation and informal conversation. Fourteen informants were interviewed, 5 were local healers. Most were elderly people.

Twenty-six medicinal plants in 24 genera from 19 families were indicated by the informants. They were listed with scientific name, vernacular name, plant parts used, method of preparation, and traditional use. The most important families in terms of taxa were Asteraceae (5 species), and Solanaceae, Lamiaceae, and Urticaceae (2). Orchidaceae, and Aextoxicaceae (1) were also considered important for other ailments.

The documented plants were used to treat rheumatic disorders, but also blood cholesterol, circulatory problems, and wound healing. Shrubs (9 species) and herbs (9) were the most used life forms followed by trees (5), and climbing plants (3). The most frequently used plant parts were leaves (58.8 %) followed by whole plant (12.1 %), twigs (9.1 %) and stems (6 %). The plants used were 12 native to Chile, 5 endemic, and 9 exotic. The most frequently used preparation methods were poultice application (93 %) followed by decoctions (7 %). Species richness were mainly Magnoliopsida (88.4%) and Liliopsida (11.5%).

The present study documents plant diversity showing that medicinal plant knowledge and cultural beliefs are still present in the Williche community for rheumatic problems. Further ethnobotanical studies on healing practices and transmission on ethno-medicinal knowledge to young generations are required for the conservation of the local forest and the communities themselves.

Orals

Greek Ethnopharmacology

20th International Congress of the International Society for Ethnopharmacology

O20. Herbal remedies in an Alexandrian manuscript of late antiquity.

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A medical manuscript with the title *Dynameron* is kept in the Marciana National Library of Venice (Cod. gr. Z. 295) and originates from a text initially written in Greek by a physician named Aelius Promotus, who lived and worked in Alexandria (1st to 2nd century AD). *Dynameron* of Aelius Promotus contains 130 chapters dealing with different diseases and their treatment. He describes 870 recipes, composed of 430 ingredients (about 300 medicinal plants, 70 animal products and 60 minerals). The large number of ingredients used in each recipe implies that Aelius Promotus was a follower of the so called "Empiric school" of medicine, although in his work are easily recognizable also influences from other theoretical sects. Most of the plants used in *Dynameron* are of well-established use and they originate from the larger Mediterranean area. However, in his recipes there are also some unusual products (such as pepper, ginger, cinnamon and cassia), showing that trade with remote regions in Asia had been highly developed in the late phase of the Roman Empire. Some of the recipes refer to ingredients that cannot be granted any apparent therapeutic reasoning. Additionally, some treatments seem more like superstitious rituals. However, when *Dynameron* is evaluated as a whole, the conclusion is that Aelius Promotus was a competent practicing physician with great experience, typical of the famous medical tradition of Alexandria during the late Roman era. There is evidence that *Dynameron* was highly estimated and was copied several times thereafter, in order to serve as a therapeutic manual, a kind of "vademecum", for the common ailments a physician might encounter in his everyday practice.

O21. *Origanum dictamnus:* phytochemical analyzes and relaxing effect on rabbit intestine.

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Origanum dictamnus L. (Lamiaceae) is a Greek species endemic of the island of Crete, nowadays cultivated in other parts of Greece as well. In 2014 the Community Herbal Monograph was issued [1] giving new opportunities to the development and sustainable cultivation of this important medicinal plant. *O. dictamnus* herb and its aqueous preparations are listed in the category of Traditional Use Herbal Medicinal Products of the EMA for the treatment of several conditions, such as wound healing of skin inflammations and bruises, mild gastrointestinal disorders and relief of cough associated with cold. Despite its importance as a medicinal herb, the first systematic analysis of the non-volatile constituents was reported recently by the work of Chatzopoulou et al., [2] which revealed a variety of phenolic compounds. Continuing this work extended studies on the chemical content of aerial parts, roots and herbal teas of cultivated *O. dictamnus* were undertaken. Targeted isolations guided by HPLC-PDA-ESIMS and NMR analyzes enabled us to create an in-house chemical library, but also revealed the presence of new constituents reported for the first time. Up to now more than 60 constituents have been identified, among them seven new natural products belonging to the classes of flavonoids, jasmonates and monoterpene derivatives. Preliminary experiments with dittany infusions on rabbit intestine confirmed the ethnopharmacological use of the herbal drug in mild gastrointestinal disorders, as they produced a dose-dependent relaxing effect both in the jejunum and colon. At the threshold concentrations of 0.1 mg/ml for the colon and 0.3 mg/ml for the jejunum, dittany produced a slight decrease in both the phasic contractions and the basal tone, while at higher concentrations it caused a significant decrease in the tone and a considerable inhibition in the amplitude and frequency of the phasic contractions.

[1] EMA/HMPC/200429/2012 Corr.
[2] Chatzopoulou et al., 2010, JAFC, 58, 6064.

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O22. Discovering herbal teas/mountain teas in Kozani-Western Macedonia-Greece, an ethnopharmacological journey.

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Wild native herbs have been widely used in alimentation and constituted the main form of traditional medicine by creating the first therapeutic approaches to treat a huge variety of symptoms. Gaining a deeper understanding of the potential benefits that herbs hold would undoubtedly aid in treating contemporary diseases. The region of Kozani, Western Macedonia in Northern Greece combines diverse altitude, multiple rivers and soil of great biodiversity value, that gives rise to a number of region-specific herbs. Moreover, only a few studies have investigated the traditional uses of medicinal herbs in Kozani, thus underlining the necessity of a systematic study of their traditional uses in health and disease.

Hence, the purpose of this study is to investigate the traditional medicinal herbs of Western Macedonia, primarily focusing on their consumption as hot drink infusions-tea in accordance with their therapeutic applications. To investigate the medical value of the herbs we have created an oral questionnaire comprising 20 questions and accompanied by photos and samples of the local herbal plants. The candidates were selected based on their age and their origin.

This study constitutes the only recent effort to register the vast variety of medicinal plants in Kozani, Western Macedonia. Among the various uses of mountain tea that came up in the study, it is noteworthy to pinpoint "Mountain tea" *Satureja montana, Stachys iva* and *Sideritis* sp for treatment of the symptoms of the common flu and cough, *Achillea coarcata* for hypertension, *Teucrium* for diabetes, while other famous herbs such as *Chamomilla recutita* and *Salvia sclarea* have also emerged. Future studies will focus on increasing the number of candidates as well as the range of the area studied. Finally, it is evident from this study that traditional knowledge and ethnopharmacology could prove to be invaluable in identifying novel therapeutic compounds.

O23. Expanding the scope of Greek Ethnopharmacology - eurasian medicinal plants on the Mediterranean.

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Analysis of Greek ethnopharmacology traditionally considers the materia medica native to Mediterranean region. A close examination of documentation dating as far back as the remotest antiquity shows, however, that this approach is limited as it does not take into consideration the materia medica introduced into the Mediterranean region from as far as China. This presentation will focus on this non-native materica medica. It will be based on a systematic screening of Greek ancient texts dating from the 5th cent. BCE to the 15th, and will trace the presence of this materia medica, made of plants and other natural substances coming not only from China, but also from all the regions either between China and the Mediterranean, or connected to the sea or land roads through which these substances were traded, that is, the whole Eurasia. This presentation will then compare the uses of this materia medica in the Greek texts and their equivalent in the pharmacopoeitic literature of the native regions of this materia medica. This will bring to light both the persistence of uses from the regions of origin of traded substances and new ones, possibly generated in the regions crossed by the materia medica. This will suggest that trade was not limited to transmission, but also included assimilation of local uses and new applications of traded substances in a thus far unsuspected way. In conclusion, when Eurasian medicinal plants reached the Greek World, they conveyed with them a multiple and diversified knowledge of their therapeutic properties and applications that benefitted and enriched Greek ethnopharmacology.

O24. Ethnopharmacological study of the medicinal plants used in the folk medicine in Drama, Nothern Greece.

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A complete ethnopharmacology survey was conducted in the Drama district of Eastern Macedonia and Thrace, Greece. Ethnobotanical knowledge is passed on orally from one generation to another. Special formed questionnaires were used to elicit information on the medicinal plants used to cure common ailments. Primary information was collected from local interviewees, mostly elderly men and women, residents of the aforementioned area. The survey subjects, natives or immigrants from Asia Minor and the Aegean Sea coastline, which used to be Greek territories before becoming Turkish after the population exchanges. Various data (information, recipes) are collected concerning plants with alleged therapeutic properties, and which are used to date as a treatment offered to both humans and livestock for various medical issues. The survey was conducted following a documented protocol specifically designed for the particular project. In some cases, animals, animal products, commonly used objects and chemicals are used alone or added to the medicinal plants to enhance their curative abilities. The findings emerged from this ethnopharmacological survey are very interesting. The verification of the data of the medicinal plants used in traditional healing practices in Drama clearly shows the need to document these practices, and the wealth of undocumented knowledge out there. The current study reinforces the importance of extending the study to other parts of Greece.

O25. In the course of thyme: the use of *Thymus* spp. in European traditional medicine & the example of *Thymus thracicus*.

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Since the beginning of human history, thyme has been an indispensable medicinal plant, employed for the treatment of various ailments, ranging from the severest to the most common ones. From the ancient Greco-Roman manuscripts to modern Pharmacopeias, knowledge, passing on from one generation to another, has survived until today with extensive studies validating the effectiveness of thyme extracts. From ancient Greco-Roman herbaria to modern Pharmacopeias, knowledge passing on from one civilization to another, has survived until today, with extensive bibliography validating the effectiveness of thyme extracts. A sub-endemic Balkan species of thyme, *Thymus thracicus* Velen. has been the focus of our research, which has resulted in the isolation of nine polyphenols so far, namely oresbiusin A (1), rosmarinic acid (2), methyl rosmarinate (3), 9″-methyl lithospermate (4), dimethyl lithospermate (5), luteolin-7-O-glucuronide (6), calceolarioside A (7), 3-methoxy-calceolarioside A (8) and grayanoside B (9). The chemical structures of the isolated compounds were established by 1D and 2D NMR analysis (^{1}H, ^{13}C, gDQCOSY, gHSQCAD, gHMBCAD), and through comparison with the literature. A series of in vitro assays followed suit, mainly focusing on the antioxidant and anti-inflammatory activities of the isolated compounds, accompanied by preliminary in silico studies, which, in turn, yielded moderate results concerning the compounds' potentiality to act as ligands for nuclear and G protein-coupled receptors, suggesting possible directions for future research.

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O26. Galen's formulas in his book on antidotes.

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According to Galen the term *antidote* is used in regard to formulas addressing a general category of internal diseases that could involve a poisoning, an attack by a venomous animal, or an improper way of living. A subgroup of antidotes, named *theriacs* from the Greek word for *wild animal*, could address all three categories. Galen's text *De antidotis* contains over 100 formulas for compound medicines, from relatively simple to very elaborate ones, as for example the one hundred-ingredients' *theriac*, designed by Galen himself. Containing formulas composed by at least 50 pre-Galenic physicians and therapists, the Galenic work is a good example of a major collection of standardized compound drugs, reaching approximately 28,000 words. A detailed textual study of these formulas revealed interesting information on the Greek and Roman pharmacy. A fifth of these formulas can be considered large, exceeding a total of 30 ingredients. Almost half of these ingredients were imported from Africa, Arabia or India, pointing to a thriving trade of medicinal substances and spices. Several of these herbal, animal and mineral drugs were expensive, rare or extravagant, even for a wealthy Roman physician. *De antidotis* represents one of the most important formularies of classical antiquity, which was instrumental for the spreading of *theriac* through the world (e.g. Tibet, China and Japan) or even adapted locally by other medical traditions (e.g. Brazil). Although the text remains underexploited, it is nevertheless a rich source of herbal knowledge with promising candidates for future research.

Orals

Pharmacological and clinical studies of medicinal plants & natural products

20th International Congress of the International Society for Ethnopharmacology

O27. *Artocarpus tonkinensis*, a remedy from Vietnamese ethnopharmacology, protects mice against collagen-induced arthritis and decreases Th17 cell function.

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Artocarpus tonkinensis (Moraceae) is a tree that grows in north Vietnam whose leaf decoction is used as a traditional remedy by the Hmong ethnic group to treat arthritis and backache. Our study evaluated the decoction's efficacy and mechanism of action in DBA/1J mice with collagen-induced arthritis (CIA). CIA are characterized by chronic joint inflammation mediated by IL-22 produced by Th17 cells, which stimulates synovial fibroblasts to induce cell proliferation and secretion of other inflammatory cytokines, and the initiation of osteoclastogenesis. Moreover, IL-6 produced by fibroblasts responding to Th17 cell-derived IL-17 amplifies inflammation and stimulates synovial tissue in an autocrine manner. Thus, IL-17 maintains the inflammatory cycle via activity of downstream cytokines.

Mice treated with the decoction (At) (5 g/100 mL, ad libitum) either from the first collagen immunization or after CIA development experienced significantly less joint edema and inflammatory infiltration, whereas CIA-induced cartilage damage could only be prevented by early At treatment. Autoimmune gene expression profiles showed that Th17 cell-associated chemokine CCL20, the chemokine receptor CCR4, the cytokines IL-6, IFN-γ, IL-17, and IL-22 and the IL-23 receptor were strongly downregulated by At. Reduced expression of IL-2, IL-17, IL-22, and FasL in lymph node cells from At-treated mice was further confirmed by real-time PCR. The decoction also inhibited polarization of Th17 cells from CD4+ splenic T cells according to levels of IL-17 and RORC, a Th17 cell-specific transcription factor. Chromatographic analysis identified At's major component as maesopsin-β-D-glucoside, which could inhibit in vitro differentiation of Th17 cells

In conclusion, although some questions remain regarding the mechanism of At-mediated Th17 cell attenuation, our findings highlight the importance of investigating traditional ethnomedicines for treatment of diseases. The potential anti-inflammatory activity of At provides an alternative RA therapy apart from methotrexate and corticosteroids that should be explored further with respect to its optimal efficacy and possible non-specific cytotoxic effects.

O28. In vitro assessment of anti-influenza activity and neuraminidase inhibition of *Cistus creticus* L. subsp. *eriocephalus* (Viv.) Greuter & Burdet grown in sea coast area of South Tuscany, Italy.

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Cistus creticus L. subsp. *eriocephalus* (Viv.) Greuter & Burdet (formerly *Cistus incanus* L.) is a perennial shrub, common on sunny or rocky soils in Mediterranean area. Leaves and aerial parts of *C. creticus* have been traditionally used in Greece and Italy to treat several respiratory infections. Also, in South Tuscany, in the sea coast area of Maremma, *C. creticus* subsp. *eriocephalus* is used as herbal tea to treat cold symptoms and flu. In this work, we chemically characterized, and we investigated two ethanolic extracts (ethanol 65% v/v) obtained from samples of *C. creticus* subsp. *eriocephalus* (CIS) aerial parts collected in two different sites of Maremma for their *in vitro* activity against influenza A H1N1 strains in pre- and post-cell infection on MDCK cells. We also evaluated anti-neuraminidase activity of the extracts by calculating IC_{50}. Oseltamivir was used as reference drug. CIS ethanolic extracts, in the same manner, were found effective as anti-influenza drugs by strongly inhibiting influenza virus entry in MDCK cells (cytotoxic/effective concentration ratio >4) and, partially, by reducing viral replication. IC_{50} in anti-neuraminidase activity test was below 0.025 mg/ml for both extracts. Chemical analyses showed that extracts are very rich in polyphenols (20.3%-23.0% m/m of extracts). Flavan-3-ols oligomers occur in CIS samples as main polyphenols subclass (11.3%-14.2%) and these constituents are likely to contribute more to the antiviral efficacy [2]. Also flavonols (4.3%-4.9%), mainly represented by myricitrin and other myricetin and quercetin glycosides, are an important chemical class of the extracts. Our findings suggest that *C. creticus* subsp. *eriocephalus* is worth to be better investigated for its anti-influenza activity and ongoing researches are aiming to investigate molecular mechanisms of CIS constituents on other viral targets such as envelope glycoproteins and hemagglutinin.

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O29. Wound-healing activity of essential oils from five *Hypericum* spp. growing wild in Greece.

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Hypericum L. is generally classified as an essential oil-poor genus [1], however studies have shown that its volatile oils possess antimicrobial, antioxidant, antiangiogenetic and gastroprotective activities [2]. The most common preparation for the treatment of wounds and skin inflammations is the infused oil obtained from the aerial parts, containing napthodianthrones, phloroglucinols and essential oil (EO); however, there is much controversy in the scientific community regarding the composition and stability of this formulation [3]. Extensive literature survey shows, that, unlike napthodianthrones and phloroglucinols, EO from *Hypericum* spp. has not yet been evaluated regarding the wound healing efficacy.

In the present study, five *Hypericum* spp. growing wild in Greece, namely *H. perforatum* (HP), *H. empetrifolium* (HE), *H. triquetrifolium* (HT), *H. amblycalyx* and *H. jovis*, were subjected to hydro-distillation to obtain their EOs. GC-MS analyses revealed the presence of 137 individual compounds representing 87.8-96.8% of the total EOs. Three taxa (HP, HE, HT) yielded enough EO for in vivo tests, which were conducted in hairless SKH mice. Wounds of 1 cm^2 created on the upper back of the mice skin under anesthesia. The wounds were evaluated by transepidermal water loss, hydration, redness, thickness and elasticity measurements, photodocumentation and histopathological examination. The EOs showed high percentages of wound-healing in comparison to the control group (untreated mice) and the Vaseline-treated group, but for HP and HT the inflammation remained. The EO of *H. empetrifolium* (HE) possesses significant wound healing properties, as it was indicated by clinical evaluation and histopathological results. This later result confirms the traditional use of HE in Greece for wounds and skin inflammations [4].

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O30. Regulation of glutathione biosynthesis by the addition of *Allium hookeri* in the diet and its role in the prevention of metabolic syndrome.

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Oxidative stress resulting from the depletion of glutathione (GSH) level plays an important role in the generation of various degenerative diseases including type 2 diabetes (T2D). We tested the hypothesis that depleted level of GSH may be enhanced through the supplementation of cysteine rich medicinal herb in High fat High fructose (HF-HFr) diet induced diabetic rats. Male SD rats were divided three groups (n=6), namely Normal diet, HF-HFr diet, and HF-HFr diet along with methanolic leaf extract (250 mg/kg) of our plant of interest. The experiment was carried out for 20 weeks. *In vitro* study was performed by using rodent L6 muscle cell line. Compared with the diabetic group, the plant extract treated group showed a significant lower blood glucose level and higher GSH and lower GSSG (glutathione disulfide) levels in skeletal muscle tissues. Plasma free fatty acid (FFA) levels were found to be higher in the diabetic group while the treated group showed a significant lower level of FFA. The protein expression of GCLC, GR, and GLUT4 were also upregulated in the skeletal muscle tissues of the treated group compared to HF-HFr group. *In vitro* studies further demonstrated the beneficial effect of the plant extract in increasing glucose uptake and GSH levels and decreasing GSSG levels in sodium palmitate (0.75 mM) treated cells. Combining all, this study suggests that exposure to HF-HFr diet caused an increased accumulation of FFA followed by increased blood glucose level and diminished GSH level. Dietary supplementation with cysteine rich herb can restore the GSH level leading to lower accumulation of FFA and controlled blood glucose level.

O31. Efficacy of honey-based ophthalmic formulation in patients with corneal ulcer: A randomized clinical trial.

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Honey is traditionally used for infectious ulcers. The aim of this study was to evaluate the efficacy of honey-based ophthalmic drop in patients with foreign body induced corneal ulcer. Honey is traditionally used for the skin, mucosal and corneal ulcers. Its use is well studied in human skin and mucosal ulcers and animal model of corneal ulcer with promising effects. In this randomized clinical trial, 50 patients with foreign body induced corneal ulcer were allocated to receive 70% sterile honey-based ophthalmic formulation or 0.3% ophthalmic ciprofloxacin, as the standard treatment every 6 hours. Moisture content, pH, electrical conductivity and hydroxymethylfurfural content of honey sample was measured according the method previously described by the association of analytical chemists international. Moisture was determined according to the correlation between honey moisture and refractive index. pH was measured by means of a potentiometric pH-metre in a 70% honey solution. Electrical conductivity was measured by a conductimeter. Hydroxymethylfurfural was determined applying reverse phase high pressure liquid chromatography and UV detection. All patients were examined for the size of corneal epithelial defect, corneal infiltration and depth and followed on a daily basis until complete healing. Duration for complete healing was considered as the outcome measure. Smear, culture, antibiogram and minimum inhibition concentration (MIC) test were performed for honey and ciprofloxacin in all patients. The average durations of complete healing of corneal epithelial defect in the honey and ciprofloxacin groups were 3.88 ± 3.44 vs. 6.32 ± 3.69 days, respectively (p=0.020). No significant difference was observed between the two groups regarding the average duration of healing of corneal infiltration (8.12 ±1.94 days vs. 8.64±2.15 days, p=0.375). MIC of honey for pseudomonas aeruginosa was 60% w/w, for E.Coli 40% w/w, and for staphylococcus aureus 30% w/w. Honey based ophthalmic drop can accelerate the corneal epithelial defect healing in patients with foreign body induced corneal ulcer, compared to ophthalmic ciprofloxacin as the standard treatment.

O32. Antiprion effects of naturally occurring compounds.

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Transmissible Spongiform Encephalopathies (TSEs) are neurodegenerative diseases afflicting a wide variety of hosts, including humans. The common pathogen, termed Prion, is believed to consist primarily or exclusively of the abnormally folded isoform (PrP^{Sc}) of a highly conserved protein (PrP^{C}). Although the exact pathogenetic mechanism remains elusive, PrP^{Sc} drives the generation of novel PrP^{Sc} molecules, through the conversion of PrP^{C} into PrP^{Sc}, thus promoting disease progression.

Neuropathological hallmarks of TSEs include spongiform degeneration, gliosis and neuronal death. Behavioral and visual disturbances, gait or limb ataxia, myoclonus and aphasia are common clinical signs. In humans clinical disease progresses rapidly (mean: 4-6months) and results in death [1].

Although several therapeutic approaches have been evaluated to prevent and/or treat TSEs, ranging from small molecules to antibodies and RNAi targeting PrP, TSEs remain incurable, rendering the need for novel therapeutics clear and still unmet [2].

In this project we evaluated the naturally occurring compounds, carnasol (CR), carnasolic acid (CA) and myrtillin for their antiprion effect. CR and CA are isolated from *Rosmarinus officinalis* and have been associated with antioxidant, anti-inflammatory and neuroprotective effects [3]. Myrtillin and other anthocyanins are abundant in *Vaccinium* and *Vitis* species and have similar beneficial health effects [4].

To evaluate the antiprion effects of the compounds, we utilized ScN2a, a cell line persistently infected with prions, and confirmed the antioxidative effects of CA and CR, exerted through upregulation of the Keap/Nrf2 pathway, by qPCR. Moreover, we established a novel antiprion effect, evidenced by the reduction in PrP^{Sc} accumulation in prion-infected cells. This effect appears to be direct, as CA, CR and myrtillin blocked *de novo* generation and reduced already formed PrP^{Sc} aggregates in RT-QuiC, cell-free experiments. These data imply a protective and possibly therapeutic role for the compounds tested, which could evolve into much-needed agents against TSEs.

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Application of hyphenated techniques in quality assurance of ethnopharmacological sources

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P001. FT-IR spectroscopy as a green analytical technique to screen the botanical origin of essential oils rich in 1,8-cineole.

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Eucalyptol (1,8-cineole) is the major volatile constituent of some essential oil (EO) products like those from eucalyptus and bay laurel that are highly appreciated in the flavor market. It is also present in a wide variety of other popular EOs like those from rosemary, sage, cardamom etc. Some of these products may differ greatly in price or bear similar olfactory notes so that may be used for fraudulent practices like admixing and mislabeling [1]. Identification of the botanical origin of the EOs is a challenging area of research for authenticity purposes mainly because of the great variability in their chemical composition [2], usually examined through GC analyses. Among the most versatile, fast and cost-effective analytical techniques, Fourier-transform mid infrared (FT-MIR) spectroscopy coupled with chemometrics has attracted great interest for evaluation of different quality aspects e.g. grading according to their content in major volatile constituents [3] but also detection of counterfeit [4]. In the present study, the transmission FT-IR spectra of commercial "eucalyptus" (*n*=5), "bay laurel" (*n*=11), "sage" (*n*=2) and "rosemary" (*n*=1) as well as "melissa" (*n*=2) EOs were pre-processed and then evaluated against a reference spectral database for authentic *Laurus nobilis* L. (bay laurel) EOs of highly varying relative contents in 1,8-cineole (ca. 25-50% v/v). The objective was to explore for possible diagnostic markers about the botanical origin of a given EO product. The FT-IR spectral bands between 1355-1375 cm^{-1}, 1680, and 1730-1750 cm^{-1} were found as the most promising ones for possible discrimination of various -rich in 1,8-cineole- EOs according to their botanical origin. Complementary GC-FID-MS analyses revealed that the above regions are associated with compounds barely found in bay laurel EO like camphor, β-thujone or citral. These findings support the exploitation of FT-IR as a green analytical technique to screen the botanical origin of 1,8-cineole-chemotyped EOs.

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P002. Investigation of *Eleutherococcus gracilystylus* adulteration / substitution with *Periploca sepium* using High Performance Thin Layer Chromatography.

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As the consumption of herbal medicines has increased globally, so too has the incidence of adverse events associated with such products [1] often linked to poor quality or adulteration. Further cases may be prevented through preemptive investigation and dissemination. One such example is the under-reported but widespread adulteration of *Eleutherococcus nodiflorus* (Dunn) S.Y. Hu (better known under the synonym: *Eleutherococcus gracilystylus* (W.W.Sm.) S.Y. Hu) with *Periploca sepium* Bunge, a medicinal plant containing cardiac glycosides.

To investigate and disseminate the widespread adulteration / substitution of medicinal plant *E. gracilystylus* with *P. sepium* using High Performance Thin Layer Chromatography (HPTLC), analysis.

31 Samples were purposefully, convenience sampled in 10 markets and pharmacies throughout China *and analysed using HPTLC Association Method (*adapted *from Monograph 2432, Ph.Eur. 8.0)*.

HPTLC analysis resulted in clear separation and differentiation of all samples and standards. 74% of the samples showed evidence for adulteration/ substitution. 52% with *P. sepium* and 10% with *E. senticosus*, 12% unidentified and 26% were found to be *E. gracilystylus.*

E. gracilystylus adulteration / substitution is common and if left unchecked has the potential to create a serious adverse event.

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P003. A Headspace Gas Chromatography-Mass Spectrometry method for the analysis of lavender essential oils' volatile content.

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In this project, we aimed to develop a comprehensive profiling method for the study of the volatile content of different lavender essential oils by static headspace gas chromatography coupled to mass spectrometry (HS-GC-MS). Analysis was performed using an EVOQ GC-TQ Bruker system with a CTC-PAL autosampler, in full scan mode (50-500 amu). Chromatographic separation was performed on a HP-INNOWax (60 m x 0.25 mm x 0.25 mm) column by applying the following temperature program: 52oC (2 min), 5oC/min to 80oC (4 min), 4oC/min to 250oC (1 min), helium constant flow rate 1 ml/min.

In order to identify the optimum parameters in terms of chromatographic separation and signal intensity, a screening Design of Experiment process was employed. More specifically, Plackett-Burman experimental design was applied using seven headspace injection parameters: incubation equilibration temperature (X1) and time (X2), agitator vortex speed (X3), post injection dwell time (X4), inlet temperature (X5), split ratio (X6) and injection flow rate (X7). Three responses (number of identified compounds, Y1; area of identified compounds, Y2; and total area of chromatographic peaks, Y3) were assessed in a representative pooled sample of lavender essential oils. Furthermore, sample volume and sample dilution ratio were studied to achieve efficient peak shapes, peak resolution and to avoid detector saturation. Data were processed by (i) MSWS Software (Bruker) and NIST 17 library and (ii) GAVIN software (MATLAB 2019b) after data treatment with AMDIS. ANOVA results showed that equilibration temperature, dwell time and injection flow rate were the most statistically significant factors affecting chromatographic efficiency and were further tested for a more detailed optimization. With the developed method we were able to identify more than 40 volatile compounds in a representative pooled sample containing equal volumes of all 12 lavender essential oils.

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P004. LC-MS analyses & biological properties of three Boraginaceae plants from Mediterranean area, *Cynoglottis barrelieri, Phyllocara aucheri* and *Symphytum anatolicum*.

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Boraginaceae is a widespread family including plants with pharmacological and medicinal value (*Borago*, Symphytum). It is also well-known for the biosynthesis of pyrrolizidine alkaloids (PAs) in all its plant species. PAs are notorious for their carcinogenic and hepatotoxic effects. In the framework of our research on this family (1, 2, 3) we report herein the survey of phytochemical composition and biological effects on three endemic plants from the Mediterranean area: Cynoglottis barrelieri, Phyllocara aucheri and Symphytum anatolicum, not previously studied. The methanolic extracts were subjected to qualitative LC-MS analysis, where nineteen phenolic metabolites have been identified as caffeic acid and its derivatives and flavonol glycosides. Moreover, six constituents have been also isolated and structurally elucidated as: caffeic acid, rosmarinic acid, isoquercitrin, rutin, astragalin and nicotiflorin. Next step in our phytochemical study was the PAs analyses through LC-MS, through which eight PAs in the form of bases and/or N-oxides (PANOs) have been identified, while two of them, heliotrine and lycopsamine N-oxide, have been also isolated. The total phenolic (TPC) and flavonoid contents (TFC) were determined in methanol extracts, which were further evaluated for their antioxidant activities by different methods, including free radical scavenging (DPPH, ABTS), reducing power (FRAP and CUPRAC), as well as metal chelating and phosphomolybdenum. Enzyme inhibitory activities were also analyzed on acetylcholinesterase (AChe), butyrylcholinesterase (BChe), α-amylase and α-glucosidase. The rich phytochemical profiles, as well as the good antioxidant (e.g 80.10- 151.97 mg TE/g extract for DPPH), neuroprotective (e.g 1.71- 2.24 mg GALAE/g extract for AChe) and potential antidiabetic (e.g 2.36- 3.91 mmol ACAE/g extract for α-glucosidase) activity of all three studied species suggest safe potential herbal sources for further phytotherapeutic applications, after the chemical elimination of their PAs content, according to European limits for safe use.

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P005. Determination of cannabidiol potency, compared to the labelled value, on commercially available hemp products.

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The use of cannabis for healing purposes has been mentioned since antiquity. The biological activity of the plant is coming from a class of compounds called cannabinoids. Cannabidiol is one of the main cannabinoids in which intense research for its properties has been conducted in the recent years. While more than 113 different cannabinoids can be isolated from C. sativa, the primary psychoactive compound is 9 D9-tetrahydrocannabinol (THC). Cannabidiol is another major cannabinoid and accounts for up to 40% of the plant extract. Due to its safety profile and lacking psychotropic action, the CBD is cannabinoid of interest, with many references to pharmacological effects in various pathological models, ranging from inflammatory and neurodegenerative diseases, such as epilepsy, multiple sclerosis, autoimmune diseases and cancer. Due to a recent reduce to the restrictions of the legislation, a wide range of hemp products are launched to the market. The aim of the present study was to assess the differences between the potency of CBD, indicated on the commercially available product and the real value, using an analytical methodology. The product samples were purchased from the Greek market and comply with the requirements of Greek law. We developed an analytical methodoly of analysis, by extracting the flowers and analyzing the extractions with HPLC analysis. We also analyzed the flowers using GemmaCert®, a novel machine for determining THC and CBD potency on flowers and grinds, using Near-infrared spectroscopy (NIRS). The results show a significant deviation between the labeled and the measured CBD potency, raising consumer protection concerns.

P006. Metabolomic analysis of Greek and Cretan sage populations and investigation of their anti-glycation activity.

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Greek sage, SF (*Salvia fruticosa* Mill.) and Cretan sage, SP (*Salvia pomifera* L.), have been used for culinary and therapeutical purposes for millennia. In this study, we analyzed metabolomically two populations of SF (Rodini Achaia, Fournoi Ikaria), two populations of SPsP (SP subsp. *pomifera*) (W Crete) and one population of SPsC (SP subsp. *calycina*) (Attica). Their dry leaves (2g) were subjected to ultrasound-assisted extraction using petroleum ether and subsequently 70% (aq.) methanol. The qualitative and % quantitative determination of the volatile constituents of the non-polar extract were carried out with GC-MS (gas chromatography-mass spectrometry), while the polar extracts were analyzed by UHPLC-DAD-MS (ultra-high performance liquid chromatography–diode array detector–tandem mass spectrometry). The extracts of SF populations had high concentrations of 1,8-cineole (34.8-39.0%) while SP ones had greater amount of a-thujone (19.7-41.0%) and b-thujone (6.0-39.1%) with distinct differences between the two subspecies. UHPLC-DAD-MS demonstrated the presence of a total of 86 compounds· 12 were present in all populations. Quantification of rosmarinic acid (RA) and carnosic acid (CA) was performed in methanolic extracts with external standards. CA was determined only in SF (21.2-39.7 mg/$g_{extract}$). RA concentration ranged from 36.4 to 142.1 mg/$g_{extract}$; the lowest values were noted in SPsP. Furthermore, we investigated their ability to inhibit the reaction of glycation of bovine serum albumin and ribose with a fluorometric method. All extracts showed inhibitory activity of at least 78% at 100 μg/mL concentration. In summary, we present the differences of secondary volatile and polar metabolites of Greek and Cretan sage populations with a novel methodology and illustrate the promising anti-glycation activities of the methanolic extracts.

