International Conference on BioActive Compounds 2026

28-29 May 2026 • Wrocław, Poland

Book of Abstracts

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Scientific Programme

Thursday, 28 May 2026

8.00–9.00 **REGISTRATION**

9.00–9.20 **CONFERENCE OPENING AND WELCOME REMARKS**

SESSION: DESIGN, SYNTHESIS AND CHEMICAL MODIFICATIONS OF BIOACTIVE COMPOUNDS

Chairpersons of the session:
Stefano Serra, Witold Gładkowski

9.20–10.00 **PLENARY LECTURE 1**
Lupane-type triterpenoid saponins for therapeutic and prophylactic applications
Charles Gauthier

10.00–10.15 **ORAL PRESENTATION 1**
Selective functionalisation of bioactive molecules with engineered ancestral cytochrome P450 enzymes
Elizabeth Gillam

10.15–10.30 **ORAL PRESENTATION 2**
Innovative lipid delivery systems for bioactive compounds
Izabela Nowak

10.30–10.45 **ORAL PRESENTATION 3**
Valorization of metal-accumulating plant biomass for the preparation of polymetallic catalysts and their catalytic application
Tomasz Olszewski

10.45–11.00 **ORAL PRESENTATION 4**
Optimization of biocatalytic cascade in the production of raspberry ketone from birch secondary metabolites
Emerik Leaković

11.00–11.30 **COFFEE BREAK**

SESSION: ISOLATION, ANALYSIS AND IDENTIFICATION OF NATURAL COMPOUNDS

Chairpersons of the session:
Elisabetta Brenna, Filip Boratyński

11.30–12.10 **PLENARY LECTURE 2**
Dissecting bioactives from multicomponent mixtures
Judith Rollinger

12.10–12.25 **ORAL PRESENTATION 5**
Bioactive compounds obtained via NADES extraction: Impact on in vitro cell migration
Kristina Radošević

12.25–12.40 **ORAL PRESENTATION 6**
Hydrolates revised: How distillation and analytical methodology fundamentally reshape chemical profile
Kamil Szymczak

12.40–12.55 **ORAL PRESENTATION 7**
From silence to signal: An integrated microgravity–metabolomics platform for discovering microbial bioactives
Yosephine Gumulya

12.55–13.10 **ORAL PRESENTATION 8**
From waste to platform chemicals: Adsorption-based fractionation of food waste-derived volatile fatty acids
Negar Baserehtaromsary

13.10–14.10 **LUNCH BREAK**

SESSION: VALORIZATION OF AGRI-FOOD BY-PRODUCTS AND WASTES

Chairpersons of the session:
Elizabeth Gillam, Agnieszka Krawczyk – Łebek

14.10–14.50 **PLENARY LECTURE 3**
Exploitation of olive oil industry products and by-products for pilot isolation and semi-synthesis of promising medicinal agents
Leandros Skaltsounis

14.50–15.05 **ORAL PRESENTATION 9**
Continuous-flow lipase-mediated synthesis of bioactive derivatives from renewable sources
Elisabetta Brenna

15.05–15.20 **ORAL PRESENTATION 10**
Flaxseed oil cakes as a source of proteins – implications for animal cell technology and cellular agriculture
Višnja Gaurina Srček

15.20–15.35 **ORAL PRESENTATION 11**
Every molecule matters: Designing biorefineries where no fraction is left behind
Swarnima Agnihotri

15.35–15.50 **ORAL PRESENTATION 12**
Fungal-based solid-state fermentation: A sustainable approach for lignocellulosic biorefineries
Marina Tišma

15.50–16.20 **FLASH POSTER PRESENTATIONS**

16.20–17.20 **POSTER SESSION COMBINED WITH COFFEE BREAK**

19.30 **GALA DINNER**

Friday, 29 May 2026

SESSION: BIOTECHNOLOGICAL PRODUCTION AND TRANSFORMATION OF BIOACTIVE COMPOUNDS

Chairpersons of the session:
Amir Mahboubi Soufiani, Aleksandra Grudniewska

9.30–10.10	**PLENARY LECTURE 4** Protein engineering of cofactor-dependent enzymes for biocatalysis *Jon Stewart*
10.10–10.25	**ORAL PRESENTATION 13** Chemo-enzymatic valorisation of industrial hemp distillation waste from essential oil production *Fabio Parmeggiani*
10.25–10.40	**ORAL PRESENTATION 14** Exploring the promiscuous activity of fatty acid hydratases *Stefano Serra*
10.40–10.55	**ORAL PRESENTATION 15** Microbial biocatalysts for the preparation of flavonoid metabolites: applications in metabolic and biological research *Kateřina Valentová*
10.55–11.10	**ORAL PRESENTATION 16** Noble metal nanoparticles as elicitors enhancing biotechnological production of bioactive metabolites in plant in vitro systems *Agnieszka Szopa*
11.10–11.40	**COFFEE BREAK**

SESSION: BIOACTIVES IN MEDICINE AND COSMETOLOGY

Chairpersons of the session:
Kristina Radošević, Marcelina Mazur

11.40–12.20	**PLENARY LECTURE 5** Are small phenolic metabolites of flavonoids responsible for the observed arterial blood pressure-decreasing effect? *Přemysl Mladěnka*
12.20–12.35	**ORAL PRESENTATION 17** Caffeic acid-functionalized silver nanoparticles enhance anti-melanogenic activity in melanocytes *Enrique Calvo Manso*
12.35–12.50	**ORAL PRESENTATION 18** Lactate: nutrient, oncometabolite, or key modulator of cellular metabolism *Małgorzata Zakłos-Szyda*
12.50–13.05	**ORAL PRESENTATION 19** Halogenated flavonoid derivatives modulating gut microbiota composition as supportive agents in colorectal cancer therapy *Martyna Perz*
13.05–13.20	**ORAL PRESENTATION 20** Pluchea grevei essential oil as multifunctional active ingredient *Alicja Surowiak*
13.20–14.30	**LUNCH BREAK**

SESSION: BIOACTIVES IN FOOD

Chairpersons of the session:
Fabio Parmeggiani, Anna Chojnacka

14.30–15.10	**PLENARY LECTURE 6** Solid-state fermentation as a platform for upcycling agro-industrial side streams *Wolfram Brück*
15.10–15.25	**ORAL PRESENTATION 21** Glycosylated mycotoxins – a hidden challenge in food safety *Vladimir Kren*
15.25–15.40	**ORAL PRESENTATION 22** Combining enzyme inhibition assays to targeted metabolomics for food bioactivity assessment *Aristeidis Tsagkaris*
15.40–15.55	**ORAL PRESENTATION 23** The BIOPEP-UWM database of peptides and proteins – opportunities available in 2026 *Piotr Minkiewicz*
15.55–16.10	**ORAL PRESENTATION 24** Circular economy in the sugar industry: waste valorisation for the discovery, development, production and application of bioactive compounds *Krzysztof Kołodziejczyk*
16.10–16.25	**ORAL PRESENTATION 25** Antihypertensive and sex-dependent metabolic effects of protein hydrolysates from agri-food by-products: Potential role of gut microbiota *Francisca Isabel Bravo*
16.25–17.00	**AWARDS AND CLOSING REMARKS**

List of Posters

P1* Synthesis and antibacterial activity of fabimycin – a novel antibiotic candidate against Gram-negative bacteria
Marcin Ochowicz

P2* Whole cell transformation of cannabidiol
Daniel Łój

P3* Upcycling spent hops: a low-cost method for the isolation of xanthohumol
Aleksandra Modzelewska

P4* Blue-biorefinery of brown macroalgae: green extraction strategies for phlorotannins recovery from *Fucus* sp.
Mariem Hamed

P5* Cannabidiol as a nefroprotective agent against ionophore coccidiostat cytotoxicity
Oliwia Kończak

P6* The effect of 4-methylcatechol on platelet aggregation in patients with asthma
Magdalena Walková

P7 Transformations of hydroxycinnamic acids with the use of fungi
Abirami Baskaran

P8 Fully enzymatic cascades for the conversion of (*E*,*E*)-farnesol into enantiomerically enriched commercial fragrances
Federico Acciaretti

P9 Growth dynamics of *Salvia atropatana* hairy root culture as a factor determining the secondary metabolite production
Wiktoria Ejsmont-Gralewska

P10 From raw material to functional snack: fermented legumes with increased bioavailability of polyphenols
Małgorzata Gumienna

P11 Synthesis of 7-azolocoumarins as fungistatic agents against fluconazole-resistant *Candida* strains
Mutiara Saragih

P12 Gene-mining the frozen frontier: keratinolytic antarctic soil bacteria for novel bioactive compounds
Marcin Sypka

P13 The impact of L-tryptophan on biochemical composition and structural properties of flax callus
Magdalena Wróbel-Kwiatkowska

P14 Leveraging food side streams for functional sweets: a Polish–Swiss research–industry initiative on microbiome-targeted snacks
Jessica Brzezowska

P15 Sustainable chemoenzymatic cascade for the synthesis of (*R*)-citronellyl nitrile from natural citral
Giovanni Cipolla

P16 Tryptophan-rich *Moringa oleifera* leaves expand plant protein potential: nutritional characteristics and spectroscopic fingerprinting
Joanna Harasym

P17 Watermelon by-products as functional ingredients: nutritional, antioxidant, and functional evaluation of seed protein and flour
Joanna Miedzianka

P18 Lacto-fermented oats as future dairy-free yoghurt
Sharmeen Mustafa

P19 DES-based protein recovery from sunflower oilseed cake
Weronika Rogowska

P20 Sensory evaluation of vanillin obtained by fungi in the Solid-State Fermentation from agri-food industry by-products
Ewa Szczepańska

P21 A plant-based agro-industrial by-product-derived protein hydrolysate reduces blood pressure and modulates gut microbiota in diet-induced metabolic syndrome
Francisca I. Bravo

P22 Cascade biorefinery of apple pomace via submerged lab fermentation: from enzymatic recovery of bioactives to binder-free biopackaging (a critical review)
Małgorzata Zawadzka

P23 Structural modification of fragrance alcohols: effects on human cells viability
Kinga Baberowska

P24 Impact of chlorination and glycosylation on the lipophilicity and membrane interactions of 2'-hydroxychalcone and dihydrochalcone derivatives
Anita Dudek

P25 Antiproliferative potential of vanillin-derived δ-iodo-γ-lactone
Anna Dunal

P26 Impact of stigmasterol-acylglicerol conjugates on nanocarrier properties - design, synthesis and physicochemical evaluation
Marcin Olesiński

P27 Click-engineered steroid bioconjugates as platforms for the development of bioactive molecules
Tomasz Pospieszny

P28 Design and evaluation of coiled-coil foldamers as nanocarrier systems
Monika Szefczyk

P29 Effect of symmetric acylglycerols containing stigmasterol residues at *sn*-1 and *sn*-3 positions on lipid model membranes
Aleksandra Włoch

P30 Metabolic profiling of *Pistacia lentiscus* L. var. *chia* via NMR and LC-MS: tracking authenticity from leaf to resin
Stavros Beteinakis

P31 Antarctic psychrophiles as bioactive factories: linking genomics with metabolite production
Iga Jodłowska

P32 Metabolomic insights into the antimicrobial effects of *Metschnikowia* yeast on phytopathogens
Zofia Perek

P33 An investigation into hop extracts: a study of volatile components and their relationship to the aroma profile
Davide Tessaro

P34 Multicomponent NADES as a sustainable alternative to conventional solvents for rosemary antioxidant recovery
Martina Železnjak

P35 Vascular effects of new bisphenol a alternatives
Ganesh Dussa

P36 Towards natural photoprotection: the greensunscreen project
Katarzyna Gaweł-Bęben

P37 Carriers of the future or just a cosmetic trend? The effectiveness of lipid nanoparticles in delivering active ingredients to the skin
Marta Marzec

P38 Vasodilatory substances from hawthorn: screening and determination of the mechanism of action
Jana Pourová

P39 Flavokawain derivatives as bioactive chalcones: effects of B-ring substitution on membrane organization and protein binding
Hanna Pruchnik

P40 Galectin-driven reorganization of ganglioside nanodomains in model membranes
Priti Sengupta

P41 Biological activity of hydroxychalcones in Merkel carcinoma cells
Monika Stompor-Gorący

P42 Linking experimental and theoretical activity of phenolic compounds in DPPH, ABTS and FRAP assays
Paulina Strugała-Danak

P43 Five novel isocoumarins protect red blood cells against copper toxicity
Lenka Táborská

P44 Modulation of multidrug resistance transporter activity, ABCB1, by combined use of antipsychotic drugs and simvastatin
Olga Wesołowska

P45 Honey as a source of bioactive compounds – a review
Grzegorz Buczkowski

P46 Properties of mayonnaises based on mustard oil
Anna Grygier

P47 Sea buckthorn: a promising source of bioactive lipids for glucose regulation
Eliza Korkus

P48 From probiotic CFS screening to food application: selection of promising candidates and preliminary evaluation in a tragacanth-based coating for refrigerated shrimp preservation
Maryam Nasri

P49 Moroheiya as a bioactive hydrocolloid: distinct functional effects of leaf powder and mucilage extract in rice noodles
Aimé Roger Raherison

P50 Liposomes encapsulated with phytosterols as a bioactive food component
Magdalena Rudzińska

P51 Nutraceutical potential of jackfruit (*Artocarpus heterophyllus.*Lam): under-utilized species from Mauritius
Joyce Govinden Soulange

P52 Looking back or into the future: which approach is better for protein design?
Ewa Kozłowska

P53 Modern detoxifying preparations: the use of biocomponents to mitigate the impact of mycotoxins in feed on animal health and production performance
Szymon Powałowski

P54 Valorization of rapeseed presscake through hydrothermal pretreatment, fractionation and fungal fermentation
Witold Pietrzak

Lupane-type triterpenoid saponins for therapeutic and prophylactic applications

Charles Gauthier

Unité mixte de recherche INRS-UQAC, Centre Armand-Frappier Santé Biotechnologie, Institut national de la recherche scientifique (INRS), 555, boulevard de l'Université, Chicoutimi, G7H 2B1, Canada
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Natural products have historically served as indispensable sources of therapeutic leads, with pentacyclic triterpenoids emerging as particularly promising bioactive scaffolds due to their structural diversity, low inherent toxicity, and capacity for advanced chemical elaboration. Among these, betulinic acid (BetA), a lupane-type triterpenoid available from the bark of white birches, exhibits broad-spectrum biological activity but suffers from negligible aqueous solubility, severely limiting its clinical translation. Saponins, amphiphilic glycoconjugates consisting of a triterpenoid aglycone linked to sugar residues, provide a versatile pharmaceutical platform to overcome this solubility barrier while preserving or enhancing biological activity and have constituted one of the main focus of our research program [1]. The plenary lecture will focus on the synthesis of BetA saponins bearing either a Lewis X trisaccharide [2], found in human milk, or the minimal trisaccharide epitopes required for adjuvant activity of QS-21 saponin [3]. These chimeric triterpenoid saponins were prepared using both linear and convergent stereoselective glycosylation strategies. We demonstrate that Lewis X-containing triterpenoid saponins rank among the most potent monovalent inhibitors reported to date of dendritic cell-specific intercellular adhesion molecule-3-grabbing non-integrin (DC-SIGN) and liver/lymph node-specific intercellular adhesion molecule-3-grabbing integrin (L-SIGN)-mediated transfer of human immunodeficiency virus 1 infection to $CD4^+$ cells, with IC_{50} values in the low micromolar range. Furthermore, these triterpenoid saponins, along with their rhamnose-modified analogs [4], were evaluated *in vivo* for their toxicological and immunological potential in both C57BL/6 and hDC-SIGN transgenic mice. Our findings reveal that, while the synthetic saponins exhibit low toxicity, replacing echinocystic acid with BetA impacts their immunogenicity profiles. This work provides a valuable foundation for the development of saponin-based therapeutic agents and highlights the potential of these glycosylation strategies for synthesizing complex and unnatural glycoconjugates for therapeutic and prophylactic applications.

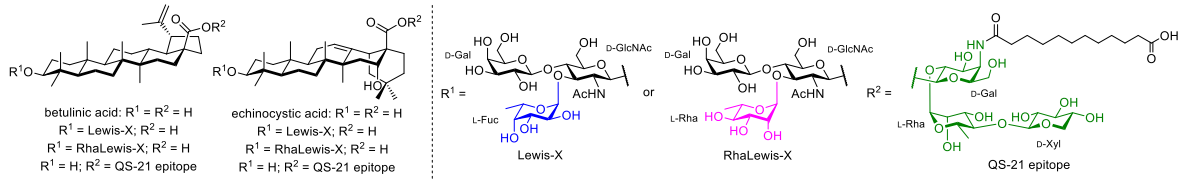

Fig. 1. Chemical structures of chimeric triterpenoid saponins.

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PLENARY LECTURE 2

Dissecting bioactives from multicomponent mixtures

Judith M. Rollinger

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Whereas classical bioactivity-guided isolation comes with several bottlenecks and limitations, biochemometric approaches applying MS- and NMR-based spectral data correlated with bioactivities open up new revenues for a fast and unambiguous identification of major and even minor bioactive constituents from complex mixtures. In my talk I will present some recent examples from ongoing research focusing on the discovery of resistance breaking anti-infectives and anti-inflammatory natural compounds.

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Exploitation of olive oil industry products and by-products for pilot isolation and semi-synthesis of promising medicinal agents

Leandros A. Skaltsounis

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Extra virgin olive oil (EVOO),), the main product of *Olea europaea* and the key ingredient of Mediterranean diet, is characterized by substantial nutritional and health beneficial value [1]. However, despite olive oil's economic and health impact, its industry is associated with environmental problems derived from the vast quantity of by-products, such as vegetation waters, olive cake, olive pulp and olive branches and leaves. [2] The amount of olive leaves produced every year exceed 18 million tons and mostly are used as animal feed, compost production or simply are burned, causing serious environmental damage. In a recent study it was found that burning of olive tree branches is a major organic aerosol source in the Mediterranean region.[3] However this material still contains high value-added compounds such as triterpenoids, secoiridois, flavonoids, phenolic alcohols, phenolic acids, and lignans which are known as olive polyphenols. All these constituents have a strong antioxidant profile and there is an increased industrial interest for possible nutraceutical and pharmaceutical applications. Our work is focused on finding alternative strategies to manage the residues of olive oil industry following two axes. Firstly, the development of liquid/ liquid or solid/liquid extraction followed by partition chromatography techniques for the isolation of these compounds in multi gram scale. Secondly the use of some of these compounds such as oleoside, EDA as starting material for the hemi-synthesis of new analogues and their evaluation as potential antitumor agents.

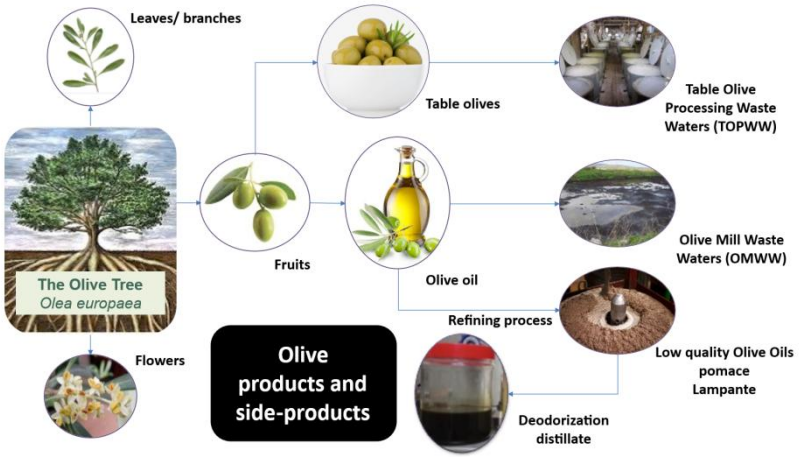

Fig. 1. Integration of extraction and chemoenzymatic valorisation to obtain valuable cannabinoids from *C. sativa* distillation waste.

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Protein engineering of cofactor-dependent enzymes for biocatalysis

Jon Stewart

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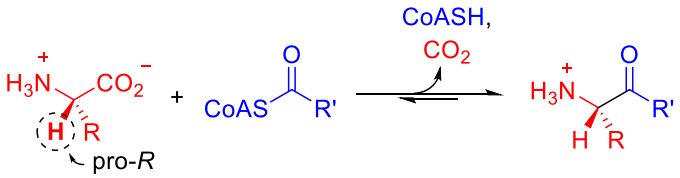
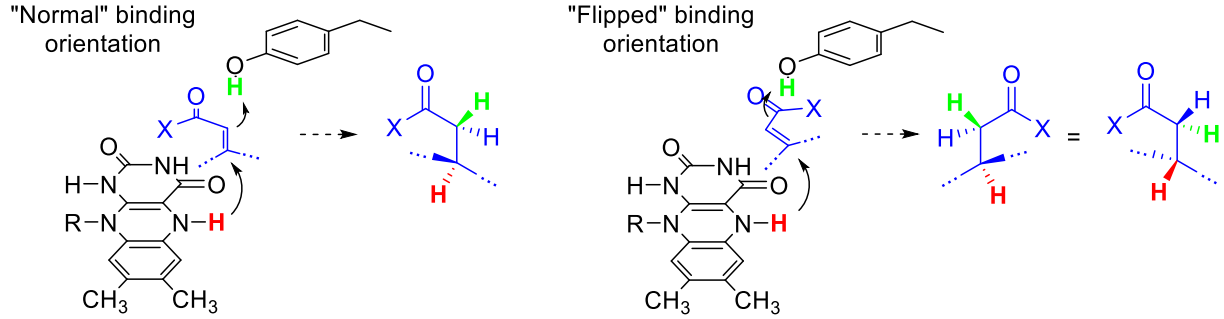
Our research group has had a long-standing interest in using cofactor-dependent enzymes for stereoselective synthesis. We have employed a number of strategies including protein engineering and MD computations to optimize catalyst properties and understand the impact of our mutations, respectively. This talk will describe some of our recent efforts in this area, focusing on pyridoxal phosphate-dependent Claisen condensation / decarboxylation enzymes and members of the flavin-dependent Old Yellow Enzyme class of alkene reductases.

