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BOOK OF ABSTRACTS

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ORAL PRESENTATIONS

OP1-Investigating the interaction of *Humulus lupulus* L. bitter compounds and gastrointestinal bitter taste receptors: *in vitro* and *in silico* study

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Keywords: *Humulus lupulus* L., Bitter Compounds, Bitter taste receptors, Gut Hormones

Bitter taste receptors (T2R in humans and *Tas2r* in mice) are G protein-coupled receptors (GPCRs) mainly linked to the detection of bitter taste in the mouth, however, recent studies have demonstrated their expression in various other tissues. Specifically, in the gastrointestinal tract the activation of T2R can determine the release of different enterohormones such as glucagon-like peptide 1 (GLP-1) and cholecystokinin (CCK) involved in regulating satiety and coordinating digestion. These enterohormones indeed play an essential role in gastric emptying, food absorption, as well as overall metabolic homeostasis, representing a potential therapeutic target for preventing overweight and obesity [1]. Considering this background, it was decided to study the bitter molecules from *Humulus lupulus* L. inflorescence as promising candidates for the prevention or treatment of overweight and obesity by regulating T2R activation thereby inducing GLP-1 and CCK secretion. *H. lupulus* bitter compounds, are α-acids (*n*-, co-, and ad-humulone, co-), and β-acids (*n*-, co-, and ad-lupulone) [2]. To date, only few studies have focused on the recognition of T2R by hop-bitter molecules, demonstrating their involvement in activating T2R1, 10, 14, 40, and 46 [1]. For this reason, this study aims to investigate the implication of hop bitter molecules such as α-acids and β-acids in inducing anorexigenic hormones secretion in intestinal STC-1 cells. Semi-preparative HPLC was applied to obtain pure bitter compound fractions from the ICE-4 standard mixture. Four fractions (FR) were obtained: FR I co-humulone (77.43 mg), FR II *n*-humulone+ad-humulone (106.45 mg), FR III co-lupulone (11.71 mg), and FR IV *n*-lupulone+ad-lupulone (18.98 mg). Due to their similar chemical properties and same retention time, for either α- or β-acids, it was not possible to separate the *n*- from the ad-forms. *In vitro* investigation in intestinal STC-1 cells, demonstrated that all bitter compounds induced the secretion of the anorexigenic hormones, GLP-1 and CCK. To show whether this action, induced by the molecules, could be related to increased expression of one of the most highly expressed bitter receptors in the intestine (*Tas2r*138), qRTPCR was used. It was demonstrated that all molecules increased the expression of *Tas2r1*38. Hence, computational studies were performed on *Tas2r*138 and its human ortholog T2R38 to predict the possible interaction with the receptor. Molecular docking experiments showed that all bitter molecules should be able to bind the active site of both bitter receptors, providing the basis for applying hop bitter compounds as lead molecules to further design gastrointestinal permeable T2R agonists.

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OP2-Natural sources of novel insecticides: the case of *Commiphora myrrha* (T.Nees) Engl. essential oil and its furanosesquiterpenoids

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The widening spread of vector-borne diseases (VBDs) is among the main causes of public health concern worldwide and the control of vectors, mainly represented by mosquitoes, remains still an everlasting and evolving challenge [1]. In this context, health organizations are progressively encouraging the research for novel and effective tools, being compounds deriving from botanical sources potential alternatives to conventional insecticides [2]. In this regard, tropical or subtropical plants turned out to be rich sources of bioactive compounds. *Commiphora myrrha* (T.Nees) Engl. (Burseraceae) is a medicinal plant especially employed for its oleo-gum resin, used for centuries as incense and for medicinal purposes, such as an antiseptic, emmenagogue, antitussive, astringent, antispasmodic, and antiparasitic agent. The oleo-gum resin is constituted of a volatile fraction obtainable under the form of essential oil (EO), mainly characterized by the furanosesquiterpenes furanoeudesma-1,3-diene (Fig. 1), isofuranodiene, curzerene, and lindestrene. Few studies investigated the insecticidal activity of *C. myrrha* extracts against mosquitoes and none explored the activity of its EO. In the presented work, the *C. myrrha* EO was chemically analyzed by GC-MS analysis, and the main furanosesquiterpenes quantified by HPLC-DAD to avoid thermal degradation of the major constituents. The EO insecticidal potential was assessed on four diverse mosquito species, namely *Aedes albopictus* Skuse, *Ae. aegypti* L., *Anopheles gambiae* Giles and *An. stephensi* Liston, demonstrating a promising larvicidal effect (LC_{50} 16.80, 4.42, 10.82, and 12.57 µg/mL, respectively). Then, the EO was fractioned and the resulting two fractions (i.e., sesquiterpenes and furanosesquiterpenes) were tested on the four mosquito species to investigate the involvement of the furanosesquiterpenes in the insecticidal activity. Indeed, only the fraction containing the above-mentioned compounds showed a larvicidal potential (LC_{50} 5.04, 3.72, 3.49, and 3.91 µg/mL). Finally, the three main furanosesquiterpenes of the latter, i.e., furanoeudesma-1,3-diene, isofuranodiene, and curzerene, were purified, characterized, and tested on larvae of *Ae. aegypti*, the most sensitive species to the EO. Furanoeudesma-1,3-diene was the most active compound (LC_{50} of 3.28 µg/mL), followed by isofuranodiene (LC_{50} of 5.58 µg/mL), and curzerene (LC_{50} of 7.44 µg/mL). These results demonstrated for the first time the outstanding potential of *C. myrrha* EO on vectors of pathogens of medical and veterinary importance and that the insecticidal activity is primarily linked to the presence of furanosesquiterpenes. All the products were also tested on human non-tumoral embryonic kidney 293 cell line (HEK293), demonstrating a moderate toxicity for the EO. Toxicity assessment on non-target organisms and in field efficacy validation are still required. Nevertheless, this work supports the potential of botanical sources for the discovery of bioactive compounds to be included in the complex and challenging management of VBDs.

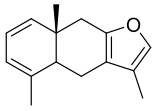

Fig. 1. Structure of furanoeudesma-1,3-diene.

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OP3-Polyphenolic composition, antioxidant and anti-tyrosinase activities of *Ficus rubiginosa* Desf. ex Vent. extracts based on the impact of its maturity stages

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Keywords: *Ficus rubiginosa;* polyphenolic compounds; antioxidant activity; anti-tyrosinase activity.