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P007. Essential oil of different populations of *Salvia officinalis* from Pindus mountain, Greece.

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Salvia officinalis L., also called common sage, has been known for its use over the centuries in cookery and in traditional medicine; principally as an anti-inflammatory, gastroprotective and antispasmodic agent. Its' essential oil is of known commercial value and numerous distinct chemotypes have been described. In this study, 49 individuals from five different populations of the North Pindus mountains were sampled in August 2017. The essential oil of the leaves of each individual was isolated by hydrodistillation using a Clevenger-type apparatus. The distillation yield did not present significant difference among populations (range: 1.95-2.44 mL / 100g of dry leaves), but it showed a high variation among individuals (1.09-3.58 mL / 100g of dry leaves). The volatile chemical components were identified and semi-quantified by gas chromatography coupled with mass spectrometry. Sixty-eight components were identified; the main of them being camphor (24,31-30,95%), *cis*-thujone (8.17-22.86%), 1,8-cineole (6.21-10.16%), camphene (4.27-7.96%) and *α*-humulene (3.32-5.22%). Statistical analysis with Kruskal-Wallis test, by non-parametric ANOVA, demonstrated that *cis*-thujone was significantly ($p<0.05$) lower in the population of Konitsa (8.17%), which on the contrary had the highest camphene participation (7.96%). Significant differences among populations were also found in other important components like borneol, bornyl acetate, *β*-caryophyllene and manool. Even though the four examined wild populations belonged to the camphor / *cis*-thujone chemotype and the one to the camphor chemotype, the analysis of their individuals demonstrated a very high variation, even within populations.

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P008. Highly glycosylated crocins in styles of wild spring-flowering crocuses from Greece.

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The genus *Crocus* L. (Iridaceae) consists of 160 taxa (sp and subsp) worldwide and among them 14 taxa are endemic of Greece. Depending on the time of the year in which they bloom and the climatic conditions of the area, *Crocus* species are separated in autumn and spring flowering taxa. In the current study, styles of *Crocus olivieri* J. Gay (from Panachaiko mountain, Achaia, Greece), *Crocus nivalis* Bory and Chaub (from Panachaiko mountain, Achaia, Greece) and *Crocus veluchensis* Herb (from Veluchi, Euritania, Greece) were analyzed by LC-ESI-MS and RP-HPLC. The major constituents were quantified by the method of external standard and compared to those of *Crocus sativus* L. (from Central Macedonia) which is the only *Crocus* species that is being cultivated. The extraction of the dried styles was performed with MeOH: water (1:1, v/v) at room temperature for 24h, in the absence of light. For the quantification of the major compounds of the four different *Crocus* species, *trans*-crocin 6, *trans*-crocin 4, *trans*-crocin 3, *trans*-crocin 3', *trans*-crocin 2 and kaempferol 3-sophoroside were isolated from *C. sativus* and *C. nivalis* extracts by semi-preparative HPLC and identified by MS/MS spectroscopy. Calibration curves were made for all the compounds. Highly glycosylated crocins in wild species were identified. *Trans*-crocin 6 was the major compound in *C. nivalis* and *C. veluchensis* at an average concentration of 88 mg/g style and 17 mg/g styles, respectively. In *C. olivieri,* the major constituent was kaempferol 3-sophoroside at an average concentration of 19 mg/g style and it was also the major flavonoid at *C. nivalis* with an average concentration of 12 mg/g style. We herein present the chemical analysis of styles of three spring-flowering crocuses for the first time.

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P009. NMR metabolic profiling of *Pistacia lentiscus* L. leaves and fruit extracts from the Greek island of Chios for the identification of biomarkers with skin beneficial effects.

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Herbal preparations of *Pistacia lentiscus* L. shrub (mainly the resinous exudate) have traditionally been used in the Mediterranean basin for their significant healing properties for more than 2500 years. The famous mastic gum from the Greek island of Chios is a Protected Designation of Origin natural product with well-known antimicrobial, anti-inflammatory, antiatherogenic, and other activities [1]. However, the fruit and leaves of *P. lentiscus* have not been thoroughly investigated and additionally, they are discarded as considered by-products of the mastic tree cultivation. In continuation of our research [2], this study emphasises on the exploration of the skin beneficial effects of fruit and leaves extracts and the comparison of the phytochemical profile of samples collected from the region of Mastichochoria and Leonidio in SE Peloponnese. From both regions, leaves (60 samples) and fruit (114 samples) were collected from different altitudes and distance from the sea during the maturing period. Samples were extracted with methanol-*d*4 and their NMR metabolic profiling spectra were acquired in a Bruker Avance III 600 MHz magnet. In parallel, their enzyme inhibition properties related to skin whitening (tyrosinase) and antiaging (e. g. elastase, collagenase) were evaluated. Finally, NMR data were introduced to SIMCA and Multivariate Statistical Analysis (MVSA) was used to evaluate the quality characteristics of the samples and facilitated their classification. In parallel, *HetCA* approach was implemented in MATLAB in order to directly identify potential biomarkers linked to their enzyme inhibition activities. Both approaches resulted in the detection of spectral features (SIMCA) and the structure elucidation of secondary metabolites (*HetCA*) in all of the extracts prior to their isolation.

To the best of our knowledge, this investigation is the first to study and compare the NMR-based chemical profiling of *P. lentiscus* fruit and leaves from two regions, Chios and Leonidio, and correlate their enzyme inhibition activity by MVSA.

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P010. The "superstitious" *Mandragora autumnalis* (Solanaceae) roots, leaves and fruits after a phytochemical investigation and extended LC-HRMS & HRMS/MS characterization.

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From ancient Greece, to medieval northern Europe, the potent pharmaceutical activity of *Mandragora* spp. (Bertol, Solanaceae) was exploited by several civilizations, due to its therapeutic, recreational, and even lethal impact. Although this forgotten genus holds a significant ethnopharmacological background, a comprehensive description of its phytochemical composition is deficient, focused mainly on the detection of tropane alkaloids and identification of volatile compounds occurring in its fruits [1,2].

Therefore, the purpose of the present research is the thorough phytochemical investigation and the isolation of secondary metabolites from *Mandragora autumnalis*, collected in Mykonos (Greece), via state-of-the-art scientific techniques. Root, leaf and fruit extracts of sequential polarities subjected to qualitative control through chromatographic and spectrometric analytical methods (TLC, HPLC-DAD, UHPLC-ESI-HRMS/MS). The methanolic extracts of roots and leaves were subjected to preliminary fractionation employing FCPC while the received fractions were evaluated qualitatively aiming towards a targeted isolation and structure elucidation (1D&2D NMR) of representative secondary metabolites.

The phytochemical investigation of the methanolic extracts of all plant parts through LC-HRMS/MS-based dereplication techniques revealed a plethora of secondary metabolites belonging to glycoalkaloids, flavonoids, phenylamides and withanosides which are detected for the first time on genus *Mandragora*. Overall, during the present study customized analytical and preparative protocols were developed for all plant parts of *Mandragora autumnalis.* This approach led to the targeted isolation of secondary metabolites, also detected for the first time in *Mandragora* genus and could contribute to a further interpretation of its ethnopharmacological uses and probably to new pharmacological targets.

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P011. Phytoindustry sourcing in nature – comparative study of the control system in two European countries.

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According to the latest data, the majority of medicinal and aromatic plants (MAPs) traded on European phytopharmaceutical market, has wild origin. Having in mind the loss of biodiversity and ecosystem degradation we claim that policy regulating wild collection is crucial to balance conservation needs and sustainable use of MAPs. To understand the situation, we compare policy regimes regulating the wild collection of MAPs in two leading exporter countries in the EU - Germany and Poland. The analysis shows mixed results. There is a variety of governing bodies granting permits and a number of unharmonised regulations on different levels in both countries.

In Germany, commercial wild collections are regulated by the Federal Nature Conservation Act - for every form of commercial wild collection, a permit is needed. In almost all German federal states these permits are granted by the Lower-Nature-Conservation-Agency, with one exception of control on a higher level. Permits set quantity restrictions and impact regulations. These regulations differ greatly among federal states. Similarly, no central entity is responsible for granting collection permits in Poland. Various authorities control wild collection. Collection of several groups of plants (e. g. forest species, protected species, and plants gathered from protected areas) requires a permit and is regulated by separate nature conservation acts. Nevertheless, a significant portion of collection remains unmonitored.

In Poland, that leads to insufficient monitoring of the MAPs collection and lack of information about the real conditions of populations. In Germany, on the other hand, lack of unified countrywide decision-process for permit granting undermines the competitive potential of single federal states as MAPs sourcing areas. To conclude, development and implementation of a coherent and consistent countrywide policy regime, including unified regulations, restrictions, and decision processes regarding commercial wild collection is needed to balance conservation and sustainable use of the wild plants for phytopharmaceutical production.

Posters

Aromatic and medicinal plants in animal health ethnopharmacological approach

20th International Congress of the International Society for Ethnopharmacology

P012. The effect of ethanolic and water propolis extracts to inhibit DPPH free radical and hydrogen peroxide.

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The various chemical compounds and their biological activities are determined to the source of apiculture and plant species such as Populus nigra. Propolis extract consisted of polyphenols and flavonoids which are well known active compounds as well as their biological activities such as the antioxidant, protective role in cardiovascular diseases or other chronic diseases. The objective of this study is to investigate the chemical substances of the ethanolic extract and water extract propolis by using HPLC. Samples were vigorously shaken in 70 % w/v ethanol and distilled water after that crude extract was evaporated until dryness. Wax was removed and crude extracts were dissolved. The antioxidant activity was determined by using the spectrophotometric method with DPPH scavenging assay and hydrogen peroxide inhibition assay. The result showed that ethanolic extract propolis was significantly higher DPPH scavenging than water extract propolis. The highest percent inhibition of EEP and WEP was about 84.56 and 49.23, respectively. In hydrogen peroxide inhibition assay, EEP and WEP completely inhibited hydrogen peroxide which affects cellular toxicity. Therefore, propolis from Thailand is the new source of polyphenols that scavenge free radicals for further research and application.

P013. An in vitro investigation into the potential benefits of *Psychotria zombamontana* as a poultry feed additive.

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The poultry industry forms the largest individual agricultural industry in South Africa. Microorganisms present in poultry feed include fungi and bacteria which may have detrimental effects on the feed or the birds consuming it. With the shift away from antibiotic feed additives and growing interest in alternative means of protecting feed from spoilage, biologically active plants may be an effective substitute. *Psychotria zombamontana* (Rubiaceae) was investigated as a possible plant-based poultry feed additive with dual benefits in protecting feed from spoilage and poultry from disease. The leaf acetone extracts of this rapidly growing shrub or small tree had promising antifungal activity in preliminary studies against *Fusarium* species.

In this investigation, leaf acetone extracts were tested against field isolates and reference strains of various fungi and bacteria implicated in poultry feed contamination and chicken infections. A rapid broth microdilution method was used to elucidate minimum inhibitory concentrations (MIC) of the extracts. Antioxidant and anti-inflammatory efficacy in terms of free radical scavenging and lipoxygenase inhibition were also determined, as was cytotoxicity against normal mammalian cells.

The crude extract had reasonable activity against various fungal and bacterial organisms including *Aspergillus flavus, Fusarium verticilloides*, *Salmonella* Enteritidis and *Salmonella* Gallinarum. The crude extract also had a high phenolic content corresponding with high antioxidant activity, as well as good anti-inflammatory activity and relatively low cytotoxicity. These are all beneficial attributes in protecting poultry against infection as well as in protecting the feed against fungal infestation. Fractionation of the extract and analysis of the most active fractions using mass spectrometry identified predominantly long chain fatty acids which are likely to be responsible for the activity, although further work is needed for confirmation of the identity of bioactive compounds in *P. zombamontana*.

P014. Antimicrobial activity and selectivity indices of acetone leaf extracts of South African *Eugenia* and *Syzygium* (Myrtaceae) species against *Staphylococcus aureus* isolated from mastitic cattle.

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Mastitis poses significant economic and welfare challenges to the livestock dairy industry globally. *Staphylococcus* species constitute the most abundant bacteria causing mastitis. Antimicrobial resistance and selective pressure for resistance development by mastitic bacteria treated with existing antibiotics complicates the control of mastitis, necessitating the search for novel effective and safe antibacterial compounds. Plants are putative sources of novel anti-staphylococcal compounds. South Africa is a hotspot for diverse useful medicinal plants. Here, the antibacterial activity and cytotoxicity of acetone crude leaf extracts of seven *Eugenia* and *Syzygium* (Myrtaceae) species native to South Africa were determined against six clinical strains of *Staphylococcus aureus* isolated from milk of cattle with subclinical mastitis, and a reference *S. aureus* strain.

A microplate serial dilution method was used to determine the minimum inhibitory concentration (MIC) of the extracts. Cytotoxicity was determined against C3A liver cells using the 3-(4,5-dimethythiazolyl-2)-2,5-diphenyltetrazolium bromide reduction (MTT) assay. The concentration causing 50% inhibition of cell viability (LC50) was calculated. Total antibacterial activity (TAA = extract yield/MIC) and selectivity index (SI = LC50/MIC) values were obtained.

The mean MIC values of the extracts ranged from 39-182 µg/mL with *Syzygium legatii* being the best (39 µg/ml) compared to gentamicin (2.23 µg/ml). All extracts had relatively low cytotoxicity (LC50 > 20 µg/ml) compared to doxorubicin (2 µg/mL). *Syzygium gerrardii* had the best SI of 6 suggesting a higher toxicity to bacteria than to mammalian cells. *S. gerrardii* also had the best mean TAA of 2912, meaning that a 2912 times dilution of the amount extracted from 1 g of the extract can inhibit bacterial growth.

These plants may be potential candidates to investigate for presence of novel anti-staphylococcal compounds to combat mastitis caused by *S. aureus*. Alternatively, a topical antibacterial herbal product may be produced from these plants.

P015. Effect of Greek aromatic plants as feed additives on laying hens' performance, and oxidative stability of egg yolk and albumen.

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An important recent area of research is the use of medicinal and aromatic plants (MARPs) in farm animal diets, aiming to increase the productive parameters and to improve the animal products quality. The objective of this experimental trial was to evaluate the effects of the dietary supplementation of a mixture of Greek dried aromatic – medicinal plants (oregano, thyme, Greek mountain tea and chamomile) on laying hens performance, egg quality and egg oxidative stability. Oregano and Greek mountain tea clones were collected from the natural environment and clonal propagation by cuttings were performed. Thyme and chamomile plants were seed propagated and all plant material was field cultivated. 432 Lohmann laying hens, 24-week-old, were randomly allocated into two dietary treatments (6 pens of 36 birds per treatment). The hens of the Control group were fed a commercial maize-soybean diet, whereas the hens of the MARPs group received the control diet further supplemented (5 g/kg) with a mixture of the examined aromatic plants. Following 6 weeks feeding, eggs were collected and analysed. Egg production and feed conversion ratio were not significantly affected. The eggs of the MARPs group showed higher ($P<0.05$) eggshell thickness and maximum load strength, although the other examined parameters (egg weight, yolk, weight, egg white weight, yolk color) were not significantly affected. Yolk resistance to oxidation under refrigerated storage (up to 60 days) was improved ($P<0.05$), as well as yolk resistance to iron induced oxidation ($P<0.05$). On the other hand, albumen carbonyl levels were not modified.

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P016. Comparison of dietary protected vs non-protected essential oil extracts on broiler chickens' performance and meat chemical composition.

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Presently, aromatic plants and their essential oils are used in the production of feed additives for various farm animal species. It is possible that the protection of these active substances, in the form of coating or encapsulation can modify or enhance their activity and their metabolism in the animal gut. Accordingly, in a 35 days trial a total of 576 male one-day-old chicks (Ross 308) were used randomly allocated into 3 groups (12 pens of 16 chickens per group). The control group was fed commercial maize and soybean meal diets. The chickens of the Arom and ProtArom groups were fed the same diets, which were further supplemented with either a mixture of unprotected essential oils (50 g/ton) or protected essential oils (40 g/ton), respectively. The main active ingredient of the mixture was carbacrol. Feed and water were provided ad libitum and conventional housing conditions were kept throughout the trial. The body weight of the chickens was recorded on days 10, 24, and 35, whereas feed consumption and mortality recorded daily. At the end of the trial, meat samples were collected and analyzed by the FoodScan apparatus. Analysis of variance (ANOVA) was used for the statistical analysis. The main findings were that the ProtArom group had higher ($P<0.001$) final body weight, better body weight gain ($P<0.001$), reduced food intake ($P<0.05$), and better food conversion index ($P<0.001$), compared to the other two groups. In addition, it was noted that the carcass protein was higher ($P<0.001$) and fat was lower ($P<0.01$) in both supplemented groups, compared to the control group. Consequently, the use of aromatic essential oils in a protected form seems to work better than the unprotected form in terms of performance increase in broiler chickens. Furthermore, feed supplementation with this essential oil mixture can modify the meat chemical composition.

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P017. Effects of different dietary plant extracts on production parameters and meat chemical composition of broiler chickens.

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Aromatic and medicinal plants contain valuable bioactive substances that can be fed to animals and benefit their health and production. In this experimental trial, three different dietary feed additives containing extract of aromatic and medicinal plants were evaluated in broiler chicken nutrition. In a 35-day trial, total of 768 male one-day-old broiler chickens were randomly allocated to 4 groups (12 subgroups of 16 chickens / group). Control group (A) was fed with commercial maize-soybean meal diets. The other three groups were fed with the same diets, further supplemented with: (B) 50 g/ton of a feed additive containing carvacrol and thymol; (C) 100 g/ton of a feed additive containing carvacrol and cinnamaldehyde; (D) 150 g/ton of a feed additive containing estragol, carvacrol and p-cymene. Feed and water were offered *ad libitum* and conventional housing conditions were kept throughout the trial. The body weight of the chickens was recorded on days 10, 24 and 35, while feed consumption and mortality were recorded daily. At the end of the trial all birds were slaughtered and samples were collected for meat chemical analysis. The statistical analysis (ANOVA) showed that: Groups B and C had higher body weight ($P \leq 0.001$) and weight gain ($P < 0.001$), as well as the best feed conversion ratio ($P \leq 0.001$), compared to the control and group D. In addition, group D performed better than the control group A, regarding final body weight ($P \leq 0.001$), weight gain ($P \leq 0.001$) and feed conversion ratio ($P \leq 0.001$). Differences were found on the meat chemical composition: Group D had lower whole carcass meat fat ($P \leq 0.001$) compared to groups A and C, while all three supplemented groups presented higher protein percentage ($P \leq 0.001$) on meat compared to the control group A. It can be concluded that the examined feed additives improved the performance and affected the chemical composition of broiler chickens to a different degree.

Acknowledgments: This research has been co-financed by Greece and the European Union (European Regional Development Fund) in context "Research–Create–Innovate" within the Operational Program (Competitiveness, Entrepreneurship and Innovation (ΕΠΑΝΕΚ) of the NSRF 2014-2020. Project Code: Τ1ΕΔΚ-03856. Acronym "GREEN POULTRY MEAT ANTIFREE".

P018. Dietary flaxseed and lupin supplementation improve fatty acid profile of milk in dairy cows.

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Dairy products (cheese, yoghurt, etc.) are an important part of the human diet worldwide, with a steadily rising production and consumption. A new strategy in the creation of novel dairy products is the enrichment of the milk nutritional profile through the dietary manipulation of the cow. Such examples of enrichment include the modification of the fatty acid profile (ratio of unsaturated to saturated fatty acids). It is possible that the enriched milk and dairy products can help in the prevention of chronic diseases such as cardiovascular disease and cancer. Today, many plants and their extracts, and by-products are examined in ruminant nutrition, such as flaxseed (*Linum usitatissimum*), cannabis (*Cannabis sativa*) and lupines (Lupinus albus). Flaxseed can be fed to cows, although its content of anti-nutritional factors, such as cyanogenic glycosides are toxic and in increased levels can be lethal for animals. Fatty acids account for 55% of the total fat content of the seed and are mainly represented by α-linolenic which corresponds to 18% of the total seed weight. Available data concerning dairy cows indicate that the addition of 0.9-1 kg of flaxseed per cow per day resulted in an increase in α-linolenic from 0.44 to 0.78 % whereas an 8% content per kilo of dry food substance increased the content of polyunsaturated fatty acids from 33.5 to 36.5 g/kg. Another new feed ingredient is the by-product of industrial cannabis (*Cannabis sativa*) that is rich in polyunsaturated fatty acids. Cannabis oil is mostly (84%) comprised of polyunsaturated omega-3 and omega-6 fatty acids (56% linolenic, 22% α-linolenic, 4% γ-linolenic and 2% stearidonate). There is yet a small number of reports on the change in the fatty acid profile of milk in animals fed with 5-10% of cannabis in the mix of concentrated feed. Moreover, a significant source of polyunsaturated fatty acids is the white lupine (*Lupinus albus*) that contains on average 8.5% oil mainly with C18-ln9 (4.36%) and C18-2n6 (1.45%) fatty acids. An important consideration in the use of sources of unsaturated fatty acid in cow diets is that large amounts of the dietary unsaturated fatty acids are hydrogenized by the rumen bacteria to saturated fatty acids which limits the effects of the dietary modifications. For this reason, feed processing methods have been proposed to protect against the oxidative action of rumen microorganisms, such as formaldehyde treatment, micronization and heating.

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P019. Dietary flaxseed and lupin supplementation alter fatty acid composition of ovine milk and Kefalograviera type cheese from sheep flocks under semi extensive Mediterranean farming system, in the area of Vonitsa, Amphilochia, Western Greece.

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In the current study, feed supplementation with flaxseed and lupin was evaluated regarding some milk quality characteristics, as well as chemical composition and fatty acid profile of Kefalograviera hard cheese originating from 20 different semi-extensive sheep farms in the area of Vonitsa, Amphilochia, Western Greece. For each farm, the diet (roughages and concentrates) of animals in the second month of milking period, was analyzed for moisture, crude protein, ether extract, crude fiber, neutral detergent fiber, starch and ash, showing that the total daily feed contained on average 17% (±1%) crude protein and 3% (±0.5%) fat. Fatty acid profile of these diets was also examined, revealing that among the most abundant fatty acids were the linoleic (C18:2), oleic (C18:1) and palmitic acid (C16:0). Further, milk samples were collected from each farm and analyzed for their chemical composition (fat, protein, lactose and total solids content) and fatty acid profile. The milk contained on average 5.9% (±0.7%) protein and 7.4% (±1.2) fat, whereas the main fatty acids detected were the palmitic (C16:0), oleic (C18:1) and stearic acid (C18:0). In addition, the analysis of the Kefalograviera hard cheese samples showed that it contained on average 26.5% (±2.4%) protein and 31.5% (± 3.1) fat and its main fatty acids were the palmitic (C16:0), myristic (C14:0) and oleic acid (C18:1). To conclude, dietary supplementation with flaxseed and lupins in sheep may improve milk production and omega-3 fatty acid composition of both milk and Kefalograviera cheese.

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P020. Production of an innovative functional product based on traditional vegetables and medicinal-aromatic plants stabilized with novel technology

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The modern trend is to consume healthy food products with functional characteristics. Co-creation plays an important role for novel products' development. Rrepresentatives of the food industry, chefs and restaurant owners, food researchers, nutritionists, conservationists and people involved with health and quality control of food products co-created a novel soup based on various vegetables and medicinal-aromatic plants (MAPs), enriched with aromatic oils, extra virgin olive oil, extractions of herbal teas and others (>30 traditional Greek food ingredients).To increase the product's functionality, encapsulated probiotics were added for stability throughout processing and storage time. High-pressure (HP) technology was used to cold-pasteurize the final product, preserving its functional-nutritional characteristics.

The product formulation was based on the high nutritional value of the ingredients used and their superior organoleptic characteristics compared to conventional products readily available. Lactic acid bacteria were used for adding functionality to the final product. The bacteria were encapsulated in a solution of sodium alginate and pectin to maintain their viability after pasteurization. The final product-soup was pasteurized using thermal (75-90°C for 5-20min) or High-Pressure technology (HP at 400-600 MPa, 25°C, for 5-15min). The results showed that encapsulated bacteria could not survive from thermal treatment, while HP treatment maintained 30-80% of their viability, depending on the conditions used.

For the finally treated product at 400MP-25°C-5min, a shelf-life test was performed at storage temperatures 0-15°C, measuring the dominant deterioration factors. The optimum shelf-life was estimated to 2 months after storage at 4°C, with 65% retention of bacteria viability (total bacteria population>10^6CFU/g) and >90% retention of ingredients' nutritional value.

The results indicated that a superior functional vegetable- and MAP-based product can be produced using appropriate ingredients and novel technology for stabilization as an alternative to thermal treatment.

Posters

Aromatic and medicinal plants of the Mediterranean basin as food and dietary supplements

20th International Congress of the International Society for Ethnopharmacology

P021. Comparison of total phenolic content of Cretan wild edible herbs (horta).

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The Greek island of Crete is well known for its traditional cuisine, which is depending exclusively on the local agricultural products and is dating all the way back to the ancient times. Medical studies revealed that Cretan diet, milestone of the Mediterranean diet, contributes to the highest longevity and low mortality rate of the Cretans. High consumption of olive oil, vegetables, herbs, fruits, fish, wine are the main principles of this diet. Moreover, wild edible green herbs, which grow in all regions of Crete, are an integral part of the traditional gastronomy. They are mainly consumed boiled or stir fried but also raw in salads. Nowadays, there is a growing interest on these plants, mainly due to their high nutritional value and their potential therapeutic properties (Cámara *et al.*, 2016; Gonçalves *et al.*, 2018).

In the present work, eight wildly grown edible green herbs, so called "horta", were collected from East Crete: aerial parts of *Dioscorea communis* (L.) Caddick & Wilkin, *Echium italicum* L., *Erodium cicutarium* (L.) L'Hér, *Glebionis segetum* (L.) Fourr., *Plantago lagopus* L., *Verbena officinalis* L., aerial parts and roots of *Scolymus hispanicus* L. and bulbs of *Muscari spreitzenhoferi* (Heldr.) H.R. Wehrh.

Different extracts of all eight studied plants were evaluated for their total phenolic content (TPC) using the Folin-Ciocalteu method. The aqueous and methanol extracts of *Erodium cicutarium* (201 mg GAE/g and 149 mg GAE/g) and *Verbena officinalis* (136 mg GAE/g, 140 mg GAE/g) showed the highest TPC among the tested samples, confirming their valuable nutritional importance in Crete, while their phenolic chemical profiles, are also in accordance to the expressed TPC as rich sources in flavonoids (flavonol and flavon derivatives) and phenolic acids (rosmarinic, chlorogenic, p-coumaric acids).

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P022. The seeds of Greek cultivar *Cynara cardunculus* hybrid, as a bioproduct of potential high nutritional value.

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Cynara cardunculus L.- artichoke, is a perennial plant of the Mediterranean basin, known since antiquity as food and for its leaves highly appreciated therapeutic properties (EMA, 2009). In our days, the plant has been widely used as a bioenergy crop. The shifting from conventional crops to artichoke cultivation has brought economical and environmental advantage according to the literature (Danalatos, 2008).

In this particular study, the seeds of a certain established Greek cultivar of *C. cardunculus* hybrid, that has been cultivated in the experimental field and used as biofuel, have been investigated. The main purpose of our study was to evaluate the chemical profile, the total phenolic content and the nutritional value characterization of its seeds, not previously studied and till now just wasted as a bioproduct. The non-polar cyclohexanic extract was saponificated, esterificated and subjected to GC/MS analyses, where to our results appeared as a rich source in ω-6 and other fatty acids (linoleic, palmitic, oleic and arachidonic acid). Several secondary metabolites have been further isolated and structurally elucidated by NMR from the methanol extract (arctiin, cynarinine, tracheloside, caffeoyl derivatives) and the dichloromethane one (arctigenin, trachylogenin, fatty acids and aryl ester derivatives). Moreover, the methanolic extract displayed the highest content of total phenolic content (142.83 mg GAE/g) followed by aqueous (88.87 mg GAE/g) and dichloromethane extract (64.32 mg GAE/g). Furthermore, the raw seeds have been shown remarkably high nutritional value (fiber, protein, vitamins, minerals, antioxidants etc) compared to the famous for their nutritional value sesame- and flax-seeds.

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P023. The adventurous trip of *Origanum vulgare* smells along a transect from its southernmost (Crete) to the northernmost (Norway) native range in Europe.

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Origanum vulgare L. is found as a native plant almost all over Europe. It is a very popular herb widely used in food flavoring, tea infusions, and dietary supplements. It is also used as an ornament in house gardens. Several therapeutic properties are attributed to the plants of that species, mainly related to the variety of their organoleptically distinct smells [1]. Through a review of scientific publications [2][3] of the last 26 years, we attempt to find out the *O. vulgare* "smell identity" along a transect of Europe, from the southernmost (island of Crete) to the northernmost part of its native range (Norway). Published information on essential oils (EO) responsible for the plant smells, were recorded in respect to their total content (intensity of smell) and their main constituents (type of smell). Plants of *O. vulgare* are highly variable in respect to their EO content, with the lowest value recorded in Bulgaria (Rhodopi Mountains, 0,03 mL 100g-1 dry weight) and the highest on the island of Crete, Greece (8,8%). The different types of plant smells are related to the dominant compounds of their total EO composition and can be described under the following four groups: (i) Plants with an oregano smell (EOs rich in carvacrol, up to 93,8% in Greece), (ii) thyme smell (rich in thymol up to 90,2%, in Greece), (iii) lavender smell (rich in linalool up to 57,9% in Austria), and (iv) marjoram smell (characterized by sabinene and isomers of sabinene hydrate, up to 28,7% in Norway). The results so far indicate that along their adventurous trip of approx. 3200Km, the native plants of the species *O. vulgare* give arise to four commercially valuable chemical "species" distinct by their smell profile.

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[2] Kokkini, S.; Karousou, R.; Hanlidou, E.; Lanaras, T. (2004) J Essent Oil Res 16:334-338
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P024. Herbal teas in Thessaloniki: From the old tradition to the modern lifestyle.

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The use of herbs in Greece has its roots in ancient times [1]. Herbal teas, particularly infusions, with both medicinal and nutritional value have a long history of traditional use among cultures and regions of Greece. Until today, several aromatic and medicinal plants are consumed because of their flavor and a long-time traditional knowledge for their therapeutic activities [2][3]. In the frame of a wider project about the most common herbs and spices used in Thessaloniki (North Greece), the plant species used for the preparation of herbal teas have been explored. Thessaloniki is a multi-cultural city populated, besides Greeks, by several ethnic communities throughout its history, with a long tradition in the herbal trade [1]. In this study, three central open markets were visited and citizens from different ages and origins were interviewed about their customs and preferences in flavors of teas, using a questionnaire. The results so far indicate that a high number of taxa is used for herbal teas and infusions, plain or mixed, most of them native in Greece and Mediterranean area. The best represented families are Asteraceae (chiefly represented by the genus *Matricaria*), Lamiaceae (mainly taxa of the genera *Mentha*, *Origanum, Thymus* and *Sideritis*), and Tiliaceae (species of the genus *Tilia*). According to citizens, the majority of the herbs are collected from the wild of the surroundings of the city areas. Besides there is a remarkable number of plant taxa are imported from the Old and New World countries. Finally, it appears that the preference of citizens is largely based on their traditional knowledge and they choose their herbal tea according to their origin, while their age is a decisive factor to whether the herbs are collected from the wild or purchased from herbal markets.

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Acknowledgements: This research has been co-financed by the European Union and Greek national funds through the Operational Program Competitiveness, Entrepreneurship and Innovation, under the call RESEARCH–CREATE–INNOVATE (project code: T1EDK-04174).

P025. Taxonomic identification and essential oils of spearmint (ηδύοσμος) plants used in the traditional cooking of North Greece.

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Among the different herbs and spices used in the traditional cooking of North Greece, spearmint leaves are the most popular. They are widely known since antiquity with the Greek name "ηδύοσμος" (derived from the two words ηδύς+οσμή = sweet odor), named to date by local people and sold in different Greek market places by the short name "δυόσμος". Fresh or dried leaves obtained from small scale cultivations (home gardens or even pots in apartment balconies) are used in a wide variety of cooking recipes, from the preparation of legume soups up to cereal and/or meat based foods. Besides, it is also used as a traditional medicine for the preparation of teas against stomach pain. The taxonomic identification of the fresh or dried leaves is very difficult to succeed with the formal identification keys which required the occurrence of inflorescence, flowers, seeds and the presence of non-glandular hairs on the leaves. On the basis of plant habit, leaf shape and their characteristic sweet odor we conclude that they are members of the genus *Mentha* and in particular that they belong to the Section *Spicatae* (included plants with a spike-like inflorescence). Research published in the last 40 years suggest that only the members of that Section may produce essential oils rich in carvone, dihydrocarvone and their relative alcohols and esters responsible for the sweet odor of plants, commercially known as spearmint. Taking into account the wild growing *Mentha* taxa in North Greece area and their surrounding countries wherefrom the cultivated glabrous "diosmos" plants are apparently selected, it is suggested that they belong to a particular chemotype of *Mentha longifolia* or *M. spicata* or the sterile hybrid between them, *M.* x *villoso-nervata*.

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P026. Chemical profiling of *Trigonella foenum* graecum extracts from different cultural practices, using NMR and multivariate data analyses.