PLP-dependent Claisen condensation / decarboxyation



Old Yellow Enzyme-mediated alkene reduction



Are small phenolic metabolites of flavonoids responsible for the observed arterial blood pressure-decreasing effect?

Přemysl Mladěnka

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Flavonoids are traditionally associated with a positive impact on cardiovascular diseases, and vasodilatory activity can be one of the underlying effects. The bioavailability of flavonoids is, however, too low to explain the decrease observed after their administration *in vivo* [1]. The missing clue can represent small phenolic metabolites produced by human microbiota in the colon. These compounds are reaching higher plasma levels, and several of them have already reported to dilate the vascular bed [2,3]. In this lecture, the effect of four flavonoid metabolites /4-methylcatechol, 3-(3-hydroxyphenyl)propionic, 3,4-dihydroxyphenylacetic acid, and 3-hydroxyphenylacetic acid), two isoflavonoid metabolites (S-equol and O-desmethylangolensin), as well as their synthetic derivatives at both *ex vivo* and *in vivo* levels on the vascular system will be reported. The mechanism of action will be suggested, and their other positive cardiovascular effects, including antiplatelet effects, will be discussed.

ACKNOWLEDGEMENTS

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PLENARY LECTURE 6

Solid-state fermentation as a platform for upcycling agro-industrial side streams

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The transition toward sustainable and health-oriented food and bio-based systems increasingly depends on our ability to transform agro-industrial side streams into high-value, functional resources. Solid-state fermentation (SSF) is emerging as a key enabling technology in this context, offering a resource-efficient and versatile platform to unlock the nutritional, bioactive, and even dermo-functional potential of plant-based by-products.

Recent research highlights both, the promise and the complexity of this approach. Fibre-rich side streams such as okara and brewers' spent grains demonstrate clear potential to modulate gut-relevant metabolites linked to satiety, metabolic health, and immune function. At the same time, variability in substrate composition and microbial dynamics continues to limit the predictability and reproducibility of these effects, pointing to a critical need for better integration of microbiology, process control, and systems-level analytics.

In parallel, emerging work on polyphenol-rich substrates illustrates how SSF can be extended beyond nutritional enhancement toward targeted bioconversion of high-value compounds. This includes the microbial transformation of complex polyphenols into bioactive metabolites with potential applications not only in functional foods, but also in adjacent sectors such as nutraceuticals and natural cosmetics. These developments reinforce the concept of side streams as versatile biochemical reservoirs rather than low-value waste.

Looking ahead, the convergence of SSF with microbiome science, metabolic engineering, and product formulation opens new opportunities to design next-generation functional ingredients and foods with defined health outcomes. However, significant challenges remain in bridging the gap from promising biological effects to scalable, standardised processes and consumer-accepted products. Addressing these challenges will require tighter coupling between upstream bioprocessing, downstream application, and physiological validation.

Ultimately, SSF has the potential to evolve into a cornerstone technology within circular bioeconomies, redefining agro-industrial side streams as strategic raw materials for sustainable innovation across food, health, and bio-based industries.

Selective functionalisation of bioactive molecules with engineered ancestral cytochrome P450 enzymes

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Enzymes offer the opportunity to introduce modifications into complex molecules in a chemo-, regio- and stereoselective manner that is difficult using traditional synthetic organic chemistry. Cytochrome P450 enzymes are amongst the most versatile enzymatic catalysts known, catalysing more than 60 specific chemical transformations by virtue of the chemistry that can occur at the haem prosthetic group. By catalysing the monooxygenation of un-activated C-H bonds, a reaction that is difficult to achieve with selectivity by purely chemical means, P450s can mediate hydroxylations, epoxidations, and C-C bond cleavage of substrates such as lipids and other organic molecules. However, their industrial application has been limited to date due to the relative instability of the naturally occurring enzymes. Our group has used ancestral sequence reconstruction to engineer highly thermostable P450s for industrial use. As well as lasting longer under operating conditions, ancestral P450s can be expressed at higher yields and using inexpensive media, which reduces the costs associated with enzyme supply. When ancestral sequence reconstruction is combined with other protein engineering techniques, the substrate specificity and regioselectivity of the enzymes can be tailored towards the production of high value chemicals from diverse feedstocks.

Innovative lipid delivery systems for bioactive compounds

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Our work focuses on developing lipid-based nanocarriers (LNPs) that protect, stabilize, and improve the delivery of sensitive active ingredients in pharmaceutical and cosmetic formulations [1–5]. These systems are designed to address two key limitations of conventional topical products: (i) the chemical/physical instability of many actives (e.g., oxidation, hydrolysis, photodegradation, recrystallization, loss of activity during storage) and (ii) limited, poorly reproducible transport across the skin barrier. Consequently, even promising actives may lose efficacy in real formulations due to exposure to light, oxygen, water, and incompatible excipients.

Topical therapy is further limited by low skin permeability, especially for hydrophilic, high-molecular-weight, or fragile compounds, while poorly soluble actives often show low bioavailability and uneven distribution. Solid lipid nanoparticles (SLNs) and nanostructured lipid carriers (NLCs) provide biocompatible matrices with tunable structures that increase loading, shield actives, enhance interaction with the stratum corneum, and enable controlled or sustained release, helping to reduce irritation linked to high burst concentrations. In addition, these carriers can improve sensory properties and formulation aesthetics (e.g., spreadability, skin feel) by stabilizing the active within a homogeneous colloidal system. They also offer practical flexibility for integrating diverse actives, from antioxidants and vitamins to peptides and plant-derived compounds, within the same technological platform.

This technology relies on advanced colloidal design supported by multi-parameter characterization, including particle size distribution (with polydispersity) and diameter, zeta potential to ensure stability and reproducible performance. We harmonized this with stability testing and functional validation through in vivo biophysical skin assessment (e.g., Visoline®, Visioscan®, and other instrumental methods as needed), linking carrier properties with measurable skin outcomes.

Overall, this integrated workflow – rational carrier design, scalable production (high-pressure homogenization), rigorous colloidal characterization, and in vivo validation – helps overcome instability, low permeability, and limited release control, enabling more effective and scalable topical formulations for both pharmaceuticals and cosmetics.

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Valorization of metal-accumulating plant biomass for the preparation of polymetallic catalysts and their catalytic application

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Human activities and industrial development have led to significant contamination of industrial and post-mining areas with heavy metals. This is a global environmental problem. The presence of metals at high concentrations is highly toxic not only to humans but also to entire ecosystems. An interesting approach for removing metals from contaminated soils is phytoremediation, a process based on the use of plants capable of accumulating metals in their above-ground parts. The resulting biomass is contaminated, as it contains heavy metals in high concentrations.

The aim of this presentation is to demonstrate a strategy for the valorization of such biomass through its transformation into polymetallic catalysts, which can be applied in a variety of organic synthesis reactions leading to high value-added compounds [1–3].

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Optimization of biocatalytic cascade in the production of raspberry ketone from birch secondary metabolites

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Raspberry ketone is a natural phenolic compound found in the red raspberry and is used as a dietary supplement to regulate body weight [1]. The natural amount of the compound in raspberries is extremely low, estimated between 0.001 to 4.2 mg per kg of fruit, which makes direct isolation of the compound economically unviable. One particularly promising route for the biocatalytic production of raspberry ketone involves a two-step enzymatic transformation of rhododendrol glycosides derived from birch bark [2]. Independent kinetic and enzyme stability studies enabled the development of robust, predictive models for both hydrolysis and oxidation steps. These models have been experimentally validated on an independent data set and used for process simulations, enabling the finding of optimal conditions and the development of efficient and environmentally responsible methods for high-value nutraceutical production.

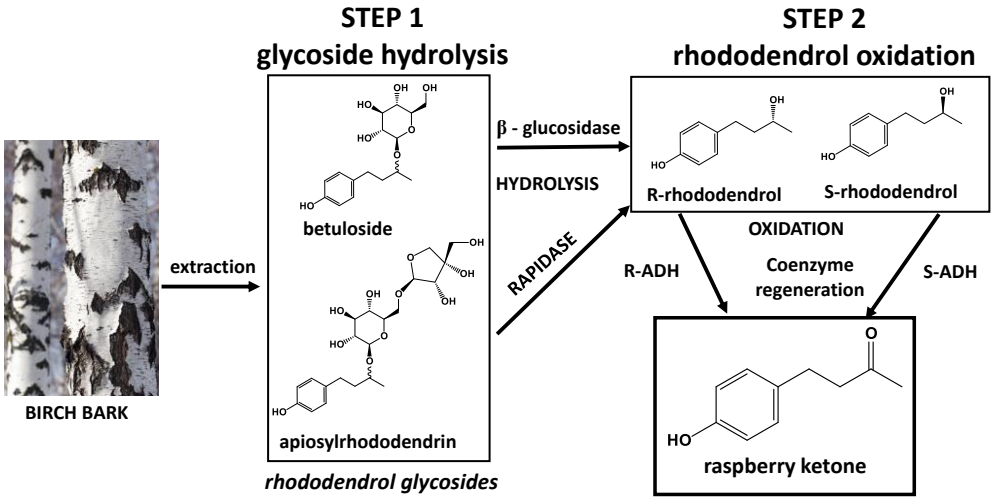

Fig. 2. Schematic overview of the optimized biocatalytic cascade [2].

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Bioactive compounds obtained *via* NaDES extraction: impact on *in vitro* cell migration

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This study explores the application of Natural Deep Eutectic Solvents (NaDES) as sustainable and environmentally friendly alternatives to conventional organic solvents for the extraction of biologically active compounds [1]. Agro-industrial oil processing by-products [2] and selected plant materials [3] were investigated as valuable yet underutilized sources of phenolic compounds and other phytochemicals [4].

NaDES systems were initially screened using COSMO-RS predictive modelling and subsequently applied for plant extraction. The obtained extracts were subjected to comprehensive chemical characterization and biological evaluation in human keratinocytes and human carcinoma cell lines. In addition to antioxidant profiling, cell viability and cell death assays were performed to assess cytotoxic effects. Further investigation was focused on cell migration and wound-healing potential. Betaine-based NaDES extracts, which demonstrated lower cytotoxicity and less pronounced induction of cell death when compared to choline chloride-based ones, were selected for detailed evaluation using scratch and migration assays.

Our findings demonstrate that NaDES enable sustainable extraction while preserving or modulating the functional bioactivity of phytochemicals. The solvent–phytochemical interplay plays a critical role in shaping the overall biological response. Importantly, NaDES-derived extracts show potential beyond conventional antioxidant applications, emerging as promising migration-modulating agents with prospective biotechnological, pharmaceutical, and cosmetic relevance.

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Hydrolates revised: how distillation and analytical methodology fundamentally reshape chemical profile

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Hydrolates are commonly regarded as simple, well-characterized by-products of essential oil distillation. However, their chemical composition remains highly dependent on both the method of production and the analytical approach applied. In this study, hydrolates obtained from selected plant materials were systematically compared with respect to three distillation techniques: hydrodistillation with plant material submerged in water, classical steam distillation in glassware, and steam distillation using a copper apparatus. In parallel, two extraction strategies prior to GC–MS analysis-headspace solid-phase microextraction (HS-SPME) and direct immersion SPME (DI-SPME) were evaluated, alongside direct liquid injection for quantitative assessment.

The results demonstrate that distillation methodology significantly influences hydrolate composition. Hydrodistillation yielded the lowest number of identified compounds, likely due to retention of polar constituents in the aqueous phase and enhanced pre-boiling extraction effects. Steam distillation increased both the number and diversity of detected compounds, while the copper apparatus provided the highest efficiency, presumably due to improved heat transfer and partial rectification effects.

Analytical methodology introduced an additional, equally strong bias. HS-SPME favoured volatile, low-molecular-weight compounds such as monoterpenes and esters, whereas DI-SPME enabled more effective detection of less volatile, more polar compounds including alcohols, acids, and higher molecular weight constituents. Consequently, qualitative profiles varied substantially depending on extraction mode.

A striking discrepancy was observed between SPME-based profiling and direct liquid injection. While SPME methods detected dozens of compounds, direct injection typically quantified only 1–6 constituents. Despite its lower sensitivity, direct injection provided the most reliable quantitative data, as it is not influenced by analyte partitioning equilibria or fiber–analyte affinity inherent to SPME techniques. This was particularly evident in the dominance of single compounds in quantitative profiles (e.g., thujenol, carvone, eugenol), contrasting with the multi-component distributions suggested by SPME.

Overall, the study demonstrates that hydrolates cannot be considered analytically trivial systems. Both the production process and analytical methodology can lead to fundamentally different qualitative and quantitative interpretations. These findings highlight the need for methodological standardization, especially when hydrolates are evaluated for bioactive properties or compared across studies.

From silence to signal: an integrated microgravity–metabolomics platform for discovering microbial bioactives

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Microbial communities in agricultural soils encode hundreds of thousands of biosynthetic gene clusters (BGCs) predicted to produce bioactive metabolites. Yet 80–90% of these BGCs remain transcriptionally silent under standard laboratory culture. This "metabolic dark matter" reflects a fundamental principle: secondary metabolism is tightly regulated by environmental sensing and stress adaptation. In nature, microbes often grow in low-shear diffusion-limited microenvironments with steep nutrient gradients (e.g., biofilm) which trigger stress-response pathways that activate silent BGCs. Standard shake-flask cultures eliminate these cues, suppressing the very pathways most likely to yield novel bioactive. We are developing an integrated platform that combines simulated microgravity and LC-MS/MS metabolite profiling to systematically activate and identify cryptic metabolites from soil microbes. Using curated soil strain collections, we apply simulated microgravity alongside nutrient limitation, osmotic stress, heat, and UV exposure to define reproducible stress recipes that induce silent BGCs expression. We then link these responses to BGCs activation through targeted transcriptional analysis, untargeted metabolite profiling, and genome-guided annotation. High-priority metabolites are validated in agricultural assays, including dual-culture confrontation tests against major plant pathogens and high-throughput growth-inhibition screens. By combining cultivation, metabolite discovery, and functional testing in a single modular workflow, we are building a reusable platform to unlock soil microbial metabolic dark matter and generate translation-ready antimicrobials and plant-growth-promoting metabolites for more sustainable Australian agriculture.

From waste to platform chemicals: adsorption-based fractionation of food waste-derived volatile fatty acids

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Volatile fatty acids (VFAs) are important platform chemicals with rapidly growing industrial demand. Conventionally, VFAs are produced from fossil-based resources, leading to high greenhouse gas emissions. In contrast, anaerobic digestion (AD) of organic waste provides a sustainable alternative, enabling the substitution of fossil-derived VFAs with waste-derived counterparts. During AD, VFAs are formed as intermediate products; however, they are obtained as dilute and complex mixtures of short-chain carboxylic acids together with microorganisms, salts, proteins, and other impurities [1]. This complexity, combined with low concentrations, poses significant challenges for downstream recovery. In particular, fractionation into individual platform chemicals remains difficult due to the similarity in physicochemical properties of VFAs [2]. Affinity-based separation methods, such as adsorption, offer a selective approach by exploiting differences in molecular interactions between VFAs and adsorbent surfaces, thereby enabling targeted VFAs isolation [3].

This study develops an adsorption-based strategy for the selective fractionation of food waste-derived VFA effluent using activated charcoal (AC), with emphasis on acetic acid (AA) and caproic acid (CA) as the dominant components. A two-stage adsorption strategy was applied. In the first stage, higher molecular weight VFAs were preferentially adsorbed onto AC, targeting CA isolation by evaluating key operating parameters, including pH (4, 4.75, 5.5, 7 and 9), temperature (10, 25, 37 and 55 ˚ C), and adsorbent to adsorbate ratio (1.56, 3.13, 6.25, 12.5, 25, 50, 100 g g^{-1}). Desorption was investigated for efficient CA recovery and adsorbent regeneration. In the second stage, conditions were adjusted to retain AA in the aqueous phase with high purity (pH 9, 50 g g^{-1}; pH 4, 6.25 g g^{-1}, room temperature).

Increasing pH from 4 to 9 decreased overall adsorption capacity while enhancing selectivity toward CA. At pH 9, the lowest adsorbent to adsorbate ratio (1.56 g g^{-1}) enabled selective isolation of CA (32.5%); with cumulative removal reaching 90.5% over consecutive cycles. Temperature had a negligible effect on CA adsorption, supporting operation at ambient conditions. Desorption was optimized using 20% (v/v) ethanol in 0.1 M NaOH, achieving near-complete CA recovery and enabling adsorbent reuse over three cycles without significant performance loss. Reducing the regeneration volume to 10 mL yielded CA concentration comparable to the feed. In the second stage, lower AA loss was observed at pH 9 and ratio of 50 g g^{-1}, with AA retained in the liquid phase at 1.2 g L^{-1} and a purity of 90.6%.

In conclusion, these results demonstrate selective isolation of CA and high-purity retention of AA, highlighting the potential of adsorption-based fractionation as an energy-efficient route for valorizing food waste-derived VFAs into platform chemicals.

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Continuous-flow lipase-mediated synthesis of bioactive derivatives from renewable sources

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Biocatalysis combined with continuous-flow processing represents a powerful strategy for the sustainable synthesis of value-added molecules from renewable feedstocks. Recently, we investigated the use of immobilized lipase B from *Candida antarctica* (Lipozyme® 435) as a heterogeneous biocatalyst for the preparation of bioactive derivatives through amidation and transesterification reactions performed in batch and continuous-flow modes.

In the first study, oleoyl ethanolamide, a biologically active fatty acid amide involved in lipid metabolism and appetite regulation, was synthesized from oleic acid contained in a mixture of fatty acids obtained by enzymatic hydrolysis of soapstock, a side-product of high-oleic sunflower oil refining. The lipase-mediated amidation with ethanolamine was carried out in limonene as a renewable solvent. After preliminary batch optimization, the process was transferred to a packed-bed reactor operated in continuous flow. Under optimized conditions (55 °C, residence time 80 min), the system reached steady state after two residence times and operated continuously for more than 150 h with stable conversion and efficient catalyst recycling, leading to a significant increase in catalyst productivity compared with the batch process.

In a second study, lipase-mediated transesterification of ethyl hydroxycinnamates with selected alcohols was investigated for the synthesis of lipophilic hydroxycinnamate derivatives with antioxidant and antimicrobial properties. Batch reactions in diisopropyl ether provided conversions up to 50–70 % within 6 h, while the implementation in a packed-bed flow reactor allowed steady conversions (44–67 %) and higher space–time yield and process productivity. The packed-bed configuration also minimized mechanical degradation of the immobilized enzyme and facilitated catalyst reuse.

The results of these studies will be discussed to demonstrate how immobilized lipases in continuous-flow packed-bed reactors enable efficient, scalable, and environmentally benign routes to bioactive derivatives starting from renewable substrates and agro-industrial side streams.

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Flaxseed oil cakes as a source of proteins - implications for animal cell technology and cellular agriculture

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Protein and peptide hydrolysates derived from oil cakes have attracted considerable interest in the biopharmaceutical and food industries due to their functional and nutritional properties. In animal cell technology, which is widely employed for the production of recombinant proteins, the use of serum-containing media presents significant safety, ethical, and regulatory challenges [1]. Consequently, significant efforts have been focused toward the development of serum-free and animal-component-free media formulations incorporating plant-derived proteins and their hydrolysates from various sources, including oil cakes. This challenge is equally relevant in cellular agriculture (CA), particularly in the production of cultivated meat (CM), where the replacement of animal-derived components is essential for safety and regulatory issues, as well as consumer acceptance.

The purpose of this report is to present the results of research projects investigating flaxseed (*Linum usitatissimum* L.) oil cake as a sustainable source of proteins (FPs) and protein hydrolysates (FPH) for reducing serum levels in cell culture media, while keeping or improving cell culture performance. The potential of FPH to support Chinese hamster ovary (CHO) cell proliferation and IgG production was investigated in low-serum conditions. Furthermore, its applicability was assessed in low-serum media for the cultivation of muscle and adipose cells, the two main cell types required for generating cell-based biomass in CM production.

The results demonstrated that adherent IgG-producing CHO cells cultured in FPH-supplemented media achieved higher cell densities and increased IgG titers compared to standard serum-containing media. In contrast, in suspension CHO cultures previously adapted to chemically defined media, the addition of FPH exerted minimal immediate effects on growth kinetics and recombinant protein productivity [2]. Evaluation of FP in muscle cell models revealed a stimulatory effect on C2C12 myoblast differentiation, demonstrated by an increase in the diameter of formed myotubes [3]. Moreover, preliminary findings in mouse preadipocyte 3T3-L1 cells indicated enhanced lipid accumulation under reduced-serum conditions, suggesting a supportive role of FPs in the adipogenic differentiation.

Overall, flaxseed-derived proteins and hydrolysates show considerable potential as a sustainable, regulatory-compliant media supplements for both biopharmaceutical production and CM technologies, while promoting environmental sustainability, animal welfare, and public acceptance of emerging technologies.

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Every molecule matters: designing biorefineries where no fraction is left behind

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Biorefineries will only fulfil their promise when every fraction of biomass i.e. cellulose, hemicellulose, lignin, and side-streams, earns its highest value. In this keynote, I outline a Fraction-to-Function vision that begins with the pretreatment of recalcitrant biomass and culminates in diversified, high-value product portfolios. The talk highlights how tailored pretreatment strategies yield clean, well-defined biomass fractions that enable downstream pathways. Central to this vision is the concept that every molecule matters. Future biorefineries will fully valorise cellulose, lignin, hemicelluloses, and other side-streams, transforming them into high-value chemicals, polymers, coatings, composites, bio-oils, and functional materials. By designing fractionation with end use functionality in mind, biorefineries can progress from linear biomass processing models to fully integrated, multi-product circular systems. Case vignettes span woody feedstocks to agro-industrial residues, demonstrating pathways that integrate microbial bioconversions into circular, multi-product biorefinery frameworks. The takeaway is simple: when substrate recalcitrance is treated as a design input rather than a barrier, lignocellulosic wastes and residues become assets, enabling resilient, market-ready biorefineries aligned with a true zero-waste vision.