The *Ficus* genus (Moraceae) includes over 800 species, comprising woody deciduous or evergreen trees, shrubs, and herbs. The majority of species within this family are native to tropical and subtropical regions. *Ficus* spp. are frequently employed as a foodstuff and in traditional medicine [1,2]. Their biological activities as anti-inflammatory, diuretic, wound-healing and antimicrobial agents have been the subject of extensive review. Despite the extensive literature on *Ficus* spp., there is a lack of research on the phytochemistry of *F. rubiginosa* Desf. ex Vent. [3]. In order to fill this gap, the present study aimed to investigate the polyphenol content, antioxidant and anti- tyrosinase properties of the extracts from *F. rubiginosa*. *F. rubiginosa*'s leaves were collected at three different maturity stages (H1, H2, and H3) and methanolic extracts were evaluated for total phenolic content (TPC), total flavonoid content (TFC), and total catechin content (TCC). The polyphenolic profile was studied by using HPLC-UV/DAD and HPLC-MS/MS analyses and the antioxidant activity was determined in vitro using DPPH, FRAP, and ABTSP assays. With the results obtained, it was demonstrated that the maturity stage exerts a significant influence on the polyphenolic content, with the extract derived from the July harvest (H2) exhibiting the highest TPC and TFC values (113.50 mg GA/g and 43.27 mg QE/g, respectively) and notable antioxidant activity. Therefore, the potential of the constituents from H2 extract against tyrosinase was investigated using the Target Binding® approach. The results demonstrated that H2 was capable of binding and inhibiting tyrosinase, with rutin being the main compound responsible for the observed activity on the enzyme. This evidence identified rutin as a prominent anti-tyrosinase agent, indicating that methanolic extracts of *F. rubiginosa* may offer a beneficial approach for the management of various skin conditions associated with tyrosinase activity.

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OP4-Chemical and biological characterization of *Salvia discolor* exudate for management of phytopathogens and pests

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Keywords: *Salvia discolor*, phytopathogens, pests, diterpene

Phytopathogenic fungi, bacteria and pests cause damage to crops, and result in economic losses. Current control of phytopathogens and pests depends on synthetic pesticides resulting in damage to the environment, and increased resistance. Considering these challenges there is more interest in exploring natural sources as safe and sustainable alternatives. The present study aims to characterise chemically and biologically the dichloromethane extract of aerial parts of *Salvia discolor* Kunth. Phytochemical investigation reveals the presence of one novel clerodane diterpene along with some known compounds 8,3'-dihydroxy-6,7,4'-trimethoxyflavone[1], 5,7-dihydroxy-3,4'-dimethoxyflavone[2], divinatorin A[3] and patagonic acid[4], which were identified through spectroscopic 1D and 2D NMR analysis. Crude extract was evaluated for its antimicrobial potential against various phytopathogenic fungi, bacteria and pests. The extract exhibited MIC values of 500 µg/mL and 1000 µg/mL against *Clavibacter michiganensis* subsp. *michiganensis*, and *Pectobacterium carotovorum* subsp. *carotovorum*, respectively. *In vitro* antifungal activity varied across different fungi at concentrations of 5, 100, 250, 500, 750 and 1000 µg/mL. The extract showed complete inhibition of mycelial growth of *Phaeomoniella chlamydospora* at 1000 µg/mL statistically equivalent to synthetic fungicide and aligned with previous research findings [5]. It shows strong activity against *Pythium dissotocum,* and *Fusarium solani*. Moderate activity was reported against *Phoma betae*, *Alternaria solani*, *Stemphylium sp*. and *Botrytis cinerea*. However, the lowest inhibition (<40%) was observed against *C. lindemuthianum* and *F. oxysporum* fsp. *lactucae* race 1. The extract also showed promising results against gray mold disease in post-harvest tomato fruit. Furthermore, bioinsecticidal activity was measured against *Tuta absoluta* [6] which exhibited an LC_{50} of 0.013 mg/mL. These results highlight the potential of surface extract of *S. discolor* as a natural source of new bioactive compounds and management of phytopathogens and pests contributing to sustainable agricultural practices.

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OP5-Structure elucidation and antioxidant activity of five new flavonol glycosides from *Atriplex halimus* L. aerial parts

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Keywords: *Atriplex halimus*, Halophyte, Flavonoids, Antioxidant activity

Atriplex halimus L. (Amaranthaceae) is a perennial halophytic shrub mainly distributed in saline and desertic habitats due its high tolerance to harsh soil and climatic conditions[1]. Fatty acids, triterpenoid saponins, alkaloids, phenolic acids, and flavonoid glycosides with potential biological activities have been previously reported as chemical constituents of *Atriplex* genus[2]. Moreover, in North African folk medicine, the use of *A. halimus* aerial parts has traditionally been linked to the treatment of several diseases such as inflammation, hormonal disorders, thyroid issues, and diabetes[3]. In light of these properties, the aim of this work was to study the polar extracts of *A. halimus* aerial parts collected near Livorno (Italy) and assess their antioxidant activity. The powdered dried aerial parts of *A. halimus* L. were extracted with solvents of increasing polarity, including *n*-hexane, $CHCl_3$, $CHCl_3$-MeOH (9:1), and MeOH. The chemical composition of all the extracts was investigated through a preliminary UHPLC-HR-ESI-Orbitrap/MS analysis, showing a series of glycosylated flavonoids previously unreported for this species in the MeOH extract. Thus, the MeOH extract was partitioned between *n*-BuOH and H_2O to give an *n*-BuOH residue, which was then subjected to size exclusion and RP-HPLC chromatographies to yield five new and three known flavonol glycosides. The chemical structure of all isolates was elucidated by spectrometric and spectroscopic analysis. New compounds showed quercetin and isorhamnetin as aglycone, with long, branched, or linear sugar chains, sometimes esterified with ferulic or *p*-coumaric acid. Finally, the antioxidant properties of *A. halimus* were investigated through cell-free assays, showing a good antioxidant potential for the plant extract and a significant activity for the isolated compounds. In conclusion, all these results suggest that this species could be an interesting research candidate for developing new antioxidant plant-based products.

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OP6-Evaluation of the activity of *Salvia interrupta* extracts against *Botrytis cinerea* on tomato

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Keywords: *Salvia interrupta*, *Botrytis cinerea*, antifungal activity

Phytosanitary defense encompasses a set of techniques aimed at protecting crops from pests. In the last decades, the optimism generated by the widespread and indiscriminate use of synthetic phytosanitary products has been tempered. Today, more environmentally friendly methodologies are gaining ground, such as integrated pest management and organic farming [1]. In the last century, it has been recognized that plant secondary metabolites play a fundamental role in plant ecophysiology. They have both a defensive role against pathogen attacks and an attractive role for beneficial organisms such as pollinators or symbionts. Thanks to these characteristics, several secondary metabolites have found application as effective starting points for discovering new phytosanitary agents [2].
Salvia is the largest genus of the Lamiaceae family, consisting of about 980 species. *Salvia* spp. contain many health-promoting phytochemicals among which polyphenols, flavonoids and terpenes [3]. For this reason, in this study, we evaluated the effectiveness of two different extracts (called "exudate" and "matrix" respectively) of *S. interrupta* against *Botrytis cinerea*, a pathogenic fungus responsible for gray mold, which causes extensive damage to a wide range of crops. Exudate was obtained immerging fresh sage leaves in dichloromethane for twenty seconds, to obtain the isolation of the leaf surface constituents, while matrix was the result of methanol extraction of the previously used dried leaves. The test was conducted in *semi-vivo* mode, on artificially infected tomato fruits, with the aim of analyzing the effectiveness of extracts at different concentrations (100, 500 and 1000 ppm). For this purpose, an incision was made on tomato fruits, subsequently treated with *S. interrupta* extracts. Then, the lesion was inoculated with *Botrytis cinerea*. The commercial products Teldor plus (fenhexamid 42,7%) and Switch (cyprodinil 37.5% + fludioxonil 25%) were used as reference standards.
Results showed that both exudate and matrix have significant bioactivity. A clear dose-dependent trend emerged between 100 and 500 ppm. However, at 1000 ppm, a reduction in efficacy was observed, presumably due to phytotoxicity effects.
Based on these first observations, *S. interrupta* appears to possess activity against *B. cinerea*, and deserves further study.