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Trigonella foenum graecum (fenugreek), is a common medicinal plant in many traditional healing systems (e.g. Ayurvedic and Chinese medicine). It is used in the form of infusions, water and alcohol extracts or tinctures, for general health improvement exerting hypoglycemic, antihyperlipidemic, antidepressant, psychotonic properties etc. [1]. Fenugreek seeds and leaves are rich in bioactive substances and secondary metabolites such as saponins, flavonoids and phenolic compounds, whereas their production is strongly depended on agricultural factors. Since it is a plant of considerable value for its nutritive composition and medicinal effects, our aim was to investigate whether different composting methods [2] could affect the yield of fenugreek crop and the metabolic profile of final products. For this reason, a field experiment was conducted for the cultivation of *T. foenum* graecum, using 4 fertilization treatments (compost, manure, NPK basic fertilization, biocyclic), as well as control with three repetitions. The duration of the experiment was 3 months and the effect on the growth, yield and quality of fenugreek crop was evaluated. Samples of the aerial parts (leaves and shoots) and the seeds of the plant were collected in the middle and in the end of the growing season. All plant material was pulverized; the aerial parts were extracted with a mixture of MeOH/H2O (50/50) and the seeds with n-Hexane and MeOH consecutively. 1D NMR spectra of the polar extracts were recorded and the samples were classified using multivariate statistical analysis (PCA and PLS-DA). Their chemical profile was further investigated with HPTLC and the n-Hexane seed extracts were subjected to GC-MS analysis. The obtained results revealed a distinct separation of the samples from different composting methods, and secondary metabolites responsible for groups clustering were identified. Hopefully these findings will contribute to reveal the best cultural practices of fenugreek, based on the chemical content.

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P027. Essential oil analysis and study of the antioxidant activity of cultivated oregano.

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The aim of this work was to study the various data concerning the growth of cultivated plants of *Origanum vulgare* L. ssp. *hirtum* (Link) Letsw (Lamiaceae). The study was focused mainly on the qualitative and quantitative characteristics of the essential oil obtained by distillation of the aerial parts and whether these characteristics are affected by the soil composition, the time of collection of the plant material and the application of two foliar fertilizer formulations to plants. The experimental plan was based on a latin square 3x3 and the statistical analysis of data was done using the ANOVA method with the SPSS 15.0 program. The volatile components were obtained by hydrodistillation and the quality and quantity control took place in a gas chromatography mass spectrometry (GC-MS). Finally, the antioxidant activity of the cultivated oregano was studied via the interaction of the samples with the stable free radical 1,1- diphenyl- 2- picrylhydrazyl (DPPH). The results of the study show that only the time of collection and not the application of various formulations to plants was statistically significant. As far as the antioxidant activity is concerned, some of the essential oils did not exhibit high radical-scavenging properties as shown in the DPPH assay, especially those of fresh plant material and thus further research is necessary.

P028. Essential oil analyses of the wild-growing *Micromeria hispida, Teucrium alpestre* and *Thymbra calostachya* (Lamiaceae), three local endemic plants of Crete, Greece.

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Micromeria hispida, Teucrium alpestre and *Thymbra calostachya* are Greek endemic plants that belong to the Lamiaceae family. They are range-restricted local endemics which are confined to parts of the island of Crete [1]. The rock-dwelling *M. hispida* is fairly rare in intermediate altitudes in Central and East Crete, *T. alpestre* is found in a variety of habitats but mainly in rocky mountainous areas in South and Central-East Crete, and *T. calostachya* is a lowland rock-dweller in East Crete [2]. Wild-growing material (dried leaves and/or inflorescences) were collected from the wild during different seasons, i.e. summer for *T. alpestre* (initiation of flowering); autumn (after the end of flowering) and spring (before the initiation of flowering) for *T. calostachya*; spring (before flowering), summer (full flowering), autumn and winter (in partial flowering) for *M. hispida*. The essential oils were isolated from each of the wild-growing species by hydrodistillation. The quality and quantity control of the obtained essential oils was performed with Gas Chromatography Mass Spectrometry (GC-MS). The essential oils of *M. hispida* were consisted mainly of borneol, caryophyllene oxide, hinesol and carvacrol. Those of *T. alpestre* mainly contained hinesol and carvacrol, while the essential oils of *T. calostachya* were characterized by γ-terpinene, p-cymene, thymol acetate and carvacrol. Only traces of essential oils were obtained for *T. alpestre*. Quantitative differences were acquired among the studied essential oils in different seasons and the yield for *T. calostachya* was comparatively higher in autumn (10.95%), while that for *M. hispida* was higher during spring before flowering (1.56%). The essential oil yield of *T. calostachya* was much higher in comparison to those of *M. hispida* and *T. alpestre* (7-fold to 11-fold, respectively).

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[2] Strid A., Englerra 33 part 2, 2016.

P029. Antioxidant activity of ethanol extracts of *Rosa pendulina* L. (Rosaceae) from Serbia.

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Rosa species are popular plants used for cosmetic preparations and to treat a variety of ordinary diseases. Teas prepared from rose hips are effective in infectious and urinary diseases, rheumatism and arthritis. This study provided information about the antioxidant activity and total phenolic content (TPC) of rose extracts. The extracts were obtained from the leaves, fruits, and nuts of *Rosa pendulina*, collected from Kopaonik Mt., Serbia. The total phenolic content and antioxidant potential for the studied 70% ethanol extracts were achieved by spectrophotometric measurements. Antioxidant activity was evaluated by DPPH, ABTS and β-carotene assays. The highest phenolic content was determined in leaves extract (280.24 mg GAE/g of dry extract), while extracts of fruits and nuts showed lower TPC (236.35 mg GAE/g and 232.15 mg GAE/g of dry extract, respectively). All tested extracts showed high antioxidant capacity. Among the tested samples the highest antioxidant activity in DPPH, ABTS and β-carotene tests was found for leaves extract (IC_{50} 0.091 mg/ml, IC_{50} 0.131 mg/ml and IC_{50} 0.848 mg/ml, respectively). The obtained activities could be attributed to the presence of high TPC. The present results are in the range of BHA (IC_{50} 0.090 mg/ml, IC_{50} 0.101 mg/ml and IC_{50} 0.017 mg/ml, respectively) and indicate the high antioxidant potential of *R. pendulina* extracts.

The study confirms the use of wild roses in the ethnobotany and ethnomedicine. Also, *R. pendulina* extracts present a rich source of natural antioxidant compounds.

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P030. Exploration of the biodiversity for the development of and sustainable use of Greek genotypes of *Rosa canina* L., rich in ascorbic acid.

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The aim of the BIOFARM project is the evaluation of Greek native aromatic and pharmaceutical plants, for the selection of genotypes rich in active molecules. In this project, *Rosa canina* L. (Rosaceae) were selected in order to identify genotypes with a high concentration in ascorbic acid. Furthermore, the development of a quick and selective assay for its determination, was another aim of this work. *Rosa canina* pseudo fruits are particularly rich in ascorbic acid- even more than citrus fruits- and therefore, a good natural source of vitamin C [1]. Moreover, rosehips also have a high content of polyphenols, which contribute to the fruits' antioxidant activity [2]. In the present study, ripe rosehips growing wild in Northern (Rhodopi Mt., Thrace and Chalkidiki, Central Macedonia) and Western (Pindos Mt., Epirus), Greece, were collected at full ripeness, during autumn. Ascorbic acid content was determined spectrophotometrically, by means of the ascorbate oxidase redox assay, which was optimized specifically for tissue's characteristics [3]. The Folin-Ciocalteu method was used for the evaluation of the total phenolic content and the radical scavenging activity was assessed by means of the DDPH assay. Our results corroborate the existing bibliographic data, proving the antioxidant capacity of the rosehip extracts. Additionally, a comparison between the three different regions, showed that the genotypes collected from Chalkidiki had a higher average content of ascorbic acid and better radical scavenging activity.

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P031. Rock samphire responds to increased salinity levels with a differentiated profile of essential oil composition when grown in an aquaponics system.

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Rock samphire (*Crithmum maritimum,* Apiaceae family) is an edible, aromatic, rock-dwelling halophyte of Mediterranean coastal areas which is considered as a functional food mainly due to its high content in bioactive compounds. In the present study we aimed at evaluating for the first time the performance of rock samphire in terms of physiology, biomass production and essential oil yield when grown at different salinity levels in co-cultivation with fish, i.e. a brackish aquaponics system (modern production method combining aquaculture and hydroponics in a recycled system which exploits fish waste for plant nutrition). Nine independent re-circulating aquaponic systems with rock samphire and sea bream (*Sparus aurata*) were used comprising three salinity treatments, i.e. 14dS/m, 23dS/m and 32dS/m. *In vivo* chlorophyll *a* fluorescence and photosynthetic pigments concentration were monitored during the two months growth period. At the final harvest, several plant growth parameters were measured and the essential oils were obtained from the aerial parts by hydrodistillation and identified by GC-MS analyses. Fluorescence parameters indicated moderate stress of the photosynthetic apparatus of *C. maritimum* in high salinity individuals compared to control plants (14dS/m). The final harvest revealed significantly stunted plant growth in plants grown in high salinities compared to control plants (80% and 60% reduction of the aerial part's fresh weight under 32dS/m and 23ds/m, respectively). Essential oil yield was highest in the plants of the 32dS/m treatment. Concerning the main compounds' relative content, limonene responded with a gradual increase to increasing salinity levels, while the opposite trend was detected for thymol methyl ether and 1-terpinen-4-ol. On the contrary, *β*-phellandrene and *γ*-terpinene exhibited the highest relative content in the intermediate salinity level. In conclusion, when grown in an aquaponics system with no external inputs and increased salinity, rock samphire experiences considerable growth inhibition but responds interestingly with a differentiated essential oil profile.

P032. Study of biological activity of beverages derived from aromatic plants of Greek flora.

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To determine the active ingredients and antioxidant capacity of beverages from five aromatic plants of Greek flora, namely: Calendula (*Calendula officinalis*), Louisa (*Aloysia citrodora*), Lemon balm (*Melissa officinalis*), Achillea (*Achillea millefolium*), and Althaea (*Althaea officinalis*). Beverages of the samples were prepared by mixing the dried herb with water, the mixture was left to boil for 5 minutes and then allowed to cool in room temperature, the water phase was collected and subsequently freeze-dried. The obtained extracts were examined with respect to their phenolic content (Folin-Ciocalteu), antioxidant capacity (DPPH), OH- radical scavenging ability, the ability to inhibit linoleic acid peroxidation (LLA) induced by AAPH and the inhibition of plant-derived lipoxygenases (LOX). Finally, chromatographic analysis (UPLC-Q-TOF-MS) was performed. This study showed a correlation between increased phenolic content and antioxidant capacity for Louisa and Calendula (Phenolic content: 81.49mg/l of extract, 77.59mg/l of extract respectively, DPPH radical scavenging activity: 70.7% and 50.5%, respectively), whereas the strongest correlation was observed in Lemon balm (Phenolic Content: 270.65mg/l and DPPH Interaction: 95.4%) whose extract showed strong interaction with DPPH radical (63.1%) even in the diluted (1:10) sample. Moderate OH- radical scavenging capacity (50% and 57.5%) was observed in the extracts of Althaea and Achillea respectively. These two extracts along with Lemon balm, showed the highest linoleic acid (LLA) and lipoxygenase (LOX) inhibitory capacity. In the LLA assay the highest capacity was shown by Lemon balm (92.6%), followed by Achillea (81.3%) and Althaea (80.8%), whereas for LOX assay values were 87.4% for Lemon balm, 38.1% for Althaea and 39.9% for Achillea respectively. Finally, more than 20 compounds were tentatively identified in the samples after chromatographic analysis. Overall, this study highlighted the antioxidant capacity of these commonly consumed beverages. Lemon balm presented the best antioxidant profile of all the samples tested. Achillea and Louisa showed close results in most experiments, but the latter had no effect on neutralizing OH- radicals. Finally, Calendula, which had a moderate phenolic content, also had minimal effect on OH- radicals and little ability to inhibit the action of LLA and LOX. These variations may be due to the composition of the samples to phenolic or other constituents, the study of which is also part of our ongoing research.

P033. Antioxidant evaluation of Virgin Olive Oil and Green Olive Oil of *"Makris"* and Oregano oil with Oregano from Samothrace and Alexandroupolis.

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To evaluate Virgin Olive Oil and Green Olive Oil of the local variety named *"Makris' olive"* (Makri, Alexandroupolis, Greece) with respect to their antioxidant properties alone and as a mixture with Oregano from Samothrace and Alexandroupolis. Two types of Olive Oil locally produced in Makri Alexandroupolis, (Virgin Olive Oil (VOOL) and Green Olive Oil (GOO)) and two types of Oregano (Oregano from Samothrace (OS) and Oregano from Alexandroupolis (OA)) were evaluated on their antioxidant and antimicrobial properties. Initially, OS, OA and dried olive leaves (OL) were investigated as aqueous extracts (AEs). Respectively, VOOL and GOO were also investigated. Additionally, four mixtures of Oregano with Olive Oil were produced (OS+VOOL, OS+GOO, OA+VOOL and OA+GOO). The mixtures were examined in 5 different times (after maceration for 24h, 72h, 10d, 20d and 30d). Assays performed included: Evaluation of total phenolic content (Folin-Ciocalteu), antioxidant capacity (DPPH and ABTS assays), antimicrobial activity and the volatile composition of the samples (GC/MS). The AEs of OS, OA and OL resulted in low antioxidant profile, although positive results were observed in phenolic content. GOO resulted in slightly lower antioxidant capacity and phenolic content than VOOL. Regarding the Oregano and Olive Oil mixtures, the combination OS/VOOL had overall higher results on antioxidant profile and antimicrobial activity. Finally, maceration time had an effect on the assays performed, however, even in the shortest maceration time examined (24h) all mixtures presented DPPH radical scavenging activity higher than 65% during the second measurement (60min). The nutritional value of olive oil and oregano is well established in the framework of Mediterranean Diet, however, the characteristics of the varieties proposed in this work as well as the combination there of has not yet been thoroughly investigated. Further analysis will provide a better understanding of the potential use of these natural products and is a part of this on-going research.

Posters

Conservation and sustainable use of herbal resources

20th International Congress of the International Society for Ethnopharmacology

P034. Preserving mature *Pyrus spinosa* Forskk. plant material through an efficient micropropagation protocol.

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Pyrus spinosa Forskk. is a native tree of Greek forests, rich in antioxidants. An effective micropropagation protocol was developed in order to preserve and maintain mature elite trees of high antioxidant content for future pharmaceutical use. Winter branches of different *Pyrus spinosa* mature trees (> 15-year-old) were selected from the area of Agia Anastasia, Chalkidiki, Greece. Plant material was maintained in a growth chamber under controlled environmental conditions (16h photoperiod, temperature 24±1°C and light intensity 35 μmol $m^{-2}s^{-1}$) and newly emerged shoots (2-3cm) were used as explants. In the multiplication stage, among the different nutrient media tested, namely: modified MS (thiamine-HCl 1 mg/l^{-1}, nicotinic acid 1 mg/l^{-1}, pyridoxine-HCl 1 mg/l^{-1}), WPM, DKW, Pear Medium 1 and 2, supplemented with 5 μM BA, Pear Medium 1 proved the most efficient. Subsequently, of the four BA variants (0, 5, 10, 20 μM) assessed in combination with Pear Medium 1, 5 μM BA was the best for shoot production (22.7±1.6/explant) and elongation (2.4±0.1 cm). Rooting evaluation included a modified rooting MS medium (½ NH_4NO_3, ½ KNO_3) and Pear Medium 1 combined with 20"-quick-dip in different IBA concentrations (0, 2.46, 4.90, 24.6, 49.0 μM). Modified rooting MS and Pear Medium 1 in combination with 49.0 μM IBA were equally effective for rooting: (79%±7.2) and (80%±7.0), respectively. A well-formed rooting system in terms of root number (2.7±0.3) and length (3.9±0.3 cm) without the presence of callus was developed on modified rooting MS, whereas Pear Medium 1 was less effective for the same parameters, (root number: 2.4±0.2 and length: 1.8±0.2 cm). Acclimatization of plantlets was successfully conducted (87.5%) on perlite under the controlled conditions of the plant growth chamber, five weeks after their establishment.

P035. Effects of nitrogen fertilization on growth and yield of milk thistle (*Silybum marianum* (L.) Gaertn).

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Milk thistle (*Silybum marianum* (L.) Gaertn.) is an annual or biennial plant that is classified as weed although, this plant can be an alternative crop since the seeds contain silymarin which has important pharmaceutical properties. A field experiment was conducted for the growing period of October 2018 through June 2019, following a randomized complete block design with three treatments and three replicates. The wild population "Mesopotamia", originated from Central Greece, were used for crop establishment. Treatments were i) control-N_0 (0 kg ha^{-1} N), ii) nitrogen fertilizer A rate-N_{75} (75 kg ha^{-1} N), and iii) nitrogen fertilizer B rate-N_{125} (125 kg ha^{-1} N), while the inorganic fertilizer was applied at two doses in late January and in early March. The aim of the study was to assess the impact of nitrogen fertilization on plant growth (rosette diameter, plant height, fresh and dry weight, chlorophyll content), silymarin and oil content, inflorescences per plant, 1000-seed weight, and seed yield. The determination of silymarin and oil content is in progress. The results showed that fresh and dry weight, height, rosette diameter, yield, and chlorophyll content were increased significantly by fertilization compared to control. Specifically, dry biomass was positively affected by both N_{75} and N_{125} fertilization rates and increased by 26.24% and 35.78% respectively. Moreover, the N_{125} fertilization rate led to the highest height (185.8 cm) and seed yield (1860.8 kg ha^{-1}). In addition, a positive correlation was observed between dry biomass and yield ($r=0.958$, $p<0.001$). To summarize, this study indicated that nitrogen fertilization could influence the growth of milk thistle, reaching greater biomass and seed yield.

P036. Development of a fast HPLC-PDA-MS protocol for the determination of carnosic acid in Greek genotypes of *Rosmarinus officinalis* L.

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In the framework of a national project aiming at exploring the biodiversity for the selection, development and sustainable use of Greek Medicinal and Aromatic plants, the species *Rosmarinus officinalis* was selected. The selection was done based on its high content of carnosic acid which is a recognized antioxidant with wide applications in the food and cosmetic industry [1,2]. The project includes collection of a large number of genotypes, genetic analysis by use of microsatellites and genotyping by sequencing, chemical analyses by HPLC-PDA-MS for the monitoring of production of carnosic acid. A fast analytical protocol was developed to ensure reliable quantification of the sensitive carnosic acid. In brief the fresh plant material is ground in liquid N2, to ensure the viability of both enzymes and carnosic acid. Optimization of the extraction showed that acetone 100% and 15 minutes of ultrasonication gave the best results. Under these conditions carnosic acid is quantitatively extracted without being degraded. A short HPLC-PDA-MS method based on a RP-C18 column was developed and optimized for the separation of carnosic acid from carnosol and its degradation products. The method was validated as required prior to metabolomic analysis and showed adequate precision (%RSD ranging from 0.20 to 4.03) and accuracy (less than 15% in three concentration levels). Quantitative results showed a great variance of the chemical content of carnosic acid among different genotypes.

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[2] Safety Assessment of Rosmarinus Officinalis (Rosemary)-Derived Ingredients as Used in Cosmetics. Cosmetic Ingredient Review, 2014. Available at: https://www.cir-safety.org/sites/default/files/rosmar032014TAR.pdf

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P037. Towards sustainable exploitation of Greek *Satureja pilosa* as a new industrial crop: Biotype selection, DNA-barcoding, clonal propagation, acclimatization and HPLC-PDA-MS studies.

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The genus *Satureja* (Lamiaceae) include more than 30 species and subspecies which are widely distributed in the Mediterranean [1] and 11 of these are found in Greece [2]. They are traditionally used for culinary purposes and as herbal teas for gastrointestinal disorders, for the treatment of cramps, muscle pain, and infectious diseases [3,4]. *S. pilosa* (range-restricted Balkan endemic savoury extending to Italy) with accession number GR-1-BBGK-04,2656-06 is a native Greek biotype selected under pilot *ex situ* cultivation for its ascending stems to be used potentially as new medicinal-aromatic crop and/or ornamental plant. Molecular barcoding was evaluated with the *18S-26S* (nuclear ribosomal DNA) and *atp*F/*atp*H, *mat*K, *rpo*C1, *pet*B/*pet*D, *rpl*16, *rpo*B, chloroplast DNA molecular markers. DNA sequences were analyzed, deposited in the GenBank and compared for genetic similarities with different *Satureja* taxa. New genetic information was generated based on new DNA sequences produced from the *pet*B/*pet*D, *rpo*B and *rpo*C1 molecular markers for the *S. pilosa* biotype. An *in vitro* clonal propagation protocol (shoot tips-nodes as initial explant material) has been developed. Axillary shoot proliferation was achieved on the AB50 basal medium supplemented with 0.4 ppm BA and 0.1 NAA. Shoots were rooted in a half strength MS medium supplemented with 4 ppm IBA. Acclimatization of the microplants was assessed: (a) *in vitro* (two systems), (b) *in vivo* (eight soil substrates, greenhouse and outdoors), and (c) in field (pilot cultivations in eight distant areas of northern Greece). An HPLC-PDA-MS method was developed for the qualitative/quantitative characterization of wild-growing *S. pilosa* aerial parts. Qualitative analysis showed the presence of depsides and flavonoids, mainly rosmarinic acid, eriodictyol-7-O-rutinoside and luteolin-7-O-rutinoside. Quantitative analysis demonstrated that *S. pilosa* is particularly rich in depsides (3.54 ± 0.10 mg%). The above are aimed to facilitate the sustainable exploitation of this potential new industrial crop in man-made environments.

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P038. *In vitro* study for micropropagation and production of secondary metabolites from callus of *Alkana tinctoria* (Boraginaceae).

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The roots of *Alkanna tinctoria* (Boraginaceae), contain large quantities of substances (alkannins and shikonins) with a wide spectrum of biological properties and excellent wound healing effect. Wild-growing plants were collected from Seich Sou forest, Thessaloniki, Greece for long-term *ex situ* conservation and sustainable exploitation. Shoot tips from the collected plants were disinfected and cultivated *in vitro*. The combined effect of three basal culture media (MS, WPM, Gamborg B5; supplemented with 1 μM BA, 0.1 μM NAA, 0.3 μM GA_3, 20 g/L sucrose) and two gelling agents (6 g/L Plant Agar, 3 g/L Gelrite) was studied for multiple shoot induction. MS medium solidified with Plant Agar proved to be the most effective exhibiting 100% shoot formation (4.7 shoots/explant, 1.82 cm height) but showed serious hyperhydricity problems. Furthermore, full and ½ strength MS in combination with IBA (0, 2.5, 5 μM) was tested for rooting *in vitro*. Full MS medium with 5 μM IBA resulted in 50% rooting. For callus induction, root segments were cultured in MS supplemented with 4.5 μM kinetin, 5.5 μM NAA, 30 g/L sucrose and 6 g/L Plant Agar. The calluses produced were re-subcultured in two different liquid media (MS + 2.5 μM kinetin + 34.5 μM IBA + 30 g/L sucrose and MS + 1 μM BA + 34.5 μM IBA + 30 g/L sucrose) shaken in a rotary shaker, in dark. The callus produced was dried and extracts were made using green extraction techniques. The chemical profiles of the callus extracts were compared with extracts from roots of wild-growing *A. tinctoria* plants from the same population and similarities occurred. Their methanolic extract appeared to contain phenolic compounds that resemble flavonoids and lignans (perhaps iridoids).

P039. EthnoHERBS: Conservation of European Biodiversity through Exploitation of Traditional Herbal Knowledge for the Development of Innovative Products.

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EthnoHERBS is the first systematic multidisciplinary attempt in South-Eastern (SE) Europe, promoting evidence that ethnobotanical information can lead to valuable drug discovery. EthnoHERBS is a H2020-MSCA-RISE project aiming to record and evaluate information on SE European traditional knowledge, explore in a high-throughput manner the biodiversity of Balkan Peninsula flora and elaborate cutting-edge technologies in Natural Products Chemistry to discover and develop innovative cosmeceutical products against skin disorders. A well-balanced multidisciplinary consortium has been established that consists of five academic and six non-academic beneficiaries from EU member states (Greece, Bulgaria, Italy, Portugal, Spain), two academic and one non-academic partners from Candidate Country (Serbia). EthnoHERBS' extensive network aims to the exchange of know-how and culture, the enrichment and broadening of scientific knowledge and enhancement of collaboration. EthnoHERBS aims to contribute to:

- Inventing and validating an integrated approach for the sustainable and effective exploitation of SE European traditional knowledge and biodiversity for the development of innovative products based on natural ingredients against skin disorders.
- Establishment of an extensive network of academic and private sector entities, exchange of know-how and culture, enrichment and broadening of scientific knowledge and enhancement of collaboration potentials.
- Widening the application of emerging techniques, novel approaches and advanced methodologies originally produced in the academic environment in order to be exploited by European biotech companies.
- Establishment of dynamic synergies between SMEs and academia through exploitation of complimentary expertise and mutual understanding in different environments and skills, stimulating entrepreneurship, creativity and innovation.
- Enhancement of the competitiveness of the European industries in the area of skin care and healing products and broadening the potentialities for the development of optimized products in favour of consumers and the society.
- Growth of the MAPs sector and strengthening the national economy of SE European countries by highlighting the unique biodiversity and the richness of Balkans' traditional medicine.

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P040. Antioxidant and antimicrobial activities and total phenolic contents of hydroalcoholic extracts from ten thyme-scented plants (*Thymus spp., Thymbra capitata*) under *ex-situ* conservation.

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Extracts and essential oils isolated both from wild and cultivated thyme-scented plants (*Thymus* spp., *Thymbra* spp.; Lamiaceae) can potentially be used in the pharmaceutical and food industries as natural preservatives due to their biological activities. The present study aimed to investigate in a comparative way the antioxidant and antimicrobial effects of hydroalcoholic extracts of 10 Greek native thyme-scented plants (species and subspecies), i.e. the Mediterranean *Thymbra capitata* and *T. longicaulis* subsp. *longicaulis,* the Balkan endemics *Thymus boissieri, T. degenii* and *T. thracicus*, the Balkan subendemics *T. sibthorpii, T. striatus* and *T. longicaulis* subsp. *chaubardii*, and the local Greek endemics *T. holosericeus* and *T. plasonii*. The aerial parts of the studied material were collected during early spring (March) at the Institute of Plant Breeding and Genetic Resources from *ex situ* cultivated mother plants of the 10 studied species and subspecies, all of which were asexually propagated from wild-growing individuals. The phytochemical profile of all the extracts was monitored using NMR spectroscopy and applying techniques for evaluation of total phenolic (TP) and total flavonoid (TF) contents which are in accordance with the results estimated using the Folin-Ciocalteu method (TP:181.06-225.18mg/g, TF:58.00-138.09mg/g of extracts). All extracts were tested for their antioxidant activity using two *in vitro* methods, i.e.DPPH and $ABTS^{\bullet+}$ radical scavenging assays. All the extracts showed good to moderate antioxidant activity, compared to the reference substance BHT. Moreover, their antibacterial activity was tested against some of the major food-borne pathogens (*Enterococcus faecalis*, *Bacillus subtilis*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella typhimurium*, *Escherichia coli* and *Klebsiella pneumoniae*). However, none of these extracts showed satisfactory antimicrobial activity against them.

P041. Isolation of non-volatile secondary metabolites from *Centaurea phyllocephala* Boiss.

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The *Centaurea* genus contains about 200-700 species, making it one of the biggest genera of the *Asteraceae* family. They are usually annual herbaceous plants or shrubs, rarely biennial and perennial, that thrive in the Mediterranean and southwest Asia, but can also be found in America or Africa. The *Centaurea* genus has a significant pharmaceutical value, as it affects many biological targets. Its plants have been used traditionally for their antioxidant, anti-inflammatory, antibacterial, antidiabetic, anti-dandruff, antirheumatic, anti-pyretic and hypotensive effects. *Centaurea phyllocephala* Boiss. is the main *Centaurea* species in Iraq and has been used traditionally as an antidiabetic. The main compounds that have been isolated from the *Centaurea* genus belong to four chemical groups: sesquiterpene lactones, lignans, flavonoids and alkaloids. In this research, we studied the aerial parts from *Centaurea phyllocephala* Boiss., which was collected from the outskirts of Peshawar, Pakistan in May 2004. Until this point, we have isolated secondary metabolites that belong to the groups of sesquiterpene lactones and flavonoids. For the isolation we used chromatographic methods, such as column chromatography and HPLC, and for the identification we used NMR spectrometry (1H, 13C, COSY, HMQC, HMBC).

P042. Phytochemical investigation of *Centaurea cyanoides* (Asteraceae) from Jordan.

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Centaurea genus includes herbaceous and mostly annual plants with several biological and pharmacological properties. They have been used many years in folk medicine as diuretic, anti-diarrheic, anti-inflammatory and emmenagogue. Many reports have mentioned the importance of *Centaurea* species in health level, mainly referring to species such as *Centaurea cyanus*, called also cornflower and many compounds have been isolated belonging to the groups of sesquiterpene lactones, lignans and flavonoids. *Centaurea* sp. is native to Europe, specifically in Mediterranean zones, in the area of Turkey and in the Middle East. *Centaurea cyanoides*, named also as Syrian cornflower is distributed in the Western part of Asia and shares common characteristics with the *C. cyanus* but it has not been further investigated. In this study *Centaurea cyanoides*, collected from Jordan, has been studied in order to isolate its non-volatile secondary metabolites. Through different chromatographic methods (CC, HPLC), phenolic acids and flavonoids like vanillic acid, and apigenin, as well as sesquiterpene lactones, have been isolated. Nuclear magnetic resonance spectrometry (^{1}H, ^{13}C, COSY, HMQC, HMBC) had been used for the identification of the structures.

P043. Flavonoids and phenolic acids from the aerial parts of *Alyssum alyssoides*.

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Most of *Alyssum* species of Brassicaceae family have been mainly studied for their contribution in ecology. In this study, *A. alyssoides* was examined for its chemical substitutes. The methanol extract of its aerial parts was fractionated with liquid-liquid extraction (distribution) with four different solvents of increasing polarity: diethyl ether, ethyl acetate, 1-butanol and water. The diethyl ether and ethyl acetate extracts were further studied for their chemical composition. So far, secondary metabolites which belong to phenolics were isolated by using several chromatographic methods (C.C. and HPLC) and were

identified by using spectroscopic methods (UV/Vis, NMR and MS): two phenolic acids: p-hydroxy-benzoic acid and 3-methoxy-4-hydroxy-benzoic acid (vanillic acid), and seven flavonoids, which are derivatives of flavonol: kaempferol 3-O-β-D-glucopyranoside (astragalin), kaempferol 3-O-(6″-α-L-rhamnopyranosyl)-β-D-glucopyranoside (nicotiflorin), quercetin 3-O-β-D-glucopyranoside (isoquercetin), quercetin 3-O-β-D-galactopyranoside (hyperoside), isorhamnetin-3-O-β-D-glucopyranoside, isoramnetin 3-O-β-D-galactopyranoside and isoramnetin 3-O-(6″-α-L-rhamnopyranosyl)-β-D-glucopyranoside (narcissin). The butanol extract was also examined for its chemical compounds. Till now, according to the NMR spectra and the TLC analysis, the main compounds of the butanol extract are phenolics.

P044. *Ex situ* conservation of *Sideritis raeseri* Boiss. & Heldr. subsp. *raeseri,* an important plant of high medicinal value: effect of 4 amino acids on *in vitro* propagation.

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Sideritis raeseri Boiss & Heldr. subsp. *raeseri* is a range restricted medicinal plant of the Balkan peninsula. Aerial flowering parts of *Sideritis* plants are widely used in the Mediterranean folk medicine as a popular tea because of its anti-inflammatory, carminative, analgesic, antitussive, stomachic, and antimicrobial properties. Conventional propagation methods (seeds, cuttings) did not allow the mass production of plant material in a short period of time. The aim was to establish an efficient protocol for mass and large-scale micropropagation, germplasm and *ex situ* conservation of the studied species. Amino acids provide a readily available nitrogen in tissue cultures, and their uptake can occur more rapidly than from an inorganic nitrogen source in the same medium. Amino acids are involved in stress defense promoting cell division, expansion, differentiation, and plant development as natural precursors of endogenous auxins or ethylene. The effect of 4 amino acids (L-arginine, L-methionine, L-tryptophan, L-proline) in different concentrations was investigated. The culture medium used was the MS enriched with 30 g/l sucrose, 0.5 mg/l NAA or 3 mg/l IAA and 3 g/l Phytagel. L-arginine and L-methionine negatively affected shoot proliferation, i.e. shoot number (3.17) and multiplication (75%) were higher with NAA only (8 weeks), however 100 mg/l methionine stimulated shoot elongation (41.63 mm). NAA + 5 mg/l arginine or 100 mg/l methionine gave better rooting results i.e. 91.67%, 12.27 roots 18.75 long and 66.67%, 20.5 roots 21.1 mm long, respectfully. L-tryptophan (50 mg/l) + IAA gave 75% multiplication with 2.42 shoots/ explant whereas 10 mg/l tryptophan better promoted shoot elongation (28.33 mm) and rooting (50%, 5.33 roots 12.29 mm long) (5 ½ weeks). L-proline (500 mg/l) + NAA enhanced shoot elongation (24.67 mm) and rooting (80%, 14.25 roots 9.55 long) (6 ½ weeks). Amino acid type and concentration affect micropropagation of *S. raeseri* ssp. *raeseri*.

P045. Effect of 5 micronuntrients and 10 vitamins on *in vitro* propagation and *ex situ* conservation of medicinal *Capparis orientalis* Veill. from Kea Aegean island, Greece.

