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Faculty of Agriculture

The 3rd International UNIFood Conference

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THE WORD OF WELCOME

Dear colleagues,

We warmly welcome you to the **3rd International UNIFood Conference – UNIFood2024** organized by the Faculty of Agriculture, University of Belgrade and University of Belgrade. This event engages not only academics, but also stakeholders from all relevant industries and business sectors, and will serve as a meeting point and platform for the dissemination of new ideas and the development of new partnerships. Food scientists, technologists, researchers, nutritionists, engineers and entrepreneurs will share their knowledge on the latest advances in all aspects of food production, processing, sustainability, safety and quality, nutrition and health, and knowledge transfer supporting environment.

The first UNIFood conference, organized as a national, was launched in 2018. as one of the events in honor of the **210th anniversary** of the **University of Belgrade** which was ranked 35th on the 2017 Shanghai list in Food Science and Technology. More than 250 proceedings from thirteen countries including 83 oral presentations (including three plenary lectures, eight invited lecturers and three section lecturers) and a round table dedicated to better cooperation between academia and industry highlighted the importance of food research in various fields of science and technology that require multidisciplinary and multistakeholder approaches. The second UNIFood conference was organized as an international in 2021. and gathered 273 participants from 23 countries, with 52 oral presentations (including four plenary lectures, five key note speakers, seven invited lectures and three section lecturers) as well as round table and workshops.

We are delighted that you have choosen to participate in this collaborative conversation, where authors from 19 countries will present their recent work in 99 poster presentations and 60 oral communications (including four plenary lectures, four keynote lectures, nine invited lectures and seven section lecturers). You will have the opportunity to participate in three educational workshops, round table and discuss current EU project results.

Belgrade, one of the oldest cities in Europe, always young, at the confluence of the Sava and Danube rivers, will be your host. At the confluence of new ideas and experiences we welcome you again and wish you a fruitful discussion and the establishment of new collaborations.

Sincerely,

Prof. Dr Mirjana Pešić
President of the Scientific Committee
of UNIFood2024

The conference organizers gratefully acknowledge the generous support provided by the following:

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PLENARY LECTURES

STRATEGIES TO INCREASE STABILITY AND BIOAVAILABILITY OF NATURAL PRODUCTS

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There is an increasing demand for natural products and plant extracts, in particular for the new generation of science-based and standardized functional botanical ingredients to formulate herbal medicinal products and healthy products, mainly represented by medical devices, cosmetics and dietary supplements. This trend is principally due to numerous health benefits of natural products and plant extracts recently reported in the scientific literature, representing new therapeutic approaches or complementary and/or alternative treatments to the current medications, and a huge opportunity to meet consumer demand. The scarce water solubility, low lipophilicity and inappropriate molecular size of many natural compounds, which undergo structural instability in biological milieu, rapid clearance and high metabolic rate, have severely limited their use. Nanomedicine represents an excellent tool to increase bioavailability and activities of natural products. Generally, nanosized delivery systems provide large surface area increasing dissolution properties and can overcome anatomic barriers. In addition, passive and active targeting can optimize the performance of the nanocarriers. Passive targeting takes advantage of the unique pathophysiological characteristics of inflamed and tumour vessels, enabling nanodrugs to accumulate in the tissues. The effect is called enhanced permeation and retention, generally obtained by the decoration with polyethylene glycol the vector surface. An intriguing strategy is to decorate the nanocarriers with special ligands in order to recognize and bind to target cells through ligand–receptor interactions. The lecture aims to describe novel nanoformulations, namely polymeric nanoparticles and lipid based-nanocarriers, which can represent successful examples to overcome these limitations of natural pleiotropic molecules, extracts and essential oils.