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OP7-Baharat: traditional middle eastern spice blends as potential antimicrobial food preservative

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Keywords: Baharat, antimicrobial, metabolomics, ICP-QQQ-MS.

For centuries, herbs and spices have been valued for their health benefits and their use as natural preservatives, flavour enhancers, and therapeutic agents. Many spices also have antimicrobial properties, making them effective in food preservation [1].
Baharat, a spice blend in Middle Eastern cuisine, highlights these qualities. Its base ingredients are black pepper, cinnamon, clove, and allspice, but regional variations exist: Lebanon adds cumin, ginger, and red pepper (blend 1); Turkey includes garlic and mint (blend 2); North Africa and Tunisia use cumin and turmeric (blend 3); and the Persian Gulf features black dry lime (blend 4).
In this study, the antibacterial activity of these four Baharat blends (1-4) was assessed by testing EtOH:H_2O extracts (3:7 v/v) against both Gram-positive and Gram-negative bacteria. Blends 2 and 4 showed significant inhibitory effects against Gram-positive bacteria, particularly *Staphylococcus aureus* and *Listeria monocytogenes*, with minimum inhibitory concentrations (MIC) of 0.390 and 3.125 mg/mL, respectively. Blend 4 was also effective against *Salmonella typhimurium* (MIC = 6.25 mg/mL). The antibacterial activity was generally less pronounced against Gram-negative bacteria, except in the case of blend 4.
Metabolomic analysis of the two most active blends (2 and 4), conducted using UHPLC-HR-Orbitrap/ESI-MS, identified 123 bioactive compounds (Fig 1). Both blends contained phenolic and hydroxycinnamic acids, flavonoids, and piperamides. In blend 2, which included garlic, organosulfur compounds were also detected, meanwhile, blend 4 contained limonoids, due to the presence of black dry lime [2]. Finally, ICP-QQQ-MS was used to analyse 12 trace elements in the spice blends, and their concentrations were compared to FAO/WHO safety limits [3].The detected levels were as follows: Al ≤ 49, Cu ≤ 9.9, Fe ≤ 588, Mn ≤ 484, Zn ≤ 17.4 mg/L; As ≤ 38, Cd ≤ 207, Cr ≤ 636, Hg ≤ 10, Ni ≤ 1266, Pb ≤ 1548, Sn ≤ 60 µL/L. All concentrations were within or below the recommended safety limits. In conclusion, this study highlights the antimicrobial potential of traditional Baharat spice blends and underscores their relevance in both food preservation and safety.

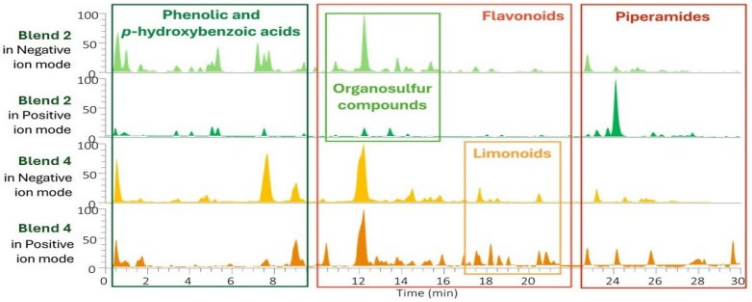

Fig 1. UHPLC-HR-ESI-Orbitrap/MS profiles of extracts blend 2 and 4 recorded in negative and positive ion modes.

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OP8-Vesicle-based formulation to optimize cannabidiol delivery

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Keywords: Cannabidiol, Nanovesicles, Drug Delivery, CBD Permeation

Cannabidiol (CBD), a natural constituent of *Cannabis sativa* L., has gained considerable scientific attention in recent years due to numerous biological and pharmaceutical properties. However, its oral administration is hindered by its very low water solubility (0.1 µg/mL) and extensive first-pass metabolism, which significantly reduce therapeutic efficacy [1]. To overcome these limitations, vesicles (VS) were developed to improve the solubility and absorption of CBD. These vesicles are bilayered structures loaded with 1 mg/mL of CBD (VS-CBD) and contain a softening compound, polysorbate 20 (T20), which allows the nanosystem to deform and pass through mucosas including nasal mucosa, thus enhancing drug permeation. [2] Physical characterization was caried out by dynamic/electrophoretic light scattering and extrusion was used to test deformability of the vesicles. A validated HPLC method was used to evaluate recovery, ebcapsulation efficiency and stability of loaded CBD. VS and VS-CBD showed the following chemical and physical properties:

	T20 (mg/mL)	CBD (mg/mL)	Size (nm)	Polidispersity Index	Z-potential (mV)	Recovery percentage (%)	Encapsulation Efficiency (%)
VS	10	-	67.72±0.84	0.288±0.008	-32.13±0.80	-	-
VS-CBD	10	1	65.27±1.27	0.230± 0.005	-30.31±0.54	99.89±0.52	96.80±0.96

VS-CBD remained stable during a one-month storage period. Since T20, as a surfactant, could also produce micelles, the formation of the bilayer was observed by SAXS analysis and the morphology of vesicles was observed by Scanning and Trasmission Electron Microscopy.
The study evaluated the performance of CBD and CBD-loaded in vesicles to cross the blood-brain barrier (BBB) through permeation experiments performed for four hours in the HCMEC/D3 cell line. After assessing cytotoxicity and cell viability, the non-inferiority of the vesicles in terms was evaluated. Apparent permeability was higher for VS-CBD than for free CBD (F-CBD) at the two CBD's concentrations tested (10 and 20 µM) and was significantly higher at the lowest concentration tested (10 µM); the values for VS-CBD at 10 and 20 µM were $(4.76\pm1.05)*10^6$ cm/s and $(4.46\pm0.93)*10^6$ cm/s, respectively, while values for F-CBD were $(3.21\pm0.08)*10^6$ cm/s and $(3.37\pm0.30)*10^6$ cm/s, respectively. In general, a lower accumulation of CBD in the lysate was observed in the case of the formulations. Analyses were performed by liquid chromatography coupled to tandem mass spectrometry. Liquid chromatography coupled with tandem mass spectrometry was used for analysis, where the mass spectrometer detected positive ion signals in multiple reaction monitoring (MRM) mode, focusing on three fragment ions of the protonated CBD ion (m/z 315.3): 1) 259.0 m/z, 2) 193.1 m/z, and 3) 134.9 m/z.

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OP9-Untargeted metabolomic analysis of *Morus alba* L. twigs extract and antibacterial evaluation of isolated metabolites

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Keywords: *Morus alba* L., antimicrobial activity, metabolomics.