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Capers can be used in cooking because of its nutritional value, pharmacology and landscape architecture. *Capparis orientalis* Veill. is a native Mediterranean chamaephyte that grows on coastal habitats, cliffs, rocks, walls, ravines and boulders. Biological studies of capers have revealed important uses such as antimicrobial, antioxidant, anti-inflammatory, immunomodulatory, anti-diabetes, antifungal, anti-proliferative and HIV-1 reverse transcriptase inhibitory activities. Still, flowering buds have diuretic properties and are used against atherosclerosis, colds and atonia. Capers seeds contain ferulic and synaptic acid being of medical value, are rich in protein, oil, and fiber and could be an alternative source of edible proteins and oils. Difficulties including low seed germination and rooting rates of cuttings and existence of lignin can be overcome by the application of tissue culture, which can improve and shorten the mass production of capers. The disinfection protocol included the immersion of shoot-tip explants in a fungicide solution (25 min) followed by 70% ethanol (20 sec) and 2% NaOCl (5 min), giving 93.18% success. Initially, 2 culture media (MS, WPM) and elevated concentrations of 5 micronutrients ($ZnSO_4.7H_2O$, $MnSO_4H_2O$, $CuSO_4.5H_2O$, $CoCl_2.6H_2O$ and NaFeEDTA), applied with 0.3 mg/l BA, 0.03 mg/l IAA, 25 mg/l ascorbic acid, 25 mg/l citric acid and 7 g/l agar, were tested. The MS medium containing 300 μM $MnSO_4.7H_2O$, i.e. 3-fold the normal concentration of MS gave 100% multiplication with 17.08 shoots 29.33 mm long (40 days). Afterwards, the application of 10 vitamins (myo-inositol, nicotinic acid, pyridoxine-HCl, thiamin-HCl, riboflavin, biotin, pantothenic acid, ascorbic acid, α-tocopherol, folic acid) was studied. Shoot multiplication was highest (66.67%) by adding 2.5 mg/l α-tocopherol to MS medium containing 2 mg/l IAA and 100% rooting with 0.1 mg/l biotin (8 weeks). After 4 ½ weeks in the greenhouse mist during summer, 98.8% *ex vitro* survival rate was achieved for rooted microshoots on a peat: perlite substrate.

P046. Development and validation of an HPLC method for the analysis of *Primula veris* flowers from Greece.

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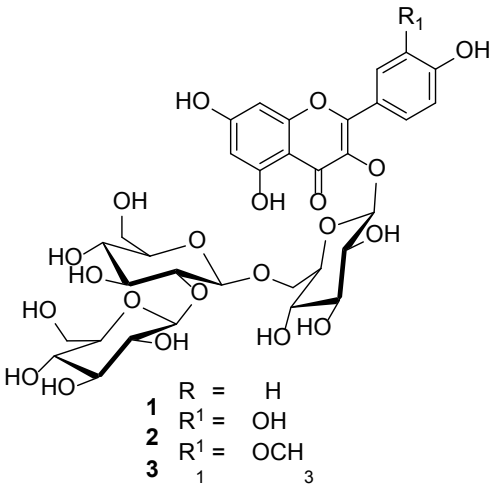
In the present study an HPLC-PDA method was developed for the determination of the flavonoids, in the flowers of *Primula veris* from Epirus, Greece. The aim was to investigate the chemical content of the over harvested *Primula veris* populations of Epirus, Greece and to develop and optimize an extraction protocol to allow fast, exhaustive and repeatable extraction. Qualitative analysis revealed that the Primula flowers from Epirus were particularly rich in flavonoids and especially in flavonol trisaccharides, derivatives of quercetin, isorhamnetin and kaempferol (1-3). Classical phytochemical analyses of the hydromethanolic (70%) extract afforded a new naturally occurring flavonoid, isorhamnetin-3-*O*-*β*-glucopyranosyl-(1→2)-*β*-glucopyranosyl-(1→6)-β-glucopyranoside (3), which is also the main constituent of the flower extracts. Its structure elucidation was carried out by means of 1D and 2D NMR and mass spectrometry analyses. An HPLC-UV method was developed and validated according to ICH guidelines. Since the main flavonols of the plant is not commercially available, rutin was used as secondary standard and the correction factor for response was determined. The HPLC method was validated for linearity, LOD, LOQ precision and accuracy in three concentration levels. R.S.D. values ranged between 0.18% and 2.67%, for the intraday variation, within the acceptable limits. The inter-day variation ranged between 0.22% and 3.37% R.S.D. Accuracy was between 96.8 and 104.4 with RSD values ranging between 0.62 and 4.20. Finally, the recovery of the extraction was proved to be over 99.5% after four extraction cycles, while the precision of the extraction expressed as RSD values varied between 1.58 and 4.66, within the acceptable limits. The developed assay was fast and simple and permitted the quality control of the herbal drug. The studied samples were particularly rich in flavonoids and the mean total content of flavonols ranged from 4.46-6.67 %mg, much higher than the 3% which is reported by the EMA [1]. Pharmacological tests on the characterized extracts are underway.



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Acknowledgment: This study is founded by the Region of Epirus

Posters

Ethnobotanical approach to drug discovery Theory, methodology and limitations

20th International Congress of the International Society for Ethnopharmacology

P047. First and quantitative ethnobotanical study of medicinal plants used by Mon people in Myanmar.

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Knowledge on medicinal plants of the earliest habitant in Myanmar, Mon people, is absent in current literature. This first ethnobotanical research is designed and implemented to fill in this gap. We aimed at: 1) documenting the diversity of medicinal plants uses by local people in Mon State and their knowledge on therapeutic usages; 2) quantitatively identifying important medicinal plant species to local people and to evaluate their status of sustainable use. Ethnobotanical survey and interviews were carried out in 10 villages in four townships of Mon State. Data was collected from interview of 166 informants through snowball sampling method. Therapeutic usages of medicinal plants were categorized into ICPC-2 standard. Quantitative indices including Use Reports, Use Values and Fidelity Level were computed using ethnobotanyR package. Voucher specimens were collected and identified by experts. In total, 158 medicinal plant species belonging to 64 families were recorded in Mon state. Thirty-seven species were newly recorded as medicinal plants in Myanmar comparing with previous literatures. These medicinal plants are applied to 79 therapy usages classified into 14 ICPC-2 diseases. Fabaceae was the most representative family and leaf is the mostly used part. Decoction and oral administration rank top in preparation and administration methods respectively. *Tinospora sinensis*, *Chromolaena odorata*, *Mimosa pudica*, *Tadehagi triquetrum*, *Alysicarpus vaginalis*, are the most important medicinal plant species with highest Use Reports and Use Values. *M. pudica* had a high-fidelity level of 91.43% for treating dysuria. Rich and diverse medicinal plants and their traditional knowledge are recorded for Mon people in Myanmar. List of medicinal plants in Myanmar can be renewed with 37 more species added. Researches on ethnopharmacology and sustainable use of important medicinal plants used by Mon people, especially for *Streptocaulon juventas*, *Tradescantia zebrina*, *Canscora lucidissima* are strongly suggested.

P048. Historical ethnobotany - some methodological considerations.

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Historical texts are increasingly scrutinized in ethnopharmacological studies as a reservoir of time-tested information that can either provide the basis for renewed uses of ancient medications or be used as leads for novel medicines developed on the basis of ancient uses as was the case with artemisinin. However promising such approach might seem, it is not free of problems, from correct identification of the natural substances referred to as materia medica to the medical conditions for which treatment materia medica is prescribed.

Based on relevant cases and recent research, this presentation will outline the problems raised by historical documentation and illustrate the results to be obtained from interdisciplinary research (from natural history, particularly botany, to (a)DNA). On this basis, it will suggest methodological guidelines that might provide the basis for further developments and the compilation of a textbook.

Considering that ancient documents represent the written record of ancient therapeutic practice, this presentation will analyze both their textual and inconographic information, that it will complement with archaeological material possibly identified though (a)DNA identification. It will consider the medical conditions mentioned in the texts in relation to the populations and their genetic profile, and will analyze the pharmaceutical techniques, including the instruments. Analysis will conclude with the examination of the expected results of therapies.

This presentation aims to illustrate the complex nature of the analysis of ancient therapeutic literature, which require multi-factorial analysis, trans-disciplinary in nature (including linguistic skills). Studied cases will provide clear keys to be possibly transferred in the future to other material and make it possible to gradually assemble a critical mass for the compilation of a reference guide in the field.

P049. Ethnopharmacological study of wild growing *Rosa* species on Vlasina plateau.

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The aim of this study was survey of wild growing *Rosa* species from Vlasina plateau (South-East Serbia) and their uses in traditional medicine by local people. This region is a landscape with exceptional features with a specific climate and diverse flora, on the slopes of which are three large settlements Vlasina Rid, Vlasina Okruglica and Vlasina Stojkovic with numerous quarters (mahalas). The collecting, processing and applying plants and plant-based products is present through generations in this region. Ethnobotanical research was done as a survey which included 104 interviewed people (58 women and 46 men) coming from 3 settlements with 45 small quarters. Semi-structured anonymous ethnobotanical interviews were used. Most respondents were between 50 and 80 years of age (70.18%). The fruits of roses are used primary for human nutrition (tea - 100%, marmalade - 67.30%, juice - 15.38%, wine - 0.96%). Also interesting is the fact that only one person uses a leaf of wild rose to make tea. It was determined that the most common conditions treated with these plants are colds (99.03%) and in nutrition (68.26%), against diarrhea (15.38%), dry cough (8.65%), stomachache and as a diuretic (2.88%).

The ethnopharmacological study showed the immense importance of medicinal and nutrition importance of species from *Rosa* genus in the daily life of a local community.

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Posters

Formulation of natural products and medicinal plants from traditional to modern phytotherapeutics

20th International Congress of the International Society for Ethnopharmacology

P050. Anti-inflammatory activity of *Ocimum gratissimum:* from traditional health practice to a modern phytotherapeutic gel formulation.

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Ocimum gratissimum, Lamiaceae commonly known as 'tchiayo' in Benin, has been reported in several ethnopharmacological surveys as a plant widely used in traditional medicine for a variety of ailments including inflammatory conditions and readily accessible to the communities for its medicinal and nutritional values [1]. Aim of the present study was to investigate the *in vitro* anti-inflammatory activity of *Ocimum gratissimum (OG)* in activated human monocyte-derived macrophages (THP-1). Furthermore, a formulation of a novel herbal anti-inflammatory gel containing *OG* extract was designed in order to develop a prototype for a potential modern phytotherapeutical product. The effect of the ethanolic *OG* leaves extract (90% V/V) collected in Cotonou, Benin was evaluated on LPS-induced cytokine (TNFα and CXCL13)-release from differentiated human macrophages (THP-1). Production of the pro-inflammatory mediators (TNFα and CXCL-13) was measured by ELISA and IC_{50} values were determined. Budesonide served as positive control and cytotoxicity controls were performed using MTT assay. The herbal gel formulation was designed by using carbopol 980, *OG* leaves extract (1%), baobab oil, shea butter, beeswax and distilled water. The skin pH (6.8-7) was maintained by drop wise addition of tri-ethanolamine. The physicochemical parameters of formulations (pH, homogeneity, viscosity, spreadability etc.) were determined. It was found that the production of TNFα (IC_{50}: 71.82 µg/mL) and CXCL-13 (IC_{50}: 8.71 µg/mL) was inhibited after treatment with *OG* extract. These effects were concentration-dependent and comparable to those of budesonide. Moreover, no alteration of cell viability was observed in the applied concentrations. The herbal gel prototype showed good homogeneity, good spreadability, good stability and no skin irritation. The results obtained reinforce the relevance of the traditional medicinal uses of *OG* and provides further information on its anti-inflammatory potential. Further studies will focus on the topical anti-inflammatory activity of the novel herbal gel formulation for the treatment of skin irritations.

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P051. Innovative electrospun PHB scaffolds as carriers for the wound healing agents Alkannins and Shikonins.

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Among the different materials used for biomedical applications and especially those intended for skin replacement, polyhydroxyalkanoates (PHAs) have attracted great interest in recent years mainly due to their non-toxic nature. One of the most common types belonging to PHA family is poly-3-hydroxybutyrate (PHB); a highly biocompatible, biodegradable and low-cost material that serves as an appealing candidate for various medical devices, such as tissue engineering scaffolds and wound dressings. The bioactive secondary metabolites Alkannins and Shikonins (A/S) are naturally occurring hydroxynaphthoquinones biosynthesized in the roots of several Boraginaceous plants. These molecules have been established in the contemporary pharmacopeia in Greece by our group as strong wound healing agents in several approved medicines (HELIXDERM®, Histoplastin Red®), while the first reports being traced back in the works of Hippocrates and Dioscorides. Since then a plethora of important biological properties have been well established, such as anticancer and regenerative ones. The aim of this work was to study for the first time the electrospinning of PHB fiber mats incorporated with a mixture of A/S pigments. Specifically, A/S-loaded and non-loaded electrospun PHB scaffolds were successfully fabricated using several polymer and drug concentrations. Scanning electron microscopy of the PHB fiber mats proved the formation of a uniform mesh with diameters ranging from 1.3-1.5 μm. The entrapment efficiency of A/S was confirmed by UV-Vis spectrophotometry and ranged between 52.6% and 92.6%, while the 72h *in vitro* release study of A/S in phosphate-buffered solution revealed that 82.6-91.5% of the entrapped drug released. Overall, these preliminary results seem promising, dictating the need for follow-up experiments aiming to develop an efficient drug release system for repairing and regenerating skin.

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P052. Natural deep eutectic solvents: an eco-friendly alternative for the extraction of naphthoquinones from *Alkanna tinctoria* roots.

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Plant derived natural products have an important role in pharmaceutical, cosmeceutical and food supplements industries. Their increasing demand is leading to the over-exploitation of plant resources and to the over-consumption of organic solvents, widely recognized to be of great environmental concern. For this reason, the design of green extraction methods of natural products is currently a key research topic. [1-3]

In order to address this issue, eco-friendly natural deep eutectic solvents (NADESs) were used instead of organic solvents for the extraction of naphthoquinones from the roots of *Alkanna tinctoria* Tausch, a plant from Boraginaceae family. More than sixty NADESs containing choline chloride-, and betaine-, as hydrogen bond acceptor combined with different hydrogen bond donors (sugars, organics acids) were investigated for their potential to extract this particular class of compounds. As a result of the statistical evaluation, the most relevant deep eutectic mixture with the highest extraction efficiency was found to be composed of levulinic acid and glucose (LeG). The optimum levulinic acid-glucose ratio and the water content were investigated *via* a single-factor experiment. With a ratio of 4:1 (w/w), and 30% of water (w/w), LeG had a high extraction yield to naphthoquinones. A further optimization step of extraction parameters was followed using response surface methodology (RSM), including the solid-to-solvent ratio, extraction temperature, extraction time as well as the application of solid phase extraction techniques for the recovery of the naphtoquinone fraction from the NADES extraction solution. Our results revealed the optimized LeG mixture as a valid green alternative for the extraction of naphthoquinones from *Alkanna tinctoria*.

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P053. Naphthoquinone rich extract of root bark of *Onosma echioides* upregulates the elastin and collagen gene in human dermal fibroblast and the formulation thereof heal burn wound in rats by upregulating VEGF gene.

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The objective of the study was to evaluate the effect of dye in expression of collagen and elastin gene in Human dermal fibroblasts and VEGF in healed burn wound model in wistar rats. The red dye was extracted from the dried root bark of *Onosma echioides* using petroleum ether (60-80°C). The marker compound 1,4-naphthoquinone dimer was isolated and the extract was standardized using HPTLC. Cytotoxicity of the extract was determined in Human Dermal Fibroblast (HDF) cell line through MTT assay. The effect of standardized extract on expression of elastin and collagen genes in HDF was evaluated at various concentrations using RT-PCR technique. The wound healing effect of formulation of the standardized extract was carried out in burn model in wistar rats. The healing potential was evaluated through expression of vascular endothelial growth factor (VEGF). Cytotoxic concentration for the pet. ether extract of *O.echioides* against HDF was not found as 50% inhibition (IC_{50}) was more than 400 ppm. Extract of *O.echioides* showed concentration dependent upregulation of collagen and elastin genes. Gene expression study of VEGF in formulation of *O. echioides* showed increased values as compared to placebo and control. The folklore uses of *Onosma echioides* for healing of wounds is justified through upregulation of collagen and elastin required for synthesis of collagen and elastin fibres in stage 3 of wound healing in HDF and upregulation of VEGF gene in burn wound created in rats.

P054. Evaluation of wound healing efficacy of phytosomal gel of *Onosma echioides* in wistar rats.

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Onosma echioides yields red colour dye, indicated presence of 1,4- naphthoquinones and is used for treatment of wounds in folklore medicine. However, the formulation of the same is not available. Hence the objective was to prepare standardized formulation and evaluate wound healing efficacy of petroleum ether extract of root bark of *Onosma echioides* in Wistar rats. Pet Ether extract of dried root bark of *O. echioides* was standardized by isolated naphthoquinone dimer using HPTLC. Phytosomes of the standardized extract were prepared by thin film hydration technique. The wound healing efficacy of the formulation was evaluated in rats by inflicting excision and incision wounds. The healing was evaluated by determination of breaking strength and tensile strength of healed skin for incision model and percentage wound contraction, hydroxyproline content, granulation tissue free radicals (lipid peroxidation), catalase in excision wound model. The formulation was applied to wounds topically.The formulation treated group showed a significant increase ($p<0.005$) in mean breaking strength when compared to control group. The phytosomal treated groups significantly ($p<0.001$) promoted wound contraction as compared to control group. Significant increase in hydroxyproline content ($p<0.0001$), collagen turnover ($p<0.0005$) and catalase activity ($p<0.0001$), as compared with control group. Granulation tissue free radical (Lipid peroxidase) decreased significantly in phytosomal treated group ($p<0.0006$) when compared with control group. Histopathological studies of the formulation treated groups also revealed the effectiveness in improved wound healing. Conclusion: The phytosomal gel of *O.echioides* effectively exhibited wound healing effect. Thus, proving the healing potential of novel drug delivery system with improved stability.

Posters

Greek Ethnopharmacology

20th International Congress of the International Society for Ethnopharmacology

P055. Phytochemical analysis of *Ganoderma lucidum* (FR.) KARST growing wild in Greece (Meteora and Lesvos island)-Biological activities.

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Ganoderma lucidum is a famous representative among pharmaceutical mushrooms, widely used in TCM (Traditional Chinese Medicine) for more than 2000 years (Wasser *et al.,* 2005). Its significant bioactivities such as anticancer, immunomodulatory and hepatoprotective, are attributed to a variety of active components, mainly polysaccharides and triterpenes (Bishop *et al.,* 2015; Wachtel-Galor *et al.,* 2011). The subject of our study was the chemical analysis of fresh and dried samples of *G. lucidum*, collected wild in Greece (Lesvos island, Meteora), to our knowledge for the first time. From the dichloromethane extract of fresh fruiting body, five (5) metabolites have been isolated (linoleic acid, 7,22-Ergosta-dien-3-one, 7,22-Ergosta-dien-3-ol, oleic acid methyl-ester, oleic acid triglyceride) while their profile on volatiles of fresh and dried material were studied through HS-SPME and GC-MS, detecting a variety of components (1-octen-3-ol, n-hexanal, 3-methylbutanal, 7,22-Ergostadien-one, sugars and phosphoric acid, to name just a few) The antioxidant activity of dichloromethane, ethanolic and water extracts of the fresh fruiting and dried bodies, evaluated through the determination of total phenolic composition (TPC), DPPH, ABTS, CUPRAC, FRAP, Metal chelating and Phosphomolibdenum method, as well as through a panel of enzymatic activities (AchE, BchE, Tyrosinase, Amylase, Glycosidase inhibition). Moreover, the nutritional composition of dried mushroom has been determined, leading to the fact that *G. lucidum* contains a high percentage of unsaturated fatty acids and proteins. Evaluating the results, it can be suggested that the rich phytochemical profile, as well as the high antioxidant and nutritional value of *G. lucidum* could support the use of Greek *Ganoderma* as a good source in cosmetics and food supplements comparable with literature from cultivated and wild mushrooms from Asia.

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P056. The sacred plants of Dionysus.

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In antique pottery pictures depicting the celebrations for Dionysus, apart from vine plants (*Vitis vinifera*) and flasks of wine, there are also ivy wreaths (*Hedera helix*) -including ivy berries- that adorned the heads of god and his worshipers, usually bacchae and satyrs. In addition, most figures hold long sticks, originating from the flowering stem of the plant giant fennel (*Ferula communis*), with a pine on top of them (*Pinus sylvestris*). Euripides, in the tragedy *Bacchae,* mentions smilax (*Smilax aspera*) as another plant of the decorative wreaths. Ivy and smilax are climbing evergreen shrubs, which have been considered as symbols of the power of Dionysus to revitalize nature every spring. Nevertheless, the furious rituals pertaining to these ceremonial orgies, with wild dances in the rhythm of drums and flutes, impose the question whether the faithful followers of Dionysus were using inebriating herbs together with wine. We have put ivy, smilax and giant fennel under bibliographical scrutiny, in order to trace any possible role in the formation of "medicinal" wines. Of these plants, only ivy seems to fulfill such criteria. In some preliminary experiments of our lab, a water extract of homogenized ivy berries was administered orally to rats, either alone or in combination with a single ethanol dose (2,5 mg/kg). The observed behavioral profile revealed that the animals were affected in terms of motor response, exploratory activity and anxiety state.

P057. Chemical analysis of essential oil and phenolic compounds isolated from ethyl acetate extract of Drama's Propolis (Macedonia, Greece).

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Propolis has been used in folk medicine since ancient times. Nowadays, almost all bees products, such as honey and propolis, use as "bio"-cosmetics and as functional foods. Meanwhile, commercial interest to propolis is growing continuously. There is not a high concentration of volatile compounds in propolis, although their aroma and versatile biological activity and characteristic pleasant aroma make them important. Chemical composition of propolis depends on the local flora, geographic location and the site of collection. Hydrodistillation of propolis collected in Drama (2014) afforded yellowish essential oils. GC-MS analysis indicated that the predominant components of this essential oil were: γ-cadinene (19.82%), α-eudesmol (12.03%), α-muurolene (9.25%), epi-α-muurolol (8.71%) and γ-muurolene (8.48%). Moreover, a phytochemical study of the propoli's ethyl acetate extract led to the isolation of five known natural products: two flavonoids: chrysin (1) and pinobanksin (2) and three esters of caffeic acids: caffeic acid phenethyl ester (3), caffeic acid benzyl ester (4) and cinnamylcaffaete (5). The chemical structures of the isolated compounds were established by 1D and 2D NMR analysis (^{1}H, ^{13}C, gDQCOSY, gHSQCAD, gHMBCAD), and through comparison with the literature.

Acknowledgements: The authors are grateful to Mr. PantelisVasilas who provides us with the sample of propolis.

P058. Evaluation of antioxidant and anti-inflammatory activities of Ranunculaceae plants and secondary metabolites from *Adonis cyllenea*.

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Several of the plants belonging to the Ranunculaceae family have been mainly studied due to their toxicity. In this study, the anti-inflammatory and antioxidant activities of extracts from different parts of Ranunculaceae's plants have been evaluated (*in vitro*): *Adonis cyllenea* Boiss. (aerial parts), *Anemone coronaria* L.(inflorescence), *Caltha palustris* L. (leaves and inflorescence), *Consolida ajacis* (L.) Schur (seeds) and *Helleborus odorus* Waldst. & Kit subsp. *cyclophyllus* (A. Braun) Maire & Petitm (aerial parts and roots).

The polar extracts of the aerial parts of *A. cyllenea* were fractionated using solvents of increasing polarity and have been purified. So far, secondary metabolites which belong to tritrpenoids (fukujusone, digitoxigenin, strophanthidine and cymarine) and a polyol (adonitol) have been isolated, by using several chromatographic techniques (C.C. and HPLC) and have been identified by using spectroscopic methods (NMR and MS). This is the first research that concerns the phytochemical composition of this endemic Greek plant.

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P059. NMR-based metabolomic analysis of four *Siederitis* spp. (Lamiaceae) hydroalcoholic extracts and their antioxidant and anti-inflammatory activities.

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Perennial *Sideritis* species have been widely used in traditional medicine since ancient times for phytotherapy. This genus includes more than 150 species worldwide, among which 16 different species and subspecies are native to Greece (including 10 perennials and 5 local Greek endemics) [1]. *Sideritis* spp. are of great interest due to their pharmacological properties, such as the antioxidant and anti-inflammatory activity [3]. According to previous phytochemical studies, *Sideritis* spp. are rich in secondary metabolites, such as flavonoids, iridoids, phenylethanoid glucosides and diterpenes [2,3]. In this study, we collected the aerial parts of four species and/or subspecies from different areas of Greece: *Sideritis euboea*, *S. clandestina* subsp. *peloponnesiaca* and *S. raeseri* subsp. *attica* (local Greek endemics) and *S. scardica* (near threatened local Balkan endemic). The phytochemical profile of hydroalcoholic extracts was monitored using NMR spectroscopy. According to the spectra obtained, the main compounds of the examined extracts were phenolics (flavonoids and phenylethanoid glucoside). All the extracts were also tested for their biological activity using three *in vitro* techniques: the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity, the inhibition of lipid peroxidation (AAPH), testing both the antioxidant activity, and the inhibition of lipoxygenase (LOX), testing the anti-inflammatory activity. All the extracts showed strong antioxidant activity (>74.8%) with the DPPH method, compared to the reference substance NDGA. Moreover, there was not observed a reduction on the levels through the 60 min of this method. The extracts with the lipid peroxidation method and the inhibition of lipoxygenase showed no or low activity.

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P060. Rapid separation of manoyl oxide and 13-epi-manoyl oxide from *Cistus creticus* leaves and their seasonal variability.

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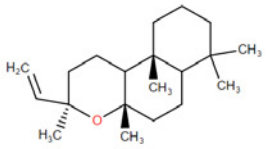
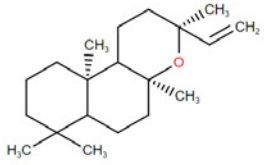
13-epi-Manoyl oxide (1) and Manoyl oxide (2) are high value diterpenoids with a broad range of pharmaceutical applications. They are naturally found in many plants. Because of their complex molecular structure, (1) and (2) are not commercially produced by chemical synthesis. In this study we present a fast and efficient method of isolation of these two components.



13-epi-manoyl oxide (1)



manoyl oxide (2)

13-epi-Manoyl oxide (1) and Manoyl oxide (2) are major components of the essential oil collected from *Cistus creticus* subsp. *creticus* (Viv.) Greuter & Burdet, (Cistaceae). Moreover, the essential oil from the plant's resin, known as "Ladano", which is collected by a traditional method, has a remarkable abundance of (1) and (2).

Leave samples from *Cistus creticus* were collected monthly from February to July 2018. The dried leaves were hydrodistilled in a Clevenger apparatus. The first fraction of the essential oil, collected after one hour, contained most of the volatile ingredients. The distillation was continued for 24 hours to yield the rest of components.

To our surprise, the essential oil of the February sample yielded (1) with purity above 99% and was completely void of (2) and any other volatile substances. The same procedure was followed for the samples of the following months, and a mixture containing only (1) and (2) was isolated. Over time, Manoyl oxide increased by up to 40% of the total mixture.

Due to the structural similarity of the two molecules it was not possible to separate them by any of the common analytical techniques. Separation was finally achieved by column chromatography using a mixture of neutral alumina-silver nitrate as filler material. All compounds were identified by 1- and 2D NMR, GC/MS, and optical rotation measurements.

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P061. Isolation and identification of components from the plant *Hypericum empetrifolium*.

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The species *Hypericum empetrifolium* (Clusiaceae) grows mostly in Turkey and Greece. It is used in traditional medicine by the locals as a therapeutic agent for wound healing, antioxidant and against psoriasis. Aerial parts of the plant were collected during its blooming period, dried and extracted in a Soxhlet apparatus via the sequential use of increasing polarity: hexane, dichloromethane and methanol. The extracts were subjected to further extractions and column chromatography. Since the methanol extract contains the greater range of components used as therapeutic agents, the methanol extract residue was re-extracted with petroleum ether, ethyl acetate and n-butanol. Analysis of the polar extracts with NMR and ESI-MS techniques led to the identification of shikimic acid as well as a number of phenolic acids, including three "chlorogenic acid" stereoisomers (5-O-caffeoyl-(-)-quinic acid, 3-O-caffeoyl-(+)-quinic acid and 5-O-caffeoyl-*epi*-quinic acid) and the methyl esters of 5-O-caffeoyl-(-)-quinic acid and 3-O-caffeoyl-(-)-quinic acid. A second group of components contained the flavonoids quercetin, hyperoside, 3"-acetyl avicularin, avicularin and guaiaverin. 3"-Acetyl avicularin, 5-O-caffeoyl-*epi*-quinic acid and 3-O-caffeoyl-(-)-quinic acid methyl ester were identified from plants of the genus *Hypericum* for the first time, while guaiaverin and shikimic acid were identified for the first time from the species of *Hypericum empetrifolium*.

P062. Essential Oil Content and Composition of Some Aromatic and Medicinal Plants from Greek Islands of South Aegean Region.

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The knowledge of the compounds in the essential oils is crucial when they are prepared for use in medicines and cosmetics. In this study the essential oil of the following plants have been analysed: *Lavandula stoechas*, *Micromeria juliana*, *Mentha piperita* and *Mentha spicata*, *Salvia fruticosa*, *Satureja thymbra*, *Coridothymus capitatus* and *Thymus vulgaris*. The above samples were selected from the Greek islands Andros, Naxos, Siros, Tinos and Crete. The plant material was extracted by hydrodistillation. Then, samples of collected essential oils were analyzed by gas chromatography-mass spectroscopy. The dominant compounds for *Lavandula stoechas* from Andros were camphor (51,0%), myrtenyl acetate (12,2%) and Fenchone (9,3%), for *Micromeria juliana* from Crete the major compounds were caryophyllene oxide (11,4%) and γ-gurjunene (8,8%). The samples of *Mentha* species were found high in carvone (54.9%) for *M. spicata* from Tinos, iso-menthol (50,7%) and menthyl acetate (10,3%) for *M. piperita*. The major compounds of *Salvia fruticosa* were 1,8-cineole (49,1%) and camphor (19,6%). The main compounds for the samples of *Satureja thymbra* from Andros were γ-terpinene (29,6%), carvacrol (28,2%) and thymol (14,4%). The principal compounds for *Coridothymus capitatus* from Crete were carvacrol (34,6%), thymol (19,5%), γ-terpinene (15,9%) and p-cymene (11,9%). Finally, the main compounds of *Thymus vulgaris* from Andros were carvacrol (76,7%) and β-caryophyllene (6,3%). All these results could be assembled in a unified system, which could be continuously expanded. Finally, this will result in higher quality products, more economic products to obtain and reducing the risk to cause health problems, even to minimize them.

P063. Ethnobotanical study of medicinal plants used for the treatment of skin disorders in Central Macedonia, Greece.

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Medicinal plants have represented, for thousands of years, the only remedy for various diseases, with the most frequent indication of use, therapy of skin disorders and minor wounds. While phytotherapy still maintains an important role in the treatment of many diseases in Greece, ethnobotanical studies on traditional uses of plants and their products are relatively scarce [1]. To this direction, our aim was to record ethnobotanical information concerning the traditional use of medicinal plants in Central Macedonia, focusing on plant species used for treating dermatological disorders, along with the development of propagation material for the conservation of the most prominent herbs. The study area was Macedonia region (Northern Greece) in the area of Edessa, Naoussa and nearby villages and ethnobotanical data were collected through semi structured interviews and summarized by indices as Fidelity Level and Informant Consensus Factor [2,3]. Among the reported plant species, we selected those reported for the treatment of wounds, warts, burns and insect bites and a number of them was cultivated in the Hellenic Agricultural Organization-DEMETER, while others were already stored in the seed bank of the Institute of Plant Breeding and Genetic Resources, HAO-DEMETER. More specifically, for species such as *Matricaria chamomilla* L. tissue culture methods were used and propagation was carried out under aseptic conditions leading to callus induction [4]. Some interesting remedies were also recorded in the study, such as the preparation of an ointment of fragmented fruits of *Momordica charantia* L., immersed in olive oil under sunlight exposition for 30-40 days, indicated for human and animal wound healing. Among the further goals of this work, is the development and the chemical profiling of the reported herbal preparations, the evaluation of their biological activity and safety using *in vitro* and cell-based assays, as well as the detection, isolation and identification of their substances.

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P064. The family Solanaceae in the Greek medical tradition.

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Introduction: several species of the Solanaceae family have been present in Greece since ancient times and have been persistently used as medicinal plants. The main source attesting to their uses though Greek history is Dioscorides' *De Materia Medica*, which was continuously reproduced, consulted and used from the 1st cent. CE to late in the 19th century, including among Greek populations in the Ottoman Empire. This presentation will discuss the therapeutic uses of the Solanaceae species attested in Dioscorides' work.

Material and method: we screened the Greek text of *De materia medica* in the original language, together with the representations of the plants in its manuscripts, including to exactly identify species, and we compared resulting data to the information to be found in earlier (Hippocratic Collection and Nicander) and later works (Galen, Oribasius, Aetius, Alexander of Tralles, Paul of Aegina, Hippiatrica, Geoponica, and edited *iatrosofia*).

Results: with the exception of Mandragora, several of the supposed species of Solanum in ancient texts refer in effect to the same species. We also confirm the presence of Datura, supposed to be native to the New World. Uses were clearly perceived and were persistent through time, from ophthalmology to psycho-activity. Interestingly, this range of uses is still present in current traditional uses of the populations of Greece.