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ORAL PRESENTATION 12

Fungal-based solid-state fermentation: a sustainable approach for lignocellulosic biorefineries

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Lignocellulosic biomass represents the most abundant renewable carbon resource on Earth and a key feedstock for the development of sustainable biorefineries. However, its complex structure, composed of cellulose, hemicellulose and lignin, poses significant challenges for efficient conversion into value-added products. Fungi play a crucial role in natural lignocellulose degradation due to their ability to produce a diverse set of extracellular enzymes capable of depolymerising plant cell wall components.

Fungal-based solid-state fermentation (SSF) has emerged as a promising biotechnological platform for the valorisation of lignocellulosic residues, particularly agricultural and food industry by-products. Under SSF conditions, filamentous fungi can efficiently grow on solid substrates with low water content, mimicking their natural ecological environment and enabling the production of hydrolytic and lignolytic enzymes involved in lignocellulose breakdown. In addition to enzyme production, SSF can facilitate the conversion of lignocellulosic biomass into a range of bio-based products, including bioactive compounds, functional food ingredients, enzymes, biofertilizers, and novel biomaterials.

This lecture will provide an overview of recent advances in fungal SSF for lignocellulosic biorefineries, highlighting the potential of lignocellulosic residues as fermentation substrates and discussing strategies for improving process standardization and products valorisation.

Chemo-enzymatic valorisation of industrial hemp distillation waste from essential oil production

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Waste from the agri-food industry is gaining increasing value in the modern economy, and exploiting the potential of these resources requires a systemic change: the use of renewable organic residues as starting material for a sustainable value creation.

The production of essential oils by steam distillation is invariably associated with large amounts of organic waste which is normally disposed of or returned to the fields, although it may still contain some valuable components. In particular, Cannabis sativa essential oil produced by steam distillation of the apical part of industrial hemp plants [1] yields a soaked biomass residue that may contain high-value cannabinoids [2]. From the perspective of sustainable exploitation of agricultural resources, a method to extract cannabidiol (CBD) from such waste was demonstrated and scaled-up, using bioethanol as a renewable bio-based solvent and without requiring chromatographic separation, with an overall yield of 10.1 mg CBD per g waste. The work paves the way to an integrated complete utilisation of industrial hemp byproducts. Furthermore, two alternative lipase-mediated chemo-enzymatic derivatisations have been designed and optimised for the conversion of the recovered CBD into cannabielsoin (CBE), an underexploited cannabinoid with attractive bioactivity [3,4]. The processes are practical and efficient, with 31–47% isolated yields and green metrics comparing well with the established chemical alternatives [5].

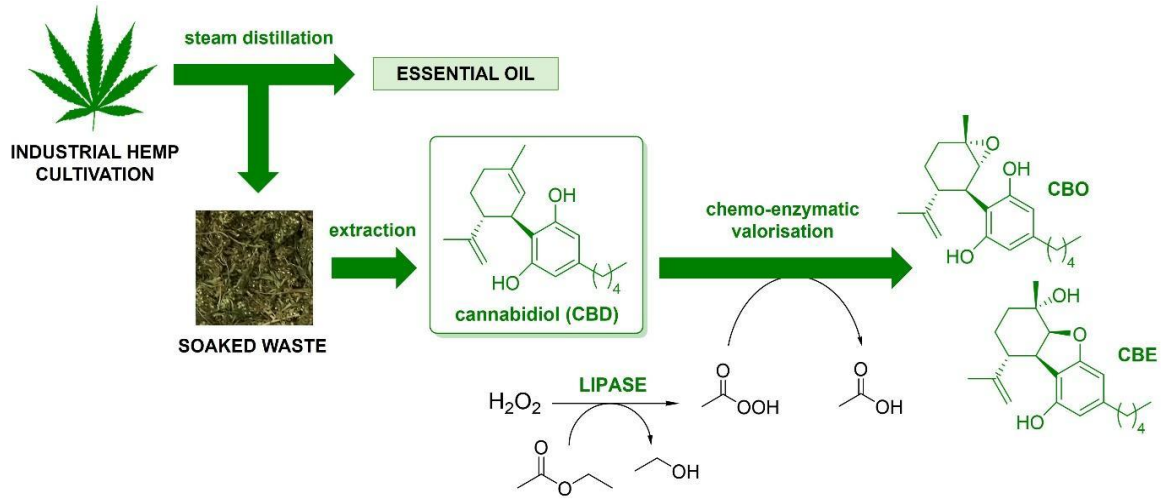

Fig. 1. Integration of extraction and chemoenzymatic valorisation to obtain valuable cannabinoids from *C. sativa* distillation waste.

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Exploring the promiscuous activity of fatty acid hydratases

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Fatty acid hydratases (FAHs, EC 4.2.1.53) are a class of enzymes that catalyze the addition of water to the non-activated *cis*-double bond of unsaturated fatty acids. FAHs are highly regio- and stereoselective leading to a single isomeric form of hydroxy-fatty acids. For example, oleate hydratase (Ohy) transforms oleic acid into (*R*)-10-hydroxystearic acid (10-HSA) with almost complete regio- and enantioselectivity [1].

Our recent studies [2] demonstrate that FAHs can catalyze biochemical reactions other than hydration, thereby exhibiting promiscuous enzymatic activity. Here, we report on the synthetic versatility of an oleate hydratase from *Lactobacillus rhamnosus* ATCC 53103, which, in addition to efficiently hydrating oleic acid, also displays epoxide hydrolase and isomerase activities (Fig. 1). More specifically, we illustrate the enantioselective hydrolysis of epoxides derived from oleic and elaidic acids, as well as the double-bond isomerization of natural polyunsaturated fatty acids such as linoleic, α-eleostearic, and punicic acids.

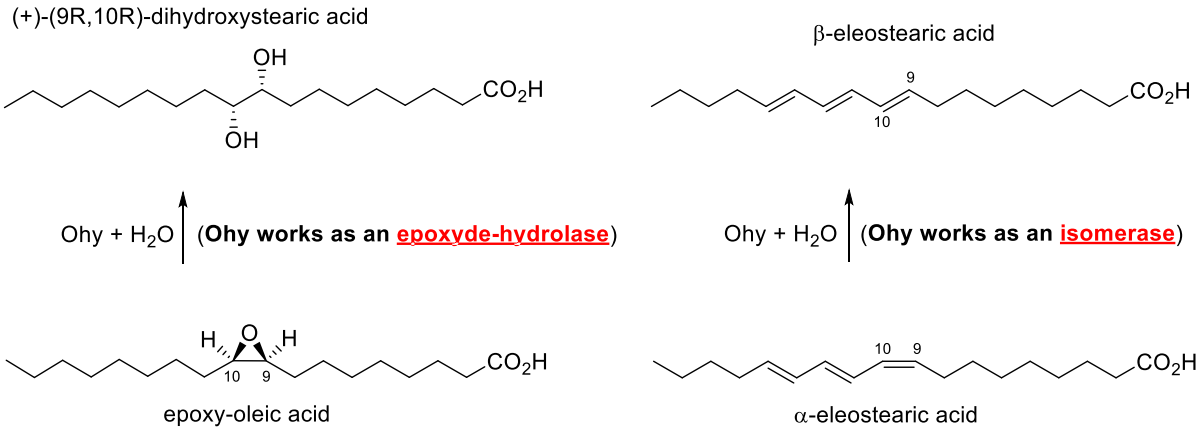

Fig. 1. Some representative examples of chemical transformations catalysed by a promiscuous oleate hydratase

ACKNOWLEDGEMENTS

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Microbial biocatalysts for the preparation of flavonoid metabolites: applications in metabolic and biological research

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Flavonoids constitute a diverse class of plant polyphenols commonly present in the human diet and linked to a wide range of biological activities. In plants and food products they are mainly found as glycosides; however, following ingestion they undergo extensive metabolic transformations mediated by intestinal microbiota as well as hepatic and intestinal enzymes. These processes generate numerous conjugated and degradation products with considerable structural diversity. Limited commercial availability of such metabolites, which are often accessible only as costly analytical standards, significantly restricts comprehensive metabolic studies, analytical characterization, and investigations of structure–activity relationships.

This presentation outlines recent progress in the use of microbial enzymes as efficient and sustainable tools for the regioselective synthesis of flavonoid metabolites. Particular attention is given to enzymatic strategies including glycosylation and deglycosylation mediated by glycosyltransferases and glycosidases, C-ring cleavage catalyzed by quercetin 2,3-dioxygenases, and conjugation reactions such as sulfation and glucuronidation performed by bacterial aryl sulfotransferases[1] and glucuronosyltransferases derived from *Streptomyces*. In comparison with conventional chemical synthesis, these biocatalytic approaches typically involve single-step or short synthetic routes carried out under mild and biocompatible conditions, frequently producing metabolites identical to those formed *in vivo*.

Libraries of flavonoid glycosides, sulfates, glucuronides, and microbial degradation products prepared using these methods have been successfully utilized as reference standards in in vitro gut fermentation experiments[2,3] and human intervention studies[4], facilitating detailed investigation of interindividual variability in flavonoid metabolism. Altogether, microbial enzymatic synthesis provides a practical and versatile platform for generating flavonoid metabolites relevant to research in food chemistry, biomedicine, and microbiome science.

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Noble metal nanoparticles as elicitors enhancing biotechnological production of bioactive metabolites in plant *in vitro* systems

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Noble metal nanoparticles (NPs) are promising abiotic elicitors for precision modulation of plant secondary metabolism [1-3]. The non-ionic Au, Pt, Ag and Cu nanoparticles (5–15 ppm; ≤5 nm) in agitated shoot cultures of kale (*Brassica oleracea* var. *acephala*) maintained on MS medium were tested. The assessment of growth, elemental bioaccumulation, glucosinolates (GLS), phenolics and antioxidant capacity were performed. The response was element-specific and dose-dependent. Platinum nanoparticles (PtNPs) delivered the strongest, system-wide enhancement: total GLS increased from 4.06 µmol/g DW (control) to 9.75 µmol/g DW at 10 ppm, with marked rises in glucobrassicin (5.10 µmol/g DW), 4-methoxyglucobrassicin (1.59 µmol/g DW) and neoglucobrassicin (2.67 µmol/g DW). In the phenolic pool, rosmarinic acid reached 58.49 mg/100 g DW (15 ppm) and caffeic acid 26.04 mg/100 g DW (10 ppm) under PtNPs, coinciding with highest FRAP, DPPH, CUPRAC and ABTS values among all treatments. AuNPs and AgNPs elicited moderate, composition-dependent remodeling of GLS as well as phenolics, whereas CuNPs favored selected shifts (e.g., chlorogenic acid 32.29 mg/100 g DW at 15 ppm) with weaker global antioxidant gains. ICP-MS confirmed dose-dependent metal uptake (PtNPs: 39.64 mg/100 g DW; BCF up to 26.43 at 15 ppm), while multivariate analyses (PCA, heatmaps) resolved clear, nanoparticle-specific metabolic fingerprints. Collectively, noble metal NPs act as tunable elicitors enabling composition-controlled biofortification of *B. oleracea* var. *acephala* biomass *in vitro*, and define scalable set-points (PtNPs 5–10 ppm; AgNPs 10 ppm; CuNPs 15 ppm) for bioreactor translation and quality-by-design control. The negligible metal carry-over and the absence of detectable free nanoparticles in extracts confirm a clean-extract profile, strengthening the safety-by-design concept and supporting the use of nano-elicited biomass in high-purity nutraceutical and cosmetic applications.

ACKNOWLEDGEMENTS
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Caffeic acid-functionalized silver nanoparticles enhance anti-melanogenic activity in melanocytes

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Hyperpigmentation disorders are driven by dysregulated melanogenesis, where tyrosinase plays a key role. Caffeic acid is a bioactive polyphenol with anti-melanogenic potential, but limited cellular efficacy. Here, we evaluated silver nanoparticles functionalized with caffeic acid (AgNP-1) in comparison with a heterofunctionalized system (AgNP-2) in α-MSH-stimulated B16F10 melanocytes. Cells were treated (0.1–20 µg/mL) and melanin content, cell viability, and tyrosinase expression were analyzed. Both nanoparticles reduced melanin production in a dose-dependent manner. AgNP-1 showed the strongest effect, reducing melanin levels to basal conditions and outperforming kojic acid at much lower concentrations. At 20 µg/mL, both nanoparticles reduced intracellular melanin similarly to kojic acid at 1000 µg/mL. AgNP-1 significantly downregulated tyrosinase expression, whereas AgNP-2 showed limited effects. Free caffeic acid was inactive, highlighting the importance of nanoparticle functionalization. These results demonstrate that nano-functionalization enhances the anti-melanogenic bioactivity of caffeic acid, supporting its application as a bioactive ingredient for skin depigmentation.

Lactate: nutrient, oncometabolite, or key modulator of cellular metabolism

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Recent advances have highlighted the pivotal role of metabolites, traditionally viewed solely as metabolic intermediates, as critical signaling molecules in diverse physiological and pathological contexts. These compounds can function as enzyme cofactors, covalently modify proteins as post-translational modifications, or act as ligands for specific receptors. Among these, lactate—historically considered a mere glycolytic byproduct—has emerged as a central regulator in multiple biological processes.

Lactate accumulates acutely during exercise and chronically within tumor microenvironments and sites of inflammation, often representing the predominant metabolite as confirmed by mass spectrometry analyses.

Novel insights have elucidated lactate sensing mechanisms involving specific transporters and G protein-coupled receptors (e.g., GPR81 and GPR132), alongside its capacity to modulate cellular functions through metabolic reprogramming [1] and a recently characterized post-translational modification termed lactylation [2]. Protein lactylation, particularly on histone lysine residues, has been shown to influence gene expression epigenetically, with emerging evidence demonstrating extensive lactylation beyond histones to numerous non-histone proteins, underscoring its broad regulatory significance [3]. Furthermore, lactate functions as a master regulator of hypoxia responses, facilitating cell proliferation and angiogenesis [1].

However, critical gaps remain concerning the identification and characterization of lactate-specific transporters and receptors, especially within gastrointestinal tissues. Importantly, targeting lactate-mediated signaling pathways holds therapeutic promise for mitigating inflammation, autoimmunity, and enhancing antitumor immunity. This presentation will synthesize current knowledge on lactate's multifaceted signaling roles and discuss future directions for research and clinical application.

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Halogenated flavonoid derivatives modulating gut microbiota composition as supportive agents in colorectal cancer therapy

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Flavonoid compounds containing additional halogen atoms exhibit documented enhanced antibacterial and cytotoxic properties as compared to their non-halogenated counterparts [1]. Our previous research has shown that examined compounds modulated the gut microbiota composition [2]. The aim of the study was to evaluate the cytotoxic, pro-apoptotic, and anti-inflammatory activity of 3'-bromo-5'-chloro-2'-hydroxychalcone, 8-bromo-6-chloroflavanone, and 8-bromo-6-chloroflavone against human colorectal cancer cell lines HCT-116, HT-29, and Caco-2, as well as normal human intestinal epithelial cells FHC. The compounds were synthesized via a Claisen–Schmidt condensation, followed by further structural modifications, typical cyclization, and oxidation steps, which led to flavanone and flavone derivatives. Cell viability in the presence of the investigated flavonoids was assessed using the colorimetric SRB (sulforhodamine B) assay, which enabled evaluation of cell proliferation based on total cellular protein content. The results demonstrated that flavonoids containing –Br and –Cl substituents exerted a cytotoxic effect on colorectal cancer cells (Table 1), while importantly showing no significant toxicity toward normal FHC cells. Such selectivity suggests their potential as promising lead structures for the development of novel anticancer agents targeting colorectal cancer. Future studies will include evaluation of compound permeability across the intestinal barrier using the Caco-2 cell monolayer model, a widely used *in vitro* model for predicting intestinal drug absorption.

Table 1. IC_{50} values of the compounds against HCT-116, HT-29 and Caco-2 cells.

	HCT-116	**HT-29**	**Caco-2**
3'-Bromo-5'-chloro-2'-hydroxychalcone	44.0 ± 13.2*	52.1 ± 1.1	97.8 ± 6.2
8-Bromo-6-chloroflavanone	38.4 ± 8.5	95.6 ± 3.8	260.7 ± 11.5
8-Bromo-6-chloroflavone	-	277.7 ± 38.1	-
2'-Hydroxychalcone	104.3 ± 9.3	116.8 ± 1.1	235.5 ± 3.3
Flavanone	213.5 ± 6.1	155.3 ± 8.1	251.9 ± 23.1
Flavone	197.9 ± 8.2	128.0 ± 4.7	274.4 ± 24.2
Quercetin	136.1 ± 6.7	204.8 ± 9.9	-

* IC_{50} values are reported in µM.

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Pluchea grevei essential oil as multifuntional active ingredient

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Natural multifunctional ingredients are increasingly sought in cosmetic and dermocosmetic formulations due to their ability to provide antioxidant protection and microbiome-supporting antimicrobial activity. The aim of this study was to evaluate the bioactive potential of a commercial essential oil (EO) obtained from *Pluchea grevei* (Famonty) as a multifunctional cosmetic ingredient and to explore its applicability in a topical formulation.

The investigated material was a commercial EO of *Pluchea grevei* obtained by steam distillation of aerial plant parts harvested in Madagascar (Astérale). Chemical and olfactory characterization were performed using gas chromatography coupled with mass spectrometry (GC-MS) and chromatography–olfactometry (GC-O). Antioxidant capacity was evaluated using DPPH and ABTS radical scavenging assays and FRAP - Ferric Reducing Antioxidant Power Assay. Antimicrobial activity was determined by broth microdilution assay against selected microorganisms relevant to skin microbiota and cosmetic contamination, including *Staphylococcus aureus (ATCC 6538), Staphylococcus epidermidis (ATCC 12228), Cutibacterium acnes (ATCC 11827), Corynebacterium minutissimum (ATCC 23348), Candida albicans (ATCC 10231), Malassezia furfur (ATCC 14521), Micrococcus luteus (ATCC 4698), Pseudomonas paraaeruginosa (ATCC 9027), Staphylococcus capitis (ATCC 35661),* and *Acinetobacter baumannii (ATCC 19606)*. A prototype cosmetic cream containing the EO was formulated to evaluate its applicability in dermocosmetic products.

The EO showed moderate antioxidant activity (IC_{50} ~32 mg/mL in DPPH, ~10 mg/mL in ABTS and ~1885 µmol TE/g in FRAP assays) and selective antimicrobial effects against skin-assosiated pathogenes and opportunistic bacteria (Table 1). A prototype cream (Aqua, Helianthus Annuus Seed Oil, Cocos Nucifera Oil, Butyrospermum Parkii Butter, Cetearyl Olivate, Sorbitan Olivate, Glycerine, Tocopherol, Dehydroacetic Acid, Pluchea gravei Oil, Benzyl Alcohol, Linalool, Limonene) confirmed compatibility with a natural emulsion system. Also skin cell cytotoxicity and ecotoxicity towards marine organisms were determined.

Overall, the results indicate that *Pluchea grevei* essential oil possesses combined antioxidant and antimicrobial properties relevant to skin care applications. These findings support its potential as a multifunctional cosmetic ingredient providing both sensory and biological benefits in dermocosmetic formulations.

Table 1. Minimum inhibitory concentrations (MIC, µg/mL) of *Pluchea grevei* EO towards pathogenic and opportunistic bacteria

	S.aureus	P. paraaeruginosa	A. baumani	C. acnes	C. minutissimum
MIC	400	800	100	800	800

Glycosylated mycotoxins – a hidden challenge in food safety

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Mycotoxins are toxic secondary metabolites produced by filamentous fungi that frequently contaminate cereal crops and represent a persistent threat to global food safety. In recent years, increasing attention has focused on glycosylated ("masked") mycotoxins, which are mainly formed in plants as part of detoxification mechanisms catalyzed by UDP-glycosyltransferases. These conjugated forms often escape routine analytical detection, remain stable during food processing, and may be hydrolyzed back to their toxic parent compounds during digestion, complicating exposure assessment and risk evaluation.

This contribution summarizes current knowledge on the formation, occurrence, and biological significance of glycosylated derivatives of major mycotoxins, including trichothecenes (e.g., deoxynivalenol and T-2/HT-2 toxins), zearalenone, Alternaria toxins, ochratoxins, and ergot alkaloids. Glycosylation is primarily catalyzed by plant enzymes that convert toxins such as deoxynivalenol to deoxynivalenol-3-glucoside or zearalenone to its 14-glucoside, thereby reducing acute toxicity to the host plant. However, these conjugates may persist during food processing and can be cleaved by intestinal microbiota, releasing the original toxic compounds and contributing to delayed exposure.

Analytical detection of modified mycotoxins, therefore, presents an emerging challenge. Modern LC-MS/MS methods increasingly include glycosylated derivatives, but the limited availability of reference standards and toxicological data still complicates comprehensive risk assessment. The synthesis and characterization of mycotoxin glycosides – using chemical, enzymatic, or microbial approaches – are essential for providing analytical standards and enabling detailed studies of their biological activity.

In summary, glycosylated mycotoxins represent an important but still insufficiently understood group of food contaminants. A deeper understanding of their biosynthesis, stability, metabolism, and analytical detection is essential for accurate exposure assessment and the development of improved strategies to ensure food safety.