Antimicrobial resistance (AMR) is a growing global concern, resulting in persistent infections, increased spread of disease, and higher mortality rates [1]. Plant natural products are promising tools in the fight against multi-drug resistant bacteria due to their diverse mechanisms of action, ability to overcome resistance mechanisms, potential for synergistic effects with existing antibiotics, and the opportunity to discover novel antimicrobials [2]. As antibiotic resistance continues to rise globally, integrating plant-derived compounds into antimicrobial strategies could be pivotal in addressing this pressing health challenge.
Morus alba L. (Moraceae), commonly known as white mulberry, is a deciduous tree native to China but widely cultivated worldwide. It offers potential health benefits, including antioxidant, anti-inflammatory, and antimicrobial properties. Rich in phytochemicals, it holds promise for managing diabetes and improving cardiovascular health, with ongoing research into its broader applications [3]. Here, we reported an untargeted metabolomic profiling and phytochemical investigation of a *M. alba* twigs extract, based on a LC-HRMS/MS and Molecular Networking combined approach. Our analysis led to the isolation of 17 secondary metabolites that were tested for their antimicrobial activity against *Staphylococcus spp.* and *Candida albicans*, highlighting the *M. alba* twigs as a valuable biomass with significant pharmaceutical potential. Due to the high chemical similarity among the isolated metabolites, structure-activity relationships for these versatile scaffolds were proposed.

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OP10-Callus culture of *Mespilus germanica* L. (Rosaceae): a biofactory for bioactive compounds

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Keywords: *Mespilus germanica*, callus biomass, LC-HRMS/MS, NMR, metabolomics

The medlar (*Mespilus germanica* L., Rosaceae) is a fruit that has experienced a decline in consumption in favour of the more cultivated *Rhaphiolepis bibas*, despite having been a popular food item in the past. Recent studies have shown a renewed interest in medlar due to its functional properties [1]. The in vitro culture of plant cells and tissues represents a promising and sustainable method for the production of bioactive compounds, which can be achieved while minimising the impact of factors such as pollution, drought, seasonal changes and the plant safety. Among these *in vitro* techniques, callus culture is particularly noteworthy in that it can serve as a precursor for large-scale cell culture [2]. The aim of this study was to establish a protocol for the production of medlar callus biomass and to analyse its chemical composition. It is noteworthy that this study employed ripe medlar pulp as the source of explants for callus induction, which is a novel approach in this field. After optimising the conditions for biomass growth, the metabolomic profile of the callus was characterised and compared with that of the fruit pulp and peel. The integration of MS and NMR data revealed that the callus is rich in ursane and oleanane triterpenoids, in contrast to the pulp and peel. The presence of flavonoids, amino acids, sugars, and phenolic acids was also confirmed. The high concentration of specialized metabolites, especially pentacyclic triterpenes, highlights the callus's bioactivity potential and its promise as a bioreactor for producing targeted functional compounds. Based on the high number of triterpenoids detected in the callus culture, its antimicrobial activity was investigated by the disc diffusion method against some bacterial strains that are common food contaminants: *Acinetobacter baumannii*, *Enterococcus faecalis*, *Escherichia coli*, *Listeria monocytogenes*, *Salmonella* sp., *Shigella* sp., *Staphylococcus aureus*. The callus showed an interesting activity against S. aureus. In the view of these results, the assay of the biofilm formation, a major virulence factor of bacteria, was performed. The callus can inhibit biofilm formation induced at different concentrations of glucose and sucrose. These results suggest that the callus from *M. germanica* represents a future resource as bioactive triterpenoids.

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OP11-Microplastics: an overview of their effects on photosynthetic organisms and the need to include lichens

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Keywords: microplastics, phytotoxic, lichen

The issue of plastics, microplastics (MPs) and related pollution is of growing global concern due to their potential impact on living organisms and human health. These small pieces of plastic waste less than 5mm in length have attracted increasing attention for their potential impact on terrestrial ecosystems, particularly on plant growth and performance. MPs and nanoplastics (NPs) have been shown to enter plant systems through root uptake, which is facilitated by their small size. Their effects on higher plants vary depending on plastic properties, plant species and environmental factors, and they can affect plant fitness in different ways and with different outcomes. Studies have found that MPs and especially NPs have inhibitory and phytotoxic effects. These include inhibition of root growth, cytotoxicity, genotoxicity, oxidative stress, inhibition of seed germination, alteration of antioxidant enzyme activity, reduction of photosynthetic activity, inhibition of gene expression, alteration of normal metabolism, reduction of biomass. Studies have been carried out on various photosynthetic organisms, including algae, macrophytes and vascular plants, including plants of phytochemical interest, but there are no studies on the effects of microplastics on lichens. Lichens have proven to be optimal biomonitors in air pollution studies, both in bioindication and bioaccumulation studies, and they are widely used as accumulators of persistent pollutants, such as metals, radionuclides and PAHs. However, these organisms are not exempt from microplastics pollution, particularly airborne microplastics, as our research has shown. In this study, airborne MPs pollution was investigated using transplants and native samples of the fruticose lichen *Evernia prunastri* in urban and remote sites. Lichen transplants were exposed outdoors for 7 weeks in parking lots and urban parks in the town of Pisa (Italy); in parallel, autochthonous samples were harvested from remote areas of the same region. The overall aim was the characterization of MPs in terms of number, shape, size and polymer composition under different experimental conditions. Microplastics were found in all lichen samples, especially fragments, fibres and tyre wear particles. The average number of MPs expressed per gram of dry lichen increased from remote areas (2 MPs/g dw) to urban parks (7 MPs/g dw) and parking lots (16 MPs/g dw). Average daily MPs deposition rates in the urban area were estimated in the range 12–143 MPs/m2. No differences emerged when comparing the length of the fibres between parking lots and urban parks, while longer fragments and shorter tyre wear particles were found in parking lots. Polyethylene terephthalate (PET) was the polymer most frequently detected in the samples.

OP12-Secondary metabolites from *Origanum majorana* L. induce bioenergetic dysfunction and redox homeostasis dysregulation in colon cancer cells

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Keywords: Lamiaceae, polyphenols, cancer, rosmarinic acid

Origanum majorana L. is a perennial hemicryptophyte plant belonging to the Lamiaceae family. Its average height ranges from 20 to 60 cm. Originating in the Cyprus area, the plant is grown throughout the Mediterranean region and is highly valued for its aroma and health-promoting phytocompounds. The most intriguing among them is rosmarinic acid (RA), a typical compound in the Lamiaceae family with antioxidant, anti-inflammatory, and anti-cancer properties [1].