Discussion: collected data will not only illustrate the continuity of uses (and, beyond, knowledge) of some Solanaceae species in the Greek World, but also assess the understanding of the physiological action of these species from antiquity to present day.

P065. Phytochemical investigation of *Paliurus spina-christi* decoction prepared according to a traditional recipe.

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Paliurus spina-christi Mill. (Rhamnaceae) is a much-branched, deciduous, thorny shrub with green fruits that become brown during maturation. The shrub is found in Mediterranean, Southwest and Central Asia and North America. Its common name is "Christ's thorn" because it is said that its spiny branches were used to make the crown of thorns which had been placed on Christ's head before his crucifixion [3]. It is reported in the historical works of Dioscurides and Pliny as a diuretic, as well as palliative against gastrointestinal pain. Today, the decoction of the fruits of *P. spina-christi* is still used in the traditional medicine of Greece and Southern France for the same properties, as well as for the treatment of type II diabetes [1, 2, 4].

The decoction of the fruits of *P. spina-christi* was prepared according to a Greek traditional recipe. The aqueous extract was treated with adsorption resin technology in order to remove sugars and other inorganic material. The resulting organic material was fractionated using Reversed Phase-Solid Phase Extraction (RP-SPE) and the interesting fractions were chromatographed by Reversed Phase-High Pressure Liquid Chromatography (RP-HPLC) to isolate the pure compounds. Structure elucidation was based on NMR spectroscopy and LC-MS.

In this study we present some preliminary results of the phytochemical study of the decoction made from the fruits of *Paliurus spina-christi*. We have so far isolated and structure elucidated two cyclopeptide alkaloids (CPAs) that are new natural products. By elaborating LC-MS dereplication methodologies, we have also identified nine known secondary metabolites, 1 flavonoid-diglycoside, 2 phenolics, 1 triterpenoid, 1 lignan, 1 phytosterol and 3 CPAs, which had been isolated during a previous classical phytochemical analysis of the fruits. The decoction extract, as well as the SPE fractions and the pure compounds are currently under evaluation for their antidiabetic activity.

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P066. Chemical investigation of the recipe "OINOS TRAGORIGANITES" [Dioscorides].

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In the Greek "Pharmacopoeia" of 1931, "Medicinal Wine" is defined as the wine in which a medicinal plant has been extracted. In the ancient texts of Plinius, Galen and Dioscorides, however, there are recipes including the co-fermentation of must with several medical plant species, resulting in wines claimed to have a broad range of pharmacological properties.

In the framework of our continuing effort to evaluate ancient Greek medical recipes, we worked on the "MATERIA MEDICA" recipe of "OINOS TRAGORIGANITES".

OINOS TRAGORIGANITES [Dioscorides]

SUGGESTED: Satureia thymbra, Thymus tragoriganum, Thymus graveolens, Micromeria thymbra — Savory of Crete, Candian Savory — Tragoriganum Wine Tragoriganites. Let down four teaspoonfuls of tragoriganum (bound in a linen cloth) into four pints of must [grape pulp] for three months and then jar it. It is good for griping, convulsions, hernias, pains of the side, the movement of winds, and difficulty to digest.

The plant material (*S. thymbra*) was collected i*n the area of Attiki* and the model must and model wine were provided by the Agricultural University of Athens (AUA). We used the upper parts of the plant and the fermentations/ extractions were carried out in the laboratories of AUA, under controlled conditions in duplicate. We performed: 1. plant extraction in model wine in 3 plant/wine mixture analogies (3%, 6%, 9% w/v) and 2. plant/must co-fermentation in 3 plant/must mixture analogies (3%, 6%, 9% w/v).

The wine extracts and the co-fermented solutions were then eluted from polymeric resins (XAD type) of different polarity (XAD4 and XAD7), in order to separate the secondary metabolites from the wine/must matrices (mainly sugars and organic/inorganic salts). The adsorbed materials were analyzed by LC-HRMS and we present herein some preliminary results concerning the chemical profile comparison between the co-fermentations, the wine extracts and the MeOH/H2O chemical extracts of *S. thymbra.*

P067. Antiseizure compounds identified from the ancient Greek medicinal plant *Helleborus odorus* subsp. *cyclophyllus* using a zebrafish epilepsy model.

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Ethnopharmacological data and ancient texts support the use of black hellebore (*Helleborus odorus* subsp. *cyclophyllus* (HOC), Ranunculaceae) for the management and treatment of epilepsy in ancient Greece. A chemical and pharmacological investigation of the root methanolic extract (RME) of HOC was conducted using the zebrafish epilepsy model to define the compounds responsible for a potential antiseizure activity. The roots and aerial parts of HOC were extracted by solvents of increasing polarity. A root decoction of HOC was also prepared (RDE). The extracts were evaluated for antiseizure activity using a larval zebrafish epilepsy model with PTZ-induced seizures. The RME exhibited the highest antiseizure activity and was therefore selected for bioactivity-guided fractionation. Isolated compounds were fully characterized by NMR and HRMS. The HRMS/MS metabolite profiling of the RME and RDE were used for dereplication and chemical profiling. The RME of HOC showed an 80% inhibition of PTZ-induced locomotor activity. The extract was fractionated and resulted in the isolation of a new glucopyranosyl-deoxyribonolactone, a new furostanol saponin, 20-hydroxyecdysone, hellebrin, deglucohellebrin and a spirostanol glycoside derivative. The antiseizure activity of the RME was associated with the new furostanol saponin and hellebrin. A standard of hellebrigenin, the aglycone of hellebrin, was also found to be active. To further characterize the chemical composition of the RME and the RDE of HOC, 30 compounds were annotated by UHPLC-HRMS/MS profiling and included bufadienolides, furostanols, spirofurostanols and alkaloids. This study is the first to identify the molecular basis of the ethnopharmacological use of black hellebore for the treatment of epilepsy, which was achieved using a microscale zebrafish epilepsy model to rapidly quantify *in vivo* antiseizure activity. UHPLC-HRMS/MS profile revealed a large chemical diversity in the constituents of the extracts. The antiseizure activity of the extract and its components should be further investigated to reveal possible mechanisms of action.

P068. Medicinal plants of Chortiatis Mountain

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This study focuses on plants used for medicinal purposes, which are grown in Chortiatis Mountain in Thessaloniki. Today hundreds of medicines are released in the world with the use of medicinal herbs. Chortiatis has a plethora of wild flora. There have been recorded 94 families with 1220 different species and subspecies (Karagiannakidou & Raus, 1996).

The sampling was carried out by hiking on May 17th, 2020 in two different routes in Chortiatis Mountain. Then, species were identified by special identification keys and a herbarium was made through strict instructions of a special manual.

14 species of medicinal plants were collected in the first route and 11 species in the second route. From those species, 4 plants were selected (*Anchusa officinalis*, *Cistus creticus*, *Galium aparine*, *Origanum vulgare* ssp. *hirtum*) for further research of their morphological characteristics, historical data, ways of usage, medicinal substances they contain and their healing properties.

Anchusa officinalis is generally used as a decoction or extract and has diuretic, antibacterial, antiviral and antioxidant properties. (Zelić et al., 2005). From *Cistus creticus* we use each extracts and each essential oil against cancer cells and pathogenic microorganisms (Dimas et al., 1999; Dimas et al., 2001; Dimas K., 2006; Demetzos et al., 1995). *Galium aparine* is generally used for the preparation of a decoction or as a poultice. It has antioxidant, antimicrobial and immunomodulatory properties (Senio et al., 2018; Goryacha et al., 2014; Bokhari et al., 2013; Ilina et al., 2019), as well as apoptotic effects on breast and colon cancer cells (Aslantürk et al., 2017). Finally, *Origanum vulgare* ssp. *hirtum* is mainly used as a condiment, which is added in cookery. It has strong antibacterial effects it is an inhibitor against the growth of various fungi and has a strong toxic effect against viruses and against carcinogenic cells (Scortichini & Rossi, 1989; Biondi et al., 1993; Scortichini & Rossi, 1993; Sivropoulou et al., 1996; Paster et al., 1990; Paster et al., 1995; Skwarek et al., 1994)

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P069. *In vitro* evaluation of antioxidant, antiprotozoal and antimicrobial activity of medicinal and aromatic plants from the region of Epirus.

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The aim of this project was focused on the study of the potential antioxidant, antimicrobial and anti-parasitic activity of sage, oregano and crithmum aromatic medicinal plants. Experimental material was consisted of leaves and flowers for sage, oregano and crithmum to obtain most of the active ingredients of the plant. Sage and oregano were collected from the mountainous region of Epirus (Souli, Paleochori, Filiates, prefecture of Thesprotia), whereas crithmum was collected from the prefecture of Preveza. Then, the herbal material was delivered to the Lab of Pharmacognosy, Department of Pharmacy, Aristotle University of Thessaloniki, where the appropriate plant materials were hydrodistilled for two hours using a modified Clevenger apparatus. The volatile extract produced was collected according to the standard procedure described in the European Pharmacopoeia [1] The extracts were then preserved at 4 °C, in airtight closed containers until their employment in the *in vitro* tests and GC-MS analysis. Further analysis of the isolated essential oils (EO), included the determination of the Total Phenolic (TP) content and their antioxidant capacity measured by the 2,2-diphenyl-1-picrylhydrazyl (DPPH) test. The EOs were examined separately to test their antibacterial activity by disk diffusion agar method. Minimal Inhibitory Concentration was performed against some lactic acid bacteria, including *Lactobacillus plantarum, Lactobacillus rhamnosus, Enterococcus faecium*, *Enterococcus faecalis*, as well as in pathogenic bacteria such as *Escherichia coli* and *Staphylococcus aureus,* by broth microdilution method. All antimicrobial tests were implemented in the Lab of Animal Health and Food Hygiene and Quality, Department of Agriculture, University of Ioannina. Finally, the antiprotozoal activity of the EOs was tested in a bovine renal cell line (MDBK) after invasion with *Eimeria tenella* sporozoites [2]. The results showed that oregano EO had the highest concentration of polar phenolic components and even the strongest antioxidant, antimicrobial and antiprotozoal activity compared to the other examined EO.

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The project / research is co-financed by the European Regional Development Fund (ERDF) under the Operational Program "Epirus 2014-2020", NSRF 2014-2020. Project Code: ΗΠ1ΑΒ-0028192. Acronym: Innochicken.

Posters

Miscellaneous

20th International Congress of the International Society for Ethnopharmacology

P070. An online survey of cannabis use comparing 'drug-type' cannabis with 'hemp and CBD' products

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The hemp and cannabidiol (CBD) market (products low in THC) is emerging globally whilst little is known about the perception and consumption of these products' usage. Due to the preparations' pharmacological, legal and commercial characteristics the market offers an opportunity to study borderlines between medicine and food-supplements with implications for harm-reduction policies.

The survey (N =1450) was distributed online (e.g., by social-media) and addressed aspects of cannabis use ranging from self-reported medical uses, indicators-of-quality, formulations used, method-of-consumption, product-sources, and concomitant medicine-use.

Responses categorised as 'drug-type cannabis' and 'hemp and CBD' were compared indicating similarities in the main self-reported medical uses such as for mood alteration, pain-relief and substitutive cannabis use (instead of prescription medicines). Over 50% of reported uses of cannabis were for 'minor self-limiting medical conditions' (e.g., pain-relief) whereas 5% and less reported use with serious medical conditions (e.g., epilepsy), a finding which adds a new perspective on our understanding of the products' use patterns. Further the presentation will consider how new and emerging blockchain technologies can contribute to our understanding of survey data related to medical products, as well as it's potential to solve big issues in medicinal plant science such as supply-chain issues (sustainability, working with big-data etc...).

Products low in THC are commonly used for minor conditions associated with chronic pain and inflammation, mostly as unregulated food supplements. Further research is needed into the fast-developing use of cannabis products in order to inform public health strategy and science.

P071. Computational investigation of the associative behavior of essential-oil ingredients in an aqueous environment.

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Our project concerns the study of candidate drug compounds found in aromatic plants of the Greek endemic flora, with the aid of experimental and theoretical-modelling techniques. In this work, we report the study of four such compounds, Carvacrol, p-Cymene, γ-Terpinene and Thymol, *via* the simulation method of Molecular Dynamics. 1M and 0.1M aqueous solutions consisting of 10^4 to 10^5 water plus compound molecules, have been simulated at atmospheric pressure and 37°C in order to quantify the molecules' tendency to cluster. General Amber Force Field (GAFF), an established model of atomistic resolution, was used in our calculations that were carried out with the aid of GROMACS molecular simulation package. Thermodynamic and mass transport properties of the systems, were also looked at in order to verify the validity of the simulations by comparing with available experimental data. Mathematical analysis (DBSCAN algorithm) of observed clustering behaviour has shown that all four compounds tend invariably to form aggregates, thus bespeaking little or no propensity to mix with water. This situation calls for the use of carrier molecules as an aid for these compounds to be transported efficiently to targeted sites for biomedical purposes. Systems of two such carrier molecules, poly(vinyl alcohol) and poly(L-lactic acid) (linear and branched) are currently under study with the same simulation techniques, in order to predict their ability to capture and release the aforementioned molecules.

Acknowledgment: This research has been co-financed by the European Union and Greek national funds through the Operational Program Competitiveness, Entrepreneurship and Innovation, under the call RESEARCH – CREATE – INNOVATE (project code: T1EDK-04174).

P072. *In vitro* assessment of strawberry tree (*Arbutus unedo* L.) honey.

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Strawberry tree (*Arbutus unedo* L.) honey (STH), a typical product of Mediterranean region, is a rich source of bioactive compounds that possess strong antioxidant activity. This study examined the phenolic profile, total phenolic content (TPC) and antioxidant activity of STH from the town of Vrgorac (southern Croatia) and evaluated the protective role of STH and its main bioactive compound homogentisic acid (HGA) against cytogenetic damage induced by the anticancer drug irinotecan in human lymphocytes. Isolated human peripheral blood lymphocytes were exposed for 2 h to three different concentrations of STH and HGA (corresponding to average daily portion of 70 g of honey, as well as five- and ten- fold higher concentration). The outcomes of the treatments were evaluated using the cytokinesis-block micronucleus (CBMN) cytome assay.

TPC was 1525 mg gallic acid equivalents per kg of honey. Antioxidant capacity, estimated by 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity was 2.96 mmol Trolox equivalents per kg of honey. Twenty-two phenolics were quantified using ultra-high-performance liquid chromatograph (UHPLC) coupled to a hybrid mass spectrometer (LTQ Orbitrap MS). Both tested substances effectively ameliorated cytogenetic damage in irinotecan-treated lymphocytes and affected lymphocyte proliferation *in vitro*. STH offered slightly better cyto-/genoprotective effects than HGA, possibly due to its complex phytochemical profile. Convincing results obtained *in vitro* speak in favour of future investigations of STH, and further development of STH-based nutraceuticals, potentially useful even in cancer patients.

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Posters

Pharmacological and clinical studies of medicinal plants & natural products

20th International Congress of the International Society for Ethnopharmacology

P073. Treatment of seborrheic dermatitis using a cream containing *Salvia fruticosa* Mill.

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Seborrheic dermatitis (SD) is a common inflammatory lifelong disease that appears in areas of skin that are rich in sebaceous glands, such as the face, the scalp and the upper torso. Its course is characterized by relapses and recessions [1]. The safety and efficacy of a cream (*SDπ*, for the face) and lotion (*Lotion-Kπ*, for scalp and the chest) containing *S. fruticosa Mill.* (Greek Sage) leaf extract rich in antioxidants [2], were evaluated in three patients (3 males, age range: 40-64 years), suffering from SD with facial (one person), facial and scalp (one person), and facial and chest involvement (one person). The patients used cream SD*π* and Lotion-Kπ, applying it to lesioned skin and around it, once at night. No concurrent therapies were used. There was a follow-up ranged from 2 to 4 years. All patients experienced a complete clearance of their disease in short time, ranged from 7 to 25 days. Regarding facial SD, common feature in all patients, the mean time to achieve this result was 9.6 days. The follow up showed that clear state lasted for a long time (range: 1-10 months). However, when relapses occurred, patients responded well to retreatment in less time than initially needed to completely clear the disease (mean time 6.6 days for facial SD). No adverse effects were observed during the treatment. In conclusion, *SDπ* cream containing *S. fruticosa Mill.* is a prominent natural remedy for the topical treatment of SD, with high efficacy as monotherapy, able to induce disease clearance and long-lasting remission, without side effects. Moreover, it showed preservation of its efficiency when treating relapses. To our knowledge, it is the first time in the worldwide literature that such a study has been reported.

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[2] Kallimanis PG. "Rosemary and sage extracts for skin diseases: Compositions and treatments method." GR1009073

P074. Plaque psoriasis therapy using a cream containing *Salvia fruticosa* Mill.

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Plaque psoriasis (PP) is a lifelong skin disease and patients have to use drugs many times in their lives, so the role of remedy safety while maintaining efficiency has become crucial [1]. The efficacy and safety of a cream (*Kπ*) containing *S. fruticosa Mill.* (Greek Sage) leaf extract rich in antioxidants [2], were investigated in three patients (3 males, age range: 29-75 years), suffering from mild PP, in various body areas (elbows, legs, neck, hands, nails). All patients used cream *Kπ* applying it to psoriatic lesions and around them, twice daily. No concurrent therapies were used. There was a follow-up ranged from 0.5 to 5 years. All patients experienced a complete clearance of their disease (mean PASI score initially=3,87 vs mean PASI score finally= 0). The mean time to achieve this result was 3,6 months. The follow up showed that clear state lasted for a long time, ranged from 4 to 48 months. However, when relapses occurred, they responded well to retreatment in less time than initially needed to completely clear the disease (range: 1-2 months). No adverse effects were observed during the treatment. In conclusion, *Kπ* cream containing *S. fruticosa Mill.* is a prominent natural remedy for the topical treatment of mild PP as monotherapy, with high efficacy, able to induce disease clearance and achieving long lasting remission without side effects. Moreover, it showed preservation of its safety and efficiency when treating relapses. To our knowledge, it is the first time in the worldwide literature that such a study has been reported.

[1] Langley RG et al., "Psoriasis: epidemiology, clinical features, and quality of life." Ann Rheum Dis. 2005 Mar;64 Suppl 2: ii18-23.
[2] Kallimanis PG. "Rosemary and sage extracts for skin diseases: Compositions and treatments method." GR1009073

P075. Guttate psoriasis therapy using a cream containing *Salvia fruticosa* Mill.

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Guttate psoriasis (GP) is a less common form of psoriasis (prevalence <10% of patients with psoriasis). It manifests with acute eruption of numerous small (0.3-1.0 cm diameter), round or slightly oval, erythematous and scaly papules and plaques which are widely disseminated, particularly on the trunk and proximal part of the extremities [1,2]. The efficacy and safety of a cream (*Kπ*) containing *S. fruticosa Mill.* (Greek Sage) leaf extract rich in antioxidants [3], were investigated in two patients (2 females, age range: 68-75 years), suffering by GP, in various body areas (back, legs). Both patients used cream *Kπ*, applying it in psoriatic lesions and around them, twice daily. No concurrent therapies were used. There was a follow-up ranged 1-4 years. Both patients experienced a complete clearance of their disease. The mean time to achieve this result was 2,7 months. The follow up showed that the clear state lasted for a long time (≥12 months). One patient experienced a slight relapse after one year, and she responded well to retreatment in less time (2 weeks) than initially needed to completely clear the disease. The other patient was in clear state one year after total cure. No adverse effects were observed during the treatment. In conclusion, cream *Kπ* containing *S. fruticosa Mill.* extract is a prominent natural remedy for the topical treatment of guttate psoriasis as monotherapy, with high efficacy, able to induce the disease clearance and without side effects. To our knowledge, it is the first time in the worldwide literature that such a study has been reported.

[1] Lisi P. "Differential diagnosis of psoriasis." Reumatismo. 2007; 59 Suppl 1:56-60.
[2] Vence L, Schmitt A, Meadows CE, Gress T. "Recognizing Guttate Psoriasis and Initiating Appropriate Treatment". W V Med J. 2015; 111(4):26-8.
[3] Kallimanis PG. "Rosemary and sage extracts for skin diseases: Compositions and treatments method." 2016; GR1009073

P076. Involvement of the benzodiazepine site in the anticonvulsant activity of *Tapinanthus globiferus* against pentylenetetrazole-induced seizures in mice.

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Tapinanthus globiferus, popularly known as Afomo among Yoruba ethnic in Nigeria, is often referred to as an all-purpose herb for the treatment of stroke and epilepsy.

The present study investigates the anticonvulsant effect of methanolic leaf extract (MET), active fractions and lupeol (isolate) of *T. globiferus* in mice as well as the underlying mechanisms.

Following phytochemical studies of *T. globiferus*, preliminary assays were performed to evaluate MET-induced toxic effect and behavioral changes. The pentylenetetrazol (PTZ 70 mg/kg, i.p.) induced seizure was evaluated in mice that were pretreated orally with vehicle 10 mL/kg, MET (4, 20 or 100 mg/kg), fractions (F1 to F6), lupeol 10 mg/kg or diazepam (DZP 3 mg/kg).

MET preserved neuron viability as well as the relative organ weight, hematological and biochemical parameters. The behavioral endpoints, neuromuscular coordination, sensory response parameters revealed a dose-dependent effect of MET. This extract, active fractions, lupeol, and DZP potentiated the hypno-sedative effect of the barbiturate and attenuated PTZ-induced acute seizure. This antiseizure effect was completely reversed by flumazenil 2 mg/kg (benzodiazepine site antagonist).

Altogether, the benzodiazepine site-mediated anticonvulsant effects of MET, active fractions and lupeol corroborate traditional application of *Tapinanthus globiferus* against epilepsy.

P077. Antihyperglycemic and antihyperlipidemic activity of the methanol extract of *Shorea roxburghii* in high fat diet/Streptozotocin induced diabetic rats.

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Shorea roxburghii (Dipterocarpaceae) is a deciduous medium to large-sized tree native to Southeast Asian countries of Laos, Cambodia and Thailand. The extract from the plant is used for treating dysentery, diarrhea and cholera [1,2]. S. *roxburghii* is rich in polyphenolic constituents mainly stilbenoids, oligostilbenoids and dihydroisocoumarins [3]. This study investigated the hypoglycemic effect of the methanol extract from *Shorea roxburghii* leaves (SRL) in high fat diet/high fructose (HFDHF) and streptozotocin (STZ) induced type 2 diabetes mellitus (T2DM) in rats as well as evaluating its ameliorative potentials in altered biochemical and hematological parameters in the treated rats. T2DM was induced in Sprague Dawley (SD) rats by feeding with HFDHF for 4 weeks and administering STZ (35 mg/kg, i.p.) Diabetic rats were given SRL extract at doses of 100 and 400 mg/kg for 30 days. The food and water intake were monitored on a daily basis, while the fasting blood glucose (FBG) levels and body weight were measured weekly. Biochemical and hematological parameters as well as histopathological studies of the pancreas were also evaluated. SRL significantly decreased FBG and improved the body weight, food and water intake of treated diabetic rats. Furthermore, biochemical such as serum triglycerides, total cholesterol, blood urea nitrogen, alanine transaminase, aspartate transaminase, glycated hemoglobin, creatinine and low-density lipoprotein and hematological parameters including white blood and red blood cells parameters were markedly ameliorated by SRL. Histopathological analyses of the pancreas indicated reconstitution of β-cells architecture in SRL treated rats. The results from this study suggests that *S. roxburghii* have antidiabetic potentials and can be considered for the treatment of T2DM Highlights.

[1] Mrikawa T, Chaipech S, Matsuda H, Hamao M, Umeda Y, Sato H, Tamura H, Kon'I H, Ninomiya K, Yoshikawa M, Pongpiriyadacha Y, Hayakawa T, Muraoka O. Bioorg Med Chem 2012; 20: 832-840.

[2] Ninomiya K, Chaipech S, Kunikata Y, Yagi R, Pongpiriyadacha Y, Muraoka O, Morikawa T. Int J Mol Sci 2017; 18: 2.

[3] Patcharamun W, Sichaem J, Siripong P, Khumkratok S, Jong-aramruang J, Tip-pyang S. Fitoterapia 2011; 82: 489-492.

P078. Biological validation of herbal drugs from Vietnamese ethnopharmacology.

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In Vietnam, two types of traditional medicine (TM) are practiced: *thuoc nam*, medicine of the South, and *tuoc bac*, medicine of the North, both of which are largely based on herbal drugs used by different Vietnamese ethnic groups. The main problem of Vietnamese herbal pharmacology is the biological validation of the traditional uses of medicinal plants and pure compounds isolated from them. To this purpose, we have performed initial validation studies on four plants of Vietnamese medical tradition: *Dacrycarpus imbricatus, Pinus dalatensis* Ferrè, *Oldenlandia pinifolia* and *Ficus hirta*.

Dacrycarpus imbricatus (Blume) de Laub: Two monocyclic diterpenes were isolated and tested for their anti-proliferative activity on acute myeloid leukemia (OCI-AML) cells. The results showed that 1 had significantly anti-proliferative activity whereas 2 was weakly active.

Pinus dalatensis Ferrè is an endemic species in Vietnam that has never been investigated. Fifteen compounds were isolated from its wood and one diterpenoid, one flavonoid, and one stilbenoid had inhibitory effects on the growth of OCI-AML.

Oldenlandia pinifolia. Thirteen compounds were isolated from the *n*-hexane, ethyl acetate and *n*-butanol extracts of whole plants *O. pinifolia*. MTT assay resulted that the *n*-butanol extract and four isolated compounds inhibited the proliferation of chronic myelogenous leukemia cells. These four tested compounds induced apoptosis and activated caspase-3, with isorhamnetin-3-*O*-*β*-rutinoside that showed the most potent activity.

Ficus hirta. A new oleanane triterpene has been isolated from the *n*-hexane extract of *Ficus hirta* leaves (Moraceae).

In conclusion, the contribution of Vietnamese TM in establishing improved medical standards via the biological validation of the traditional remedies *Dacrycarpus imbricatus, Pinus dalatensis* Ferrè, *Oldenlandia pinifolia* and *Ficus hirta* has been analyzed. Specifically, several compounds isolated from different parts of the four medicinal plants were able to inhibit cell cycle progression and to increase apoptosis. Additional studies will clarify their mechanisms, toxicity and therapeutic importance.

P079. Effect of Fusarubin, a naftoquinone isolated from the endophytic fungi of *Cladosporium* spp., on senescence of acute myeloid leukemia cells.

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Endophytic fungi interact with host plants and alter aspects of their physiology, conferring increased tolerance to stress, improving immune system function, bolstering defenses against disease, and aiding substance absorption. Thus, the possibility of exploiting endophytic fungi as biocontrol agents has received increasing attention. *Cladosporium* species are endophytic fungi that grow on organic matter and are considered food contaminants and the anti-microbial and anti-tumor naphthoquinones fusarubin (FUS) and anhydrofusarubin (AFU) were isolated from a Cladosporium species residing inside *Rauwolfia* leaves. These compounds are not used in the ethnopharmacology but they can affect the action of ethnopharmacological plants such as *Rauwolfia*. The impact of FUS and AFU on cell growth was assessed in acute myeloid leukemia (OCI-AML3) and other hematologic tumor cell lines (HL-60, U937, and Jurkat). Treatment with FUS or AFU reduced the number of OCI-AML3 cells as evaluated by hemocytometer. Flow cytometry analyses showed that this effect was accompained by diverse impairments in cell cycle progression. Specifically, FUS significantly decreased the percentage of cells in S and increase the percentage of cells in G2/M phases, whereas AFU increased the percentage of cells in G0/G1 phase and decreased the percentage of of cells in S and G2/M phases. Both substances significantly increased apoptosis at higher concentrations. FUS, that was more potent than AFU, up-regulated p21 expression in a p53-dependent manner, as detected by Western blot analyses, likely the consequence of decreased ERK phosphorylation, increased p38 expression (both of which increase p21 stability) and decreased Akt phosphorylation. The up-regulation of p21 was possibly a mark of senescence since it was not accompained by and increase of caspase-9-dependent apoptosis. However, FUS was also able to induce apoptosis in 8% of cells by the production of Fas ligand, thus promoting caspase-8/3-dependent apoptosis. These results suggest that FUS induces a differential effect on the same cell line: senescence on the large majority of cells or apoptosis on a small fraction of them.

P080. Exploring bioactive components in different parts of *acer truncatum* for sustainable development.

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Acer truncatum (Sapindaceae) has been used as an ethnomedicine by the Mongolian for a long history. Traditionally its leaves have been consumed as a substitute for tea in northern China. Previous phytochemical research has focused on the leaf (AL) due to its long history as a tea for healthcare. The oil from *Acer truncatum* seeds (ATO) is rich in unsaturated fatty acids, especially in nervonic acid (NA). Its branch (ABr), bark (ABa), fruit (AF), root (AR), or seed residue extracts (ASR), however, have not been paid attention. Principal component analysis showed the close clustering of ABr, ABa, and AR, indicating that they share similar chemical components, while AL and AF clustered more distantly. Using UPLC-QTOF-MS analysis, 15 compounds were putatively identified from ASRs, including *N*-acetyl-D-tryptophan, nonanedioic acid, 10*E*-heptadecenoic acid, pregnanolone, phosphatidylglycerol (34:2), and phosphatidylinositol (34:1), which have not been reported in *Acer* prior to this study. The flavonoids miscanthoside and isovitexin/vitexine were also identified in *A. truncatum* for the first time. Furthermore, five samples each of ATO and ASR were selected to estimate the cytotoxic activities against four human tumor cell lines (A-549, SMMC-7721, MCF-7, and SW-480) and the AChE inhibitory activity. All ASRs were cytotoxic on all tested cells and exhibited higher inhibitory activity than ATOs at 100 μg/mL. ASRs exhibited stronger cytotoxicity on SMMC-7721. Similarity, the ASRs exhibited higher AChE inhibitory activity than the ATOs. All ASRs significantly inhibited AChE activity, and the inhibition rates were close to 90% at 20 μg/mL. Our results indicate ASRs were rich in bioactive components with cytotoxic and AChE inhibitory activities. These findings suggest that ATO and ASR could be a promising resource in the nutraceutical and pharmaceutical fields, which will support the sustainable uses of *Acer truncatum*.

P081. Cytotoxicity of selected medicinal plant species used by traditional healers in treating people living with HIV/AIDS in Uganda.

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Many people living with HIV/AIDS (PLHIV) in Uganda widely use herbal medicines. This is fueled by the belief in their therapeutic benefits and the fact that fewer PLHIV in Uganda have access to antiretroviral drugs than those eligible. However, the toxicity and safety of the medicinal plants used have not been investigated, posing a threat to patients' health. The objective of this study was to determine the cytotoxicity of medicinal plant species commonly used for HIV treatment in Uganda.

The cytotoxicity of the plant extracts was determined using the AlamarBlue cell viability assay with the human glioblastoma cell line: U87.CD4.CXCR4. The cells were treated with varying concentrations of the ethanol and Dimethyl Sulphoxide (DMSO) extracts of *Warbrugia ugandensis* Sprague*, Erythrina abyssinica* DC., *Cryptolepsis sanguinolenta* (Lindl.) Schltr., *Albizia coriaria* Oliv, *Psorospermum febrifugium* Spach, *Gymnosporia senegalensis* (Lam.) Loes., *Zanthoxylum chalybeum* Engl., *Securidaca longipendunculata* Fresen., *Acacia hockii* De Wild., *Gardenia ternifolia* Subsp. *jovis-tonantis* (Welw.) Verdc. and *Bridelia micrantha* (Hochst.) Baill. In addition, the half maximal cytotoxic concentration (CC_{50}) of the plant extracts was determined. Both the ethanol and DMSO extracts of *W. ugandensis* and *A. coriria* were highly cytotoxic with CC_{50} values of 7.6 and 1.5 µg/ml and 6.4 and < 4 µg/ml respectively. Most of the plant extracts (15/22) tested were moderately cytotoxic (CC_{50} = 21-200 µg/ml). The ethanol extracts of the plant species tested were generally less toxic to the cells than the DMSO extracts suggesting that a significant fraction of toxic substances are non-polar. The DMSO extracts of the plant species tested were generally more cytotoxic to the cells than the ethanol extracts. Some of the medicinal plant species such as *C. sanguinolenta, P. febrifugium* and *S. longipendunculata* had high CC_{50} values with correspondingly low cytotoxicity levels.

P082. Chemical and biological investigation on *Calluna vulgaris* (l.) Hull. Grown in Italy for its use as innovative skin-protective agent.

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Calluna vulgaris (L.) Hull. (Ericaceae) is a perennial shrub common in many European countries, from mountainous to flat areas. In ethnobotany, *C. vulgaris* aerial parts decoction has been used for urinary complaints. More recently, photo-protective activity was also investigated.

In order to better define the role of this medicinal plant in the modern phytotherapy, we focused our attention on chemical and biological characteristics of the few studied Italian *C. vulgaris*. First, we chemically characterized different extracts of *C. vulgaris* collected in different periods and areas of Italy. Then, we studied a standardized water extract evaluating red/ox properties, antiradical capacity, antimicrobial activity and the effectiveness in modulating collagen synthesis in human fibroblasts.

Italian *C. vulgaris* aerial parts were found to mainly contain polysaccharides and polyphenols; arbutin was also detected. *C. vulgaris* samples collected in Tuscany, Piedmont and South Tyrol were found to have a similar quali-quantitative phytochemical profile; the flowering stage was confirmed to be the "balsamic" period. Water extracts were found to be rich in all chemical markers, being the richest in polysaccharides and arbutin and having a good content in polyphenols. We prepared a standardized water dried extract of *C. vulgaris* collected in Tuscany (CVE): >10% polysaccharides, >4% total flavan-3-ols, >0.5% chlorogenic acid, >0.5% flavonoids (as hyperoside) and >0.05% arbutin.