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Combining enzyme inhibition assays to targeted metabolomics for food bioactivity assessment

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Enzymes are proteins acting as biochemical catalyst providing high specificity towards their substrates and resulting in products that can be used in different ways, i.e., correlating product rate synthesis to enzyme activity. Within the food bioactivity scope, identifying bioactive edible plant sources with enzyme inhibitory potential has attracted significant attention. Considering that enzymes tend to be hyperactive during a disease state, identifying natural bioactive inhibitors that can be potentially exploited for nutraceutical purposes is of topical interest. Within this work, we will present our research activities in the field focusing on three different pillars. First, in-house assay optimisation is of utmost importance to acquire optimal analytical performance for an applied enzyme assay [1]. Commonly, published studies cite outdated protocols without any discussion or insight on how to achieve reliable and repeatable results hindering the reproducibility of such methods. Secondly, a comprehensive summary of applied enzyme assays will be presented including enzymes related to: i) cognitive function (acetylcholinesterase and butyrylcholinesterase, ii) digestive function (pancreatic lipase, alpha-amylase and alpha-glucosidase) and skin health (tyrosinase and collagenase). In all cases, published [2, 3] and under implementation case-studies will be discussed focusing on food sample preparation strategies and implementation of bioactive extracts into functional foods. Last but not least, the hyphenation of enzyme inhibitory assays with targeted metabolomics increases the capacity and impact of such approaches. By using ultra high-performance liquid chromatography coupled to high resolution mass spectrometry (UHPLC-HRMS), the tested extract composition can be tentatively identified without the need of analytical standards. This significantly reduces the analytical cost while targeted screening allows to develop quantitative methods focused on molecules identified in the extracts of interest. So far, selecting analytes for quantitative method development has been mostly based on literature findings that not necessarily apply to one's under-investigation samples. Additionally, targeted metabolomics generate a wealth of data indicating the need to implement chemometric tools, both non-supervised and supervised models, for result interpretation [2, 3]. Importantly, chromatographic and enzyme assay data can be fused potentially revealing correlations and connecting the in vitro inhibitory effects to certain identified bioactive molecules. In conclusion, combining enzyme assays to targeted metabolomics can indicate edible plant sources with bioactive potential paving the road for functional food development.

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The BIOPEP-UWM database of peptides and proteins – opportunities available in 2026

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Peptides belong to the most extensively studied classes bioactive compounds of food origin. The BIOPEP-UWM database [1,2] serves as popular source of information about above compounds (c.a. 85 thousands visits to date). The database is continuously modified and improved. The last improvements concerned the repository of amino acids and modifications as well as database of allergenic proteins and their epitopes.

The BIOPEP-UWM repository of amino acids and modifications enables annotation of peptides containing not only canonical (proteinogenic) amino acids, but also other amino acids (e.g. phosphorylated or non-proteinogenic) and other monomers (carboxylic acids, alcohols etc.). The above residues are annotated using special biological code. Sequences written using set of symbols beyond canonical amino acids are used in the databases of biologically active peptides, sensory peptides and proteins, available as a queries for the search engines of particular databases, conversion into SMILES code, creation of profiles of potential biological activity as well as proteolysis simulations. Examples of processing of protein sequences, annotated using e.g. symbols of modified amino acids have been published [3,4].

Framework for structural classification of allergenic proteins, based on standard classification of protein families is the last innovation introduced in the BIOPEP-UWM (Database of allergenic proteins and their epitopes [2]). Insertion of data to new version of above database is recently in progress. Links between pages of individual peptides in various databases have been added to the databases of bioactive, sensory and virtually bioactive peptides.

The BIOPEP-UWM database is used also as a tool for education, among bioinformatics tools applied in during studies at the Faculty of Food Science of UWM in Olsztyn [5]. The exercise applied within biochemistry course within Food Safety and Certification field of study includes experimental hydrolysis of casein using trypsin, chymosin and zingipain, followed by in silico simulated proteolysis by the BIOPEP-UWM proteolysis tool. Computer simulations of casein hydrolysis by chymosin and zingipain may serve for explanation of milk clotting by both above enzymes in contrast to trypsin.

ACKNOWLEDGEMENTS

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Circular economy in the sugar industry: Waste valorisation for the discovery, development, production and application of bioactive compounds

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The sugar beet (*Beta vulgaris L.*) industry generates substantial quantities of by-products – including molasses, sugar beet pulp (SBP), thick juice, and beet leaves – that represent an underexploited reservoir of bioactive compounds with significant pharmaceutical, nutraceutical, and industrial potential. Presented work reviews the application of circular economy principles to the systematic valorisation of these by-product streams, focusing on five major classes of bioactive compounds: fructooligosaccharides (FOS), triterpenoid saponins, betaine, structural polysaccharides, and polyphenols. Enzymatic transfructosylation of sucrose-rich feedstocks such as molasses and thick juice yields prebiotic FOS at 49–56% conversion efficiency, rivalling pure sucrose substrates while dramatically reducing production costs. Sugar beet pulp contains 75–85% polysaccharides on a dry weight basis, amenable to sequential fractionation into arabinose, galacturonic acid, and glucose streams for conversion to platform chemicals including arabitol, mucic acid, and levulinic acid. Triterpenoid saponins accumulate preferentially in beet fibre (12.7 g/kg), while betaine concentrates in molasses (3–8% w/w) and exhibits clinically validated homocysteine-lowering and hepatoprotective activities. Integrated biorefinery approaches combining FOS, pectin, ethanol, and biogas co-production can achieve 20–45% reductions in global warming potential compared to conventional processing. This review synthesises recent advances in extraction technologies, enzyme engineering, life cycle assessment, regulatory frameworks, and industrial applications – including bioplastics, 3D bioprinting, and functional foods – to present a roadmap for transforming the sugar industry from a linear commodity model into a circular bioeconomy paradigm.

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Antihypertensive and sex-dependent metabolic effects of protein hydrolysates from agri-food by-products: Potential role of gut microbiota

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Gut microbiota (GM) modulation and the consumption of bioactive peptides present in protein hydrolysates (PHs) have emerged as promising preventive strategies against hypertension (HTN) and metabolic syndrome (MetS). In addition to their effects on key MetS pathways, bioactive peptides may also influence GM by acting as nitrogen sources and exerting antimicrobial effects. Importantly, these peptides can be generated from agri-food by-products, thereby contributing to circular economy approaches. The aim of project PID2020-114608RA-I00 was to obtain PHs from agri-food by-products with the ability to modulate GM and exert antihypertensive effects. A total of 100 PHs were generated from animal- and plant-based by-products under different enzymatic hydrolysis conditions. Several PHs showed relevant angiotensin-converting enzyme inhibitory activity and promoted the growth of bacterial groups considered beneficial for health after *in vitro* fermentation with GM from hypertensive rats (SHR). Six selected hydrolysates were evaluated after acute oral administration (55 mg/kg) in SHR. Two PHs, one of animal origin and one of plant origin, showed antihypertensive effects. Notably, these effects disappeared when animals were previously treated with an antibiotic cocktail to drastically reduce GM, suggesting, at least in part, a GM-mediated mechanism. In addition, four-week administration of the two PHs (55 mg/kg/day) to SHR lowered BP during the first two weeks, and modified cecal GM composition, mainly in the group receiving the animal-derived PH, in which *Akkermansia* was the bacterial group most clearly associated with the biological effect.

Considering these findings, project PID2023-151563OB-I00 aimed to validate the antihypertensive effect of the two selected PHs after prolonged administration in a diet-induced animal model of MetS, to evaluate additional metabolic effects, and to determine whether the response was sex-dependent. Wistar rats were fed a cafeteria (CAF) diet for 12 weeks and during the last 4 weeks, animals received daily either vehicle (water) or PHs (55 mg/kg/day). Food intake, body weight, and systolic BP were recorded weekly, and oral lipid and glucose tolerance tests were performed during weeks 2 and 3 of supplementation, respectively. The CAF diet increased SBP to hypertensive values, whereas both PHs significantly reduced SBP in CAF-fed animals of both sexes. They also exerted other sex-dependent metabolic effects that differed between PHs. The plant-derived PH showed the strongest overall effect: in females, it reduced adiposity index and plasma leptin levels and tended to decrease postprandial triglycerides, whereas in males it improved glucose tolerance.

These results show that PHs obtained from agri-food by-products have potential as functional ingredients for the prevention of HTN and MetS, with a multitarget mode of action that includes BP-lowering effects and possible involvement of the GM. They also highlight the importance of considering both sexes in these studies while supporting the valorization of agri-food by-products.

ACKNOWLEDGEMENTS

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Synthesis and antibacterial activity of fabimycin – a novel antibiotic candidate against Gram-negative bacteria

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Antibiotic resistance represents a growing global health threat and significantly limits the effectiveness of currently available antibacterial therapies. Gram-negative bacteria are particularly problematic due to the presence of an outer membrane that restricts the penetration of many antibiotics and contributes to multidrug resistance. Consequently, the development of new antibacterial agents targeting these pathogens has become an urgent priority in modern medicinal chemistry. Fabimycin is a recently developed antibiotic candidate designed to selectively target Gram-negative bacteria by inhibiting the FabI enzyme involved in fatty-acid biosynthesis. The aim of this study was to synthesize Fabimycin and evaluate its antibacterial activity against selected Gram-negative bacterial strains. The compound was obtained through a multistep synthetic procedure and subsequently characterized using standard analytical techniques Subsequently, its antibacterial activity will be evaluated using established microbiological assays. The results are expected to provide insight into the antibacterial potential of Fabimycin against Gram-negative pathogens. This study may contribute to the development of new therapeutic strategies targeting antibiotic-resistant bacteria and highlight the potential of FabI inhibitors as promising candidates for next-generation antibacterial drugs.

Whole cell transformation of cannabidiol

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Interest in naturally derived bioactive compounds has increased markedly in modern medicine and cosmetology [1]. The discovery of novel derivatives of established lead structures, including cannabinoids, represents an important direction in pharmaceutical research. Biotechnological approaches, particularly microbial biotransformation, provide efficient and environmentally sustainable tools for forming structurally modified molecules with potentially improved biological properties. The diverse enzymatic machinery of microorganisms enables highly regioselective transformations that are often difficult to achieve by conventional chemical methods. A major limitation of many phytochemicals, including cannabinoids, is their poor bioavailability, which arises from their pronounced hydrophobicity [2].

The aim of this study was to generate more hydrophilic cannabidiol derivatives through microbial biotransformation as a strategy to improve their physicochemical properties [3]. Filamentous fungi were shown to catalyze the hydroxylation and glycosylation of cannabidiol **(1)**, the principal non-psychoactive cannabinoid. A key methodological challenge was the isolation of biotransformation products from the complex culture broth containing numerous endogenous secondary metabolites. To overcome this limitation, multi-step purification procedures based on preparative flash liquid chromatography with UV detection were optimized. In parallel, analytical methods using high-performance liquid chromatography (HPLC) coupled with UV–Vis and charged aerosol detection (CAD) were developed. These methods enabled accurate monitoring of the biotransformation process and reliable assessment of fraction purity.

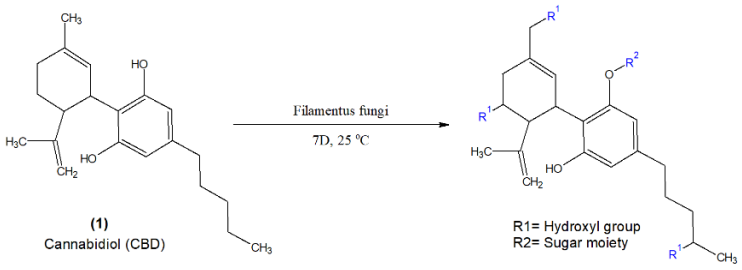

Fig. 3. Microbial transformation of cannabidiol (1).

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Upcycling spent hops: a low-cost method for the isolation of xanthohumol

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Xanthohumol (Fig. 1) is a prenylated flavonoid present only in hops. It is widely known for its therapeutic potential in the prevention and treatment of a large number of diseases, including oncologic diseases, metabolic disorders, neurogenerative diseases and many more [1]. Despite it being the most prominent hop flavonoid, providing 80–90% of the total flavonoid content [2], its extraction from hops its highly limited due to its high thermal and light sensitivity [3].

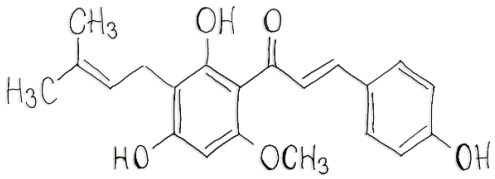

Fig. 1. Chemical structure of xanthohumol.

Supercritical hop extraction is a method which allows highly effective extraction of hop acids from hop pellets. It utilizes CO_2 in its supercritical state, in which xanthohumol is very poorly soluble [4], therefore the majority of this compound remains in the by-product. The main goal of this research is to obtain highly xanthohumol-enriched extract using accessible and economical methods. This study details pretreatment of spent hops, optimization of the extraction process and purification of the obtained extract. The final method utilizes triple-phase extraction system and macroporous resin purification, which consequently allows for achieving a significant improvement of the purity level of xanthohumol, presented in Fig. 2.

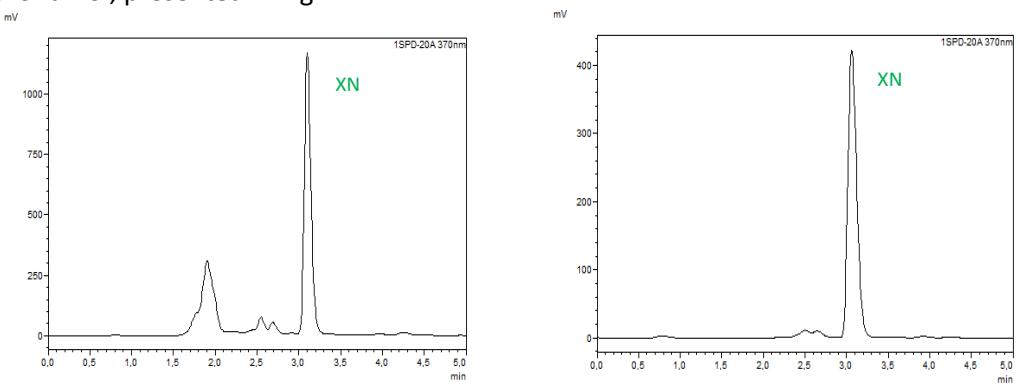

Fig. 2. HPLC chromatogram of the xanthohumol (XN) extract before and after purification.

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Blue-biorefinery of brown macroalgae: green extraction strategies for phlorotannins recovery from *Fucus* sp.

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Marine biomass is gaining increasing attention within blue economy and circular biorefinery strategies as a sustainable source of high-value bioactive compounds. Brown macroalgae, particularly Fucus sp., synthesize phlorotannins, unique polyphenols with significant antioxidant and biotechnological potential. However, their efficient and sustainable extraction remains a technological bottleneck for industrial valorisation. This study aimed to develop and optimize green extraction strategies for phlorotannins recovery and compare them with conventional ethanol maceration. The conventional method yielded 17.30% extract (DW basis), corresponding to 30.9 mg GAE/g DW total phenolic and 1.91 mg PGE/g DW phlorotannins.

Enzyme-assisted extraction (EAE) using Viscozyme® and Alcalase® was evaluated. Omitting thermal enzyme inactivation significantly improved phenolic recovery, confirming the thermal sensitivity of phlorotannins. Viscozyme without heat inactivation produced the highest phlorotannins yield among enzymatic treatments, reaching 1.50 mg PGE/g DW.

To further enhance sustainability, natural deep eutectic solvents (NADES) based on betaine: urea and lactic acid: urea were investigated. Optimized lactic acid: urea extraction at reduced solvent volume (2.5 mL) and mild temperature (30 °C) achieved the highest phlorotannins recovery (5.23 mg PGE/g DW) and high total phenolic content (37.52 mg GAE/g DW), demonstrating improved selectivity and reduced energy input. These results demonstrate that NADES-based extraction offers a rapid, low-energy, and scalable alternative to conventional solvent extraction, supporting sustainable phlorotannins valorisation within a blue-biorefinery framework.

Cannabidiol as a nefroprotective agent against ionophore coccidiostat cytotoxicity

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The presence of veterinary drug residues in the environment poses a threat to public health and animal welfare. Ionophore coccidiostats (lazalocid (LAS), monensin (MON) and narasin (NAR)) are commonly used in animal production. Overdosing, errors in feed mixing or interactions with other drugs pose a risk of nephrotoxicity in animals, which has also been observed in humans as a result of accidental poisoning. Cannabidiol (CBD), known for its antioxidant and cytoprotective properties, may have a protective effect on kidney cells by mitigating the toxic effects of veterynary drugs.

The aim of this study was to evaluate the effect of CBD on cytotoxicity induced by ionophore coccidiostats in human kidney cells (HEK-293). HEK-293 cells were exposed to LAS, MON, and NAR, either alone or in combination with CBD. Cytotoxicity was assessed using mitochondrial activity (MTT), lysosomal activity (NRU), total protein content (TPC), and membrane integrity (LDH) assays. The nature of interactions between CBD and the coccidiostats was determined using interaction analysis.

The study ionophores induced dose-dependent the cytotoxicity. LAS was the most potent coccidiostat, significantly impairing mitochondrial function and membrane integrity at 1 µg/mL. Co-treatment with CBD significantly attenuated the toxic effects of the coccidiostats. Interaction analysis predominantly revealed antagonism effects, indicating a robust protective effect of CBD. However, an additive effect was observed at high concentrations of the LAS-CBD combination. CBD exerts a protective effect against coccidiostats nephrotoxicity in human renal cells.

CBD could potentially modulate cellular responses to ionophore-induced cytotoxicity, offering a basis for further in vivo studies on its detoxifying potential.

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The effect of 4-methylcatechol on platelet aggregation in patients with asthma

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Asthma bronchiale is a respiratory disease characterised by chronic inflammation that affects the lower respiratory tract. There are some reports connecting asthma and allergic immune-mediated conditions with alterations in blood coagulation and platelet aggregation [1]. Methylcatechol (4MC) is a metabolite of natural flavonoids produced by the human microbiota and has recently been shown to have potent antiplatelet effects [2,3].
This study aimed to investigate the antiplatelet effects of 4MC in a group of patients suffering from asthma.

A total of 40 patients were recruited at the Institute of Clinical Immunology and Allergy of the University Hospital in Hradec Králové. Heparinised whole blood was collected and examined for platelet aggregation using an impedance aggregometer. The obtained data were also compared to our recent group of generally healthy volunteers.

4MC decreased platelet aggregation triggered by both arachidonic acid and collagen in the asthma group. Its effects on the former inducer were comparable in both the asthma and control groups, in contrast to that of acetylsalicylic acid, which was less potent in asthma patients. In the case of collagen-induced aggregation, 4MC as well as acetylsalicylic acid were more potent in the asthma group compared to healthy individuals.

4MC can be a suitable alternative to common antiplatelet drugs also in asthma patients.

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Transformations of hydroxycinnamic acids with the use of fungi

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In this study, a diverse panel of endophytic and basidiomycetous fungi was systematically evaluated as whole-cell biocatalysts for the transformation of *p*-coumaric, caffeic, ferulic, and sinapic acids into corresponding vinyl phenols, aldehydes, and alcohols. These compounds exhibit antimicrobial, anti-inflammatory, and antioxidant activities, making them attractive for applications in the cosmetic, food, and pharmaceutical industries [1]. A number of endophytic and basidiomycete strains were screened, revealing distinct substrate-dependent distributions of phenolic acid decarboxylase (PAD), carboxylic acid reductase (CAR) and alcohol dehydrogenase (ADH) activities (Fig. 1).

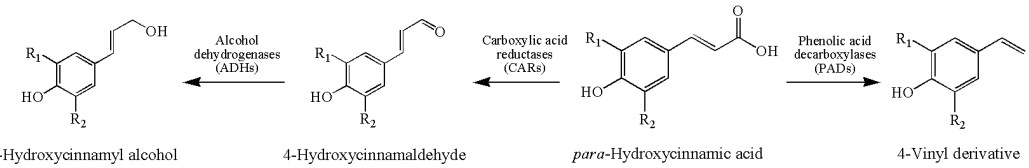

Fig. 1. Products of PAD-, CAR- and ADH-mediated transformations of *p*HCA substrates: $R_1 = R_2 = H$ (*p*-coumaric, **1**), $R_1 = OH$ and $R_2 = H$ (caffeic, **2**), $R_1 = OCH_3$ and $R_2 = H$ (ferulic, **3**), $R_1 = R_2 = OCH_3$ (sinapic, **4**).

Fourteen strains exhibiting consistent biotransformation profiles were advanced to preparative-scale evaluation and monitored by UHPLC-based time-course analysis. *p*-Coumaric acid and ferulic acid emerged as reliable substrates, with *Umbelopsis* sp. JAR-T achieving near-quantitative conversion to 4-vinylphenol and 4-vinylguaiacol with minimal by-product formation, rivaling yields reported for engineered bacterial systems [2]. Notably, *Trichoderma harzianum* BUK-T demonstrated unique ADH-mediated reduction of sinapic acid to sinapyl alcohol, addressing a major gap in fungal biotransformation literature. This integrated screening-to-preparative workflow provides a scalable framework for sustainable lignin-derived phenolic compound production and expands the biocatalytic toolbox for renewable aromatic valorization.

ACKNOWLEDGEMENTS

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Fully enzymatic cascades for the conversion of (*E*,*E*)-farnesol into enantiomerically enriched commercial fragrances

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In recent years, most companies working in the Flavour & Fragrance sector are collectively taking action to improve sustainability in their production processes and biocatalysis can offer beneficial synthetic solutions to satisfy this quest [1]. Among the key ingredients for modern perfumery, Biomuguet and Biocyclamol (Takasago) are 100 % readily biodegradable lily of the valley odourants made from 100 % renewable raw materials, however their chemical synthesis relies on transition metal catalyst that could raise environmental concerns [2].