Two extracts from *Origanum majorana* L. leaves and flowers (MF) and twigs (MT) were obtained by applying a green extraction technique (solvent: EtOH/H2O 50:50; temperature 40°C; time 4h; ratio 1:30 g/mL). The phytochemical analysis performed by HPLC-ESI-MS analysis showed that the two extracts have identical profiles, composed of 21 different identified compounds, that differ only from the quantitative point of view. Subsequently, we characterised MF and MT also for their antioxidant properties in *in vitro* cell-free systems: DPPH test, SOD-Like activity assay, and Catalase-Like activity assay. The results revealed that the two extracts possess similar antioxidant activity in these tests. Then, we tested MF and MT for their cytotoxic activity on four cancer cell lines (CaCo-2; HepG-2; A549; MCF-7), finding that CaCo-2 cells were the most sensitive to the treatment, with MF as the most effective. Since mitochondrial functionality is known to be a potential target of natural compounds in colorectal cancer toxicity [2], we investigated this possible mechanism of action mediated by MF extract on CaCo-2 cells. For this purpose, we performed Mitostress test using the Seahorse XFe24 Analyzer, which revealed that the extract interferes with mitochondrial respiration and lactic acid efflux, two parameters critical for cancer survival and proliferation. Coherently, we observed that MF exerts a dose-dependent inhibition of lactic dehydrogenase activity, and a reduction of glucose consumption rate and intracellular pyruvate levels after 6h, and of intracellular ATP after 24h of treatment. The impairment of mitochondrial respiration, probably, is the cause of the increased ROS levels detected over a period of 24h of MF exposure and the consequent dysregulation of cellular redox homeostasis. We also demonstrated that MF directly supplies electrons to cytochrome C in an *in vitro* cell-free model, suggesting a possible interference with the mitochondrial electron transfer chain. Lastly, the specific modulation of the expression levels of BCL-2 and BAX induced by MF exposure suggested that mitochondrial dysfunction and oxidative stress trigger apoptotic cell death in CaCo-2 cells.

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OP13-Spinach baby leaves and “Carciofo Bianco di Pertosa” leaves as source of functional ingredients

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Keywords: eco-sustainable extracts, *Spinacia oleracea* L. baby leaves, “Carciofo bianco di Perosa” leaves, bioactives

Vegetable and fruit byproducts are recognized as having health-promoting effects due to the functional properties of their nutrients and specialized metabolites [1]. Herein, *Spinacia oleracea* L. baby leaves and “Carciofo bianco di Pertosa” leaves, as source of bioactives, were investigated, by green chemistry approaches.
Conventional and unconventional eco-sustainable extractions of *S. oleracea* L. baby leaves were performed using EtOH and EtOH: H_2O mixtures as solvents. The obtained extracts were analysed by LC-ESI/HRMSMS. In this way, 42 compounds, mainly flavonoids, and 20-hydroxyecdysone were identified along with primary metabolites detected by NMR analysis. In order to highlight the chemical differences between the extracts, LC-ESI/HRMS and NMR data were analysed using Principal Component Analysis. The results revealed how 20-hydroxyecdysone and flavonoids occurred in the highest amount in the EtOH and EtOH:H_2O (70:30) extracts obtained by SLDE-Naviglio extraction, respectively. Considering the interesting biological activity reported for 20-hydroxyecdysone [2], it was quantified the extracts by LC-ESI/QTrap/MS/MS using the MRM method. Our attention was focused on “Carciofo Bianco di Pertosa” leaves. EtOH: H_2O (75:25) extract obtained by SLDE-Naviglio extraction preliminarily submitted to LC-ESI/HRMSMS, in negative ion mode, allowing the identification of polar fatty acids and specialized metabolites belonging to flavonoids, and sesquiterpenes of which the structural elucidation was performed by 1D- and 2D-NMR experiments as well as FIA-MS analysis. In this way, in addition to the most well-known caffeoyl-, dicaffeoyl quinic acid derivatives and flavonoids, eleven sesquiterpenes lactones were unambiguously characterized. Among these, one compound is here described for the first time and two compounds were never identified before in *Cynara* genus. Based on the ability to prevent skin photoaging processes reported for cynaropicrin [3], for the most abundant sesquiterpene identified in the extract the tyrosinase inhibitory activity was evaluated by a spectrophotometric assay. Kojic acid, a known tyrosinase inhibitor, was used as a positive control (IC_{50} = 30.09 μg/mL). The obtained results showed how extract, with an IC_{50} = 77.78 μg/mL, and pure compounds inhibited the tyrosinase enzyme. In particular, dodesacylcynaropicrin-8-glucoside and the new sesquiterpene showed the highest inhibition (IC_{50} = 41.26 and 43.55 μg/mL, respectively).

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OP14-Antifungal potential of carnosic acid from *Salvia somalensis* againstphytopathogenic fungi

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Keywords: *Salvia somalensis*; carnosic acid; phytopathogenic fungi; *Botrytis cinerea*

Plant pathogens and pests cause a loss of up to 40% of the yield of economically important crops every year [1]. Among them, fungi cause more economic damage than any other group of microorganisms [2]. These phytopathogens includes *Colletotrichum coccodes*, a species involved in solanaceous anthracnose diseases, *Fusarium oxysporum*, responsible of vascular wilt in several growing scenarios, *Sclerotinia sclerotiorum*, causing basal and stem rot or white mold in a wide range of host, *Rhizoctonia solani*, a soil-borne pathogen leading to damping-off and basal rot in many crops, and *Botrytis cinerea* the causal agent of gray mold and one of the most important plant pathogens of worldwide interest [3, 4, 5]. The aims of the present study were (i) to characterize the dichloromethane extract of the fresh aerial parts of *S. somalensis*, cultivated in Liguria (Italy), (ii) to quantify carnosic acid production and (iii) to find an eco-friendly alternative approach to control diseases caused by phytopathogenic fungi. The phytochemical investigation yielded several known terpenoids, as well as a diterpene, 4α,9α-epoxy-2H- dibenzo[a,d]cyclohepten-7(5H)-one, not previously described as a plant metabolite before. Resultsshowed a noteworthy quantity of carnosic acid (113.90 µg/mg of dried extract). The potential antifungal activity of the plant surface extract and carnosic acid against five phytopathogenic fungi was considered. A complete inhibition of *C. coccodes*, *S. sclerotiorum*, and *R. solani* mycelium growth was observed by carnosic acid at 500 µg/mL. High inhibition values were observed against *B. cinerea* and *F. oxysporum* compared to reference active ingredients. Four different *B. cinerea* strains exhibited a pronounced sensitivity to carnosic acid, also those originating from agricultural crop scenarios where high load of active ingredient for gray mold control was historically adopted. Additionally, the formation and development of the germinative tube in *B. cinerea* were greatly slowed down, highlighting that potential interesting use of new active ingredients to be use in organic farming.

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OP15-Lignans in *Schisandra chinensis* L. green extracts: quantitative analysis and evaluation of tyrosinase inhibitory activity by a spectrophotometric assay and STD-NMR

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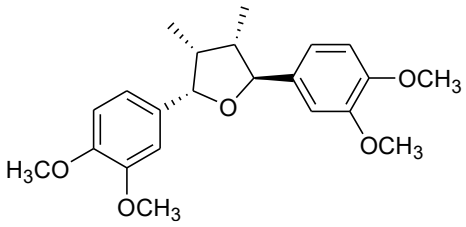
Keywords: Lignans, quantitative analysis, tyrosinase, STD-NMR