In DPPH test, IC_{50} of CVE was below 50 µg/ml; oxidative processes at low potential (<+0.50) were detected by cyclic voltammetry measurements. CVE exerted a moderate antimicrobial effectiveness against *Streptococcus pyogenes*. In fibroblasts, CVE stimulated pro-collagen I synthesis at very low concentrations.

Due to the abundance of antioxidant constituents such as polyphenols and arbutin and of hydrophilic polysaccharides and given the activity on human fibroblasts, *C. vulgaris* growing in Italy possesses several positive features to be better investigated as innovative agent for skin anti-aging and protection.

P083. Polyphenols inhibits the growth and angiogenesis of breast cancer cells in vitro and in vivo.

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Phenolic compounds, for example, flavonoids, lignans, stilbenes, and phenolic acids, are plant metabolites found in herbs, beans, fruits, tea, cocoa, and coffee and may contribute to inhibition breast cancer cells. Chlorogenic acid, the main phenolic compound in coffee, has also been found in the leaves of Hibiscus sabdariffa and select herbs such as Lonicera japonica (honeysuckle) and Eucommia ulmoides. Chlorogenic acid (CGA) exhibits potentials towards liver, breast and skin cancer. Cancer cells stimulated with CGA exhibits differential expression of transcriptional factors and regulatory molecules. In the present study, we investigated the effects of chlorogenic acids (CGA) and cisplatin (CDDP). on proliferation, apoptosis, and angiogenesis of breast cancer cells in vitro and in vivo. It was found that co-treatment of CGA and CDDP could not only induce tumor cells into apoptosis through activating the mitochondria pathways, but also suppress the angiogenesis in xenograft animal model. These effects were associated with downregulation of the expression of MMP2/9, VEGF, and VEGFR-2, up-regulation of P53 signaling. CGA suppress the angiogenesis in xenograft animal model. These effects were associated with downregulation of the expression of MMP2/9, VEGF, and VEGFR-2, up-regulation of P53 signaling. Thus, CGA and CDDP in combination showed a better anti-tumor effects in breast cells than either CGA or CDDP presence alone and might represent an effective therapeutic strategy for breast cancer therapy.

P084. Physalin a attenuates inflammatortory responses through inhibiting the toll-like receptor 4-mediated c-jun n-terminal kinase pathways and up-regulating the antioxidant activity.

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Physalin A (PA) is a withanolide isolated from *Physalis angulata* L. In this study, and PA could inhibit the production of inflammatory cytokines, including prostaglandin E_2 (PGE_2), nitric oxide (NO), interleukin-1β (IL-1β), interleukin-6 (IL-6), and tumor necrosis factor-α (TNF-α) in lipopolysaccharide (LPS)-induced RAW 264.7 cells. However, PA inhibited the level of pro-inflammatory factor, including inducible nitric oxide synthase (iNOS) and cyclooxygenase-2 (COX-2), and suppressed the inhibitor of nuclear transcription factor κB (IκB)/nuclear factor-kappa B (NF-κB) and toll-like receptor 4 (TLR4)-mediated c-Jun N-terminal kinase (JNK) /activator protein 1 (AP-1) inflammatory signaling pathways in LPS-stimulated RAW 264.7 cells. Moreover, in the carrageenan-induced hind paw edema in mice, PA has been shown to inhibit the production of inflammatory mediators NO, malondialdehyde (MDA), and TNF-α production. In contrast, PA increased the antioxidant factor levels of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx). According to the result data, suggest the anti-inflammatory effects of PA may be through the suppression of the TLR4/JNK/AP-1 and IκB/NF-κB signaling pathways and up-regulation of the anti-oxidative activity.

P085. *Antrodia salmonea* induces autophagic and apoptotic cell death in human glioblastoma cells.

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We investigated the *in vitro* anticancer properties of *Antrodia salmonea* (AS), as well-known edible/medicinal mushroom in Taiwan, on human glioblastoma (GBM8401) cells; and revealed the underlying molecular mechanisms involved in autophagic- and apoptotic-cell death. Treatment of GBM8401 cells with fermented culture broths of AS (0-200 μg/mL) inhibited cell viability/growth. The results showed that AS significantly decreased the cell viability of GBM8401 cells with IC_{50} values of 75.5 μg/mL. AS-induced autophagy was evidenced via increased LC3-II accumulation and AVOs formation in GBM8401 cells. These events are associated with increased SQSTM1/p62, decreased ATG 4B, and dysregulated Beclin-1/Bcl-2 ratio. Inhibition of autophagy by 3-methyladenine (3-MA)/chloroquine (CQ) diminished AS-induced death in GBM8401 cells. AS-induced apoptosis through increased cleaved form Caspase-3, PARP cleaved and Bax expression. Further, inhibition of apoptosis by Z-VAD-FMK suppressed AS-induced cell death. Interestingly, blockade of AS-induced reactive oxygen species (ROS) production by N-acetylcysteine (NAC) pretreatment substantially attenuated AS-induced cell death. We also demonstrated that NAC pretreatment inhibited AS-induced autophagic cell death by decreased LC3-II. Taken together, *Antrodia salmonea* crucially involved in execution/propagation of ROS-mediated autophagic or apoptotic cell death in human glioblastoma cells, which can be used in the development of novel anticancer drugs.

P086. *Mikania micrantha* Inhibits Melanogenesis By Regulation of Autophagy In Melanocytes.

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With the increasing interest in skin beauty, the development of new skin brighteners has attracted widespread attention, and environmental awareness has increased. Modern people like to be able to use green resources as raw materials to develop products. The alien invasive species *Mikania micrantha* is growing too fast, causing serious damage to Taiwan's native ecosystem. The research team found that the entire plant, rich in polysaccharides and antioxidant phenols, is rich in polysaccharides and antioxidant phenols, whether in flowers, roots, stems, or leaves. It has a strong ability to keratinocyte proliferation and wound healing. Activity and telomerase activity, it is found that its anti-inflammatory effect is much higher than the chemical drug L-NAME. Another study showed that *Mikania micrantha* has antioxidant and anti-blood cancer effects, and can be developed into a health food to prevent free radical diseases and anti-cancer. Autophagy is a cooperative process between autophagosomes and lysosomes that degrades organelles. Recent research suggests that autophagy regulators may play an important role in the initial formation of melanosomes, a lysosomal-related organelle that synthesizes melanin pigments. In this study, we found that *Mikania micrantha* has the effect of increasing autophagy mechanism and reducing tyrosinase activity to inhibit melanin production. The purpose of this study was to determine whether *Mikania micrantha* inhibits melanin production by activating autophagy.

P087. Public's perceptions of the emerging role of Cannabidiol (CBD) products.

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Cannabidiol (CBD) products, derived from hemp (*Cannabis sativa*) are becoming more popular as people seek alternative ways of managing their health; sales rose 333% in the US in 2018[1]. The market value of CBD is estimated to be worth £1billion by 2025[2]. The objectives were to explore the public's perceptions and use of CBD products including where products are sourced, how knowledge is obtained and opinions on quality, safety and efficacy of CBD products. A questionnaire comprising open and closed questions was developed to consider quantitative and qualitative perspectives. An online platform, SurveyMonkey, was used to distribute the survey using social and professional networks (September - November 2019), which led to snowball sampling. A total of 596 responses were collected (63% female, 40% White-British) internationally. In total, 11% (n=66) claimed to use CBD products including oils, balm, tinctures and vapes for a range of ailments such as pain, stress and anxiety. Participants who did not personally use CBD products (n=530), 35% claimed they would like to try them, while 42% of participants knew of someone who was already using a CBD product for conditions such as arthritis and epilepsy and "*generally improving quality of life*". Knowledge of CBD was commonly attributed to social media and news articles. Perceptions on safety and efficacy were positive with the notion of natural medicines being superior. Quality of products was associated with well-known brands or products which had a marketing authorisation; participants identified it was difficult to determine the quality of products brought online and better regulation is required. This study identifies that people continue to seek natural alternatives to supplement and maintain their health, thus the popularity of CBD continues to rise. The lack of clinical evidence to help make informed decisions about using CBD products needs to be addressed with more research.

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P088. Chemical investigation and Antioxidant activity of cultivated *Sideritis cypria* Post: A preliminary study.

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Sideritis species (Lamiaceae) have been used for many years in the folk medicine. In Cyprus, three endemic *Sideritis* species have been found; *S. curvidens* Stapf, *S. perfoliata* L. and *S. cypria* Post[1]. *S. cypria* is a perennial herb belonging to the section *Empedoclia* Rafin., growing in Pentadactylos Mountains (Cyprus). Traditionally, the infusion of *S. cypria* is locally used as diaphoretic, tonic, as well as against stomach disorders, headache and common cold[2,3]. Previous studies reported the chemical characterization of wild *S. cypria* of its essential oil[4] and extracts[1]. In the present study, we report, here in, the first study of the cultivated *S. cypria*. The infusions of flowers and leaves were examined separately for their contents. So far, eleven compounds have been isolated using chromatographic techniques; four flavones, five phenylethanoid glucosides, one iridoid and one quinic acid derivative. Furthermore, the antioxidant activities, as well as the total phenolic and flavonoid contents from both the infusions and the decoctions of flowers and leaves were investigated. As far as we know, no literature concerning the antioxidant activity of the specific species has been reported previously. The compounds were identified using 1D and 2D NMR spectroscopy (COSY, HSQC, HMBC, NOESY).

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P089. Essential Oil Composition of Ginger Rhizomes and Its Ethnopharmacological Validation.

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The essential oils of *Zingiber officinale Roscoe*, *Zingiber zerumbet Smith*, *Curcuma amada Roxb*. and *Z. montanum* Koen available in Bangladesh were obtained through hydro-distillation. They yielded 0.22%, 0.228%, 0.18%, 0.20%, and 0.24% on a fresh weight basis, respectively. The chemical compositions of different fresh ginger essential oils were identified, quantified, and validated by GC-MS analysis. The major chemical constituents characterized as monoterpene, diterpene and sesquiterpene hydrocarbons such as camphene, *beta*-phellandrene, neral, geranial, Zingiberene, 1,5-heptadiene-3-yne, *alpha*-curcumene, (1R)-2,6,6-Trimethylbicyclo[3,1,1] hept-2-ene, *delta-3*-carene, limonene, *alpha*-humulene, bicyclo[2,2,1]heptan-2-one-1,7,7-trimethyl-(1S), zerumbone, 2,6,9,9-tetramethyl-2,6,10 cycloundecatrien-1-one, 2(3H)- naphthalenone, 4,4a,5,6,8-hexahydro and 3,7-cyclodeccadien-1-one,10-(1-methylethenyl-). The LC50 values in brine shrimp lethality bioassay of volatile oils obtained from four samples exhibited a significant mortality rate at 1.614, 1,351, 1,514, and 0.796, respectively, in comparison to reference standard vincristine sulfate (0.474µg/ml). The DPPH free radical scavenging activity of the fresh volatile oil samples was 46.67 µg/mL, 71.01 µg/mL, 22.02 µg/mL, and 19.55 µg/mL, respectively, where the reference standard butylated hydroxytoluene (BHT) showed the IC50 value of 21.10 µg/mL. The thrombolytic activity of oils samples 10.05%, 2.92%, 17.90%, and 29.48% was also evaluated, where streptokinase showed 40% clot lysis. The samples exhibited no promising antimicrobial and antifungal activity due to low concentration selection. Among the test samples, the most encouraging results obtained in *Zingiber montanum* Koen (Bon ada) in chemical composition showed the highest cytotoxicity with LC50 of 0.796 µg/ml free radical scavenging activity with 19.55 µg/mL and potential thrombolytic activity with 29.48% clot lysis. The cytotoxic activity, antioxidant activity, thrombolytic activity of the volatile oils exhibited in the study may be due to major compounds present in their compositions. The above findings validated the ethnomedicinal use of ginger as an herbal spice in food or condiment, diet supplements, drug additives, therapeutic substances. Thus Bangladesh can preserve foreign currency, preventing importation from foreign countries, thus commercial exploration of the potentials of Bangladeshi varieties.

P090. Chokeberry fruit in gastrointestinal motility disorders therapy.

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In recent years, the interest of pharmaceutical and medicinal science has been focused on the research of berry fruits and their possible therapeutic effects and nutritional value. The chokeberry (*Aronia melanocarpa* (Michx.) Elliott) has been used for centuries as a rich source of phenolic compounds, mainly anthocyanins. Functional gastrointestinal and motility disorders are considered to be the most common gastrointestinal disorders amongst the human population.

The aims of the study were to analyze the chemical composition and the antispasmodic effects of the chokeberry ethanolic extract, waste extract and the juice in the isolated rat ileum. The HPLC method was used for quantification of the anthocyanins and flavonoids in extracts and juice. The study examined the influence of the chokeberry preparations and cyanidin-3-*O*-galactoside, the main anthocyanin compound, on the spontaneous ileum smooth muscle contraction, as well as on contraction induced by acetylcholine and potassium chloride.

Results demonstrated that anthocyanins were the most represented compounds in all tested chokeberry preparations. Chokeberry waste extract contained the highest amount of examined bioactive compounds. The chokeberry preparations, in a concentration dependent manner, significantly relaxed the rat ileum spontaneous and induced contractions, due to the concentration of bioactive polyphenolic compounds. The dominant anthocyanin, cyanidin-3-*O*-galactoside, also expressed the spasmolytic effect and therefore may be responsible for the relaxant activity.

Based on the results obtained, our study offers an important insight into the gastrointestinal activity and the potential use in phytotherapy of not only the chokeberry ethanolic extracts and juice but also its waste extracts. Hence, the chokeberry could be used as a herbal remedy for the control of gut motility. However, it would be necessary to carry out clinical studies to confirm its spasmolytic effects in humans.

P091. Leaf Methanolic extract of a traditional medicinal plant of North-East India prevents hepatic steatosis via regulating AMPK mediated pathways.

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Non-alcoholic fatty liver disease (NAFLD) has attracted scientific attention in the pathogenesis of metabolic syndrome and its related health disorders. NAFLD includes a wide spectrum of liver dysfunction ranging from hepatic steatosis (intracellular triglyceride accumulation) to steatohepatitis. The present study has been undertaken to examine the beneficial effect of an indigenous medicinal plant of North East India against hepatic steatosis by using both cell culture and animal models. Male SD rats were used for this study (20 weeks' experiment). A total of 18 SD rats were divided into 3 groups (n=6), Normal chow diet (G1), High-fat high-fructose (HFFD) diet with treatment of vehicle (G2) or methanolic plant extract (G3). The extract formulated in 0.3% w/v Carboxy Methyl Cellulose (CMC) was administered to the animals through oral gavage at a dose of 250 mg/kg. Bioactivity guided fractionation of the extract has been carried out by using rodent hepatocyte (CC1) cell line treated with sodium palmitate (PA, 0.75 mM) with or without different extracts/fractions. Molecular mechanism has been dissected by using immunoblotting studies. Animal studies showed a positive effect of the plant extract in upregulating AMPK phosphorylation and reducing the levels of both triglyceride (TG) and total cholesterol (TC) in hepatic tissues compared to HFFD-fed animals. Cell culture studies further demonstrated a decrease in intracellular TG level upon treatment with methanolic leaf extract in PA-treated cells. Interestingly, ethyl acetate fraction of the methanolic extract also showed a significant decrease in TG level in PA-treated cells compared to others fractions, namely hexane, butanol, and water. Chemical profiling demonstrated the presence of verbascoside as one of the bio-active principle(s). The present study for the first time demonstrates a positive effect of the leaf methanolic extract of a traditional medicinal plants of North East India in reducing hepatic steatosis via upregulating AMPK signaling pathway.

P092. Zerumbone promotes hyaluronic acid production and enhances aquaporin-3 expression in human keratinocytes.

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Solar ultraviolet (UV) radiation is the most important external factor causing skin photoaging. We evaluated the skin hydration/moisturizing efficacies of Zerumbone (Zer, 2.5-10 μM), a natural sesquiterpene of Zingiber zerumbet, with UVB-irradiated human skin keratinocyte (HaCaT) cells and underlying molecular mechanism. We found that Zer promotes skin hydration by increasing the expression of hyaluronan synthase-2 (HAS-2) and aquaporin-3 (AQP-3) with or without UVB (30 mJ/cm2)-irradiated HaCaT cells. Notably, Zer dose-dependently activates ERK pathways and pre-treatment cells with the Pharmacological ERK inhibitor (U0126), HAS-2 and AQP-3 expression was attenuated. Further, the increased Src phosphorylation was confirmed by treatment with Zer in HaCaT cells. Notably, silencing of Src (siRNA transfection) significantly diminished Zer-mediated skin hydration, as evidenced by impaired HAS-2 and AQP-3 expression. We may suggest that Zer-induced HAS-2 and AQP-3 expression was mediated by the Src and ERK signaling cascades. RT-PCR confirmed that Zer increased the expression of natural moisturizing factor-related genes HAS-2 and AQP-3. Together, these results suggest that Zerumbone might be used as a cosmetic ingredient with positive effects on skin hydration and moisture retention.

P093. Components of a herbal combination against inflammatory disorders from Benin: In vitro anti-inflammatory and phytochemical screening of *Combretum collinum* Fresen, *Piliostigma thonningii* (Schum.) Milne-Redh. and *Ficus natalensis* Hoechst.

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A herbal combination of *Combretum collinum* Fresen, *Piliostigma thonningii* (Schum.) Milne-Redh., *Ficus natalensis* Hoechst., *Fadogia agrestis* Schweinf. Ex Hiern, *Entada Africana* Guill. & Perr. and *Chasmanthera dependens* Hoechst. is traditionally used for the treatment of musculoskeletal disorders and pain in the North of Benin and has already shown anti-inflammatory effects in a preliminary observational trial [1]. The present study aims to characterize the phenolic composition of *C. collinum*, *P. thonningii* and *F. natalensis* leaf extracts and to investigate the *in vitro* anti-inflammatory effects in TNFa-stimulated immortalized human keratinocytes (HaCaT). To mimic an inflammatory process on skin, HaCaT cells were stimulated with 20 ng/ml TNFá for 24 hours to overproduce interleukins 8 and 6. Dilutions of the plant extracts were tested for their anti-inflammatory effects by co-incubation with TNFá. Subsequently, cytokine levels in supernatants were measured by ELISA. The phytochemical analysis of the hydrophilic extracts by HPLC, LC-MS and partially NMR after fractionation identified the following major phenolic compounds: myricetin-3-*O*-rhamnoside and myricetin-3-*O*-glucoside *(C. collinum*), quercetin-3-*O*-rhamnoside (*P. thonningi*) and quercetin-3-*O*-rutinoside and chlorogenic acid and its isomers (*F. natalensis)*. Aqueous leaf extracts of *C. collinum* (CCL) and *P. thonningii* (PTL) decreased IL-6 and IL-8 release in TNFá-stimulated HaCaT cells (PTL: IC_{50} values of 74.37 µg/mL for IL-8 and 89.28 µg/mL for IL-6, CCL: IC_{50} of 142.5 µg/mL for IL8 and a decrease of IL-6 release by 29.3 % for the highest extract concentration of 200 µg/mL). For *F. natalensis* aqueous leaf extract no anti-inflammatory effect could be demonstrated. Regarding the identified phenolic compounds, only myricetin-3-*O*-rhamnoside showed an anti-inflammatory effect. Quercetin-3-*O*-rhamnoside, however, showed no influence on IL-6 or IL-8-release. The *in vitro* anti-inflammatory effect of *C. collinum* and *P. thonningii* supports the application of the herbal combination in traditional Beninese medicine and represents a potential alternative treatment strategy for inflammatory diseases.

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P094. *Sideritis* genus: Traditional medicine herbs for thousands of years and a source of various therapeutic agents.

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Sideritis L. genus consists one of the most important ethnopharmacological genus around the world[1]. Considering as highly beneficial source for human health, *Sideritis* species are widely used as pharmaceutical herbal products in food and cosmetic industries. Being known as Greek mountain tea, has been listed by European Medicine Agency (EMA) as a traditional medicine for the relief of mild gastrointestinal discomfort and against the common cold[2]. In the past few years, vigorous research studies have provided significant pharmacological activities related to the phytochemical profile of the specific genus[1,2]. A currently clinical study carried out by Wightman et al. (2018) showed that Greek mountain tea improved the aspects of cognitive function and mood in a group of healthy, older adults, leading to new therapeutic approaches[3]. A challenge, however, is to provide the market with standardized medicinal plants with known concentration of interesting bioactive ingredients. To address the aforementioned challenge, our research goal is to thoroughly identify the chemical profiles, as well as to examine the biological activities of cultivated *Sideritis* species. Up to date, we have studied three cultivated *Sideritis* species; one endemic species of Greece *S. euboea* Heldr. and two endemic species of Cyprus, *S. perfoliata* L. subsp. *perfoliata* and *S. cypria* Post. Their total extracts and infusions were investigated and their major compounds were isolated using chromatographic techniques. In total, so far, from the methanol extract of *S. euboea* 16 compounds were isolated[4], from the traditional infusion of the *S. perfoliata* subsp. *perfoliata* 12 compounds were isolated[5], while from the traditional infusions (both flowers and leaves) of the *S. cypria* 11 compounds were isolated. All the structure elucidations were carried out by means of 1D and 2D NMR methods (COSY, HSQC, HMBC, NOESY). [1]

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P095. Antioxidant potential of *Schisandra rubriflora* – extracts from parent plant material and from biomass of *in vitro* cultures

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Antioxidant activity of plant extracts is a very desirable attribute in the modern phytotherapy. Recent studies proved that fruits and leaves of *Schisandra chinensis* (Turcz.) Baill. show considerable antioxidant activities [1,2]. Biological properties of this plant are attributed mainly to dibenzocyclooctadiene lignans and phenolic compounds [2].

The object of this research was another, endemic for Sichuan province of China, less known species - *Schisandra rubriflora* Rehder & E.H.Wilson [1]. The aim of the study was to investigate the antioxidant potential of extracts from *S. rubriflora* fruit and leaf extracts of male (M) and female (F) specimens collected in May 2017 ("Clematis" company, Poland), using *in vitro* models. The estimations were performed also for microshoot culture extracts (M and F lines) growing on agar Murashige-Skoog [3] medium with 1 mg/l 6-benzylaminopurine and 1 mg/l indole-3-butyric acid (for 30 days). The antioxidant potential was evaluated using: CUPRAC (cupric ion reducing antioxidant activity), QUENCHER-CUPRAC (quick, easy, new, cheap and reproducible treatment, involving forced solubilization of bound phenolics by oxidizing TAC (total antioxidant capacity), FRAP (ferric reducing ability of palsma) and DPPH (1,1-diphenyl-2-picrylhydrazyl) methods. Additionally estimation of total polyphenol content using Folin-Ciocalteu method was performed.

Using all applied methods the highest antioxidant potential was confirmed for leaf extracts. The antioxidant activity of fruit and *in vitro* microshoot (M, F) extracts were comparable, but lower than for leaf extracts.

This is the first report documented the antioxidant potential of *S. rubriflora* extracts.

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[2] Mocan A., Crisan G., Vlase L. et al. Molecules 2014, 19, 15162–15179.
[3] Murashige T, Skoog F. Physiologia Plantarum, 1962, 15:473–497

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P096. Wound healing potential of crocin and safranal, main saffron (*Crocus sativus* L.) active constituents, in excision wound model in rats.

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Saffron is traditionally suggested for wound healing in Persian medicine. It is investigated for wound healing effect in multiple studies with promising results. It is not examined that which ingredient of saffron contributes more to this effect. This study is aimed to evaluate and compare the wound healing potential of saffron and its active constituents, crocin and safranal, in rats. Forty female adult rats with induced excision wound were randomly divided to 4 groups to receive topical formulation of saffron, crocin, safranal and placebo for 7 days. The Wound area and histopathologic stage of wound healing was evaluated as outcome measures. The wound area was significantly lower in treatment groups (saffron, crocin and safranal) compared to control group in the 7th day of intervention. Compared to control group, the wound in all treated groups (saffron, safranal, and crocin) showed decreased inflammatory response and more progression to proliferation phase. The saffron group showed more advanced healing phase with complete epithelialization of the wound in 7th day of study when partial and no epithelialization was observed in safranal and crocin groups. The study showed the wound healing properties of both safranal and crocin with the slight superiority of safranal. However saffron seems to be more potent in wound healing effect compared to safranal and crocin as its main active constituents.

P097. Biochemical, histopathological and physiological effects of *Citrullus colocynthis* L. on peripheral neuropathy in streptozotocin-induced diabetic rats.

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Ethnopharmacologic relevance: *Citrullus colocynthis* L. (*C. colocynthis*) is traditionally used for nerve originated pain in Persian medicine.

This study aimed to investigate the biochemical, histopathological and physiological effects of *Citrullus colocynthis* (*C. colocynthis*) on peripheral neuropathy in streptozotocin (STZ)-induced diabetic rats.

Seventy male adult Norway rats were included in the present study. Diabetes was induced using a single intraperitoneal injection of STZ (65 mg/kg) in sixty rats. After four weeks, the diabetic rats were randomly divided to treated groups by gabapentin, oral administration of *C. colocynthis* fruit pulps powder (100 and 300 mg/kg/day), topical preparations as oil-based solution and ointment, and placebo for four weeks. The change in metabolic, physiological, biochemical and histological parameters was considered as outcome.

Metabolic outcomes (body weight and blood glucose level) were improved in *C. colocynthis* treated groups as compared to placebo. Tail flick and hot plate tests, as physiological tests, had lower latency in *C. colocynthis* treated groups. Measurement of oxidative stress markers (malondialdehyde, superoxide dismutase and catalase) showed the antioxidant effect of *C. colocynthis*. Histological evaluation of sciatic nerve revealed that *C. colocynthis* decreased the number of demyelinated and degenerated nerve fibers. Among *C. colocynthis* treated groups, 100 mg/kg/day oral powder showed the best outcomes.

Results obtained from the present study showed that *C. colocynthis* fruit, by antioxidant and hypoglycemic activity, has a positive effect on the treatment of diabetic neuropathy (DN). It can enlighten the physiopathologic background of previously observed positive results of *C. colocynthis* in DN.

P098. Antispasmodic activity of the fungus growing termite *Macrotermes bellicosus* used in traditional medicine.

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Insects and insect-derived products play a vital role in traditional medicine in many parts of the world since ancient times. Among these insects, fungus-growing termites like *Macrotermes bellicosus* (*M. bellicosus*) is used in Benin for the treatment of infectious and inflammatory diseases such as diarrhea and dysentery [1]. *In vitro* studies reported antimicrobial and anti-inflammatory activities of *M. bellicosus* [1,2]. However, the occurrence of anti-spasmodic effects remains largely unexplored. This study aimed to investigate the spasmolytic activity of *M. bellicosus* in isolated rat small intestinal preparations using an organ bath equipment with isometric transducer in order to examine the influence on intestinal motility. An ileum/jejunum segment (1.5 cm) was prepared, cleaned and suspended in 20 mL organ baths containing aerated (95% O_2, 5% CO_2) modified Krebs solution maintained at 37°C. The preparations were allowed to equilibrate for 20 minutes with a preloaded tension of 10 mN. Thus, acetylcholine (ACh, 1 mM) was used to induce control contractions which were set 100%. *M. bellicosus* extract in ascending concentrations (0.05 – 2.0 mg/mL) was applied into the organ bath two minutes prior induction of contraction. The influence on basal tone directly after extract application as well as the intensity of ACh-induced contractions in relation to control applications was assessed. As a result, a concentration-dependent decrease of the induced contractions down to 41.85%±6.5 (% control) in the highest concentration (IC_{50} = 1.54 mg/mL; IC_{25} = 0.81 mg/mL) was observed. Moreover, application of *M. bellicosus* extract induced a significant decrease in basal tone down to -1.15±0.7 mN (1 mg/mL) and -2.07±2.4 mN (2 mg/mL). The effect was most pronounced in the highest concentration (2 mg/mL, -6.37 mN±5.5). The observed spasmolytic effect with a suppression of induced contractions and relaxation of basal tone reinforces the relevance of the traditional medicinal uses of *M. bellicosus* in Benin.

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[2] D Hammoud Mahdi, J Hubert, A Schubert, V Ahyi, K Nieber, C Vissiennon. Ethnomedicinal use, chemical composition and *in vitro* pharmacology of the termite species *Macrotermes bellicosus* used in traditional medicine in Benin. Congress abstract, Z Phytother 2019; 40(S 01): S38 DOI: 10.1055/s-0039-1697321.

P099. *Corydalis hendersonii* Hemsl, a Traditional Tibetan folk medicine, protects against acute myocardial ischemia in mice.

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Corydalis hendersonii Hemsl (CH), belonging to the family Papaveraceae, is mainly distributed in the area at altitude of 4200–5200 meters in Tibetan. As one of well-known characteristic Tibetan folk medicine, CH has the function of lowering blood pressure and has been used clinically to treat high altitude polycythemia and vasculitis. Our studies revealed that CH total extract, from 80% ethanol reflux extraction, exerts anti-acute myocardial ischemia (AMI) effects in LAD mice by echocardiography method (EF and FS values), detecting CK-MB and LDH levels, and HE and MASSON pathological examinations. The anti-AMI effect was closely related to anti-inflammatory through regulating NFκB and JAK2-STAT3 pathways. Moreover, the total extract was separated to obtain rich alkaloids fraction (RAF) and poor alkaloids fraction (PAF), through strong acid anion exchange resin. A pharmacological evaluation by LAD mice experiment showed RAF of anti-AMI effect at the dosages of 25, 50 and 100 mg/kg by increasing EF and FS values in a dose-dependent manner. The study further expands the potential of clinical application and excavates valuable traditional medicinal resources, enrich the basic research of the traditional Chinese medicine for the treatment IHD, and enhance the national drug innovative research.

P100. Bioassay guided isolation of 8-(2"-pyrrolidinone-5"-yl)- quercetin from *Senecio candicans* DC and its gastroprotective effects.

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Peptic ulcer disease (PUD) encompassing gastric and duodenal ulcer is the most prevalent gastrointestinal disorder. Duodenal, gastric ulcers and gastric cancer are common and serious diseases all over the world. It is estimated that every year nearly four million people are affected worldwide due to peptic ulcer, which accounts to nearly 10% of world population with different aetiologies. Excessive stress, smoking, chronic alcohol intake, *H. pylori* bacterial infection and chronic usage of non-steroidal anti-inflammatory drugs are main causes of PUD. The flavonoidal alkaloids or flavo alkaloids build a quite special group of natural products, where the typical flavonoid backbone (two aromatic rings are combined with an oxygen heterocycle) is linked with a nitrogen containing moiety too. Such group of natural products exhibit interesting gastroprotective effects. *Senecio candicans* DC is an endemic shrub of the Western Ghats, The Nilgiris, India. The hot water decoction of *S. candicans* leaf is being traditionally practiced as a remedy for gastric ulcer. The gastroprotective activity of the extract has been scientifically validated and reported that the decrease in lipid peroxidation and subsequent oxidative damage or by free radical scavenging activity may be the underlying mechanism. Further, focusing on the free radical scavenging activity, a bioassay guided approach was adopted to isolate a flavoalkaloid, 8-(2"-pyrrolidinone-5"-yl)- quercetin from *S. candicans* by screening the in vitro antioxidant ability of different extractions and fractions. The isolated was characterized using different spectral studies. The compound was further evaluated for its gastroprotective effects in experimental animal models.

P101. Greek edible *Crepis* species: evaluation of the phytochemical profile, antioxidant and anti-inflammatory activities.

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The genus *Crepis* L. belongs to the Cichorieae tribe of the Asteraceae family and comprises approximately 200 species most of them edible. Our studies are focused on the chemical and biological investigation of Greek species of the genus *Crepis*. So far, *C. dioscoridis* L. [1], *C commutata* (Spreng.) Greuter [2], *C. incana* Sm. [3], have been studied by our laboratory, while *C. heldreichiana* (Kuntze) Greuter and *C. crocifolia* Boiss. & Heldr. (recently renamed *Phytosia crocifolia* (Boiss. & Heldr.) Kamari & Greuter) are currently under investigation. The plant materials were left to dry and then extracted with cyclohexane: Et_2O:MeOH (1:1:1) and MeOH:H_2O (5:1) at room temperature separately. The nonpolar extracts of the aerial parts were set under investigation for isolation of sesquiterpene lactones (SL). The structures of all isolated compounds were elucidated by NMR spectroscopy. The extracts gave a plethora of sesquiterpene lactones. Although, the genus is characterized by guaiane-type SL [4] the last three under investigation species afforded also eudesmanolides and germacranolides.

In addition, the antioxidant activity of selected extracts was evaluated by the DPPH free radical scavenging assay. Activity was found in the polar extracts with IC_{50} values ranging between 0,06-0,26 mg/mL depending on the phenolic content of the extract as measured by the Folin-Ciocalteu method. Further, the most active extract exhibited anti-inflammatory activity in vivo, reducing mouse paw edema by 32%.

[1] Tsoukalas et al., 2014
[2] Kotti et al., 2019
[3] Barda et al., 2018
[4] Zidorn 2008

P102. Wild edible mushrooms in the order *Cantharellales* induce apoptosis to A549 cancer cells. Piceatannol was identified as a bioactive compound.