In this work, we propose a novel biocatalytic approach for the synthesis of Biomuguet by ER-mediated reduction of (*E*,*E*)-farnesal, prepared in turn by enzyme mediated oxidation of commercial (*E*,*E*)-farnesol. The two reactions were combined in a telescopic cascade where the first step is performed in homogeneous phase, and the second in a biphasic medium after recovering the product of the first reaction with heptane. Alternatively, if the two reactions were combined in a two-step one-pot cascade in homogeneous solution, Biocyclamol was produced instead (Fig. 1). The short-chain dehydrogenase AaSDR1 from the mosquito *Aedes aegypti* was selected as the catalyst for the first step [3]. After identifying the best conditions and implementing a cofactor regeneration system with an alcohol dehydrogenase (SyADH) and acetone as sacrificial substrate, we achieved a > 90 % conversion of 15 mM farnesol in 24 h. For the subsequent step of stereoselective hydrogenation of farnesal, we performed a screening of the ERs available in our group in biphasic system and with a modified formate dehydrogenase (FDH) and ammonium formate as the cofactor recycling system, identifying NemA from *E. coli* as the enzyme of choice with a 51 % conversion and 76 % ee (*S*). The telescopic cascade was therefore implemented starting from15 mM (*E*,*E*)-farnesol using heptane in the second step, resulting in the formation of 90 % Biomuguet with an ee of 93 % (*S*). Adopting instead the one-pot homogeneous system, a mixture of 54 % Biocyclamol and 28 % Biomuguet was obtained, suggesting that further improvements are required.

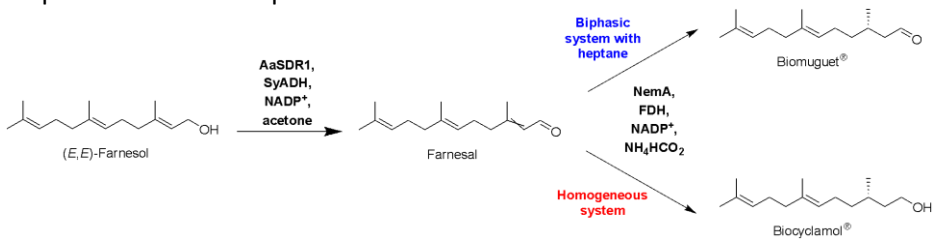

Fig. 1. General scheme of the two enzymatic cascades to obtain Biomuguet and Biocyclamol.

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Growth dynamics of *Salvia atropatana* hairy root culture as a factor determining the secondary metabolite production

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In the context of the increasing demand for standardized pharmaceutical raw materials, plant in vitro systems, particularly hairy root cultures, represent a stable, controllable, and environment-independent platform for the production of bioactive secondary metabolites. Owing to their genetic and biochemical stability, rapid growth, and high biosynthetic capacity, hairy roots constitute an excellent model for studying the interplay between primary and secondary metabolism. *Salvia atropatana*, a medicinal species native to the Middle East, is traditionally used in the treatment of inflammation, infections, wounds, and diabetes [1].

The aim of this study was to determine the growth kinetics and metabolite production profile of *S. atropatana* hairy root cultures to define the optimal harvest time corresponding to maximal accumulation of bioactive compounds.

Hairy roots of *S. atropatana* were cultivated in previously optimized half-strength SH medium [2], supplemented with 3% sucrose, without plant growth regulators, under rotary shaking conditions. Biomass growth was monitored over a 60-day cultivation period, with sampling at 5-day intervals. Growth parameters, including specific growth rate (μ) and doubling time (dt), were determined, and phytochemical profiling was performed using UPLC-DAD/ESI-MS.

The growth curve exhibited a distinct lag phase up to day 10, followed by an exponential phase (day 10–30) characterized by $\mu = 0.1\ day^{-1}$ and dt ≈ 6 days. This phase was followed by growth deceleration and the onset of the stationary phase at day 45 (dry weight), with biomass reaching 12 g/L DW (growth index approximately 45). Metabolite profiling revealed eleven phenolic compounds and twelve diterpenes. The majority of identified metabolites, including rosmarinic acid (RA) as the dominant compound, reached peak accumulation during the stationary phase, although selected metabolites exhibited more complex, non-growth-associated biosynthetic patterns.

These results indicate that growth curve determination constitutes a critical tool for defining optimal harvest strategies in *S. atropatana* hairy root cultures. Importantly, harvest timing may be selectively adjusted to maximize the accumulation of specific metabolites, reflecting their distinct biosynthetic profiles. Furthermore, optimization of harvest timing provides a foundation for future studies aimed at enhancing metabolite biosynthesis through elicitation strategies and for the development of scalable production systems for pharmaceutical applications.

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From raw material to functional snack: fermented legumes with increased bioavailability of polyphenols

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The increase in interest in plant-based diets is conducive to the development of functional foods with high biological value. Legumes are a valuable source of protein, fibre, and phenolic compounds with antioxidant and potentially prebiotic properties [1,2]. The fermentation process can further increase the bioavailability of bioactive ingredients and modulate their impact on the gut microbiota. The study aimed to develop vegetable snacks classified as convenience foods based on fermented broad bean seeds (*Vicia faba* L.) and to evaluate changes in the content of bioactive compounds during simulated in vitro digestion, taking into account the effect on the enzymatic activity of the intestinal microflora.

The research material consisted of snacks obtained from fermented legume seeds with the addition of marjoram (2%), carrots (30%), and red beet (15%). Fermentation was carried out with the participation of lactic acid bacteria, and then the products were freeze-dried in order to preserve and preserve biological activity. Simulated in vitro digestion was performed, including the gastric, small intestine, and large intestine stages. After each step, the total content of phenolic compounds (mg GAE/g s.s.), antioxidant activity (mg TE/g s.s.), the content of reducing substances, and total protein were determined. In addition, the activity of bacterial β-glucuronidase and β-glucosidase was determined, and microbiological analyses were performed for bacteria of the genera *Lactobacillus*, *Enterococcus*, *Enterobacteriaceae,* and *Bifidobacterium*.

The results showed that fermentation significantly increased the bioavailability of polyphenols and the antioxidant activity of the tested products. The highest values were recorded after the stage simulating the conditions of the large intestine (up to 5.6 mg GAE/g s.s. and 28.8 mg TE/g s.s.), which indicates beneficial transformations of phenolic compounds involving the intestinal microbiota. Modulation of bacterial enzyme activity was also observed – reduction of β-glucuronidase activity while maintaining a favorable intestinal microflora. The survival rate of fermentation bacteria after freeze-drying exceeded 80% of the initial population.

The results indicate that fermented legume snacks may be an innovative functional product combining high nutritional value with the potential to modulate the activity of the gut microbiota. The use of fermentation as an element of the technological process increases the bioavailability of polyphenols and may contribute to reducing risk factors for colorectal diseases.

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Synthesis of 7-azolocoumarins as fungistatic agents against fluconazole-resistant *Candida* strains

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The increasing prevalence of antifungal resistance on *Candida* species pose a crucial public health challenge, particularly in immunocompromised patients [1]. Addressing this challenge, the discovery of novel molecular scaffolds and structural modifications remains an essential approach in antifungal drug discovery. Coumarin scaffolds are well known for their diverse biological activities, including antiviral and antimicrobial effects [2]. Furthermore, azole-containing compounds are known as important antifungal agents due to their capability against fungal sterol biosynthesis [3]. Therefore, the combination between coumarins and azole moieties may represent a potential approach for the development of antifungal candidate compounds.

In this research, the 7-azolocoumarins were synthesized through a series synthetic route involving the functionalization of the coumarin followed by azole substitution at the C-7 position. The structure of the synthesized compounds was determined using NMR. The antifungal activity was evaluated *in vitro* against fluconazole-resistant and non-resistant *Candida* strains. The IC_{50} evaluation of tested azolo-coumarins compounds represented inhibitory effects varying among compounds and strains. The IC_{50} value ranged approximately from ~70 to >300, indicating moderate antifungal activity and implying that structural differences among the azolo-coumarins may influence their biological activity. Further studies will focus on structure-activity relationship and optimization of the most active compounds to improve potency and pharmacological properties.

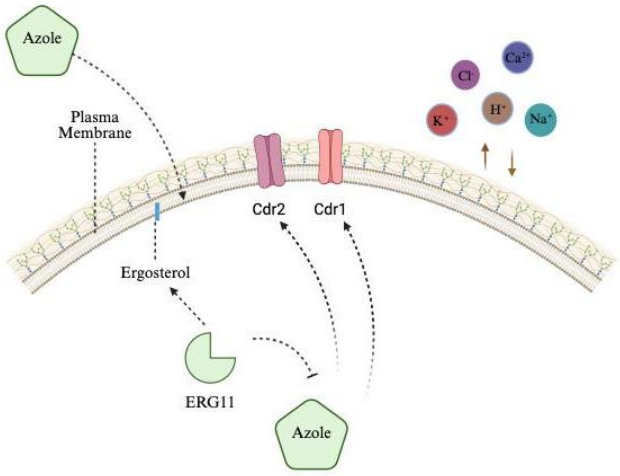

Figure 1. Mechanism of Antifungal action for the Azole drugs

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Gene-mining the frozen frontier: keratinolytic antarctic soil bacteria for novel bioactive compounds

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Extremophilic microorganisms are a rich source of industrially important biomolecules, including enzymes, anti-freeze proteins, compatible solutes, unsaturated fatty acids, pigments, etc. [1]. With more than 80% of Earth's biosphere permanently experiencing temperature below 5°C, cold-adapted microorganisms, including both psychrophilic and psychrotolerant microbes, are potentially one the most numerous and diverse group of extremophiles on the planet [2]. Until recently, research on their biodiversity, novel compounds discovery and industrial potential of cold-adapted microorganisms was relatively limited and typically focused on single metabolites or proteins [3]. The main bottlenecks being the lack of sufficient laboratory cultivation methods, costs of genome sequencing and scarcity of databases for adequate genome annotation. However, due to the development of novel analytical techniques applied in -omics sciences and their integration with AI-driven discovery tools, cold-adapted bacteria are experiencing renewed and intensified interest, allowing to gather and process previously unattainable data [3,4]. The six Antarctic bacterial isolates, *Paeniglutamicibacter sp.* B7_Z, *Filibacter sp.* B8_B, *Arthrobacter sp.* B11, *Leifsonia sp.* B16_Z, *Sporosarcina sp.* B18_K, *Kocuria sp.* B30_Z were selected from IBMP's robust collection based on their ability to degrade keratin-rich waste poultry feathers and subjected to phenotypic characterization using Gen III MicroPlates™ (BIOLOG, USA) to obtain 'metabolic fingerprints' for each isolate. Subsequently, whole genomes of selected bacteria were obtained via MinION™ nanopore sequencing (Oxford Nanopore Sequencing, UK), assembled and analyzed using several gene-mining tools and databases, including Conserved Domain Database, MEROPS database, antiSMASH 8.0 and BAGEL4, for discovery and characterization of novel or previously unexplored metabolic pathways, biosynthetic gene clusters (BGCs), bioactive compounds and enzymes. Phenotypic characterization of selected bacteria revealed distinct patterns of carbon sources utilization and capability to withstand increased salinity (up to 8% of NaCl) and other growth inhibitors. Similarly, whole-genome bioinformatic bioprospecting of keratinolytic Antarctic bacteria uncovered significant biosynthetic diversity, including gene clusters putatively involved in pigment, peptide and terpenes synthesis, with numerous genes currently lacking functional characterization.

ACKNOWLEDGEMENTS

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The impact of L-tryptophan on biochemical composition and structural properties of flax callus

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The study investigates the impact of L-tryptophan on callus shaken cultures of flax. Two different concentrations of L-tryptophan were tested: 0.1 mM and 1 mM. They resulted in lower callus biomass, but the levels of bioactive compounds such as carotenoids and polyphenols were increased. The influence of L-tryptophan on structural properties of plant cell walls in tested callus was determined using the FTIR method. Thus, the decrease in the cellulose and lignin amounts was observed after treatment with L-tryptophan. The supplementation of flax callus cultures with this amino acid resulted in higher levels of pectin. The possible relationship between tryptophan and auxins (which are derived from tryptophan) and the role of these metabolites in shaping the structure of the plant cell wall were discussed.

Leveraging food side streams for functional sweets: a Polish–Swiss research–industry initiative on microbiome-targeted snacks

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Despite growing interest in valorising food industry side streams, converting nutrient-rich by-products into functional, consumer-accepted foods remains challenging. Plant residues such as spent grains and fruit pomace are rich in fibers, polyphenols, and bioactive compounds with microbiome-modulating potential [1,2], yet processing is crucial since many current products fall short in taste, texture, and convenience [3]. Additionally, the stability of wet by-products remains a key limitation, often reducing their application potential in final products [4]. Scaling these innovations from lab to industrial levels is particularly difficult, highlighting the need for joint initiatives between academic and industrial partners to bridge scientific knowledge with practical, scalable solutions.

The **SWEETReclaim** initiative aims to address these challenges by developing scalable, plant-based food formulations using promising pretreatments i.a., extrusion and spray drying, to preserve bioactivity and improve sensory quality. A comprehensive approach will guide the workflow from by-product selection to industrial-scale prototype production. A structured process of testing and refinement will ensure technical feasibility, functional efficacy, and consumer acceptability, while academic–industry collaboration will support scientific rigour, scalability, and market alignment. This integrated strategy will efficiently translate laboratory research into real-world food applications.

The project is expected to deliver plant-based snack prototypes that combine gut microbiota benefits, sensory appeal, and industrial scalability. **SWEETReclaim** is going to provide a science-based roadmap for next-generation functional snacks, combining sustainability, validated health benefits, as well as industrial scalability and, ultimately, bringing microbiome-focused, market-ready foods to the health and wellness sector.

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Sustainable chemoenzymatic cascade for the synthesis of (*R*)-citronellyl nitrile from natural citral

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In recent years, major flavour and fragrance (F&F) companies have increasingly focused on the sustainability of their products and processes, largely driven by rising consumer awareness. Biocatalysis, especially multistep cascade reactions, is a promising approach since enzymes are renewable and operate under mild, water-based conditions.

(*R*)-Citronellyl nitrile (Levocitrile) is an important chiral fragrance with a stronger and cleaner lemon scent than its racemic form. However, its industrial production relies on a five-step chemical synthesis from myrcene using transition-metal catalysts, raising environmental issues.[1]

In this study, a more sustainable chemo-enzymatic cascade was developed to produce Levocitrile starting from natural citral, a mixture of geranial and neral derived from essential oils (Fig. 1). The key step is the stereoselective reduction of citral to (*R*)-citronellal,[2] catalyzed by an ene-reductase along with a formate dehydrogenase for cofactor recycling. A rational engineered KmOYE showed the best performance with higher enantioselectivity and efficiency.

Next, (*R*)-citronellal aldoximes were formed in situ via direct addition of hydroxylamine to the reaction mixture. Finally, these intermediates were dehydrated into the desired nitrile using an aldoxime dehydratase. The most effective enzyme for this step was OxDBr1, and the reaction was carried out in a biphasic system to avoid inhibition from residual hydroxylamine.[3]

Overall, the complete cascade enabled the production of Levocitrile from citral with good yield and high enantiomeric purity, offering a more environmentally friendly alternative to traditional methods.

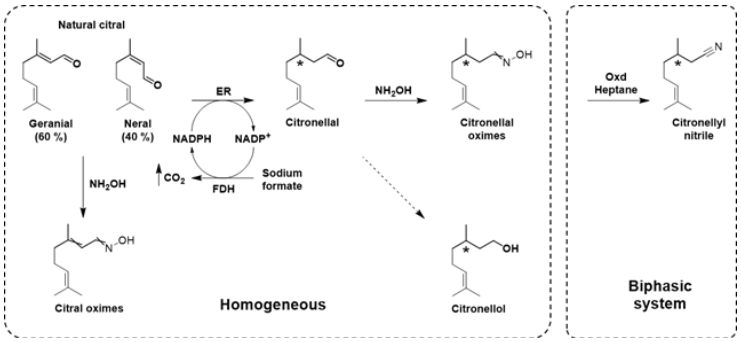

Fig. 1. Schematic representation of the chemo-enzymatic cascade.

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Tryptophan-rich *Moringa oleifera* leaves expand plant protein potential: nutritional characteristics and spectroscopic fingerprinting

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Moringa oleifera leaves are recognized as a nutrient-dense plant material with significant potential for functional food applications. This study comprehensively characterised the bioactive profile of *M. oleifera* dried leaves through integrated nutritional and spectroscopic analyses. Amino acid profiling, antioxidant activity assessment using FRAP, DPPH, ABTS, and ORAC assays, chromatographic analysis of organic acids and sugars, and vibrational spectroscopy (FT-IR/ATR and Raman) were employed. The crude protein content was 16.13 ± 0.43%. The amino acid analysis revealed that *M. oleifer*a protein contains all essential amino acids, with an exceptionally high tryptophan content (Amino Acid Score, AAS = 200%), representing a distinctive nutritional advantage among plant protein sources. Sulfur-containing amino acids (AAS = 49.57%) and lysine (AAS = 49.79%) were identified as the primary limiting amino acids. Antioxidant evaluation demonstrated solvent-dependent behaviour: the 80% ethanolic extract showed significantly higher FRAP activity (27.05 ± 1.05 mg TE/g DM), while ORAC values reached 96.68–107.24 mg TE/g DM across extracts. Total phenolic content was approximately 19.5 mg GAE/g DM in both water and ethanolic extracts. Spectroscopic fingerprinting provided direct molecular evidence for proteins, lipids, carbohydrates, and flavonoid glycosides, with the C–O–C stretching band at 1138 cm^{-1} confirming flavonoid glycoside content [1]. These findings support the use of *Moringa* leaves as a multifunctional ingredient for formulating balanced functional foods, particularly when combined with complementary protein sources to address limiting amino acid deficiencies [2].

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Watermelon by-products as functional ingredients: nutritional, antioxidant, and functional evaluation of seed protein and flour

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The growing interest in plant-based diets drives the search for alternative protein sources to complement animal-derived proteins. Watermelon (*Citrullus lanatus* L.) seeds, a by-product of the food industry, are rich in protein and represent a promising raw material for functional food ingredients. This study aimed to evaluate the nutritive, antioxidant, and functional properties of watermelon seed protein concentrate and watermelon flour to explore their potential valorisation. Protein concentrate was obtained using an alkaline extraction method, and watermelon pulp was processed into flour by drying. Analyses included determination of protein and fat content, polyphenol levels, antioxidant activity (ABTS+ assay), solubility, water and oil absorption, foaming capacity, and emulsification.

Results demonstrated that the seed protein concentrate and flour exhibited high protein content and significant antioxidant activity, as well as favorable functional properties, including good solubility and water/oil absorption, while foaming and emulsifying capacities were limited. These findings indicate that watermelon by-products can be valorized as functional ingredients, supporting both waste reduction and the development of nutritionally enhanced food products.

This study provides a comprehensive evaluation of watermelon by-products, integrating nutritional, antioxidant, and functional properties of both seed protein concentrate and flour in a single analysis. This research highlights the combined potential of multiple by-products as functional food ingredients and emphasizes valorization of food industry waste to support sustainable food development.

Table 1. Key nutrition and functional properties of watermelon seed protein concentrate.

	Dry matter	Total protein	Fat	Carbohydrates
Flour	95.75 ± 0.02 a	32.08 ± 0.01 b	6.83 ± 0.07 a	53.31± 0.06 a
Protein concentrate	95.16 ± 0.05 a	82.52 ± 0.29 a	1.51 ± 0.03 b	8.63 ± 0.41 b

Values are means ± standard deviation. n = 3; a–b the same letters in the columns mean homogeneous groups (Duncan's test $p \leq 0.05$).

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Lacto-fermented oats as future dairy-free yoghurt

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Oats (*Avena sativa*) grown in the Nordic countries are being increasingly used to produce dairy replacements. This study aimed to cultivate the exopolysaccharide (EPS)-producing probiotic strain *Lactobacillus delbrueckii* subsp. *bulgaricus* on enzyme hydrolysed oats to achieve a yoghurt-like product and assess its the macronutrient profile. Bacterial growth was investigated on two different concentrations of oat. The highest viable count of approximately 1×10^8 cells/mL was recorded at 37°C. EPS production was confirmed by the presence of peaks at absorption bands at 3400 cm^{-1}, 2925 cm^{-1}, and 1640 cm^{-1} that correspond to O–H, C–H, and C=O functional groups, found typically in bacterial EPS[2]. The total lipid content of the fermented oats increased (approximately 20.00 ± 0.20%), including an increase in mono- and polyunsaturated fatty acids such as oleic and linoleic acids [1]. The crude protein content also increased to approximately 50.00 ± 1.00% with a modest improvement in the essential amino acid contents, especially those that are a limiting amino acid in grains. Thus, these post-fermentation nutritional improvements, together with potential probiotic properties and the prebiotic EPS production denote that oats could in future be a nutritious dairy-free yoghurt alternative.

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DES-based protein recovery from sunflower oilseed cake

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As the global population continues to grow, the demand for food, including protein-rich products, is also increasing. Consequently, there is growing interest in developing protein recovery methods that are both efficient and environmentally friendly [1]. Oilseed cakes, which are by-products generated during the mechanical pressing of oil from oilseed crops, represent a valuable yet underutilized source of proteins, containing approximately 30–40% protein, depending on the plant source [2].

In this context, deep eutectic solvents (DESs) have recently attracted considerable attention as alternative green solvents. DESs are mixtures composed of two or more components, in which one acts as a hydrogen bond donor (HBD) and the other as a hydrogen bond acceptor (HBA). In addition to their low toxicity and biodegradability, they exhibit low volatility and high thermal stability. Moreover, they can be prepared using simple and cost-effective procedures, and their physicochemical properties can be tailored for specific applications through the appropriate selection of components and compositions [3].

In this study, a DES composed of choline chloride and glycerol was used as a sustainable and environmentally friendly solvent system for the extraction of proteins from sunflower oilseed cake. Following extraction, protein-rich fractions were obtained by antisolvent precipitation. The resulting precipitates were characterized using ATR-IR, ^{13}C CP-MAS NMR, CHN elemental analysis, and SDS–PAGE. Furthermore, the efficiency of the DES-based extraction process was compared with that of a conventional protein extraction procedure (ASAP).

Overall, the results of this study demonstrate that deep eutectic solvents can serve as an effective and promising alternative to conventional extraction methods, highlighting their potential for sustainable biorefinery applications.

ACKNOWLEDGEMENTS

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Sensory evaluation of vanillin obtained by fungi in the Solid-State Fermentation from agri-food industry by-products

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Vanillin is the compound widely used in the food industry as a flavoring agent. Currently, chemically synthesized vanillin provides the majority of the world's supply. Due to the increase in consumer awareness, there is a change in preferences towards natural food additives.