The fruits of *S. chinensis* are widely used in Traditional Chinese Medicine for their sedative, antistress and cognitive function improvement action attributable to the main bioactive constituents, belonging to the lignan class. It is possible to classify lignans into eight different subgroups depending on the cyclization mode and how oxygen is incorporated into the skeleton. These eight subgroups are: furofuran (FF), furan (FR), dibenzylbutane (DB), dibenzyl-butyrolactone (DBL), aryltetraline (AT), arylnaphthalene (AN), dibenzocyclooctadiene (DCO), and dibenzylbutyrolactol (DBLL)[1]. In this work, after carrying out several green extractions of *S. chinensis* (Turcz.) Baill. (Schisandraceae) fruits including SLDE-Naviglio and UAE extractions using EtOH:H_2O (100:0, 75:25, 50:50) as bio-solvents, lignans were isolated and quantified in the different extracts. Moreover, considering that the overexpression of the tyrosinase enzyme is involved in the process of neurodegeneration, the tyrosinase inhibitory activity of all 26 isolated lignans was tested by a spectrophotometric assay. Among these lignans the most active compound was granschisandrin which showed an IC_{50} value of 53.49 ± 2.18 µg/ml, comparable to that of kojic acid (IC_{50} = 28.18 ± 2.49 µg/ml), used as reference compound. To obtain a confirmation and understand the kind of binding between the lignan ligand and protein, STD-NMR experiments were also performed.



granschisandrin

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OP16-*Corylus avellana* L. by-products: an "optimized" antioxidant source

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Keywords: hazelnut skins, RSM, antioxidant, phytochemical profile

In a world where wastes are a growing problem, the shift from linear to circular economy - based on reduction, reuse and recycling - is inevitable. The aim of this work is to valorise the skins of *Corylus avellana* L. as food industry by-products. Many studies have highlighted that hazelnut and its by-products are rich in polyphenol compounds, which possess strong bioactive properties [1].The hazelnut skins correspond to the cuticle that covers hazelnuts representing the 2.5% of the total kernel weight. Normally, they are discarded after the process of roasting hazelnuts due to their bitter taste, but considering the high content of specialized metabolites, it is important to give them a second life. The extraction process was conducted using ultrasound assisted extraction and three variables, temperature (30-50-70 °C), time (1-2-3 h) and solvent (100% EtOH, 50% EtOH, 100% H_2O), resulting in 27 extracts. Subsequently, five different *in vitro* assays were used to evaluate the Total Phenolic Content (TPC) by Folin-Ciocalteu reagent and the antioxidant capacity by 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), Ferric Reducing Antioxidant Power (FRAP) and *β*-Carotene Bleaching assay (BCB). The optimal extraction conditions (30 °C, 74 minutes, 21% EtOH) were determined using Response Surface Methodology [2]. This is a powerful mathematical and statistical tool that consider the influence of multiple extraction variables on each test, allowing the identification of the most favorable combination of factors to maximize the desired outcome. Once the optimized extract was obtained, tests were performed to confirm the results predicted by the methodology. For example, in the TPC assay a result of 365.41 mg GAE/g DW was obtained, which perfectly fits within the predicted confidence interval (363.9-410.9 mgGAE/g). The work continued with the analysis of the phytochemical profile of the optimized extract using an UHPLC-DAD-ESI-Orbitrap Exploris^{TM}120 Mass Spectrometer to evaluate the present compounds that are in particular phenolic compounds. After that, the optimized extract was tested on HepG2 cells, showing cytoprotective effect against oxidative stress by reducing oxygen free radical levels. Considering these results, hazelnut skins are a rich source of bioactive compounds which can still be exploited and further studies will be needed to investigate the mechanisms responsible for the antioxidant activity for their potential use in the pharmaceutical and cosmeceutical fields.

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POSTER PRESENTATIONS

P1-Metabolomic analysis of *Suaeda vera* and *Lavatera agrigentina* from the Maccalube natural reserve

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Keywords: *Lavatera agrigentina*, *Suaeda vera,* Phytochemical profiling, Maccalube

Amaranthaceae family, which includes the former Chenopodiaceae family, consists of approximately 178 genera and 2052 species [1]. Meanwhile, the Malvaceae family encompasses roughly 245 genera and 4,465 species, predominantly thriving in tropical and subtropical regions worldwide [2]. This study investigated the secondary metabolism of *Lavatera agrigentina* and *Suaeda vera*, two plant species collected from the unique, stressful environment of the " Maccalube " protected reserve, which is dominated by methane-producing mud volcanoes and high salinity/heavy metals [3]. *Suaeda*, a halophytic genus in the Amaranthaceae family, comprises over 100 species known as Seablites or Seepweeds due to their high salt tolerance. These plants possess antibacterial and antiviral properties and have been traditionally used to treat symptoms of heart and liver diseases [1]. Comprehensive analysis of the hydroalcoholic extract from *S. vera* aerial parts using untargeted LC-MS revealed a rich phytochemical profile. This analysis identified a diverse range of specialized metabolites, including flavonoids, various phenolic compounds (phenylpropanoids, lignans, and phenolic acids) and their glycosylated forms, phenylethylamine alkaloids and their glycosides, fatty acids, and sulphated flavonoids. We also investigated *L. agrigentina*, a species endemic to Southern Sicily and belonging to the Malvaceae family. Though cultivated for ornamental value, these plants offer more than visual appeal. They hold potential as food and remedy for ailments, from digestive to skin issues, and throat infections [2, 4]. HR-ESI-MS annotation of the hydroalcoholic extract of *L. agrigentina*, revealed a diverse metabolites encompassing various classes of compounds, including flavonoids, phenolic compounds, and their respective glycosides, coumarins, terpenes, fatty acids, indole alkaloids, sulfated flavonoids, and sulphated phenolic acid. The abundance of flavonoids, phenolic compounds, and alkaloids, including glycosylated and sulphated forms, in *S. vera* and *L. agrigentina* suggests their potential for mediating stress responses and contributing to their resilience in the challenging environments. This diverse chemical profile and environmental tolerance, especially to heavy metals and salinity, suggests potential applications in agriculture and phytoremediation.

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P2-Exploring the impact of environmentally relevant ibuprofen levels on specialized metabolite production in *Cymodocea nodosa*

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Keywords: Ibuprofen; specialized metabolites; stress response; seagrass

The presence of active pharmaceutical ingredients in marine environments is a growing global concern due to their potential adverse effects on organisms and ecosystems. One of the most frequently detected pharmaceutical in coastal seawaters is the non-steroidal anti-inflammatory drug ibuprofen (IBU). It gained significant attention recently due to its widespread use during the COVID-19 pandemic [1]. With an estimated annual global consumption exceeding 10,000 metric tons, its presence in marine environments is expected to increase, leading to greater contamination of coastal habitats in the future [2]. Seagrass meadows, essential for coastal protection, carbon sequestration, and water quality, are increasingly exposed to these contaminants. Despite their ecological importance, the effects of pharmaceuticals on seagrasses remain underexplored. This study focused on *Cymodocea nodosa* (Ucria) Ascherson (Cymodoceaceae), a seagrass species prevalent in the Mediterranean, to evaluate the impact of environmentally relevant concentrations of IBU. Using mesocosms to simulate near-natural conditions, the study examined the effects of short-term IBU exposure (12 days) at three different concentrations (0.25, 2.5, and 25 µg/L) on specialized metabolite production respect to IBU untreated control. Chemical quali-quantitative analyses were performed by UHPLC-Orbitrap/ESI-HRMS on the *C. nodosa* methanolic extracts. Chicoric acid for phenolic acids, rutin for flavonoid glycosides, and catechin for (epi)catechin and their derivatives were used as pure external standards and statistical analyses were carried out to assess significant differences among samples. A total of 39 compounds were tentatively identified: a tricarboxylic acid; 23 phenolic acids, mainly caffeic/ferulic/*p*-coumaric acid derivatives; 13 flavonoids such as quercetin, kaempferol and isorhamnetin derivatives; and two dihydrochalcones. Among phenolic acids, *p*-coumaroylmalic acid and *p*-coumaroyl hexoside significantly decreased in plants exposed to medium or high IBU concentrations compared to control. Instead, gallic acid and *p*-coumaric acid showed a significant increase in plants treated with high IBU concentration. Among flavonoids, only rutin increased in plants treated with high IBU concentration. Previous studies demonstrated that abiotic stresses can modulate the expression of genes involved in the biosynthetic pathways of some specialized metabolites. The changes in the production of these specialized metabolites could be considered as a response of *C. nodosa* to the stressful conditions induced by IBU.