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Mushrooms constitute a kingdom containing more than 140,000 species. 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. In Greece, there have been recorded about 3,500 mycetes, 1,800 from which are mushrooms The aim of the present study was to examine *in vitro* beneficial bioactivity of mushroom methanol extracts and to investigate the molecular identity of the bioactive ingredients. Thus, 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 DPPH, MTT and PI/annexin V-flow cytometric assays. 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 furthermore induced apoptotic cell death to A549 cells. Ultrahigh Performance Liquid Chromatography coupled with tandem Mass Spectrometry analysis, using the Orbitrap High Resolution and Mass accuracy analyzer, 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. According to our best knowledge, piceatannol 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.

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P103. Chios mastic improves blood pressure haemodynamics in patients with arterial hypertension: Implications for regulation of proteostatic pathways.

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The aim of this study was to assess the acute effects of Chios mastic (CM) on peripheral and aortic haemodynamics and associated changes in gene expression of molecules involved in pathways related to hypertension. In a randomised double-blind case-controlled crossover design, 27 consecutive eligible volunteers (13 hypertensive patients) were assessed at two consecutive visits one week apart. Participants were randomly assigned to first visit oral administration of 2800 mg of CM (four tablets of 700 mg IASIS Pharma Hellas S.A.) or placebo. At each visit, haemodynamic parameters were assessed at three distinct time points (baseline, 2 and 3 hours after administration). Haemodynamic assessment included non-invasive measurement of aortic systolic blood pressure (SBP) and diastolic blood pressure. The results showed hypertensive patients presenting acute decreases in peripheral and aortic SBP and in peripheral pulse pressure after CM administration. After adjustment for age, gender and body mass index (BMI), these differences retained significance while as compared to placebo. Finally, from gene expression analyses, CM administration in hypertensive patients tended to reduce the expression of proteasomal (PSMB6, PSMB7, RPN6), chaperone HSP27 and the pro-oxidant NOX2 genes ($P < 0.1$ for all); as compared to controls, in hypertensive patients PSMB6 and NOX2 expression was significantly reduced ($P < 0.05$) suggesting that CM exerts disease-specific regulatory effects on genes involved in proteostatic and pro-oxidant pathways. In conclusion, this is the first study to demonstrate favorable effects of CM on peripheral and aortic blood pressure (BP) haemodynamics in hypertensive patients. The gene expression analysis pointed towards downregulation of the proteasome system and the NOX2 pro-oxidant pathway, which merits further investigation. Taken together, these findings should trigger further research of the role of CM in the management of hypertension.

P104. Anti-inflammatory and antioxidant effect of cannabidiol, in an Acute Autoimmune Hepatitis mice model.

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Cannabidiol (CBD) is a major active component of the Cannabis plant (*Cannabis Sativa L.*), which unlike tetrahydrocannabinol (THC) is devoid of euphoria-inducing properties. Broadly, CBD has demonstrated anxiolytic-like, anti-inflammatory and immunomodulatory benefits. Concanavalin A (ConA) is a lectin found in the jack bean (*Canavalia ensiformis*) and was studied for the first time by Jones and Johns (1916). ConA has been associated with a variety of toxicological effects (upon them mitogenic, cytotoxic and hepatotoxic). Intravenous administration of ConA is widely used for the induction of acute autoimmune hepatitis (AIH) in mice and the injury is mainly driven by activation and uptake of T-cells in liver.The aim of the present study was to investigate the anti-iflammatory and antioxidant effects of CBD on adult mice, with induction of AIH by ConA administration. Mice were pre-treated with CBD (20 mg/kg, oral administration) for five days and challenged with saline or ConA (20mg/kg; i.v.) on the fifth day. Anti-iflammatory analysis was assessed by determining the IL-2, IL-4, IL-10 and INF-γ levels on plasma serum by enzyme-linked immunosorbent assay. Antioxidant analysis was assessed by determining the GSH/GSSG ratio and SOD activity in liver tissue.The results reveal that IL-2, IL-4 and INF-γ levels on plasma were increased after ConA intoxication (inflammation index) and were reduced when mice were pre-treated with CBD. The detected IL-10 levels where increased in pretreated with CBD mice, suggesting a protective anti-inflammatory effect. Moreover, GSH/GSSG ratio increases after CBD administration, indicating an increase in antioxidant potency. The SOD activity is increased at ConA group and reduced at the group that has been treated with CBD and ConA. In conclusion, CBD administration shows an anti-inflammatory and antioxidant effect when mice are pretreated with CBD, before the ConA injections.

P105. Extract of *Salvia haenkei* presents anti-ageing activity in an *in vitro* human epidermal model.

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Skin is a very complex organ that acts as a barrier for the body and plays various roles including homeostatic regulation, immune surveillance and temperature maintenance. Endogenous factors, in concert with external assaults, continuously affect the skin, leading to structural changes that influence its appearance and its various physiological functions. Skin aging is a biological phenomenon hugely related to exogenous factors such as ultraviolet radiation, pollutions, etc that results in cumulative structural and physiological skin changes. This process has been firmly related to a variety of skin disorders such as benign and malignant neoplasm, degenerative disorders, etc. It is thus clear that the maintenance of the integrity of the epidermal barrier and the slowdown of the skin aging processes are important in the prevention of skin diseases. Previous studies have demonstrated the potential anti-senescence activity of an extract of the Bolivian plant *Salvia haenkei*, which acts by reducing senescent cells affecting the IL1a release and reducing the ROS formation. This study was designed to investigate the effect of an extract of *Salvia haenkei* (SH) on HaCat human keratinocyte cell line as a model of the epidermis. HaCat cells were exposed to stress factors related to premature aging of cells such as ultraviolet radiation and radicals. The ROS scavenger activity was firstly confirmed in keratinocytes; interestingly it was found that SH can restore the barrier integrity compromised by UV-induced stress acting by reinforcing the cytoskeleton structure and sealing the tight junctions. Moreover, SH was found to be able to increase the migration velocity of the cells, suggesting an interesting role in improving the barrier functions of the skin. The results of this study identify useful roles of the extract of *Salvia haenkei* for anti-aging skin treatment and open new perspectives for its clinical use.

P106. Antioxidant analysis of plant extracts' beverages prepared at home conditions.

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Aromatic plants took their name from the aroma that comes from those plants. These plants were known to people from ancient times. They were used as flavorings and medicinal potions.

Today aromatic plants are again becoming popular. Their antioxidant and antimicrobial properties have found application in the food industry as natural preservatives, as well as in medicine as inexpensive drugs with a low number of side effects.

Also, the problem of antibiotic resistance is pushing humanity to look for new methods of fighting bacteria. Today's treatments for infections are not as effective as before, this increases the duration of treatment and mortality.

Smoking, UV radiation, environmental pollution, all this can increase the level of free radicals adversely affecting our health. Free radicals damage DNA, and as a result, damaged protein forms. These speeds up the aging process and stimulates the development of certain diseases. Antioxidants act as a natural shield against free radicals. And especially widely antioxidants are present in the plant world.

In this diploma thesis will be studied 5 extracts and 5 mixed extracts from 5 aromatic plants. Samples which ware used in this study were extracted from plants grown in Thrace, namely: *Althaea officinalis*, *Achillea millefolium*, *Calendula officinalis*, *Aloysia citrodora*, *Melissa officinalis*. Mixed samples contain extracts of the above plants in different ratios: *Aloysia citrodora* & *Melissa officinalis* (50%/50%), *Achillea millefolium* & *Calendula officinalis* (30%/70%), *Achillea millefolium* & *Melissa officinalis* (20%/80%), *Althaea officinalis* & *Aloysia citrodora* (50%/50%), *Althaea officinalis* & *Melissa officinalis* (70%/30%). The purpose of sample mixing was to enhance the antioxidant and antimicrobial properties.

For research antioxidant activity, such methods were used as: DPPH, stable free-radical molecules, common antioxidants assay, the process of lipid peroxidation of linoleic acid by method of the linoleic acid (LLA) and the lipoxygenase inhibition (LOX).

P107. Silybin, curcumin and quercetin as metabolic modulators in human resistant cancer cells.

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Cisplatin and doxorubicin are chemotherapeutic agents used to treat human cancers. Despite their toxicity, the other issue that consequently leads to therapy failure is the onset of drug resistance. Among the main known resistance mechanisms, in the last decades also metabolic reprogramming has emerged as a possible one. For this reason, our laboratory has started research into natural compounds as possible modulators of metabolism to overcome resistance. Previously, we have already demonstrated that cisplatin and doxorubicin-resistant cells shift their metabolism toward a glycolytic and a lipogenic phenotype. With this in mind, after an initial screening of different natural compounds, our laboratory identified silybin, curcumin, and quercetin for their ability to modulate glycolytic and lipid metabolism. We performed cell viability assays to demonstrate the good toxicological profile of the three compounds. Moreover, the synergic effect of the combined treatment of silybin and doxorubicin was demonstrated on human adenocarcinoma cell lines. It was investigated the role of silybin in the modulation of glycolytic metabolism, evidencing that it was able to overcome doxorubicin resistance by reducing GLUT1 protein expression and glucose uptake. In parallel, we demonstrated the potential effect of the combination of curcumin or quercetin with cisplatin in human ovarian cisplatin-resistant cancer cell lines. We investigated the ability of these two compounds to modulate both glycolytic and lipid metabolism, concluding that curcumin and quercetin are able to sensitize cisplatin resistance acting on lipid metabolism. In light of the fact that these two compounds are able to potentiate the effect of cisplatin, our laboratory is still working on elucidation of the specific mechanism of action involved in the modulation of lipid metabolism.

P108. Anti HSV-1 effects of 14-deoxyandrographolide from *Andrographis paniculata*.

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Andrographolide and its analogs are diterpene lactones isolated from the medicinal plant *Andrographis paniculata* (Burm.f.) Nees, (Acanthaceae). These compounds have been reported to have a wide range of pharmacological activities, including anti-diabetic, anti-inflammatory, anti-tumor, antibacterial, anti-parasitic, anti-malarial, and anti-viral effects. In this report we describe a comprehensive isolation process, which allowed the rapid isolation of the three main components of Andrographidis herba, i.e. andrographolide (A), 14-deoxyandrographolide (DHA) and neoandrographolide (NA). The protocol included extraction with methanol, fractionation by vacuum liquid chromatography followed by column chromatography over Sephadex LH-20. Purity of compounds was assessed by NMR and HPLC-PDA-MS. The antiviral activities of the three isolated andrographolides, i.e. A, NA and DHA, were accessed using a recombinant herpes simplex virus (HSV) type 1 expressing the luciferase reporter gene under the control of the HSV thymidine kinase promoter. Consistently with previous observations, we find that none of the three compounds possesses virucidal activity nor inhibits the early stages of virus infection, i.e. virus host-cell attachment or penetration. However, two of the compounds (A and DHA) were found to possess antiviral activity, although the results are complicated by the cytotoxic activity of one of the compounds (A). Further studies, using wild-type HSV-1 strain 17, verified the anti-HSV-1 effects of A and DHA and suggested that the effect takes place during the later stages of the virus life-cycle.

P109. *In vivo* antimalarial activities of artemisinin and *Vernonia amygdalina* Del. leaf in mice.

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Combination therapy in the treatment of malaria and other diseases is a common practice in endemic regions. Recent alarming spread of drug-resistant *Plasmodium falciparum* (Pulcini *et al*., 2013; WHO, 2018) calls for a renewed intensive effort to develop new antimalarial drugs. This study investigated antimalarial activities of *Vernonia amygdalina* leaf (VA) alone and in combination with artemisinin on *P. berghei* ANKA-infected mice.

Fifty mice were infected with malaria parasite (*P. berghei*) and later grouped into 10 groups of 5 mice 2h before administration. Groups I-IV were orally administered with distilled water (0.2mL), Chloroquine (10 mg/kg), Artemether-Lumefantrine (4 mg/kg) and artemisinin (10 mg/kg), respectively as negative and positive controls while groups V-VII were administered with infused extract of VA at 50, 100 and 200 mg/kg. Also, groups VIII-X were orally administered with infused extract of VA at same doses and concurrently administered with Artemisinin at 10 mg/kg. Administration of drugs was repeated for 4 days and the smears were taken on day 5 to determine the parasitaemia level with corresponding suppression effect.

The result showed a dose dependent chemosuppression of 50-68% at 50-200 mg/kg. At 50 mg/kg, VA with artemisinin gave chemosuppression (60%) that was not significantly different from that of VA alone at 200 mg/kg (62%). The animals dosed with VA together with artemisinin were able to survive more than those dosed with VA alone and with artemisinin alone.

The combination of artemisinin and VA in the study suppressed the parasitaemia growth in *P. berghei* infected mice. This combination can therefore be explored for antimalarial drug development.

Pulcini S, Staines HM, Pittman JK, Slavic K, Doerig C, Halbert J, Tewari R, Shah F, Avery MA, Haynes RK, Krishna S (2013) Expression in yeast links field polymorphisms in PfATP6 to *in vitro* artemisinin resistance and identifies new inhibitor classes. Journal of Infectious Diseases 208(3):468-478

WHO, 2018. Global report on insecticide resistance in malaria vectors: 2010–2016. Geneva: World Health Organization; 2018 (http://www.who.int/malaria/publications/atoz/9789241514057/en/, accessed 15 October 2018).

P110. Lavender (*Lavandula angustifolia*) and lemon balm (*Melissa officinalis* subsp. *officinalis*) cultivated in Greece: Biological activity of essential oils and leaves.

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The biological activity of two Lamiaceae members, *Lavandula angustifolia* Mill. and *Melissa officinalis* L. cultivated in Greece is examined. Both are of high economic importance since they are used in a variety of products from foods to cosmetics, perfumery and aromatherapy. The genus *Lavandula* includes 22 species grown in countries around the Mediterranean. Among them *L. angustifolia* commercially known as English lavender, is widely exploited with cultivations of local varieties and artificial hybrids. It has been used for centuries as a therapeutic agent and its essential oils became very popular after the 1st World War, then used as antimicrobial and later as an ingredient in perfumery and cosmetics. *Melissa officinalis* grows wild in the northern part of the Mediterranean basin. Three morphologically and chemically distinct subspecies are recognized. Subsp. *officinalis* plants are commercially exploited almost all over Europe, under the vernacular name lemon balm. In Greece it is found in the wild but mainly in small scale cultivations. With the common names "μέλισσα" and "μελισσοβότανο", it is used in traditional foods and herbal teas. Twelve essential oils from *L. angustifolia* cultivated in Greece were tested for their: (i) antioxidant activity in two different *in vitro* assays presenting significant antioxidant profile (38-71%), and (ii) inhibitory activity against lipoxygenase (42-63%) and acetyl-cholinesterase (17-51%). *M.officinalis* subsp. *officinalis* leaves from hydroponic crop plants were tested *in vitro* as antioxidants and were found to possess high activity in both assays (71-84% and 66-83%). The anti-lipoxygenase and anti-acetylcholinesterase activities were low (23-29% and 26%) compared to standard reference compounds. The present results on the biological activity of the two examined species are very promising.

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P111. Protective effect of Chios Mastic Gum on bone mineral density of ovariectomized rats.

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The aim of this study was to evaluate the potential effect of Chios Mastic Gum (CMG) [*Pistacia lentiscus* var. *chia*] on bone mineral density (BMD) of the lumbar vertebra of ovariectomised rats, a site used for BMD measurement in humans. Thirty 10-month-old Wistar rats were randomly allocated into three groups of ten, Control, Ovariectomy and Ovariectomy-plus-CMG in their food. The BMD of the sixth lumbar vertebra was evaluated in all groups before ovariectomy (baseline) and after 3 and 6 months post ovariectomy. For the characterization of CMG, Gas Chromatography-Mass Spectrometry (GC-MS), Ultrahigh Performance Liquid Chromatography-High Resolution Mass Spectrometry (UPLC-HRMS) and High Resolution Mass Spectrometry/Mass Spectrometry (HRMS/MS) were used. The results showed that both acidic and neutral triterpenoids were identified while special attention was given to the characteristic triterpenic acids of CMG e.g. masticadienonic, isomasticadienonic, moronic, oleanonic acids. Comparison of the absolute values of the sixth lumbar vertebra BMD revealed no statistical difference between groups at baseline. At three months, the BMD of the Ovariectomy-plus-CMG group was significantly higher than the BMD of the Ovariectomy group ($p=0.001$), while there was no statistical significant difference with the Control group ($p=1.000$). At six months, the BMD of the Ovariectomy-plus-CMG group was significantly higher than the BMD of the Ovariectomy group ($p=0.010$), with no statistical significant difference versus the Control group ($p=0.599$). In conclusion, according to the results of the present study, the administration of CMG with the above-mentioned triterpenoids and acids exerted a significant beneficial impact on the BMD of the sixth lumbar vertebra.

P112. Chemical analyzes of *Matricaria pubescens* and *Matricaria recutita* polar extracts and study of their anti-inflammatory properties.

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Matricaria pubescens (Desf.) Schultz (Asteraceae), known as hairy chamomile, is endemic in North Africa and is used in rheumatic and muscular pains, coughs, allergies, ocular affections, dysmenorrhea, scorpion stings, dehydration and toothaches [1]. In Europe *M. recutita* L is the officially recognized species used for mild gastrointestinal problems, ulcers and inflammations of mouth and throat, for irritated skin and mucosae and for the relief of common cold [2]. As a part of a project aiming at studying *Matricaria* sp. the analysis of *M. pubescens* from Algeria and *M. recutita* from Greece was undertaken. Methanolic and hydroalcoholic extracts were prepared and studied by HPLC-PDA-MS and NMR. In *M. pubescens* more than 20 compounds have been identified up to now, among them quercetagenin-3-O-glucopyranoside, reported for the first time in *Matricaria* sp. and two polyamines previously reported in other Asteraceae sp. [3]. Fingerprint analysis and comparison of this species to the officially recognized in Europe *M. recutita* L. shows many similarities in the chemical content and justifies the ethnopharmacological uses in Algerian traditional medicine. *M. recutita* extracts from Greece were also characterized by HPLC-PDA-MS and were rich in phenolic constituents. Structure elucidation was carried out by HPLC-PDA-MS by use of reference standards, by classical phytochemical analyzes followed by 1D and 2D NMR and by combination of NMR and HPLC-PDA-MS on sub-fractions. The characterized extracts were screened for their protective effects on 3T3 fibroblasts. The cells were exposed to UVA light to 5-7J/cm2 in presence of different doses of the extracts. Cell viability and oxidative stress were evaluated by neutral red absorption (540nm) and 5-(and-6)-chloromethyl-2',7'-dichlorodihydrofluorescein diacetate, acetyl ester fluorescence (excitation at 485 and emission at 520nm). *M. recutita* methanolic extract had the best protective effect in mild inflammation (60 minutes) The best extract will be further tested in vivo, on hairless mouse skin, as topical anti-inflammatory agent.

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P113. Total phenolic and total flavonoid analysis of hydroalcoholic extracts from plants belong to *Achillea* species and their antioxidant and antimicrobial activities.

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Achillea species are known for their healing properties since ancient times. There is extensive literature on their pharmacological action due to their bioactive compounds. The present study aimed to investigate the antioxidant and antimicrobial effects of hydroalcoholic extracts from the inflorescences and leaves of the species *Achillea crithmifolia* Waldst. & Kit., *A. grandifolia* Friv. and *A. millefolium* L. The phytochemical profile of all extracts was monitored using NMR spectroscopy and applying techniques for evaluation of total phenolic (TP) and total flavonoid (TF) contents which are in accordance with the results estimated using the Folin-Ciocalteu method (TP:125.42-191.98mg/g, TF:47.34-180.02mg/g of extracts). All extracts were tested for their antioxidant activity using two *in vitro* methods, DPPH and ABTS $^{\cdot+}$ radical scavenging assay. All extracts showed good to moderate antioxidant activity, compared to the reference substance BHT. Moreover, the antibacterial activity was tested against some of the major food-borne pathogens (*Enterococcus faecalis*, *Bacillus subtilis*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella typhimurium*, *Escherichia coli* and *Klebsiella pneumoniae*). None of extracts showed satisfactory antimicrobial activity.

P114. Natural products isolated from *Thymus thracicus* Velen. (Lamiaceae) exert strong antiplatelet action.

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Today, the need for antiplatelet therapy with similar biological effects of aspirin without its drawbacks, is the objective of major pharmaceutical companies. Aim of this study was to investigate antiplatelet effects of oresbiusin A (1) and methyl ester of rosmarinic acid (2), isolated from the methanolic extract of *Thymus thracicus*, compared to aspirin in whole blood. Experiments for antiplatelet action were performed on whole blood from healthy donors who have not taken medication for more than 20 days. Platelet activation in the presence of compounds (1) or (2) vs. aspirin was analyzed by flow cytometry, quantifying CD61PerCP, CD62-P PE and CD41aFITC, present in activated platelets. Welding experiments were performed in Multiplate assay with coagulant factors ADP, Collagen and arachidonic acid. Microparticle quantification was done with flow cytometry. The IC_{50} values calculated for different agonists (ADP, Collagen and arachidonic acid) by flow cytometry, using the expression of CD62-P were 1.6-3.5mM for (1), 0.75-2.45mM for (2), significantly lower than the respective ones of ASP (2.5-5.03mM). Multiplate assay confirmed antiplatelet action of (1) and (2) in all the conditions used. We analyzed the presence of microparticles after activation of platelets with ADP, which, in the presence of substances (1) and (2) showed a striking reduction. In order to explore their ability to act as antiplatelet drugs, and to explain their described *in vitro* activity, we adopted *in silico* molecular docking studies. The computational approach revealed that both constituents possess antiplatelet activity inhibiting the proteins cyclooxygenase-1 (COX-1), human protease-activated receptor 1 (PAR1), P2Y12 receptor (P2Y12R), P-Selectin (CD62P), transcription nuclear factor kappa B (NF-kB) and protein kinase C alpha (PKCa), all involved in platelet aggregation. Furthermore, we explored the binding capacity of both compounds to Human Serum Albumin, as a drug transport mechanism. Based on our results we conclude that both compounds exert strong antiplatelet action combined with reduction of microparticle formation.

P115. Investigation of the anti-inflammatory potential of the Himalayan ethno-medicinal plant *Sauromatum venosum*.

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Western Himalaya hosts a diverse flora comprising of many ethno-medicinally important plants. Most of the plants have not been investigated for scientific evidence of the medical claims set by the local ethnic communities [1]. One of these Himalayan plants is *Sauromatum venosum* (Dryand. ex Aiton) Kunth (Araceae). Plants from this family are known to be used for the treatment of pain and inflammation. The local residents have also reported *S. venosum* for the treatment of cancerous diseases [2]. Since inflammation is linked with the development and progression of various cancers [3], the anti-inflammatory effects of this plant were investigated by using cyclooxygenase inhibition assays with COX-1 and COX-2 enzymes. Plant tubers collected from Kotli Azad Kashmir (a district in Himalaya) were extracted, and dried tuber extracts were analysed by LC-MS and used for the *in vitro* testing. Both, COX-1 and COX-2 inhibition studies revealed that the plant extracts (at 20µg/ml) are exceptionally active against the COX-2 isoenzyme with a maximum inhibition by the dichloromethane extract of 77%, while this extract showed only 10% inhibition of COX-1. Other extracts (*n*-hexane, ethylacetate and methanol) showed COX-2 inhibition of 70%, 61%, and 75%, while these extracts showed COX-1 inhibition only of 6%, 9%, and 13%, respectively. Selective COX-2 inhibitors are more useful for the treatment of inflammatory conditions as they do not inhibit COX-1, leading to less side effects like ulcer and bleeding. Isolation and characterization of the active constituents will be reported.

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P116. Screening for tyrosinase inhibitors from actinomycetes; Identification of trichostatin derivatives from *Streptomyces* sp. CA-129531 and scale up production in bioreactor.

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In the frame of MICROSMETICS EU project 56 potential candidate actinobacteria strains of global biodiversity were selected to be studied using OSMAC strategy. In total 614 extracts were produced and evaluated for their skin-whitening bioactivity. Among them, the EtOAc extract of the fermentation broth of the strain *Streptomyces* sp. CA-129531 originated from Martinique, exhibited in cell free and cell-based assays the most promising activity (IC_{50} 63 μg/mL).

The scale-up production of the strain was performed in a bioreactor Biostat C+ using the culture medium DNPM with some modifications in order to adhere to the industry regulations. Chemical investigation and bioguided isolation of the extract, led to the isolation of one new trichostatic acid analogue, namely trichostatic acid B, along with six known trichostatin derivatives, four diketopiperazines, two butyrolactones and one hydroxamic acid siderophore. Among them, trichostatin A (TSA) showed six times stronger anti-tyrosinase activity (IC_{50} 2.18 μM) than kojic acid (IC_{50} 14.07μM), while deoxytrichostatin A displayed also strong inhibitory activity (IC_{50} 19.18 μM). In order to investigate the mechanism of mushroom tyrosinase inhibition by TSA, kinetic studies were conducted based on the Lineweaver-Burk and Dixon plots. Moreover, for the monitoring of TSA production, its evolution was investigated in a 30 L bioreactor until dextrose exhaustion, showing that its production started together with the exponential phase of growth (day 4) and the maximum concentration was reached at day 9. Despite the cytotoxicity of some individual components, the EtOAc extract showed no cytotoxic effect on HepG2, A2058, A549, MCF-7 and MIA PaCa-2 cell lines and against BJ fibroblasts at the concentrations where the whitening effect was exerted, reassuring its safety and great tyrosinase inhibitory potential.

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P117. Identification of new phytochemicals that inhibit α-synuclein fibrilization to prevent or delay the progression of Parkinson disease.

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Alpha-synuclein is a 140 amino-acid residue protein that is naturally unfolded. Aggregates of α-synuclein are found in Lewy bodies, a defining neuropathological characteristic of Parkinson's disease (PD) and other synucleinopathies (Vekrellis et al. Lancet Neurol 2011). A-synuclein transmits PD pathology cell-to-cell with a prion-like mechanism. Thus, the identification of compounds that inhibit α-synuclein fibrilization or induce the depolymerization of aggregates to prevent PD progression would be of great interest for pharmacological intervention. Here, natural compounds from various classes (steroids, flavonoids, terpenes and terpenic acids) were screened to identify specific inhibitors of α-synuclein fibrilization. For screening assays, we expressed recombinant α-synuclein in *E. coli* BL21 and purified it by two-step FPLC. The purified recombinant α-synuclein was aggregated to fibrils in PBS at 37°C under intense agitation for 7 days (Patterson et al. JoVE 2019). We confirmed that these fibrils are biochemically and cellularly active and when injected into the striatum of mice, they evoke phosphorylation of the endogenous α-synuclein. All compounds tested were purified from various plants and characterized by LC-MS, ^{1}H-NMR. Inhibition of α-synuclein was assessed: [1] by reading the OD600 *vs* time and [2] by the thioflavin T fluorescence assay, that both measure the formation of α-synuclein fibrils. Further, the antioxidant potential of the compounds was assessed with the Briggs-Rauscher reaction and correlated with their potential fibril inhibition effect. The results on the inhibition on α-synuclein fibrilization will be presented. ^{15}N/^{13}C-labelled α-synuclein is generated to shed light on the binding site of lead compound(s) onto α-synuclein associated with inhibition of fibril formation. The isolated compounds can potentially be used as new nutraceuticals to prevent or delay PD.

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P118. Extracts from felina 32 hemp inflorescences in cancer: an *in vitro* study.

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Cannabis sativa L. is a plant traditionally cultivated as a source of fibers and nutrients. Recently, the interest toward its medical uses is growing, owing to the highlighted therapeutic potential of its numerous phytoconstituents, including anticancer properties [1]. In this context, our work aimed at evaluating the antiproliferative properties of *C. sativa* var. Felina 32 inflorescences, collected in June and September, in relation to non-psychoactive cannabinoids and caryophyllane sesquiterpenes content.

Inflorescences were subjected to Bligh-Dyer extraction, to obtain two terpenoid-rich organic fractions, namely JOF (June Organic Fraction) and SOF (September Organic Fraction). Extracts were analysed by gas chromatography/mass spectrometry (GC/MS) to determine the major non-psychoactive cannabinoids and caryophyllane sesquiterpenes. The cytotoxicity of JOF, SOF and the major compounds detected at GC/MS, alone or in combination, was evaluated in different cancer cell lines. The endocannabinoid system involvement in the antiproliferative effects of the samples was assessed using CB1 and CB2 receptor inhibitors. Immunofluorescence and western blotting analysis were performed to confirm the mechanistic hypothesis.

Results highlighted cannabidiol, cannabichromene, β-caryophyllene, β-caryophyllene oxide and α-humulene as the major compounds, with higher amount in SOF compared to JOF. The extracts and the pure compounds inhibited cancer cell proliferation, with higher potency towards MDA-MB-468 cells. Interestingly, the pure compound combinations induced cytotoxic effects similar to those of the extracts. After treatment with CB1 and CB2 inhibitors, the cytotoxicity of JOF, SOF and cannabidiol was not reduced; conversely, a slight lowering of β-caryophyllene and cannabichromene cytotoxicity was highlighted, thus suggesting that CB2 receptor activation can partly contribute to their activity. This hypothesis was deepened by evaluating the effect of JOF, SOF and pure compounds on the CB2 protein expression.

In conclusion, Felina 32 hemp inflorescences can be an interesting bioactive phytocomplex with antiproliferative properties, likely due to the synergistic interactions between nonpsycoactive terpenoids and caryophyllane sesquiterpenes.

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P119. Evaluation of the antiproliferative/cytotoxic action of olive oil phenolic ingredients on human breast cancer.

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Olive oil consumption has been associated with the protective effect towards several malignancies, especially breast cancer. This chemopreventive activity has been attributed to olive oil polyphenols (OOPs) such as hydroxytyrosol and tyrosol, along with their derivatives oleuropein aglycone, ligstroside aglycone, oleacein and oleocanthal. Several studies have demonstrated that certain of these compounds inhibit breast cancer cell proliferation by inducing apoptosis, while in combination with chemotherapeutic drugs improve the sensitivity to treatment. However, further investigation is necessary to clarify cytotoxicity and the mechanism of action not only of single OOPs, but also of their combination as isolated compounds and in natural extracts.

We evaluated the antiproliferative action of OOPs on three human breast cancer cell lines (MDA-MB231, MCF7 and SKBR3) and a non-cancer cell line (MCF10A). Two-dimensional cultures were treated with increasing concentrations of each OOP (10-100μM) and cytotoxicity was determined by an adenosine-triphosphate (ATP)-based luminescence assay. Half-maximal effective concentrations (EC50) were calculated in conditions of atmospheric O2 (20% v/v). Combinations of two, three or four different OOPs were examined for synergistic effect through 3-(4,5-*di*methylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) method. Finally, olive oil extracts of determined OOPs content were tested in order to simulate the olive oil effect on cell proliferation.

Our results reveal that single OOPs suppress cancer cells proliferation in a time- and concentration-dependent manner. Specifically, oleocanthal, oleuropein aglycone and ligstroside aglycone were the most effective polyphenols among the OOPs tested. Oleocanthal and oleuropein aglycone caused the highest cell inhibition with EC50 values lower than 30μM, while ligstroside aglycone presented EC50 lower than 60μM. Most combinations of OOPs had a synergistic inhibitory effect, whereas OOPs' extracts showed a significant antiproliferative effect in all cancer cell lines. The triple negative MDA-MB231 cells showed the highest sensitivity to polyphenolic treatments. The mechanism of action of OOPs and polyphenolic extracts are currently under investigation.

P120. Evaluation of antiproliferative/cytotoxic action of olive oil phenolic ingredients on human melanoma cancer.

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Olive oil polyphenols (OOPs) have been associated with the prevention of many types of cancers. Recent studies have demonstrated that certain of these compounds such as hydroxytyrosol, oleocanthal, oleacein and oleuropein, inhibit cell proliferation and induce apoptosis in human melanoma, a highly invasive skin cancer. Due to the need for an effective treatment, we aimed to clarify the cytotoxic potential and the mechanism of action of single OOPs and their combination as isolated compounds and natural extracts.

We evaluated the antiproliferative/cytotoxic effect of OOPs on two metastatic human melanoma cell lines (SK-MEL-28 and A2058) as well as on normal human keratinocytes (HaCaT). Two-dimensional cell cultures were treated with increasing concentrations of oleuropein aglycone, ligstroside aglycone, oleacein, oleomissional and oleocanthal (10-100μM) and cytotoxicity was determined by an adenosine-triphosphate (ATP)-based luminescence assay. Half-maximal effective concentrations (EC50) were calculated in conditions of atmospheric O2 (20% v/v). Then, combinations of two, three or four different OOPs were examined for synergistic effect through 3-(4,5-*di*methylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) method. Finally, polyphenolic extracts of determined OOPs content were tested in order to simulate the olive oil effect on cell proliferation.

Our results reveal that single OOPs suppress proliferation of melanoma cells in a time- and concentration-dependent manner. Specifically, oleocanthal, oleuropein aglycone and ligstroside aglycone showed the strongest inhibitory effect among the OOPs tested. Interestingly, oleocanthal was the most effective polyphenol (EC50 <20μM), while aglycone oleuropein was the second one (EC50 <40μM). Most combinations of OOPs showed a synergistic inhibitory effect, whereas polyphenolic extracts had a significant antiproliferative effect on both cancer cell lines. The highly metastatic A2058 cells were more resistant to the effect of OOPs compared to SK-MEL-28. The mechanism of OOPs' action and polyphenolic extracts starting with early apoptosis and ending by the following necrosis, as well as actin cytoskeleton polymerization state, are currently under investigation.

P121. Unrevealing the hypoglycemic mechanism of *Eryngium cymosum* F. Delaroche.