The main goal of this research was to obtain vanillin through Solid-State Fermentation on agri-food by-products such as brewer's spent grain, wheat bran, and linseed oil cake (Fig. 1). A specially designed SSF culture single-use bag bioreactor made of a poliamide-6 foil sleeve was used to conduct the process on a bench-scale (600 g of dry medium). After extraction and purification, obtained vanillin samples were subjected to sensory analysis, to determine whether the origin of microbiologically obtained vanillin affects its aromatic properties. The panellists assessed that the extracts obtained from the cultures of *P. chrysosporium* CBS246.84 and *F. culmorum* MUT5855 proved to be attractive flavors as they showed more attractive sensory properties than synthetic vanillin and were comparable to commercially available vanilla bean extract. This is the first study to include sensory analysis of vanillin obtained biotechnologically by the SSF method.

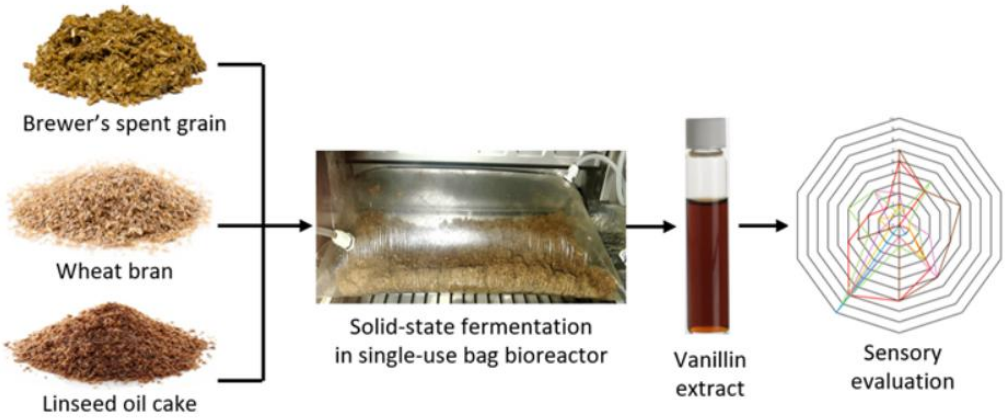

Fig. 1. Methodology used in this research.

ACKNOWLEDGEMENTS

This research was funded by the project "Development of biotechnological production of vanillin with the use of agroindustrial by-products" financed by the LIDER XII programme financed by the National Centre for Research and Development (Poland) under the contract No. LIDER/44/0228/L-12/20/NCBR/2021.

A plant-based agro-industrial by-product-derived protein hydrolysate reduces blood pressure and modulates gut microbiota in diet-induced metabolic syndrome

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Hypertension is a leading cause of cardiovascular disease and premature death worldwide. The gut microbiota (GM) has emerged as a potential target for its prevention due to its involvement in blood pressure (BP) regulation. In a previous study, we demonstrated that acute administration of a protein hydrolysate derived from a plant agro-food by-product (PH) modulated fecal microbiota and exerted antihypertensive effects in spontaneously hypertensive rats, effects that were abolished by antibiotic treatment. Therefore, this study aimed to evaluate the antihypertensive effect of prolonged PH administration in a diet-induced model of metabolic syndrome (MetS) and to explore the contribution of gut microbiota to these effects. Hence, sixteen 12-week-old female Wistar rats were fed a cafeteria (CAF) diet for 12 weeks. During the final 4 weeks, animals received daily an oral dose of vehicle (VH, water) or PH (55 mg/kg/day). Food intake, body weight, and systolic blood pressure (SBP) were recorded weekly, and gut microbiota composition was analyzed at the end of the study. Obese rats showed increased SBP to hypertensive levels, whereas PH administration significantly reduced SBP, restoring normotensive values. No significant differences were observed in alpha diversity between groups. In contrast, compositional analysis of the microbiota revealed a separation between groups based on centered log-ratio (CLR) transformation and principal component analysis (PCA), indicating shifts in microbial community structure. Among the taxa contributing to this separation, the genus *Prevotella* showed a markedly higher relative abundance in CAF animals compared to PH-treated rats, with a large effect size. Although this difference did not remain statistically significant after multiple testing corrections when considering all taxa, it was consistently detected across analyses and was reduced in PH-treated animals. Importantly, *Prevotella* abundance positively correlated with SBP, such that lower levels were associated with lower blood pressure.

These findings demonstrate that prolonged PH administration exerts antihypertensive effects in a diet-induced model of MetS and is associated with compositional shifts in the gut microbiota. The consistent reduction of *Prevotella* and its association with SBP suggest a potential role for this genus in BP regulation, although further studies are required to confirm its causal involvement.

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Cascade biorefinery of apple pomace via submerged lab fermentation: from enzymatic recovery of bioactives to binder-free biopackaging (a critical review)

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Apple pomace is a rich but underutilized resource due to the low bioavailability of its cell-wall-bound phenolics [1], [2]. The aim of this study is to critically evaluate Submerged Fermentation (SmF) using Lactic Acid Bacteria (LAB) as a highly efficient, zero-waste cascade biorefinery strategy. The methodology involves a systematic review of recent literature (mainly 2023-2026) focusing on the sequential enzymatic recovery of bioactives (liquid phase) and the direct conversion of residual biomass into biodegradable packaging (solid phase).

Key results indicate that SmF offers profound engineering advantages over alkaline extractions through natural pH dynamics. LAB-generated organic acids lower broth pH (3.0-3.8), preserving delicate protein-polyphenol complexes [3]. *Lactiplantibacillus plantarum* emerges as the most robust candidate. Its enzymatic cascade utilizes feruloyl esterases and β-glucosidases to liberate deeply bound aglycones [2], [4], generating massive surges in Total Phenolic Content and radical scavenging capacities. Furthermore, reutilizing the wet fermented solid phase represents a major material innovation. Hydrolytic enzymes biochemically deconstruct the rigid lignocellulosic network [5], minimizing synthetic additives. Compression molding these biologically pre-treated matrices (80-160°C) yields robust, "binder-free" biofilms reaching tensile strengths of 16.49 MPa with complete soil degradation within 15-30 days [6].

Main conclusions reveal that while this cascade approach is highly effective, critical sustainability bottlenecks persist. Thermally drying high-moisture fermented biomass consumes 50-70% of the total energy demand, posing a major industrial challenge [7]. Additionally, SmF-induced acid stress can trigger the accumulation of toxic biogenic amines, requiring rigorous genomic screening of starter cultures [8]. Future implementation demands "precision fermentation" to selectively dictate metabolic pathways [2], alongside integrating nanotechnology to bridge barrier performance gaps in bio-based packaging.

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Structural modification of fragrance alcohols: effects on human cells viability

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Fragrances compounds are widely used in perfumes and cosmetic products, but some traditional compounds raise safety concerns due to potential toxicity and environmental impact. [1] The continual interest in developing aroma compounds for novel fragrance formulations has driven the search for molecules that maintain the desired scent while offering improved safety. In this study, a series of compounds structurally related to lily-of-the-valley alcohol were synthesized via the preparation of acid derivatives followed by esterification and reduction, and their cytotoxicity was evaluated using human A-375 melanoma cells. The compounds were tested at different concentrations, and cell viability was measured using the MTS assay, with results compared to selected commercially available lily-of-the-valley fragrance ingredients.

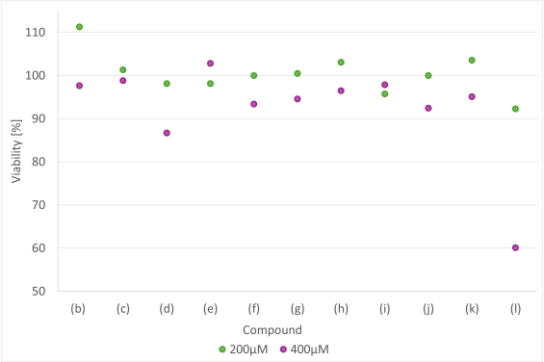

Fig. 1. Cell viability of A-375 cells after exposure to synthesized alcohols and commercially available compounds at different concentrations

The conducted studies showed that most of the newly synthesized alcohol derivatives did not significantly reduce the viability of A-375 cells at the selected concentrations. In contrast, one of the reference commercial odorants caused a greater decrease in cell viability under similar conditions. These results suggest that the synthesized compounds may represent a promising alternative to traditional fragrance ingredients, combining attractive olfactory properties with a potentially lower toxicological risk, as indicated by their effects on cell viability.

ACKNOWLEDGEMENTS

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Impact of chlorination and glycosylation on the lipophilicity and membrane interactions of 2′-hydroxychalcone and dihydrochalcone derivatives

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Flavonoids, including chalcones and their derivatives, constitute a broad group of naturally occurring polyphenolic compounds exhibiting diverse biological activities, largely determined by the structure of their core scaffold and the nature of their substituents [1]. Among them, chlorinated derivatives are of particular interest, as the presence of a halogen atom may significantly modulate the physicochemical and biological properties of a molecule, including its lipophilicity, membrane affinity, and biological activity [2,3]. In plants, flavonoids commonly occur in the form of glycosides, and glycosylation strongly influences their solubility, transport, and behaviour in biological systems.

The aim of this study was to evaluate how the structure of chlorinated dihydrochalcone glycosides—specifically derivatives of 2′-*O*-*β*-D-(4′′-*O*-methylglucopyranosyl)-dihydrochalcone substituted with chlorine at positions 2, 3, 4, and 5′—and their corresponding 2′-hydroxychalcone aglycones affects their lipophilicity and interactions with model lipid membranes. Partition coefficients (log P), determined experimentally using UV–Vis spectroscopy, revealed a pronounced shift in amphiphilicity, with values ranging from −0.56 for the more hydrophilic glycosylated derivatives to 1.88 for the lipophilic aglycones. To investigate membrane-modulating effects, fluorescence spectroscopy was employed using Laurdan (6-dodecanoyl-2-dimethylaminonaphthalene) and DPH (1,6-diphenyl-1,3,5-hexatriene) fluorescent probes incorporated into POPC (1-palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine) and cholesterol-containing liposomes. The results demonstrated clear structure-dependent differences in lipophilicity among the studied compounds. Fluorescence measurements indicated perturbations in both the polar headgroup region of the membrane (monitored by Laurdan) and the deep hydrophobic core (probed by DPH), suggesting alterations in membrane order and fluidity. These effects are consistent with the amphiphilic nature of the investigated molecules and their experimentally determined membrane affinities. Overall, the results provide insight into how glycosylation and chlorine substitution influence the physicochemical behaviour of dihydrochalcone derivatives and contribute to the rational design of compounds with desirable physicochemical and biological properties.

ACKNOWLEDGEMENTS

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Antiproliferative potential of vanillin-derived δ-iodo-γ-lactone

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Lactones are intramolecular esters of hydroxy acids naturally produced by plants, insects and microorganisms as secondary metabolites. They exhibit diverse biological activities, including antibacterial, antifungal, anti-inflammatory, analgesic and anticancer effects. Aromatic lactones can inhibit cancer cell proliferation by targeting enzymes essential for cancer growth. Both natural and synthetic lactones are thus widely applied in the pharmacy for developing antibiotics, antifungal and analgesic drugs and potential anticancer agents [1].

δ-Iodo-γ-lactone (**3**) was synthesized from the natural aromatic aldehyde vanillin (**1**) through a seven-step synthetic pathway. The key step of the synthesis was iodolactonization of carboxylic acid **2** followed by deprotection of the phenolic group (**Fig. 1**). Antiproliferative studies on selected human and canine cancer cell lines, as well as assessment of hemolytic activity against human erythrocytes, identified iodolactone **3** as the most active derivative among the synthesized iodolactones [2].

The aim of this study was further evaluation of iodolactone **3** against a broader panel of human cancer cell lines. The strongest activity, in some cases comparable to doxorubicin, was determined against gastric adenocarcinoma (EPG85-257RDB), melanoma (LM-MEL-75) and ovarian carcinoma (A2780) cell lines. Importantly, no significant cytotoxicity against normal human dermal fibroblasts (NHDF) was observed. These findings indicate that iodolactone **3** is a promising candidate for further development as a potential anticancer agent.

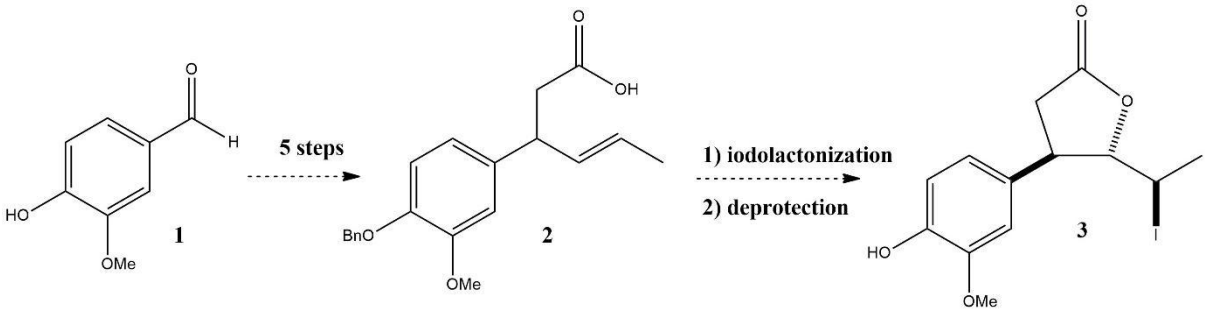

Fig. 1. Synthesis of δ-iodo−γ-lactone **3** from vanillin (**1**).

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Impact of stigmasterol-acylglicerol conjugates on nanocarrier properties - design, synthesis and physicochemical evaluation

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Nanocarriers are nanoscale systems designed to enhance the stability, bioavailability, and controlled release of bioactive compounds. Among them, liposomes play a particularly important role and are widely applied in pharmaceutical, cosmetic, and food technologies as carriers of drugs, antioxidants, and lipophilic molecules. Their functionality is largely determined by the physicochemical properties of the lipid bilayer, especially its fluidity, rigidity, and thermal stability. Membrane fluidity governs the mobility of acyl chains and bilayer permeability, and is closely associated with the main phase transition temperature (T_m), corresponding to the transition from the ordered gel phase to the liquid-crystalline phase.

Stigmasterol is a naturally occurring phytosterol structurally related to cholesterol, exhibiting antioxidant, anti-inflammatory, and hypocholesterolemic properties. Owing to its rigid steroidal ring system, it can modulate lipid ordering and influence the dynamic parameters of the membrane. The aim of this study was the design, synthesis, and evaluation of the effects of two novel stigmasterol–acylglycerol conjugates: 1,3-dicapryloyl-2-stigmasterylsuccinoylglycerol and 1,3-distearoyl-2-stigmasterylsuccinoylglycerol [Fig. 1] on the the physicochemical properties of the liposome using fluorimetry and Fourier transform infrared spectroscopy (FTIR) as well as differential scanning calorimetry (DSC). The results of the study show that the compounds affect the physicochemical properties of the liposome.

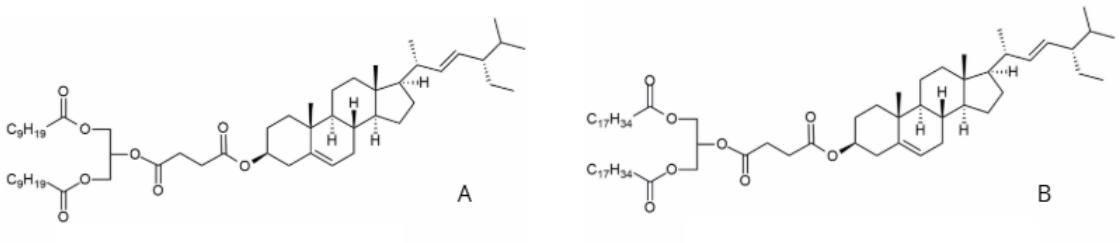

Fig. 1. Structures of the synthesized stigmasterol–acylglycerol conjugates: 1,3-dicapryloyl-2-stigmasterylsuccinoylglycerol (A), 1,3-distearoyl-2-stigmasterylsuccinoylglycerol (B).

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Click-engineered steroid bioconjugates as platforms for the development of bioactive molecules

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Natural steroidal frameworks provide a privileged structural motif for the development of bioactive compounds due to their structural rigidity, lipophilicity, and ability to interact with biological membranes. Inspired by aminosterols such as squalamine, significant efforts have been devoted to expand the structural diversity of steroid-based systems with potential antimicrobial and therapeutic relevance. Herein, we present the synthesis of structurally diverse steroid bioconjugates assembled via copper-catalysed 1,3-dipolar cycloaddition reactions. This click chemistry strategy enabled the efficient preparation of steroidal dimers, quasi-podands, and hybrid conjugates derived from bile acids and β-sterols. The incorporation of 1,2,3-triazole rings provided chemically stable linkers while enhancing hydrogen-bonding capacity and potential target interactions.

The obtained conjugates were characterised using NMR spectroscopy, FT-IR analysis, and mass spectrometry. Their structural diversity translates into varied physicochemical parameters and an enhanced propensity for supramolecular organisation. Preliminary computational studies suggest promising interactions with selected biological targets, indicating their potential as scaffolds for further development of bioactive agents. Overall, this work highlights steroid-based click conjugation as a powerful approach for generating multifunctional molecular systems of interest in medicinal and bioactive compound research.

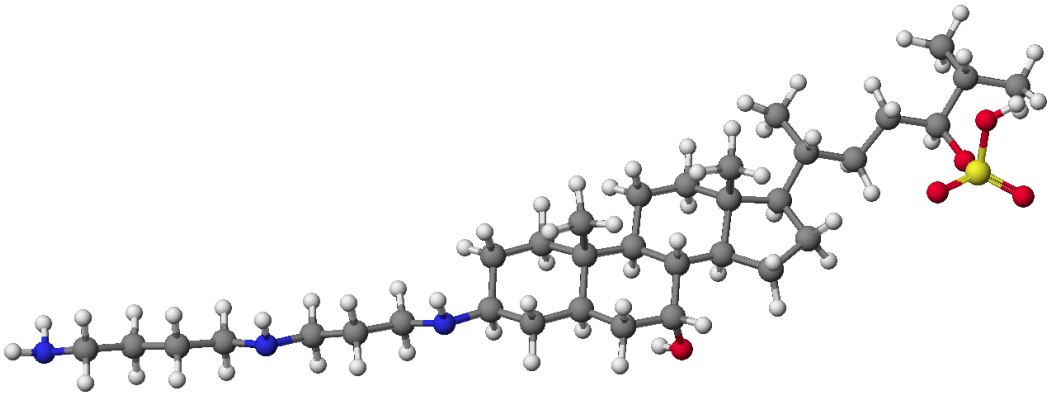

Fig. 1. Structure of squalamine.

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Design and evaluation of coiled-coil foldamers as nanocarrier systems

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Coiled-coils (CCs) constitute a highly promising peptide-based platform for nanocarrier applications, as they display one of the most clearly defined sequence–structure relationships among protein motifs and readily self-assemble into stable, tunable, and functional architectures. These features allow CC systems to efficiently transport, protect, and release therapeutic cargo in a controlled and biocompatible manner [1]. CC peptides containing cyclic β-amino acids, such as *trans*-(1*S*,2*S*)-2-aminocyclopentanecarboxylic acid (*trans*-ACPC), offer an even more versatile scaffold for the development of foldamers with programmable self-assembly behavior and adjustable photophysical properties [2-4].

In our previous work, we investigated foldameric bundles based on CC architectures using a combination of computational modeling and experimental characterization [5]. In the present study, we further evaluate their biological properties through cytotoxicity, hemolysis, and proteolytic stability assays, enabling assessment of both their biocompatibility and resistance to enzymatic degradation. Our results show that CC-based foldamers incorporating cyclic β-amino acids assemble into stable and well-defined supramolecular structures (Fig. 1) with favorable biological profiles, underscoring their potential as robust and adaptable nanocarrier systems for therapeutic applications.

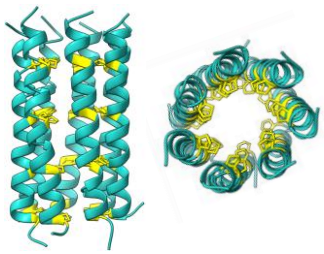

Fig. 1. Coiled-coil foldamer-based potential nanocarrier.

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Effect of symmetric acylglycerols containing stigmasterol residues at *sn*-1 and *sn*-3 positions on lipid model membranes

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Phytosterols are highly regarded for their broad health-promoting properties, including anti-inflammatory, antibacterial, antifungal, and anticancer activities, as well as their ability to lower cholesterol levels. Among these compounds, stigmasterol deserves special attention—it is one of the most common phytosterols in the human diet [1]. However, despite its significant biological potential, this compound is characterized by low bioavailability. In order to increase the efficiency of delivering stigmasterol to target tissues, its conjugation with acylglycerols was undertaken. Due to the natural presence of acylglycerols both in food products and the human body, they represent a promising delivery system for active substances [2-4].

The aim of the study was to develop four new phytosterol carriers consisting of acylglycerols containing stigmasterol residues at positions *sn*-1 and *sn*-3, with natural palmitic or oleic acid at the *sn*-2 position. Furthermore, the obtained molecules were incorporated into liposomes to create new nanoformulations in a mixture with a model phospholipid. Their impact on the physicochemical properties of the membranes – including polarization, fluidity, and main phase transitions – was determined using differential scanning calorimetry (DSC) and fluorometry.

The results of the study showed that the conjugates obtained significantly affect the fluidity, degree of order, and temperature of the main phase transition of the liposomal membrane. The nature and scale of these changes are closely dependent on the chemical structure of the compound. The data obtained are crucial for the practical use of these substances in the design of modern functional foods and as components of innovative drug delivery systems in the pharmaceutical industry.