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P3-Hydroalcoholic extracts of three *Mentha* species from Campania region: chemical profile and antibacterial potential against food strains

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Keywords: *Mentha*, Hydroalcoholic extract, chemical composition, antibacterial activity

Lamiaceae family includes aromatic plants that grow in many regions of the world. Some of them are widely used to improve the flavour and aroma of foods and the overall quality of the product. *Mentha* is a genus of aromatic perennial plants distributed in temperate regions of Europe, Asia, Australia and South Africa [1]. In literature, this plant is reported for its medicinal and commercial importance [2]. The proposal of this work is to study the chemical profile and antibacterial activities of hydroalcoholic extracts obtained from three species of the genus, *Mentha spicata* L., *Mentha pulegium* L., and *Mentha longifolia* L., collected in Campania region. The chemical analysis of the extracts, performed by LC-HRESIMS/MS, revealed a total of 21 compounds, mainly belonging to flavonoids. The highest number of components is identified in *M. longifolia* (17 compounds), followed by *M. pulegium* (12 compounds); finally, 9 compounds are identified in *M. spicata*. Tuberonic acid, luteolin-7-O-rutinoside, sideritiflavone and hymenoxin are common compounds in the three extracts. Diosmina, salvianolic acid G, jaceosidin, narigenin and sakuranetin are found only in *M. longifolia*, nobiletin is solely present in *M. pulegium* and 5- hydroxyauranetin is identified in *M. spicata*. The analysis of total phenolic content (TPC) was estimated by Folin-Ciocalteu spectrophotometric method, and the evaluation of total flavonoid content (TFC) was conducted by aluminum chloride colorimetric assay: both *M. pulegium* and *M. longifolia* extracts showed a higher phenol content than *M. spicata*, while *M. pulegium* had the highest amount of flavonoids.
The antibacterial activity of hydroalcoholic extracts was evaluated using crystal violet and MTT tests against some Gram positive (*Listeria monocytogenes* and *Staphylococcus aureus*) and Gram negative (*Acinetobacter baumannii*, *Pseudomonas aeruginosa* and *Escherichia coli*) foodborne strains. The crystal violet test revealed a notable antibiofilm action, exhibited by all three extracts, on the *A. baumannii* strain: the percentage of inhibition exceeds 42.95% (10 µl/mL of extract) and at the highest concentration (20 µl/mL) reaches up to 50.78%. Furthermore, at the concentration of 20 µl/mL, the samples showed biofilm inhibitory activity, although less effective, also against *P. aeruginosa*. These results highlighted the potential of *Mentha* extracts as natural ingredients, not only to improve the flavour profile of ailments, but also to contribute to their protection against pathogenic strains, opening new perspectives in the field of food safety.

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P4-Il progetto AGRIL per la tutela e la valorizzazione di varietà locali di agrumi minori della riviera ligure

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Keywords: Agrumi; Agrobiodiversità; Liguria

Gli agrumi hanno avuto una grande importanza nell'economia ligure e hanno disegnato il paesaggio agrario prosperando in modo non omogeneo nelle zone più riparate dai venti e maggiormente esposte al sole in tutto l'arco costiero trovando un microclima ideale (Cougnet, 1879). L'introduzione della floricoltura ha segnato un certo abbandono dell'agrumicoltura, con il rischio di estinzione di intere varietà locali. Per questo motivo, il progetto "AGrumi minori della RIviera Ligure (AGRIL)" del Programma di Sviluppo Rurale della Regione Liguria (PSR 2014-2020, sottomisura M10.2: "Sostegno per la conservazione, l'uso e lo sviluppo sostenibili delle risorse genetiche in agricoltura") ha come obiettivo principale la tutela e la valorizzazione delle seguenti varietà di importanza storica: Arancio Pernambucco di Finale ligure, Limone del Ponente Ligure, Chinotto di Savona e Arancio Amaro del Ponente Ligure.

Un importante obiettivo del progetto è l'iscrizione dei materiali reperiti e propagati all'Anagrafe Nazionale della Biodiversità di interesse agricolo e alimentare, a tal fine è necessaria la loro caratterizzazione dal punto di vista morfologico, ma anche fitochimico. A questo scopo, sono state avviate e, tuttora in corso presso il CREA-OFA di Acireale, analisi per la valutazione di proprietà antiossidanti (fra cui contenuto in polifenoli e vitamina C), acidità totale, solidi solubili totali e zuccheri totali per evidenziare peculiarità distintive di queste varietà. Sono inoltre previste iniziative di promozione per rendere note al pubblico queste importanti piante del patrimonio agricolo e culturale del Ponente Ligure.

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P5-The phenolic leaf extract of *Ptilostemon casabonae* (L.) Greuter after *in vitro* simulated gastrointestinal digestion

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Keywords: *Ptilostemon casabonae*; HPLC-PDA-MS/MS; INFOGEST; caffeoylquinic acids

For centuries, plants have played a central role in traditional medicine and have been used for the treatments of numerous diseases. They are still important for healthcare today, because many modern medicines derive from plant compounds. This study is focused on *Ptilostemon casabonae* (L.) Greuter, an endemic species in Sardinia. This wild, thistle-like plant is traditionally consumed for its flavor and biological effects. It is harvested and eaten either raw or cooked and is used medicinally to treat liver disorders and for its diuretic and digestive properties [1]. The aim of this work was to evaluate the complex digestion process of the hydroalcoholic extract obtained from the leaves of *P. casabonae* by applying an *in vitro* simulation model, to determine if there were quali-quantitative differences in the extract after in vitro simulation of gastrointestinal digestion. The INFOGEST protocol was used for this purpose [2].
Primarily, HPLC-PDA combined with MS was utilized to characterize and quantify the compounds present. The HPLC-PDA-MS/MS analysis revealed a complex polyphenolic fraction. In agreement with existing reports on related species, flavonoids, especially *O*-glycosides of quercetin, and derivatives of caffeoylquinic acid were found [1]. Chromatograms obtained in PDA mode revealed both qualitative and quantitative variations in the profile after *in vitro* digestion of the extract. In the intestinal phase, notable differences were observed for chlorogenic and cryptochlorogenic acids, as well as dicaffeoylquinic acids, with the appearance of new compounds and variations in their abundances, probably due to isomerization and degradation phenomena caused by changes in pH, high temperatures and stirring [4], [5]. Based on the phytochemical composition, antioxidant properties were evaluated by spectrophotometric methods such as ABTS and DPPH, [3] showing that the digested extract had lower antioxidant activity than the undigested extract.