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Type 2 diabetes (T2D) is a complex disease that negatively impacts overall metabolism due to insulin malfunction and deficient insulin secretion, leading to chronic hyperglycemia. Controlling blood glucose levels is one of the main goals of every therapeutic approach used to treat the disease in order to avoid complications that could result in premature death of T2D patients. In developing countries such as Mexico, diabetic patients consume infusions of medicinal plants to control this disease. In particular, the usage of the aerial parts of *Eryngium cymosum* F. Delaroche by locals of Tlanchinol and Huejutla de Reyes municipalities was detected in the Hidalgo state. Hence, we aimed to assess the hypoglycemic effect of its traditional extract and then it was evaluated on two of the main mechanisms responsible of fasting and post-prandial hyperglycemia: gluconeogenesis and carbohydrate breakdown by alpha-glucosidases. In addition, insulin was measured in the two conditions to stablish a link between the hypoglycemic effect and insulin-related mechanisms. The results showed that the *E. cymosum* extract exerted a significant hypoglycemic effect in STZ-NA rats that could be related to the inhibition of gluconeogenesis since the extract held a significant anti-hyperglycemic effect in pyruvate tolerance tests (AUC reduction of 38%). Moreover, it powerfully inhibited the activity of glucose-6-fosfatase and fructose 1,6-bisphosphatase (91% and 96%, respectively). On the other hand, it was able to significantly decrease the post-prandial hyperglycemia in maltose tolerance tests (AUC reduction of 46%) regardless of a modest inhibition of alpha-glucosidase activity (32%). Finally, insulin levels were unchanged after the administration of the extract indicating that its hypoglycemic effect is not associated to insulin-related mechanisms. In conclusion, the traditional consumed form of *E. cymosum* presents a hypoglycemic effect that is mainly related to its inhibitory capacity of gluconeogenesis, contributing to the reduction of fasting and post-prandial hyperglycemia.

P122. Amelioration of insulin resistance and inhibition of gluconeogenic rate-limiting enzymes by 3 Mexican medicinal plants.

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Type 2 diabetes (T2D) is a disease that affects the metabolism of carbohydrates, lipids and proteins partially due to insulin resistance. The liver of a diabetic patient produces 35 g extra of glucose per day because of hepatic insulin resistance; therefore, inhibiting the hepatic glucose production is a good target for the control of high glucose levels. In traditional medicine, *Ageratina petiolaris* (Ap), *Cecropia obtusifolia* (Co) and *Equisetum myriochaetum* (Em) have been used by people in some communities of the Hidalgo State in Mexico for the treatment of T2D. In previous works we demonstrate their hypoglycemic effect as well as the effect over gluconeogenesis in a pyruvate tolerance tests in STZ-NA hyperglycemic rats. Therefore, the aim of this study was to stablish whether this effect was caused by allosteric inhibition of rate-limiting enzymes of the gluconeogenic pathway. Additionally, the effect of the extracts on insulin resistance was assessed in STZ-NA hyperglycemic rats. The results showed that, after 42 days of treatment, extracts of Ap, Co and Em reduced both glucose and triglyceride levels of the hyperglycemic rats. The extracts also improved glucose tolerance by reducing the AUC glucose vs the control. Furthermore, Ap and Co reduced HOMA-IR indices. The enzymatic assays showed that while the extracts of Ap and Co inhibited the enzyme glucose-6-phosphatase (G-6-Pase) (IC50 = 223 and 146 µg/ml, respectively), Em did not. Moreover, fructose-1,6-bisphosphatase (F-1,6-BisPase) was inhibited by the three extracts (IC50 = 41, 238 and 64 µg/ml, respectively). In conclusion, the inhibition of gluconeogenesis exerted by these plants could be explained by the reduction of the activity of either G-6-Pase or F-1,6-BisPase and additionally they decreased insulin resistance, which contribute to diminish the glucose levels.

P123. Nerolidol induced apoptosis in blader carcinoma (TCC SUP cell line) by activation of p38 MAPK signaling pathway.

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Nerolidol is a sesquiterpene alcohol occurring in many essential oils. In recent years it has been a subject of numerous studies showing many beneficial properties which could be used to treat broad range of health problems including viral, bacterial, fungal and parasitic infections, inflammation and cancer. The aim of present work is to investigate the effect of nerolidol on human bladder carcinoma (TCC SUP cell line) and the pathway leading to apoptosis.

In order to assess the pathway leading to apoptosis of TCC SUP cell line induced by nerolidol, series of tests were performed. Cell proliferation assay, conducted by means of MTT staining, was used to determine cytotoxicity of nerolidol on investigated cell line. As a reference, well known chemotherapeutic drug cisplatin was used. To find out the mode by which nerolidol is decreasing the number of the cells, apoptosis quantification using Annexin/PI staining was used. In order to provide additional information about apoptosis induced in the cell culture, impact of nerolidol on two signaling molecules (Ca2+ and cAMP) was determined. To further demonstrate that apoptosis is the main way involved in nerolidol-induced cell death, immunostaining of p38 MAPK proteins was done.

Results suggest nerolidol induces apoptosis by activating p38 MAPK signaling pathway. Cell proliferation assay suggests nerolidol has a cytotoxic effect on tested cells in concentration and time dependent manner. Anexin/PI test done for nerolidol concentration of 0.05 mg/mL after 48-hour incubation, confirms that nerolidol induces apoptosis in TCC SUP cells. Immunostaining results show increase in active p38 enzyme forms suggesting inducement of apoptosis by this signaling pathway. Nerolidol induces a strong calcium release from endoplasmic reticulum after drug administration. Administration of nerolidol increases level of cAMP suggesting nerolidol activates adenylate cyclase.

P124. Chemical and Pharmacological Progress on a Tibetan Folk Medicine Formula *Bawei Chenxiang Powder.*

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Bawei Chenxiang Powder (BCP) is a traditional Tibetan folk medicine formula which has been used in clinic for more than 1300 years. It includes the resinous wood of *Aquilaria sinensis*, kernel of *Myristica fragrans*, fruit of *Choerospondias axillaris*, travertine, resin of *Boswellia carterii* or *B. bhaw-dajiana*, stem of *Aucklandia lappa*, fruit of *Terminalia chebula* (roasted), and flower of *Gossampinus malabarica*. BCP has the function of clearing heart heat, nourishing heart, tranquilizing mind, and inducing resuscitation in traditional medicine. Modern research shows that the chemical composition of BCP mainly contains terpenoids (sesquiterpenes, triterpenes), polyphenols (flavonoids, lignans), and tannins, which exert the pharmacological activities such as anti-myocardial ischemia, anti-cerebral ischemia, spatial learning and memory promotion. However, most chemical studies are the single herbs of BCP and lack of the overall analysis of this formula considering the cross-reaction of compound ingredients. Based on the multi-component, multi-path and multi-target effects, the modern technologies such as metabolomics, system pharmacology, system biology and others were used to investigate the material basis and mechanism of action of BCP. In short, as a typical representative of valuable resources of Chinese medicine, BCP is widely used in Tibetan and Mongolian traditional medicine that are worthy of further study and beneficial to the solution of cardiovascular and cerebrovascular diseases.

P125. Anti-ovarian cancer activity of Zaleya decandra against the human ovarian teratocarcinoma cell line PA1 by impairing mitochondrial activity.

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Ovarian cancer is a major challenge to public health and globally stands seventh in the most common cancers and eighth in most cancer deaths among women. Despite its rare occurrence, ovarian cancer remains the deadliest form of gynecological malignancy. Even though treatments such as surgery, chemotherapy, immunotherapy, radiotherapy are available, the percentage of adverse effects is light with low survival rate. In the era of new therapeutic drugs for various ailments, medicinal plants and natural compounds are promising alternatives for cancer treatment. Zaleya decandra is one such medicinal plant attracting researchers to explore the biomedical importance of the plant. Considering the pharmacological significance of Z. decandra, the current research work was put forth for the first time to validate the in vitro anti-proliferative activity of ethanolic root extract of Zaleya decandra (EEZD) against the human ovarian teratocarcinoma cell line (PA1). The preliminary phytochemical analysis showed the presence of alkaloids, terpenoids, phenolics, flavonoids, tannins, and sterols. The increase in the generation of ROS in Z. decandra treated cells indicate that the phyto-constituents present in the extract alone or in combination could have targeted this enzyme while exerting anti-proliferative effects. The mitochondrion membrane potential (MMP) in Z. decandra treated PA-1 cells using cationic fluorescent dyes displayed a drastic reduction in the rhodamine binding indicating reduced mitochondrial activity. ROS induced DNA damage by Z. decandra in ovarian cancer cell is evident as stained by AO/EtBr which could probably be mediated by ROS induced inhibition of PARP-1 by the constituents present in Z. decandra. Therefore the present study warrants extensive investigation using pre-clinical model system to substantiate the findings. Since the toxicological studies report the safety of Zaleya decandra, it promises to emerge as a possible alternative drug to treat ovarian cancer.

P126. Evaluation and analysis of clinical value of phytotherapeutic potential of natural herbal medicines in the treatment of asthma.

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Asthma is one of the most common chronic diseases with a multifactorial etiology that is characterized by chronic airway inflammation. Affecting approximately 358 million people worldwide. Over the past decades, the global prevalence, mortality, morbidity, and economic burden of asthma have increased. The treatment response for a particular patient may be difficult to predict due to the heterogeneous nature of asthma. For many decades, natural herbal medicines, polyherbal formulations and decoctions of plant-derived materials have widely been accepted as alternative complementary therapies for the treatment, cure or prevention of a wide range of chronic airway inflammation diseases including chronic sinusitis, chronic otitis media, chronic pharyngitis, chronic rhinitis, and chronic laryngitis and asthma. This review was aimed to summarize and critically discuss about the therapeutic viability and clinical applicability of natural herbal medicines for the treatment of asthma in human. The critical analysis of the literature revealed that oral and acupuncture application of natural herbal medicines exhibit remarkable potential for the treatment of asthma in adults. In this review, the clinical efficacy of various herbal formulations such as Chinese herbal formula therapies, single herb therapies, acupuncture and diet therapy for the treatment of asthma has been discussed. Conclusively, we recognized that natural herbal medicines demonstrate remarkable clinical efficacy when used alone or in conjunction with other complementary therapies for the treatment of asthma patients.

P127. Insights into the distribution and diverse applications of southern African indigenous Kei-apple as a potential candidate for commercialization.

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Globally, there is an increasing awareness of the importance of indigenous plants in new product development. However, existing basic and fundamental information on interesting indigenous plants need to be critically examined in order to provide a foolproof pathway for commercialization. Kei-apple known as *Dovyalis caffra* L. is a member of the family Salicaceae. Presently, Kei-apple is found in many southern African countries such as South Africa, Swaziland, Lesotho, Malawi, Mozambique, Namibia and Zimbabwe. Furthermore, the plant is widely reported in most of the provinces in South Africa. An estimated 28 different genotypes from different parts of South Africa are currently maintained in a National facility (Agricultural Research Council farm, Friedenheim, Nelspruit, Mpumalanga Province; 25.4884° S; 30.4028°) in South Africa. Kei-apple is recognized as an underutilized wild fruit with great potential and high commercial prospect. In ethno-medicine, the value of different parts of Kei-apple for treating different disease conditions in humans and livestock is well-recognized. Kei-apple is used for producing jelly jam, nectars and fruit leather and home-made beverage. As a result of its durability and presence of thorn, it is highly preferred for fencing (forming an impenetrable hedge) gardens in rural communities. Sales of the fruit also serve as source of income in some parts of South Africa. These aforementioned diverse applications provide the basis for more focused and co-ordinated research to fully explore the commercial potential locked in Kei-apple. Thus, critical aspects especially detailed basic research in the botany for selection of genotype(s) with desired traits is paramount. In addition, effective propagation approach/protocol and efficient orchard management need to be devised in order to optimize fruit yields for the anticipated commercialization demands.

P128. Evaluation of the antiproliferative effect of the *Pinaropappus roseus* Less. Phenolic aqueous extract on MCF-7 and HeLa cells.

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The biological activity of phenolic compounds against some cancer types has been reported. The use of *Pinaropappus roseus* Less. (*Asteraceae*) is advised by Mexican ethnopharmacology against some medical pathologies as inflammatory events, gastrointestinal and skin infections and this is was advised to use in some cancer pathologies. It must be noted that there is not scientific evidence to prove its therapeutic effect. The objective of this work was to evaluate the antiproliferative effect of the aqueous phenolic extract of *Pinaropappus roseus* Less. (APE) on MCF-7 and HeLa cells. The total phenols of the APE was 0.368±0.00 mg gallic acid equivalents g^{db-1} by Folin-Cicolteau method. The characterization of phenolic compounds by HPLC-DAD-MS2 allowed to identify six phenolic compounds: 3-hydroxybenzoic acid; 2′,6-dihydroxyflavone; 5,6,7-trihydroxyflavone; apigenin; 4′,5,7-trihydroxy-3′-methoxy-flavone and luteoline-7-glucoside .The antiproliferative effect (cell death) was evaluated by the violet crystal assay. The cellular inhibition was of 60% (24h) and 84%(48h) on MCF-7 cells; and of 60%(24h) and 55%(48h) on HeLa cells. The half maximum inhibitory concentration of aqueous phenolic extract (IC50) on MCF-7 was 2.24±.02µgGAE mL^{-1} and .047±.04µgGAE mL^{-1}, and 0.04±.02 µgGAE mL^{-1} on HeLa cells.

P129. Evaluation of the antimicobacterial activity of the aqueous phenolic extract of *Pinaropappus roseus* Less.

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One of the plants used in traditional medicine in the state of Oaxaca,Mexico is *Pinaropappus roseu*s Less.(Asteraceae); it is used as a topical infusion for the treatment of dermatological infections colonized by *Staphylococcus aureus*, and another microorganism such as *Candida albicans*. Currently, it is unknown if its effect is related to the inflammatory response or an antimicobacterial process. The objective of this work was to evaluate the antimicobacterial activity of the aqueous phenolic extract of the leaf tissue of *Pinaropappus* ro*seu*s (APE) on *Staphylococcus aureus* and *Candida albicans*. The total phenolic compound content of the PAE was 0.368±0.00 mg gallic acid equivalent/ g^{db}, quantificated by the Folin-Cicolteau method. The antimicobacterial activity of the APE was evaluated by the disk diffusion method, and the minimum inhibitory concentration (MIC) was determined by macrodilution method. The aqueous phenolic extract has not bactericidal activity. But this showed antifungal activity with a MIC of 0.296±02 mg/mL. The phenolic aqueous extract was evaluated against antibiotic as sulfamethoxazole and trimethoprim mix, ceftriaxone, benzyl penicillin and gentamicin (with inhibition percentages corresponding to 136%, 78%, 71% and 48% respectively). The characterization of the APE was carried out by HPLC-DAD-MS/MS^2 allowed the identification of six phenolic compounds (mainly flavones as Luteolin and apigenin).

P130. Phenolic metabolites profile of *Schisandra rubriflora* male and female plant raw material and their *in vitro* cultures.

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Schisandra chinensis (Turcz.) Baill. is an East-Asian medicinal plant species which fruits have been known in Europe from TCM as e.g. hepatoprotective, antioxidant, antitumour and adaptogenic phytopharmaceuticals [1]. Under our previous research of soil-grown plant material as well as in *in vitro* cultures of this species we studied phenolic acids and flavonoids [2].

Our current research deal with phenolic compounds biosynthetic potential of plant material and of *in vitro* cultures of less known Chinese endemic Schisandra species – *Schisandra rubriflora* Rehder & E.H.Wilson.

The aim of the study was chromatographic qualitative and quantitative analysis of fruit, leaf and shoot extracts of male (M) and female (F) specimens collected in different vegetation periods ("Clematis" company, Poland). The estimations were performed also in microshoot culture extracts (M and F lines) growing on agar Murashige-Skoog [3] medium with 1 mg/l 6-benzylaminopurine and 1 mg/l indole-3-butyric acid (30 days).

In the all analyzed extracts the presence of 27 phenolic compounds was confirmed by UHPLC-DAD-MS [4]. The quantification of 3 phenolic acids: chlorogenic, neochlorogenic and cryptochlorogenic acids, and 8 flavonoids: 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): 430.66 in fruits, 2814.42 (F) and 3452.16 (M) in leaves, and 3282.59 (F) and 2266.06 (M) in shoots.

The total content of estimated compounds in extracts from microshoot cultures were equal (mg/100 g DW): F – 515.49 and M – 631.48.

Our results documented for the first time, that both, *S. rubriflora* soil-grown plants and *in vitro* cultures, are a rich source of phenolic compounds of potentially high antioxidant properties.

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P131. Phenolic acid and flavonoid studies, and antioxidant activity of *Akebia quinata* steam and fruit extracts.

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Akebia quinata (Houtt.) Decne is an East-Asian origin, medicinal plant species which steams in Europe have become a new pharmacopoeial raw material since 2019 [1]. In traditional phytotherapy *A. quinata* is recommended as: diuretic, antiphlogistic and analgesic remedy. Moreover is used as the stimulant of the menstrual flow and to promote the milk-secretion [2].

Under our study we preformed the qualitative and quantitative HPLC-DAD [3] analyses of phenolic acids and flavonoids of steam and fruit extracts of *A. quinata* cultivated in Europe ("Clematis" company, Poland). In the both analyzed extracts the presence of 6 phenolic acids was confirmed: caffeic acid, cryptochlorogenic acid, 3,4-dihydroxyphenylacetic acid, isochlorogenic acid, neochlorogenic acid, and rosmarinic acid. Additionally, in the steam extracts the presence of 3 flavonoids was confirmed: isoquercetin, quercitrin and rutoside.

The highest total contents of estimated phenolic compounds was confirmed for steam extract – 4244.21 mg/100g DW for phenolic acids, and 1268.82 mg/100g DW for flavonoids. The main compounds (mg/100g DW) were: cryptochlorogenic acid (2045.13), neochlorogenic acid (918.14) and isochlorogenic acid (845.96).

The performed for studied extracts estimations with: CUPRAC (cupric ion reducing antioxidant activity), FRAP (ferric reducing ability of palsma), DPPH (1,1-diphenyl-2-picrylhydrazyl) and Folin-Ciocalteu (total polyphenol content) methods, reveal the high antioxidant potential of steam extracts.

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P132. Antiprotozoal activity of compounds isolated from the root bark of *Ziziphus jujuba*.

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Human protozoal diseases such as malaria, sleeping sickness, Chagas disease and leishmaniosis remain important health problems, with significant morbidity and mortality, particularly in tropical countries. For centuries, plants have been used by local populations to prevent and cure human diseases in many parts of the world. According to the WHO, approximately 80% of the African population uses traditional medicine for their health care [1]. In a screening of Niger plants for antiprotozoal activity, the dichloromethane extract of *Ziziphus jujuba* Mill. root bark exhibited inhibitory activities against *Trypanosoma cruzi* (IC_{50} = 7.4 µg/mL), *Leishmania donovani* (IC_{50} = 2.7 µg/mL), *Plasmodium falciparum* (IC_{50} = 1.7 µg/mL), and *Trypanosoma brucei rhodesiense* (IC_{50} = 10.2 µg/mL). *Z. jujuba,* commonly known as jujube, is traditionally used against diarrhea, ulcers, vomiting, indigestion, pulmonary ailments, dysentery and fever [2,3]. Eight cyclopeptide alkaloids and eight triterpenes were isolated from *Z. jujuba*. Among these, two compounds have not yet been described in the literature. The antiprotozoal activity was determined for thirteen of the compounds, and their cytotoxicity in L6 rat myoblast cells was measured. Mauritine M is a previously described cyclopeptide alkaloid and is the most active compound with IC_{50} values around 3-4 µM against *T. b. rhodesiense* and *P. falciparum*. Some of the triterpenes showed inhibitory activities against *L. donovani* (IC_{50} < 3 µM) and *P. falciparum* (IC_{50} < 0.2 µM). Active compounds should be further tested to assess their possible use as antiprotozoal drugs.

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P133. Mechanisms of *Helleborus cyclophyllus* Boiss apoptotic activity on A549 cancer cell line.

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Helleborus cyclophyllus Boiss, locally known as "skarfi", is a wild grown rhizomatous, poisonous plant of rocky Greek mountains, known for its pharmaceutical properties since ancient times. In a previous study its possible anticancer potential was examined. Morphological examination of exposed A549 cancer cells and primary lung fibroblasts to *H. cyclophyllus* methanol extracts, showed after appropriate staining, that the extract led selectively A549 cancer cells to apoptotic cell death, contrary to primary lung fibroblasts which were used as a model of normal cells. In particular, a few hours after treatment, the cell membrane of A549 cancer cells became waveform and intracellular material was exocytosed through vesicles (ectosomes). Subsequently, the cells were detached, adopted a round, swollen shape including a large vesicle into the cytoplasm, a structure looking like thanatosome. This structure marginated the cell nucleus at the periphery of the cell. Later, the apoptotic action was completed with the formation of numerous apoptotic bodies. In this work we first examined through MTT assay, the cytotoxic activity on A549 cancer cells after 24 h treatment with the extract, in comparison with primary lung fibroblasts. High cytotoxicity against A549 cancer cells was exerted, even at 0.02 mg mL^{-1}, while cleavage of procaspase 3 and PARP1 indicated apoptotic cell death. The apoptosis was mediated through the mitochondrial pathway as indicated by the cleavage of caspase-9 and not of caspase-8. Primary lung fibroblasts were resistant to treatment with *H. cyclophyllus* Boiss extract. Collectively, all our findings provide evidence for selective apoptotic activity of *H. cyclophyllus* Boiss extracts towards A549 cancer cells. Further investigation on the structure of bioactive compounds is of interest.

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P134. Do bitter phytocompounds target inflammation-related macromolecules?

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An association between the bitter taste of phytochemicals and their anti-inflammatory activity (AIA) was shown in previous studies [1,2]. In order to unravel the potential molecular background of this effect, SwissTargetPrediction was employed, an online tool developed by Swiss Institute of Bioinformatics that provides an estimation of the most probable macromolecular targets (MMT) of a small molecule [3]. Using BitterDB, a database of bitter phytocompounds [4], SuperSweetDB, a database of sweet phytocompounds [5], and some other sources, 141 bitter and 38 sweet phytocompounds were selected and fed to SwissTargetPrediction, which found a total of 1097 MMT for these bitter and sweet phytocompounds. The statistical analysis was performed by means of Fisher's exact test with the aim of finding out significant differences between the MMT of bitter compared with sweet phytocompounds. Given the 1097 calculations performed, the significance threshold was lowered (according to Bonferroni criterion) to 5E-5. As this resulted in only three MMT, the significance threshold was relaxed to 1E-4. Out of the 9 resultant MMT only one (brain glycogen phosphorylase) was preferentially targeted by sweet phytocompounds, while all the rest (epidermal growth factor receptor erbB1, matrix metalloproteinase 3, acetylcholinesterase, cytochrome P450 19A1, androgen receptor, cyclooxygenase-2, glycogen synthase kinase-3 beta, matrix metalloproteinase 9) were preferentially targeted by bitter phytocompounds. There is no information linking brain glycogen phosphorylase (the „sweet target") to a putative AIA, while all the „bitter targets" are more or less involved in AIA. Bitter phytocompounds (by comparison with sweet ones) have a higher probability of targeting molecules involved in anti-inflammatory pathways.

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P135. Bioassay-guilded isolation anticancer compounds from *Stahlianthus thorelii*.

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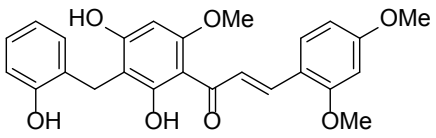
Stahlianthus thorelii Gagnep. is a medicinal plant used wildly to treat the diseases related to inflammation, cancer, and osteoarthritis. Bioassay-guided isolation of ethanolic extract of rhizomes of *S. thorelii* has yielded two new phenolic compounds, thorechalcone 1 and thorelignan 2, along with eight known analogs (3- 10). All the compounds were assayed for cytotoxicity on four human tumor cell lines (A549, MCF-7, WiDr, and HepG2); thorechalcone 1, showed the most anti-proliferative effect on HepG2 cells and was further evaluated its effect on cell cycle progression at 12.5 μM. Besides the structures of 1 and 3 were elucidated by IR, UV, and X-ray analyses, all the isolates were identified their structures by NMR and MS data. Moreover, the quantification of two major compounds 4 and 5 by using HPLC-DAD was also validated. This study presented the isolations of antiproliferative potentials of new chalcone and known flavonoid derivatives from S. *thorelii*. The validated simple, accurate, and rapid HPLC method could be deployed for the quality control of herbal drugs.



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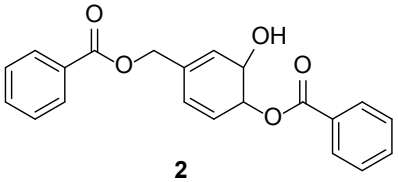

2

P136. Code of taste in traditional Chinese medicine - a genetic key?

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In Traditional Chinese Medicine (TCM), the taste of herbs is classified into five flavors, which are related to certain ethnopharmacological activities and target organs (sweet- Spleen/Pancreas, pungent- Lungs, salty- Kidney, sour- Liver, bitter- Heart). The ancient concept of taste-based selection of herbs was suggested to have a biological basis in the widespread extraoral distribution of taste receptors (TASR) and chemosensors (CHEMS) [1,2].

The aim of the present work was to check whether the TCM theory of flavors might have a correspondent in the tissue specific TASR gene expression levels and phenotypes.

A systematic search with bioinformatic tools [3-6] using the name of TASR and CHEMS genes as keywords, was performed.

The most striking finding was that among all the organs, the highest expression of TAS1R3 gene was reported in spleen (TAS1R3 is constituent of both sweet and umami TASR). Not unexpected, the highest expression of SCNN1A and SCNN1G (salty TASRs) was reported in the kidneys. Concerning the 25 bitter TASRs, only TAS2R19 and TAS2R38 expression was highest in the heart among the 5 TCM organs, but certain phenotypes (e.g. TAS2R16) were associated with cardiopathies. Only indirect data (e.g. phenotypes, eQTLs) supports the traditional association of pungency with lungs. No genetic evidence supports the association sour- liver.

In conclusion, at least some of the TCM taste-organ associations are supported by modern genome data. Further experimental studies are required to explore the potential biotechnological applications of these ethnopharmacological concepts.

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[3] Nat. Genet. 45 (2013) 580–585 (https://gtexportal.org)
[4] M. Uhlen et al, Science 347 (2015) 1260419 (www.proteinatlas.org)
[5] F. Bastian et al, Data Integr. Life Sci. (2008) 124–131 (www.bgee.org)
[6] Nucleic Acids Res. 47(2019): D506-515 (www.uniprot.org)

P137. Gallic acid inhibits melanogenesis by activating the autophagy system in B16F10 cells.

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Gallic acid (GA) is a natural phenol compound that is commonly found in green plants and fruits. It has been shown to have a variety of biological properties, including anti-oxidant, anti-cancer, anti-inflammatory and other activities. GA has been reported to inhibit melanin production via down-regulation of MITF and its downstream signaling pathway. In this study, we intended to delve into the molecular mechanisms involved in the anti-melanogenic activity of GA in induction of autophagy in melanoma B16F10 cells. We found that GA-induced autophagy was evidenced via increased LC3-II accumulation and acidic vesicular organelle (AVO) formation in B16F10 cells. These events are associated with decreased p62 and ATG4B, and dysregulated Beclin-1/Bcl-2 ratio. Further, GA was found to suppress the PI3K/AKT/mTOR signaling cascade, activating autophagy in B16F10 cells. Notably, pretreatment of cells with the autophagy inhibitors 3-MA diminished GA-mediated anti-melanogenic activity. Gallic acid is expected to become a significant skin-whitening agent in cosmetic applications in the future.

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P138. Bioassay-guilded isolation anticancer compounds from *Euphorbia neriifolia*.

S.S. Chang[1,2], N.-L. Nguyen[1,2], H. Thanh Vo[1,2], Y.-C. Lin[2], C.-C. Liaw[2,4], B.-H. Lee[5,7], C.-Y. Chung[5,7], M. Liu[6], Z.-H. Lin[2], C.-J. Tai[1,5,7], Y.-H. Kuo[1,2,3]

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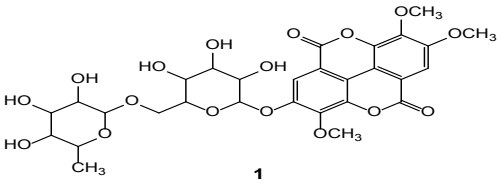
Euphorbia neriifolia, a medicinal herb widely distributed in Asian countries, used in folk medicine to treat the diseases related to inflammation, diabetes mellitus, and cancer. Bioassay-guided isolation of methanolic extract of stems of *E. neriifolia* using open columns, and RP-HPLC resulted in the isolation has yielded three ellagic acid derivatives, 3,3′,4-tri-*O*-methyl-4′-*O*-rutinosyl ellagic acid (1), 3,3′-di-*O*-methylellagic acid (2) and 3.3′.4′-tri-*O*-methylellagic acid (3), along with another six known compounds as (6S,9S)-roseoside (4), 6,7,8 trimethoxy coumarin (5), 7-hydroxy-6-methoxy-2*H*-chromen-2-one (6), Scopoletin (7), Kaempferol-3-*O*-rhamnoside (8), Indole carboxylic acid (9). All the compounds were evaluated by their inhibition of NO production in LPS-induced RAW264.7 macrophages and assayed for cytotoxicity on human tumor cell lines A549 and MCF-7; in there, three ellagic acid derivatives showed the anti-proliferative effect on A549 and MCF-7 cells. Base on the structures of compounds 1 to 3, we also found 3.3′.4′-Tri-*O*-methylellagic acid with the most anticancer effect. Notably, compound 3 demonstrated potent anti-NO activity (IC_{50} = 17.41 ± 0.73 µM) and cytotoxicity against A549 and MCF-7 with the EC_{50} value of about 58.14 µM with dose-dependent and led to apoptosis.



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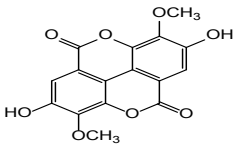

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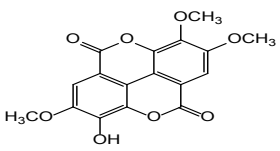

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Posters

Traditional use of plants and herbal pharmacopoeias the contribution of the Mediterranean flora

20th International Congress of the International Society for Ethnopharmacology

P139. Ethnopharmacological perspective on *Pulicaria* species (Compositae).

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The genus *Pulicaria* Gaertn. is represented about 127 species in the world. The main distribution areas are from Europe to North Africa and Asia, particularly around the Mediterranean. *Pulicaria* species are traditionally used in folk medicine. For instance, species are used for bronchitis, cold, inflamed wound in Turkey, as cicatrizant, antiinflammatory and for muscular- skeletal diseases, skin diseases in Spain, for diabetes, ankle sprain, headache, flatulent colic in Yemen, for abdominal pain in Mauritania. Besides that, some species are used in folk veterinary in Italy as antiparasitics, repellents and for respiratory ailments.

Different types of chemical compounds have been reported on the genus *Pulicaria*, such as phenolic derivatives, monoterpene derivatives, sesquiterpenes, diterpenes, flavonoids, triterpenes, steroids, essential oils, and others. Besides that flavonoids and sesquiterpenoids are the dominant compounds. A few studies refer to the composition of the essential oils of *Pulicaria* species. For examples, major compounds of essential oil of *Pulicaria* taxa in Turkey are found as cyclohexenylpiperidine, 7-methoxy-6-ethoxy-2,2dimethlchromanone, epibicyclosesquiphallandrene, 1,3,5-cycloheptatriene,2,3,4,5,7,7-hexamethyl and 1,3-cyclopentadiene,1,2,5,5 tetramethyl. On the other study, 2-isopropyl-4-methylphenol and isobutyric acid 2-isopropyl-4-methylphenylester are isolated from the essential oil of *P. odora*.

There are many biological activity studies on the genus *Pulicaria* in the literature. Different extracts of *P. dysenterica* are shown to be active against *Vibrio chlorea*. The antimicrobial activity of *Pulicaria* species has been studied against different microorganisms. Antiinflammatory activity of *Pulicaria wightiana* which is used as traditional folk medicine in India has been studied and found efficacy for treatment of inflammation.

In this presentation, recent studies on biological activity and traditional uses of *Pulicaria* species are evaluated.

P140. An ethnobotanical study on the traditional uses of medicinal and aromatic plants in the mountainous area of Troodos in Cyprus.

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Agricultural Research Institute, Nicosia, Cyprus

Troodos mountain range can be considered as a cradle of biodiversity for the island of Cyprus and local people as the guards of an important portion of the island's cultural heritage. Medicinal and aromatic plants (MAPs) are a distinctive component of the Mediterranean vegetation and their traditional uses a valuable cultural element that should be safeguarded and promoted. The aim of this ethnobotanical study was to collect and document information about MAPs traditionally used in Troodos and identify the most popular and important taxa. Semi-structured interviews were conducted in 2019–2020 and exponential discriminative snowball sampling was employed for candidate respondents' detection and selection. The knowledge on the traditional plant uses obtained through interviews was classified in general and medicinal use categories, whilst the most popular and important taxa were identified using well-known ethnobotanical indices. Through the study, it was realized and documented that MAPs had a vital role in the daily life of the local people in the past. It was also realized that the transmission and adoption of the knowledge regarding traditional MAPs uses between successive generations (intergenerational continuity) diminishes in time. However, society's shift towards a healthier lifestyle implies the importance of documenting traditional uses in combination with the current scientific knowledge on MAPs appropriate utilization.

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