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Metabolic profiling of *Pistacia lentiscus* L. var. *chia* via NMR and LC-MS: tracking authenticity from leaf to resin

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Natural products science focuses on the chemical constituents synthesized by organisms like plants and microbes. Within this field, pharmacognosy leverages this vast molecular diversity to identify bioactive compounds and structural scaffolds for drug development. Integrating 'omics' technologies into this workflow can significantly expedite the discovery of bioactive markers while strengthening quality control frameworks.

By combining omics-based approaches and bioinformatics, this research sought to characterize the secondary metabolites of *Pistacia lentiscus* L. var. Chia leaves and its signature secretion, Chios Mastic Gum (CMG). A primary objective was to develop a dependable framework for verifying CMG authenticity. Despite its long history of health benefits and its EMA-certified status as a traditional medicine for digestive and dermatological issues, there is currently a lack of in-depth metabolic profiling and standardized authenticity testing for this unique resin.

The leaf profiling was initially conducted using integrated NMR and LC-HRMS platforms, utilizing multivariate analysis and advanced statistical tools such as Statistical TOtal Correlation SpectroscopY (STOCSY) and Statistical HeterospectroscopY (SHY). Significant metabolic variations were observed, with the annotation of galloyl derivatives and flavonoid glucosides driving the classification based on tree age and geographic subregions of Chios. Furthermore, an NMR-based prediction model was developed for CMG authentication, incorporating diverse samples including Iranian mastic, pine, and frankincense resins. The synergistic levels of masticadienonic acid, isomasticadienonic acid, and oleanonic aldehyde emerged as definitive biomarkers for distinguishing genuine CMG from other resins. These findings underscore the transformative role that omics and bioinformatics are poised to play in future natural products research.

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Antarctic psychrophiles as bioactive factories: linking genomics with metabolite production

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The search for novel bioactive compounds increasingly turns toward extreme environments, where unique metabolic adaptations may drive chemical diversity. The aim of this study was a comprehensive phenotypic, genotypic, and metabolomic characterization of Antarctic soil bacteria to evaluate their potential for the production of bioactive compounds with antioxidant activity.

Taxonomic identification of bacterial isolates was performed using 16S rDNA sequencing. The antioxidant potential of the isolates was assessed under different temperature conditions (20°C, 5°C, and 5°C/−5°C) using DPPH and FRAP assays. Selected high-potential strains were further characterized, including phenotyping analysis, metabolite-oriented analyses: pigment extraction, and fatty acid profiling using GC–MS to evaluate membrane adaptations to oxidative and temperature stress. Genomic sequencing using Ion Torrent technology was initiated to complement metabolomic insights. The results demonstrated that several Antarctic strains, particularly representatives of the genus *Sporosarcina*, *Dietzia*, and *Cryobacterium* exhibit high antioxidant potential. Notably, bioactive metabolite production was strongly influenced by cultivation temperature, indicating adaptive metabolic responses to environmental stress. Furthermore, the selected isolates mostly produced pigments belonging to the carotenoid family. Fatty acid analysis indicated temperature-dependent changes in membrane composition, particularly in the proportion of unsaturated fatty acids.

In conclusion, Antarctic psychrophilic bacteria constitute a promising and largely untapped source of bioactive compounds. The integration of genomic and metabolomic approaches provides valuable insight into their adaptive mechanisms and biosynthetic potential, supporting their future application in biotechnology and natural product discovery.

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Metabolomic insights into the antimicrobial effects of *Metschnikowia* yeast on phytopathogens

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Metschnikowia are unconventional yeasts with high industrial potential. One of the most important features of these microorganisms is their strong antimicrobial properties, especially against phytopathogenic fungi, which cause significant damage in the agri-food industry. The use of *Metschnikowia* yeasts in biocontrol processes can bring substantial benefits in crop production. However, to develop an effective biopreparation that protects plants from phytopathogens, research focusing on the poorly understood mechanisms of the antimicrobial activity of these yeasts is required. The aim of this study was to investigate the antimicrobial activity of *Metschnikowia* yeasts against phytopathogenic fungi using metabolomic approaches. The first step of the research involved performing antimicrobial activity tests of five selected yeast isolates against eleven phytopathogenic molds. Based on these results, yeast–pathogen combinations (12 systems) were selected, from which co-cultures were prepared in liquid media, and the culture supernatants were used for metabolomic analysis using UHPLC-QToF-UHRMS. Additionally, metabolite analysis was performed using LARAPPI/CI MSI for co-cultures grown on agar media.

The results showed that the studied yeast strains exhibit strong antagonism against phytopathogens. LARAPPI/CI MSI analysis detected a range of antimicrobial compounds, such as lactic acid, cinnamic acid, indole, and its derivatives, while UHPLC-QToF-UHRMS analysis confirmed that different yeast-pathogen combinations induce distinct metabolic responses.

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An investigation into hop extracts: a study of volatile components and their relationship to the aroma profile

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Hop is a complex and multifunctional climbing plant, whose inflorences are mostly used for beer brewing. They are employed to impart bitterness, flavor, and aroma to beer, as well for their antiseptic and stabilizing properties. Since the brewing industry began to grow unabated, interest in this plant has grown, causing the number of genetic experiments conducted and thus the number of varieties on the market to increase exponentially. Hop can be used in different forms: whole cones, pellets, or extracts. The latter are gaining interest in recent years thanks to their useful properties with respect to other options, such as better control on bittering and flavor profile, longer shelf-life, lower liquid absorption and reduced vegetable matter. During this research, thirty-two extracts were distilled from hop pellets and analyzed through MS-GasChromatography in order to identify and quantify volatile components present in each hop variety. A total of one-hundred thirty-two different components were found, the majority constituted by terpene hydrocarbons, along with esters, alcohols, epoxides, and other oxygenated compounds. The most common ones were β-myrcene, caryophyllene and α-humulene. All the samples were successfully mapped and between fifteen and twenty components were found in each extract. Additionally, a kinetic study and a comparison between hydrodistillation and microwave extraction were performed. From the former, it was found that, by distillation, as early as the first hour, 90% of the terpene content is extracted mostly as monoterpenes, and that, after the first half hour, sesquiterpenes and oxygenated components are successively extracted. From the latter, however, it was investigated how different heat transfer mechanisms affect the quality and composition of the extract. The microwave system, relying on faster heating, favored the extraction of monoterpenes, especially in the first part of the process; it also recorded a higher yield in terms of volume of extract per amount of hops used and process time. Finally, Hierarchical Cluster Analysis has been performed, showing that the considered hops, analyzed by their volatile component profile, can be grouped into four main clusters.

Multicomponent NADES as a sustainable alternative to conventional solvents for rosemary antioxidant recovery

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The industrial extraction of bioactive compounds for dietary supplements is often limited by a trade-off between safety, efficiency and cost. While water is the most economical and tax-free solvent, its extraction yield for many bioactives remains low.[1] Conversely, ethanol provides high efficiency but is burdened by significant excise duties and regulatory taxes and the requirement for additional evaporation steps, all of which substantially increase the overall production cost.
This study presents the engineering of a novel food-grade multicomponent system – a natural deep eutectic solvent (NADES), designed according to the acceptable daily intake (ADI) of each constituent. This ensures the solvent's safety for direct integration into final formulations, eliminating the need for costly downstream processing.

The extraction efficiency was benchmarked against water as the baseline industrial solvent. Determination of Total Phenolic Content (TPC) via the Folin-Ciocalteu method revealed that the designed NADES significantly outperforms water solutions in terms of TPC yield. Furthermore, HPLC analysis confirmed a rich polyphenolic profile and suggested that the multicomponent matrix provides a stabilizing environment for the extracted molecules, protecting them from degradation.

By leveraging this NADES system, manufacturers can achieve enhanced extraction yields while maintaining the economic advantages of water-based processes by avoiding the costs associated with conventional solvents.

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Vascular effects of new bisphenol a alternatives

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The widespread use of bisphenol A (BPA) in epoxy resins and plastics has raised health concerns, leading to bans in children's products and a shift toward alternative "BPA-free" bisphenol analogues[1,2]. However, there is little information regarding their biological impacts. Early research indicates that at least some of them may have negative health impact on humans[3, 4, 5]. In this project, the vascular effects of ten important new bisphenol A alternatives were tested. Screening of vasodilatory activity was performed on isolated rat aortas, the most active bisphenol S-MAE (EC_{50} < 0.5 µM) was selected for mechanistic studies. First, experiments excluded the role of functional endothelium. Hence, the vasodilatory mechanisms were studied on the vascular smooth muscle. The direct role of K^+ channels was excluded as well as that of the cGMP/PKG pathway. In contrast, presence of bisphenol S-MAE significantly decreased the effect of a Ca^{2+} channel opener in a dose-dependent fashion.

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Towards natural photoprotection: the greensunscreen project

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Ultraviolet radiation (UVR) is a major environmental factor affecting skin health and overall human well-being. Its harmful effects include sunburn, pigmentation disorders, premature skin aging, immunosuppression, and skin cancer [1]. Prevention of UVR-induced damage relies primarily on the use of sunscreen products containing UV filters, which are classified as mineral (inorganic, reflecting UV radiation) or synthetic (organic, absorbing UVR) [2]. However, synthetic filters raise concerns regarding human safety and environmental impact. Evidence indicates that both organic and inorganic filters can exert toxic effects on marine ecosystems and contribute to coral reef bleaching [3]. Moreover, synthetic filters are not compatible with natural cosmetic formulations, highlighting the growing demand for safe, effective, and environmentally friendly photoprotective agents.

The Green Sunscreen Project—an international research network funded by the Polish National Agency for Academic Exchange (NAWA) under the Strategic Partnerships Programme (Project No. BPI/PST/2024/1/00119) —aims to address these challenges by identifying natural plant-derived ingredients with photoprotective potential. The project focuses on plant species from Croatia, Mongolia, and Greece, which are adapted to high solar exposure and have a history of traditional skin-related applications. Investigated species include *Eryngium amethystinum*, *Limonium vulgare*, *Crithmum maritimum*, and *Arundo donax*. Plant extracts were obtained using low-energy, environmentally friendly extraction methods and non-toxic solvents suitable for natural cosmetic formulations. In the preliminary stage, small-scale extracts were evaluated using standard *in vitro* assays, including Folin-Ciocalteu, DPPH and ABTS scavenging, *in vitro* SPF measurement, and tyrosinase inhibitory activity, to assess their photoprotective potential. Preliminary studies confirmed that selected plant species are promising ingredients for photoprotective formulations. Further studies will involve advanced experimental models, including 3D human epidermal models and zebrafish embryos, to elucidate the safety and efficacy of selected extracts.

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Carriers of the future or just a cosmetic trend? The effectiveness of lipid nanoparticles in delivering active ingredients to the skin

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Effective delivery of active ingredients into the skin remains a major challenge in cosmetic formulation, making lipid nanoparticles promising carriers for improving the stability and performance of cosmetic actives [1,2]. This study aimed to evaluate the effectiveness of lipid nanoparticles (LNs) as delivery systems for selected cosmetic ingredients and to compare the performance of nano-based and conventional formulations. Lipid nanoparticles loaded with selected iridoid glycosides, namely aucubin (I) and catalpol (II) [3-5], as well as sesquiterpene lactones, costunolide (III) and dehydrocostus lactone (IV); were prepared using different methods: high-pressure homogenization (HPH) for LN systems containing sesquiterpene lactones, and high-shear homogenization (HSH) combined with sonication based on a multiple-emulsion approach for LNs loaded with iridoid glycosides. The resulting systems were characterized in terms of mean particle size (Z-Ave), polydispersity index (PDI), and zeta potential (ZP), which are key parameters related to physicochemical stability of the LN dispersions. Particle sizes below 100 nm were obtained for LNs loaded with aucubin and catalpol, whereas carriers containing costunolide and dehydrocostus lactone reached approximately 350 nm. Subsequently, the nanoparticle dispersions were incorporated into selected cosmetic vehicles, including hydrogels for I and II, as well as oil-in-water (O/W) emulsions and oleogels for III and IV. The obtained formulations were then evaluated *in vivo* in volunteers using set of Courage+Khazaka instruments: Corneometer® CM 825 (skin hydration), Tewameter® TM 300 (transepidermal water loss, TEWL), Mexameter® MX 18 (skin pigmentation and erythema), Visioscan® VC 98 (skin topography), Visioline® VL 650 (skin macro relief). Their effectiveness was compared with that of corresponding formulations containing the same active ingredients in a conventional, non-nano form. The results demonstrated a consistent trend showing that nano-based formulations produced greater improvements in skin macro relief parameters than the corresponding conventional formulations. Moreover, in the case of iridoid glycosides, nanoformulations exert a more pronounced effect on key skin parameters, particularly epidermis hydration and TEWL. Overall, the study provides comparative insight into the potential of lipid nanoparticles as cosmetic delivery systems and highlights the importance of both formulation type and the physicochemical characteristics of the carrier in determining product performance.

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Vasodilatory substances from hawthorn: screening and determination of the mechanism of action

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INTRODUCTION: Standardized extract from hawthorn (*Crataegus* spp.) dilates blood vessels, improves their elasticity [1, 2], and reduces blood pressure *in vivo* in hypertension [3]. The active ingredient is unknown.
OBJECTIVE: To test the vasodilatory effects of selected substances contained in hawthorn.
MATERIAL: 21 substances with diverse structures (flavonoids, glycosides, pentacyclic triterpenes) and absorbable metabolites (small phenolic acids).
METHODOLOGY: *Ex vivo* testing on three types of isolated blood vessels from two animal species (pig, rat).
RESULTS: The two most effective substances, isorhamnetin and tamarixetin, induced vasodilatation in all selected models - porcine coronary arteries (EC_{50} = 21.7 and 30.9 µM); rat aorta (16.7 and 23.3 µM) and rat mesenteric artery (22.2 and 16.8 µM). Both isorhamnetin and tamarixetin blocked vasoconstriction induced by activation of L-type calcium channels, and this effect was dose-dependent. By contrast, their effects did not depend on the presence of functional endothelium, nor were they directly related to potassium channels or the cGMP/protein kinase G cascade.
CONCLUSION: The vasodilatory effects of isorhamnetin and tamarixetin from hawthorn are likely mediated by inhibition of calcium influx into vascular smooth muscle cells or related intracellular cascades.

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Flavokawain derivatives as bioactive chalcones: effects of B-ring substitution on membrane organization and protein binding

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Flavokawains A, B, and C are chalcone-type natural products characterized by a 1,3-diaryl-2-propen-1-one scaffold, predominantly found in the root extracts of *Piper methysticum* (*kava*), where they occur alongside kavalactones as major bioactive constituents with preparation-dependent relative concentrations [1]. The literature indicates that flavokawains exhibit significant pharmacological potential, and their activity profile is closely dependent on the substitution pattern of the aromatic rings. Structure-activity relationship analyses reveal that the α,β-unsaturated enone moiety is critical for biological activity, as cyclization to the corresponding flavanone markedly reduces cytotoxicity, and comparative studies indicate that flavokawain B generally exhibits higher anticancer potency than flavokawain A in many in vitro assays, with aromatic substituent patterns modulating lipophilicity, reactivity, and cellular efficacy [2].

The biological and physicochemical properties of five flavokawain derivatives, differing in the pattern of methoxy substitution on the B ring, were evaluated in this study. The hemolytic activity of the compounds was investigated using human erythrocytes. Interactions with model lipid membranes were analyzed using fluorescent probes (Laurdan, DPH, and TMA-DPH), allowing the localization of the molecules within the bilayer. The integrity and monodispersity of liposomes after incubation with the compounds were monitored by dynamic light scattering (DLS). Binding affinity to HSA was studied using the fluorescence quenching technique.

All investigated derivatives exhibited low hemolytic activity toward human erythrocytes, indicating a lack of toxicity toward blood cells within the studied concentration range. Fluorescence studies suggest that the derivatives preferentially localize within the hydrophobic core of the lipid bilayer, causing concentration-dependent membrane rigidification. DLS analyses confirmed that derivatives A1–A3 did not significantly affect vesicle size, whereas A4 and A5 induced an increase in both the size and polydispersity of the liposomes. All derivatives formed stable complexes with HSA, and the measured binding affinities indicate that the introduction of additional methoxy groups weakens albumin binding.

The study demonstrated that the substitution pattern on the B ring of flavokawains plays a key role in determining their biophysical properties. These findings provide important insights into the molecular mechanisms and structure–activity relationships, which are essential for the rational design of new therapeutics based on the chalcone scaffold with potentially improved optimized pharmacokinetic properties.

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Galectin-driven reorganization of ganglioside nanodomains in model membranes

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Galectins are soluble glycan-binding proteins that regulate a wide range of cellular processes, including cell adhesion, immune regulation, and transmembrane signaling, through their ability to organize glycoconjugates at the plasma membrane[1]. Galectin-1, which belongs to the prototype class of galectins, forms non-covalent homodimers that can cross-link glycan counterparts on the plasma membrane, leading to the formation of multibranched networks known as glycan-galectin lattices[2]. Such assemblies have been proposed to influence membrane organization and receptor clustering during cellular signaling. The glycosphingolipid GM1 ganglioside is known to oligomerize in membranes and form ganglioside-enriched nanodomains that may act as templates for lectin binding[3]. In this study, we investigate how the nanoscale organization of GM1 influences galectin-mediated membrane organization. Giant unilamellar vesicles (GUVs) containing GM1 were prepared with lipid compositions designed to generate nanodomains of different sizes, providing controlled membrane platforms. A series of galectin-1 constructs, including the native homodimer as well as engineered dimers and tetramers with different linkers, were introduced to probe how protein architecture affects membrane organization. Galectin-1 induced changes in nanodomain size and surface coverage were quantified using Förster resonance energy transfer (FRET) combined with mcFRET simulations. Also, the flexibility of proteins with different linkers were assessed by AFM and their binding to the GM1-containing lipid bilayers were characterized using the AFM tool. These experiments aim to provide insight into how galectin architecture and GM1 nanodomain organization together influence glycolipid clustering at membrane surfaces.

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Biological activity of hydroxychalcones in Merkel carcinoma cells

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Chalcones, known as precursors of flavonoids, represent an important class of natural bioactive compounds widely distributed in plants. Due to their relatively simple chemical structure and ease of modification, they have become an attractive scaffold in medicinal chemistry and pharmacological research. Numerous studies have demonstrated that chalcone derivatives exhibit a broad spectrum of biological activities, including antioxidant, anti-inflammatory, antimicrobial and anticancer effects [1,2]. Among them, hydroxychalcones have attracted particular attention because the presence and position of hydroxyl groups in the aromatic rings may significantly influence their biological properties, including redox activity and interactions with cellular targets. These structural features may affect their ability to modulate intracellular signaling pathways, oxidative balance and cell death mechanisms [3,4].

The aim of presented study was to evaluate the biological activity of selected hydroxychalcones differing only in the position of the hydroxyl group in the aromatic ring: 2'-hydroxychalcone, 4'-hydroxychalcone and 4'-hydroxychalcone. Particular attention was paid to their influence on oxidative stress and mitochondrial function during the early stages of apoptosis, with emphasis on their potential role in the modulation of reactive oxygen species production and mitochondrial membrane integrity.

The experiments were performed using Merkel cell carcinoma cells, a rare and aggressive type of human skin cancer. Cells were exposed to the tested compounds at several concentrations for 24 and 48 hours. The biological effects of hydroxychalcones were evaluated by assessing reactive oxygen species (ROS) generation and changes in mitochondrial membrane potential, which are key indicators of mitochondrial involvement in apoptosis initiation.

The obtained results demonstrate that hydroxychalcones are capable of inducing early apoptotic events, including phosphatidylserine externalization, accompanied by significant alterations in mitochondrial membrane potential and modulation of intracellular ROS levels in a dose-dependent manner. These findings suggest that the position of the hydroxyl group in chalcone derivatives may influence their biological activity and ability to modulate mitochondrial-dependent apoptotic pathways.

Overall, the results highlight the potential of hydroxychalcones as promising bioactive molecules that may contribute to the development of novel therapeutic strategies targeting cancer cells.

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Linking experimental and theoretical activity of phenolic compounds in DPPH, ABTS and FRAP assays

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Phenolic compounds constitute a large and structurally diverse group of natural antioxidants widely distributed in plants. Their antioxidant activity is strongly related to molecular structure, including the number and position of hydroxyl groups and other substituents. This study aimed to conduct a comprehensive analysis of the antioxidant properties of 70 phenolic compounds representing distinct structural classes: anthocyanidins, flavones, flavonols, flavanones, flavanonols, isoflavones, and phenolic acids.

The antioxidant capacity of the investigated compounds was evaluated using three complementary *in vitro* assays: DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging, ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)) radical cation decolorization, and FRAP (Ferric Reducing Antioxidant Power). The results were expressed as reaction rates in Total Antioxidant Units per micromole of compound, representing the parameter TAU.

The analysis revealed clear differences in the antioxidant activity profiles obtained across the three methods, reflecting their distinct mechanistic bases. A high level of mutual coherence was observed between the DPPH and ABTS assays, consistent with their shared neutralization mechanism. Among the analyzed compounds, flavonols and phenolic acids containing catechol or pyrogallol moieties exhibit the highest antioxidant activity, characterized by both high radical scavenging capacity and significant reducing ability. The theoretical calculations indicate that the mechanism of action of the investigated polyphenols involves both formal hydrogen atom transfer and single electron transfer, with the dominant pathway depending on the assay used.

The theoretically estimated rate constants correlated significantly with the experimentally determined TAU values, with the strongest agreement for DPPH (Spearman $\rho=0.677$, $p<0.001$) and FRAP ($\rho=0.522$, $p<0.001$), while ABTS showed a weaker, borderline correlation ($\rho=0.234$, $p=0.051$), likely due to the formation of a pre-reaction complex in the processes. For DPPH, a strong linear correlation was found exclusively for log-transformed rate constants (Pearson $r=0.545$, $p<0.001$), indicating a logarithmic relationship between theoretical and experimental activity

The obtained results confirm the strong structure-dependence of antioxidant activity in phenolic compounds. The forthcoming QSAR models will constitute a valuable tool for predicting and screening new bioactive phenolics with elevated antioxidant potential.