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P6-Investigation on the metabolomic profile of *Malus domestica* varieties from Molise region of Italy

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Keywords: *Malus domestica*, metabolomics, chemometrics, metabolic profile

Apples (*Malus domestica* Borkh.) are one of the most extensively produced and consumed fruits worldwide [1] and represent an important source of bioactive compounds like polyphenols which are responsible for their antioxidant and anti-inflammatory properties and possess a role in the prevention of degenerative diseases [2]. This project is based on the investigation of the metabolomic profile of several autochthonous cultivars belonging to *Malus domestica* from Molise region of Italy such as Limoncella, Annurca, Gelata and Zitella varieties: although the nutritional characteristics of the apples are well known, knowledge of the Molise products is still incomplete compared to other Italian regions (e.g., Annurca apple of Campania region). Thus, the comprehensive phytochemical investigation of metabolomes of these cultivars could be helpful to trace and/or complete their fingerprint. This work has started with a specific extraction protocol obtaining a hydroalcoholic and lipophilic extract of both pulp and peel which were previously accurately separated and freeze-dried. By applying NMR spectroscopy with a 600 MHz, 1D ^{1}H and 2D NMR spectra were acquired and processed. Using different DataBases, main metabolites belonging to various classes including carbohydrates, amino acids, organic acids and other compounds such as polyphenols were identified in the hydroalcoholic extract. A powerful bioinformatic tool will be applied to quantify the detected compounds. Moreover, preliminary results of the differentiation of the selected varieties were obtained with the PCA technique which showed the separation of four clusters on the plot. The application of LC-MS/MS will be able to identify fatty acids and polyphenols that cannot be well observed and discriminated by NMR in an extract. The antioxidant activity will be evaluated and will represent an index of the potential application in cosmetics of the selected varieties of apples with the goal to obtain a cosmeceutical formulation which represents one of the main goals of this project.

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P7-Nanoformulations based on *Pistacia vera* L. with immunomodulatory activity

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Keywords: plant waste, specialized plant metabolites, nanoformulations, immunomodulation

The use of specialized plant metabolites represents a novel approach in modern medicine, aimed at harnessing naturally occurring resources, including waste products from processing industries. Given the growing interest in *Pistacia vera* L., in this study pistachio stems, which are typically discarded, were examined for the first time. The aim is to maximize the use of raw materials, reduce waste, and enhance their benefits, aligning with the concept of "circular bioeconomy." Furthermore, given the numerous biological properties of the plant, with particular attention to the anti-inflammatory activity due to compounds such as α-pinene and myricetin (1), this study reports the phytotherapeutic use of two different extracts obtained from pistachio waste products with particular focus on their immunomodulatory effects. To address issues of cytotoxicity and low bioavailability, the potential for incorporating these extracts into nanoformulations, such as liposomes and transferosomes, was assessed. In this study, hydrolate and essential oil were obtained from the stems through hydro distillation. The extracts were characterized using liquid and gas chromatography coupled with mass spectrometry, identifying 21 specialized metabolites, 13 of which were common among the two extracts. These extracts were then formulated into liposomes and transferosomes and characterized for polydispersity index, size, and negative charge. Notably, encapsulation of the extracts within the vesicles reduced their size compared to empty vesicles (2). The prepared transfersomes were larger and more homogeneous than liposomes. Moreover, transfersomes were found to be more stable than liposomes, as they showed no changes in size, homogeneity, and charge after 90 days from the preparation. The encapsulation efficiency of the nanoformulations was evaluated by quantifying the main compounds present in the extracts (naringenin, myristic acid, and catechin), achieving over 80% efficiency. Following this, the immunomodulatory activity of the essential oil and hydrolate on U937 cells stimulated with LPS was investigated. Specifically, the incorporation of the oil into transferosomes significantly enhanced its immunomodulatory effect on macrophages by inhibiting the expression and activity of the ATP citrate lyase (ACLY) enzyme. Additionally, the hydrolate in solution and the essential oil in liposomes reduced the production of ROS and NO• induced by LPS stimulation of the cells. The extracts also demonstrated the ability to inhibit prostaglandin E2 (PGE2) production. Based on these results, it can be concluded that *P. vera* extracts may play a crucial role in modulating macrophages induced by various stimuli, such as LPS endotoxin, by acting on immunometabolism. Most interestingly, the formulation in transfersomes abolished the cytotoxicity of the essential oil while increasing its ability to inhibit inflammatory mediators via the immunometabolic citrate pathway.

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P8-Il progetto "MI.GA.FLOR - l'agrobiodiversità floricola imperiese: mimosa e garofano"

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Il progetto MI.GA.FLOR mira alla salvaguardia e alla valorizzazione di varietà floricole locali: tracciando la storia delle varietà, identificandone il germoplasma ed evidenziando le caratteristiche estetiche l'obiettivo è di evitare che varietà storiche vengano soppiantate da varietà commerciali, conservarle per evitare dispersione genetica, sensibilizzare floricoltori e consumatori per evitare la perdita di patrimonio floricolo locale, inserire le varietà nell'Anagrafe Regionale Prodotti Agroalimentari Tradizionali (PAT) e nell'Anagrafe Nazionale della Biodiversità di Interesse Agricolo e Alimentare.
Le specie interessate dal progetto sono: *Acacia* sp. e *Dianthus cariophyllus*.
All'interno del progetto si sviluppano molte attività, tra cui:

- Caratterizzazione morfologica: le selezioni sono state studiate attraverso la stesura di schede di descrittori morfologici, che saranno propedeutici per l'iscrizione all'Anagrafe Nazionale;
- Caratterizzazione genetica: attività che viene svolta mediante approcci diversi, per l'*Acacia* sono state eseguite analisi di barcoding che identificano le specie, per il *Dianthus* sono stati analizzati specifici marcatori molecolari.
- Caratterizzazione storica: attraverso lo studio di fonti storiche sono state ricostruite origine, insediamento e diffusione delle selezioni progettuali.
- Caratterizzazione agronomica: attraverso l'utilizzo di descrittori, lo scopo della caratterizzazione è quello di fornire l'identificazione dei parametri agronomico-colturali, di adattamento alle differenti condizioni edafiche, di verifica dei tempi di sviluppo fenologico, di valutazione dei dati meteoclimatici e dello stato di salute delle piante per l'iscrizione delle risorse genetiche all'Anagrafe Nazionale della Biodiversità.
- Risanamento e produzione di materiale di propagazione sano precommerciale: indagine preventiva e di risanamento atta a produrre un insieme di buone pratiche per lo sviluppo futuro di produzioni controllate dal punto di vista della qualità fitosanitaria.
- Realizzazione e gestione di campi collezione delle risorse genetiche *in situ* e *ex situ* e realizzazione della banca del germoplasma.
- Predisposizione schede colturali e dossier per l'iscrizione all'Anagrafe Nazionale della Biodiversità per le varietà vegetali
- Predisposizione del dossier "Agricoltori Custodi della Biodiversità Vegetale".

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