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Five novel isocoumarins protect red blood cells against copper toxicity

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Isocoumarins are originally natural compounds containing a lactone moiety. Based on substitutions, they might exhibit numerous pharmacological activities, such as antioxidant, anti-inflammatory, hepatoprotective, and neuroprotective properties. Additionally, they can block α-glucosidase [1], an enzyme transforming complex carbohydrates into absorbable monosaccharides. Its inhibition delays carbohydrate digestion, reduces glucose plasma levels, and hence represents a treatment modality in diabetes mellitus [2].

In this study, twenty-two newly synthesized isocoumarins were tested. First, their potential toxicity was examined on human erythrocytes using the lactate dehydrogenase activity assay. Second, their effect on the copper-triggered hemolysis of human erythrocytes was studied employing the same method. Subsequently, the effect of these compounds on the activity of yeast and mammalian α-glucosidase was tested employing a kinetic spectrophotometric assay.

At a concentration of 100 µM, none of the included compounds exhibited toxicity to red blood cells, but 5 of them reduced red blood cell lysis induced by copper. Four compounds significantly inhibited yeast α-glucosidase at this concentration, two of which showed potent activity with IC_{50}s of 23 and 29 µM. None of the investigated isocoumarins, however, blocked mammalian α-glucosidase significantly, nor at a concentration of 100 µM.

The newly prepared isocoumarins do not seem to be good candidates for novel antidiabetic drugs, but they might be interesting for further development as protective agents against copper toxicity (e.g., in Wilson´s disease). The connection between the effect and the chemical structure is not yet fully understood and should be investigated in more detail in the future.

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Modulation of multidrug resistance transporter activity, ABCB1, by combined use of antipsychotic drugs and simvastatin

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Our previous results demonstrated that antipsychotic drug, trifluperazine (TFP), possessed potent anticancer activity in colon cancer cells which was enhanced when TFP was combined with anti-hyperlipidemic drug, simvastatin (SIM). Both drugs exhibited strong synergy. There are multiple molecular mechanisms that might be responsible for the observed effect. One could be connected to cellular uptake/efflux systems such as ABCB1 protein (P-glycoprotein). This multispecific transmembrane transporter is frequently overexpressed in multidrug-resistant (MDR) cancer cells. TFP is an inhibitor of ABCB1 [1]. Moreover, ABCB1 is sensitive to the lipid composition of the surrounding membrane [2]. Therefore the combination of an ABCB1 inhibitor (e.g., TFP) and cholesterol-lowering drug could potentially increase their MDR-modulating potency. Changes in lipid membrane composition and disturbed cholesterol homeostasis are observed in cancer cells, especially MDR ones [3]. Antipsychotics also up-regulate expression of genes engaged in cholesterol biosynthesis [4]. The aim of the present work was to investigate whether phenothiazine-type antipsychotic drugs in combination with SIM could affect ABCB1 transporter activity to a greater extent than both types of drugs applied separately. Madin-Darby Canine Kidney (MDCK) cells and MDCK-MDR1 cells transfected with the human *ABCB1* gene were used as a model system. Flow cytometric functional test, based on the measurement of the accumulation of ABCB1 substrate (rhodamine 123) was employed. Inhibition was expressed as the calculated FIR factor. The results showed that combination of TFP, and other phenothiazine-type antipsychotics with SIM increased the degree of ABCB1 modulation but only when the cells were cultivated in the presence of the drugs for a long enough period. It was concluded that the observed effect was not due to straightforward inhibition of the transporter protein by the drugs but rather due to the alterations in cellular microenvironment (e.g., membrane composition) induced by the combination of antipsychotic and cholesterol-lowering drugs.

Table 1. Inhibition of ABCB1 transport activity by TFP and TFP combined with SIM at 5 µM.

TFP concentration [µM]	FIR	FIR TFP + SIM (1 hour)	FIR TFP + SIM (48 hours)
5	1.73	1.27	2.59
10	2.16	2.21	2.83
25	2.30	2.38	3.00

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Honey as a source of bioactive compounds – a review

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Honey is a natural product which owes its health benefits to its numerous bioactive compounds. Indeed, the nutritional and therapeutic properties of honey have been known for centuries. The composition of honey is highly diverse and depends on the type of honey and its origin. The main bioactive compounds in honey include phenolic acids, flavonoids, enzymes, and organic acids. The therapeutic properties of honey include antibacterial, antifungal, antiviral, antioxidant, antidiabetic, antiobesity, anticancer, anti-inflammatory, and wound-healing qualities. In this review, I will present a comprehensive analysis of the medicinal properties, health benefits, and physiochemical properties of various honeys.

Figure 1. A summary of bioactive compounds in honey and their therapeutic properties and health benefits.

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Properties of mayonnaises based on mustard oil

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Fat is an essential component of the diet. However, it is important to remember to choose fats characterized by a high content of unsaturated fatty acids. The presence of other bioactive components in the fat contributes to increasing its nutritional value. At the same time, the presence of unsaturated fatty acids is a factor that accelerates fat oxidation and the formation of undesirable fat oxidation products.

Mayonnaise is one of the products characterized by a high fat content. The use of oils with a high content of unsaturated fatty acids can increase the quality of mayonnaise. Most often, refined rapeseed oil is used for the production of mayonnaises. However, in this study, cold-pressed mustard oil was used to prepare mayonnaises. White mustard variety Warta, used for mayonnaise production, is a new mustard variety characterized by a higher content of n-3 fatty acids and a lower content of glucosinolates.

The aim of the study was to compare the fat quality and the content of bioactive compounds in mayonnaise prepared with refined rapeseed oil versus mayonnaise prepared with cold-pressed oil from Warta variety mustard.

Gas chromatography with a flame ionization detector was used to analyze n-3 fatty acids and phytosterols. Fat quality and antioxidant potential were determined using chemical methods. Viscosity was measured with an oscillatory rheometer.

In the sensory analysis, mayonnaises based on rapeseed were rated the highest. Increasing the amount of mustard oil used to prepare the mayonnaise decreased the overall rating of the mayonnaise. Mayonnaise made from mustard oil was characterized by the highest content of bioactive compounds such as n-3 fatty acids and phytosterols. However, rapeseed-based mayonnaises exhibited the highest antioxidant potential.

Mayonnaises based on mustard oil provide a better source of n-3 fatty acids and phytosterols than mayonnaises based on rapeseed oil. The quality of mayonnaise made from mustard oil, stored for 2 weeks, met the fat quality requirements.

Sea buckthorn: a promising source of bioactive lipids for glucose regulation

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Obesity and associated type 2 diabetes mellitus (T2DM) represent major global health challenges, driving the search for novel therapeutic and nutritional strategies. Sea buckthorn (*Hippophae rhamnoides* L.) has emerged as a promising dietary component due to its high content of bioactive compounds, particularly fatty acids such as palmitoleic acid (POA, 16:1n-7). However, the role of POA remains controversial, and the biological activities of its cis and trans isomers have not been sufficiently compared.

The aim of this study was to evaluate the antidiabetic potential of sea buckthorn oleosomes and oils, as well as to assess the activity of POA isomers. Oleosomes from five cultivars and oils obtained using different extraction methods were analyzed. Additionally, *in vitro* lipolysis products, mimicking gastrointestinal digestion, were examined. Digested oils, especially those obtained by hexane extraction, significantly enhanced glucose-stimulated insulin secretion (GSIS) via activation of G protein-coupled receptors (GPR40, GPR55, GPR119, and GPR120). Free fatty acids released during digestion were identified as key contributors to this effect. Among them, palmitoleic acid showed the strongest insulinotropic activity and high affinity for the studied receptors, particularly GPR40. Comparative analysis revealed that *trans*-POA exhibited lower cytotoxicity than the *cis* isomer, while maintaining similar insulin-secretory activity. Both isomers activated GPCRs but differed in their signaling pathways. Additionally, lysophosphatidylcholines containing POA were identified as novel ligands for GPR119.

These findings suggest that sea buckthorn oils and oleosomes may serve as valuable functional food components supporting T2DM management, primarily through the activity of free fatty acids acting as GPCR ligands.

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From probiotic CFS screening to food application: selection of promising candidates and preliminary evaluation in a tragacanth-based coating for refrigerated shrimp preservation

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Pacific white shrimp (*Litopenaeus vannamei*) is highly valued for its sensory and nutritional quality, yet its shelf life is limited by microbial spoilage during refrigerated storage. Natural bioactive compounds and edible coatings are increasingly explored as safer alternatives to conventional preservatives [1]. In a previous in vitro screening study, nine probiotic strains were evaluated for antimicrobial activity, leading to the selection of *Lactobacillus acidophilus*, *Pediococcus acidilactici*, and *Lactobacillus rhamnosus* as the most promising cell-free supernatant (CFS)-producing candidates for further application-oriented investigation [2]. The present study represents the next stage in this progression, focusing on the preliminary evaluation of selected probiotic CFS in a tragacanth-based coating system for refrigerated shrimp preservation. For application-stage clarity, the current work emphasizes formulations derived from *P. acidilactici* and *L. acidophilus*, while *L. rhamnosus* remains part of the background selection framework. Shrimp samples were treated with representative coating formulations and stored at 4 °C for 10 days, with microbial quality monitored at regular intervals. Microbial counts increased in all groups during storage; however, coated treatments showed a slower increase than the untreated control. Among the two main application strains, both demonstrated preliminary preservation potential, but the *P. acidilactici*-based formulation showed the clearest advantage in total bacterial count control under the tested conditions. On day 10, total bacterial count reached 4.68 log CFU/g in the *P. acidilactici* treatment, compared with 7.00 log CFU/g in the untreated control, while the *L. acidophilus* treatment reached 6.40 log CFU/g. Similar control-relative reductions were observed in selected spoilage-associated bacterial groups. Overall, this study demonstrates a clear transition from screening to application, supporting the potential of probiotic-derived metabolites as natural bioactive components in seafood preservation systems. Further optimization and validation are required for broader practical applications.

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Moroheiya as a bioactive hydrocolloid: distinct functional effects of leaf powder and mucilage extract in rice noodles

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This study extracted and characterized Moroheiya (*Corchorus olitorius* leaves) mucilage extract (MHE) as a bioactive-rich hydrocolloid and evaluated its functionality in rice noodles in comparison to Moroheiya leaf powder (MHP). Hot water extraction was used to extract mucilage polysaccharides under varying temperatures (70 and 90 °C) and extraction times (3, 5, and 10 h). The best extraction process was achieved at 90 °C for 10 h, resulting in the extraction of 12.11% polysaccharides, which had a high content of flavonoids at 27.17 mg QE/g extract, total sugars at 0.96 g/g crude extract and antioxidant capacities of 0.08 mM TEAC/g crude extract (2,2-diphenyl-1-picrylhydrazyl, DPPH•) and 0.04 mM TEAC/g crude extract (2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid), ABTS•$^{+}$). In addition, molecular size analysis showed a broad range of polysaccharides, with a predominance of high molecular weight components (>22,800 Da), alongside minor low-molecular-weight components. Rheologically, MHE and MHP (1%, w/v) exhibited weak elastic gel characteristics and shear-thinning viscosity, which is indicative of pseudoplastic non-Newtonian behavior. The incorporation of MHE (0.5, 1 and 1.5%, w/w) into rice noodles boosted antioxidant activity; however, higher inclusion levels reduced firmness and increased cooking loss. MHE and MHP at the same studied concentrations showed similar effects on noodle quality, differing primarily in visual appearance and antioxidant enhancement. Therefore, appropriate Moroheiya incorporation could be a promising approach for the development of better nutritional value and the creation of healthier noodles.

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Liposomes encapsulated with phytosterols as a bioactive food component

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Plant sterols and stanols, collectively referred to as phytosterols, are currently consumed in several approved specialty foods to reduce low-density lipoprotein (LDL) cholesterol and prevent cardiovascular disease. Compared with cholesterol absorption (50–60%), phytosterol absorption is much lower, ranging from <7% for sitosterol to <16% for campesterol, and depends on the medium and the addition of emulsifiers. The bioavailability of many biologically active compounds in the digestive tract can be enhanced by encapsulating them in liposome-based delivery systems. It is crucial for consumers' health and safety to develop new forms of plant sterols and stanols that are thermo-oxidatively stable and have higher gastrointestinal tract absorption.

The main goal of our research was to incorporate liposomes containing encapsulated phytosterols and their esters into mayonnaise and to analyze their stability during storage and frying.

Mayonnaise samples were prepared in the laboratory using rapeseed oil, egg yolk powder, sugar, acetic acid and salt. It was divided into samples, and 2%, 4%, 6%, and 8% of stigmasterol oleate liposomes were added. The resulting samples were heated at 60 °C for 8 hours and at 180 °C for 30, 60, 90, and 120 minutes. Fat was extracted from the tested samples using the method described by Folch et al. [1]. Subsequently, the fatty acid and sterol content was determined by GC-FID gas chromatography [2,3]. The control sample was mayonnaise without liposomes. The experiment was performed in triplicate.

Based on the results, the encapsulation of stigmasterol oleate in liposomes protected both oleic acid and stigmasterol from degradation during heating at 60 °C. During heating at 180 °C, stigmasterol degraded after just 30 minutes, while oleic acid remained stable.

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Nutraceutical potential of jackfruit (*Artocarpus heterophyllus.*Lam): under-utilized species from Mauritius

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Underutilized jackfruit (*Artocarpus heterophyllus*) represents a promising source of functional foods and nutraceuticals. This study evaluated the pulp and seed extracts of two Mauritian varieties (Jaune and Laboue), revealing significant ($p<0.05$) levels of bioactive compounds including phenols, flavonoids, terpenes, and tannins, with the highest values in seed extracts *(Laboue: TPC = 73418 mg GAE/g, TFC = 153.52 mg quercetin/g; Jaune: TPC = 8370 mg GAE/g, TFC = 44.25 mg quercetin/g)*. Seeds extracts also showed high vitamin A and C content *(Jaune: 4.18 µg β-carotene/g; 1235.78 mg/100 mL ascorbic acid)* lower than the control (2318 mg/100ml ascorbic acid)(Table 1). Seeds and pulp extracts were evaluated using FRAP and DPPH assays, with seed extracts highest (DPPH= 0.60 mg/mL; FRAP=536.05mg Fe^{2+} eq/g) (Table 1) and antimicrobial activity screening against *B.cereus, E.coli, S. aureus, P. aeruginosa and C.albicans* showed potent antioxidant and antimicrobial profile *(MIC in Seeds Extracts = 0.39 mg/ml similar as antimicrobial controls)* (Table 2). Overall, methanolic seed extracts showed higher nutraceutical activity than pulp, with the Laboue variety exhibiting the strongest antibacterial, antifungal, and bioactivities, highlighting jackfruit's potential as a functional food.

Extracts	Jackfruit Varieties	Vitamin A (Mean beta carotene/g)	Vitamin C (mg/100 mL ascorbic acid)	DPPH (IC50-mg/mL)	FRAP (mg Fe 2+ eq/g)
Seeds	**Jaune**	4.18	1235.78	0.65	536.05
	Laboue	3.00	1306.22	0.60	579.30
Pulp	**Jaune**	3.67	58.71	12.2	173.96
	Laboue	2.70	132.09	4.90	503.27

Extracts	Variety	Minimum Inhibitory Concentration (MIC) (mg/mL)				
		E.coli	***P.aeruginosa***	***S.aureus***	***B.aureus***	***C.albicans***
Seeds	**Jaune**	**0.39 ± 0.00**	**1.56 ± 0.00**	5.21 ± 0.00	0.78± 0.00	6.25± 0.00
	Laboue	3.13 ± 0.00	**1.56 ± 0.00**	**1.56 ± 0.00**	**0.39± 0.00**	**3.13± 0.00**
Pulp	**Jaune**	3.13 ± 0.00	3.13 ± 0.00	**1.56± 0.00**	2.08± 0.00	6.25± 0.00
	Laboue	6.25 ± 0.00	12.5 ± 0.00	6.25± 0.00	6.25± 0.00	6.25± 0.00
Controls		0.39 ± 0.00	3.13 ± 0.00	0.39 ± 0.00	0.39 ± 0.00	0.39 ± 0.00

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Looking back or into the future: which approach is better for protein design?

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The engineering of robust biocatalysts remains a cornerstone of industrial biotechnology, with Oleate Hydratases (OAHs) gaining significant attention for their ability to convert unsaturated fatty acids into high-value hydroxy acids [1]. However, optimizing these enzymes for industrial conditions often requires a delicate balance between catalytic activity and thermodynamic stability. In this study, we explore two contrasting yet potentially synergistic strategies for protein design: Ancestral Sequence Reconstruction (ASR) [2] and automated stability-driven design (PROSS) [3].

Using the GRASP algorithm, we performed a comprehensive ASR of the OAH phylogenetic lineage to rediscover "extinct" structural motifs that may confer superior stability and broader substrate tolerance. In parallel, the PROSS (Protein Repair One-Stop Shop) server was employed to design a stabilized version of a contemporary OAH through an energy-based, evolutionary-filtered approach.

Our comparative analysis reveals a striking divergence between the two methods: the modifications suggested by PROSS and the substitutions identified through ASR occur at distinct, non-overlapping sequence positions. We discuss the implications of these disparate results for the design of OAH variants, questioning whether "restoring" the past or "repairing" the present is more effective for enhancing biocatalytic performance.

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Modern detoxifying preparations: the use of biocomponents to mitigate the impact of mycotoxins in feed on animal health and production performance

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Mycotoxins are toxic compounds produced by filamentous fungi such as Fusarium, Aspergillus, Penicillium, that may contaminate food and agricultural products. The most important mycotoxins considering their health and economic impact are aflatoxins, ochratoxin A, patulin, trichothecenes, fumonisins, deoxynivalenol, and zearalenone. These metabolites can appear at various stages of the food chain, posing a threat to humans and animals [1]. The health problems of animals exposed to mycotoxins impact their development, weight gain, reproductive problems, and productivity, resulting in economic losses for farmers. Various preventive measures are taken at the plant cultivation stage, but they do not sufficiently eliminate these compounds. Furthermore, their resistance to various environmental factors means they cannot be destroyed by simple physical procedures, such as high temperatures, which are standard in many technological processes. As a result, mycotoxins enter plant-derived products, including feed. Therefore, various adsorbent materials, such as activated carbon, aluminosilicates, and cholestyramine, are increasingly being used as feed additives to minimize the risk of mycotoxin presence. Microbiological methods, including the use of microorganisms safe for both humans and animals, such as yeast, have also gained interest in recent years [2]. As a biological eco-component, yeast cell walls with a high content of mannanoligosaccharides (MOS) and β-glucans, which also have adsorption properties, are often added. Despite their widespread use, the above-mentioned ingredients have their limitations, which were analysed in this paper. The data indicate that mineral sorbents are effective in sequestering aflatoxin B1, but are significantly less effective in adsorbing fusarial toxins, including zearalenone, deoxynivalenol, and fumonisin. Another problem is the potential for non-selective binding of other ingredients, such as nutrients, which reduces the viability of their wider use.

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Valorization of rapeseed presscake through hydrothermal pretreatment, fractionation and fungal fermentation

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Rapeseed cake is a solid residue originating from crude rapeseed oil pressing. Its high protein content (~30%) makes it a valuable animal feed or, recently, a source of plant protein isolates for food production. However, industrial rapeseed cakes are heat processed prior to oil pressing, therefore proteins in the seeds are desaturated which makes their further extraction through alkaline solubilisation and isoelectric precipitation extremely inefficient.

In this study, a process for extraction of proteins from industrial rapeseed cake was developed using mild-conditioned hydrothermal pretreatment for recovery of three fractions: fiber rich solids, protein concentrate and pretreatment liquid which was subsequently used as cultivation media for edible *Aspergillus oryzae* fungus. Screening experiments (300 mL scale) showed that pretreatments under alkaline conditions (pH 9-11) does not provide efficient protein extraction from the feedstock and only alkaline conditions provide effective separations of fibers and proteins in studied process temperatures (90-180°C). The experiments were then scaled to 1.5 L using a bench scale hydrothermal reactor with 75 g/L solid load and process pH of 1 and 3. The fiber rich fraction was obtained by filtration and the filtrate was then fermented with *A. oryzae* with or without recovery of remaining suspensions by centrifugation which were recovered prior to and after cultivation process.

The liquid and solids recovery after filtration showed that higher liquid filtrates can be obtained under lower pH of the pretreatment process which corresponded with lower recovery of solids that contained ca. 18 and 27% crude protein (CP) with ca. 25 and 19% crude fiber for pretreatment pH of 1 and 3, respectively. The CP contents in filtered liquids ranged 12-20 g/L which corresponded with CP extraction efficiencies of 40.5-82.6% in relation to the liquid recovery and CP input. The fungus did not grow in samples pretreated at 150 and 180°C (pH 1) and at 180°C. Cultivation of *A. oryzae* showed that biomass yield was not depending on whether the media were centrifuged or not, yielding ca. 9 g/L biomass regardless of pretreatment conditions. CP and crude lipid (CL) content in biomass ranged 30.6-38.1% and 6-19% respectively. Unfermented protein isolates were recovered with the yields ranging 1.6-18.5 g/L crude liquid and had CP and CL contents of 27.7-35.6 and 15-25% respectively. Recovery of protein concentrates (by drying) after fermentation showed high yields of 26-59 g/L with CP and CL contents of 28-43% and 11-18% respectively. In addition, total glucosinolates (GS) contents were determined for all solid samples obtained in the experiments. A reduction of GS content in relation to the feedstock was observed for solid samples after filtration (from 12.2 to 7-11 µM/g) while slight increase was observed for various protein concentrates (15-17 µM/g) and only one sample GS content shown values exceeding EU limit of 18 µM/g in feed. Analysis of GS in fungal biomass did not yield any result, probably due to unsuitability of the method to such a sample.

The research shows the potential of hydrothermal pretreatment for hot pressed rapeseed cake for recovery of its components and fermentation with *Aspergillus oryzae*.

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