Keywords: natural products, solubility and stability, bioavailability, nanocarriers

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MEDITERRANEAN TRADITIONAL INFUSIONS AS A VALUABLE SOURCE OF BIOACTIVE COMPOUNDS

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Herbal infusions are commonly consumed in the traditional Mediterranean diet. Some examples of these are mountain tea, lemon balm, and sage which are valuable sources of bioactive compounds. Mountain tea is recommended as a traditional medicine for the relief of mild gastrointestinal disorders and against the common cold. Many studies have unveiled its benefits on human health which are attributed mainly to their rich content of polyphenols. Lemon balm is considered a traditional herbal medicinal product for the relief of mild symptoms of mental stress and to aid sleep, as well as for the symptomatic treatment of mild gastrointestinal complaints including bloating and flatulence. The presence of a wide range of chemical constituents (such as triterpenoids and polyphenols) has been reported to be responsible for its pharmacological effects. Sage infusions as traditional herbal medicines are used for addressing mouth and throat disorders, skin disorders, minor wounds, and gastrointestinal ailments. Their rich phytochemical content, including flavonoids, depsides, and phenolic acids could be responsible for the reported broad range of pharmacological properties such as antioxidant and anti-inflammatory activities. Observational epidemiological studies support the benefits of the Mediterranean dietary pattern to increase life expectancy, reduce the risk of major chronic disease, and improve quality of life and well-being. As part of the Mediterranean diet, representing the gold standard in preventive medicine, probably due to the antioxidant and anti- inflammatory properties of the consumed herbal products, the present study aimed to explore the phytochemical content and the bioactive compounds of mountain tea, lemon balm, and sage.

Keywords: mediterranean diet, mountain tea, lemon balm, sage

ENZYME INHIBITORS: FOES AND FRIENDS?

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Enzyme inhibition theory is a prominent topic in scientific research. The increasing prevalence of global health problems underscores the urgent demand for therapeutic options to combat these diseases. Enzymes are vital components in the discovery of new drugs. Some key enzymes are implicated in the development of diseases. By inhibiting these enzymes, we can reduce the harmful effects of the diseases and effectively manage them. By inhibiting cholinesterase in Alzheimer's disease, acetylcholine levels in the synaptic clefts can be elevated, leading to enhanced memory function in patients. This principle is known as the cholinergic hypothesis, which serves as the foundation for the majority of Alzheimer's medications. Furthermore, blocking amylase and glucosidase can effectively postpone the increase in blood sugar levels for individuals with diabetes. Utilizing tyrosinase, the primary enzyme in melanin formation, can lead to targeted skin therapies. In the pharmaceutical industry, various chemicals like galantamine, kojic acid, and acarbose have been developed as enzyme inhibitors for diseases like Alzheimer's, skin diseases, and diabetes. However, their usage is restricted, primarily because of toxicity issues. Therefore, there is a demand for new and harmless inhibitors to replace these synthetic ones. In the study, the recent developments, advantages and disadvantages of enzyme inhibition methodologies and their application were summarized. The information can open on new horizons for further applications on enzyme inhibition and further applications.

Keywords: enzyme inhibition, amylase, obesity, cholinergic hypothesis, cancer

NATURAL INGREDIENTS OBTAINED FROM PLANTS AND MUSHROOMS WITH APPLICATION FOR FOOD INDUSTRY

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Lately, there has been a surge in scientific validation highlighting a multitude of applications and benefits stemming from the utilization of natural ingredients and edible matrices worldwide. The utilization of plants and mushrooms to enhance health not only represents a rich cultural legacy but also serves as a traditional source of micro and macronutrients and various medicinal preparations. These matrices comprise natural ingredients of significant added value, serving as both natural colorants and preservatives, while imparting bioactive properties upon integration into other products.

Presently, diverse innovative technologies have been employed to optimize extraction systems, thereby enhancing the purity of natural target compounds and extraction yields. Notably, colorants extracted from *Beta vulgaris* L., *Gomphrena globosa* L., *Bixa orellana* L., and *Curcuma longa* L. have found successful application in a variety of food formulations. Conversely, a wide array of biowastes and/or by-products have proven effective reservoirs of bioactive molecules. For instance, extracts from *Agaricus bisporus*, abundant in pro-vitamin D2 (ergosterol), and fruit residues have demonstrated diverse bioactivities. Acorn peels have been utilized as preservatives, while by-products from the fish industry contribute to the formulation of wholesome pet food. Furthermore, the extraction of bioactive compounds from all components of figs and pumpkins, followed by their incorporation into derived products, has yielded satisfactory outcomes, bolstering the circular economy of these constituents. In parallel, bioactive molecules sourced from olive pomace have been reclaimed for further integration into cosmeceutical formulations, concurrently enhancing sustainable extraction processes and their optimization.

These findings underscore the efficacy of natural ingredients sourced from various matrices, advocating for their valorization as reservoirs of naturally derived ingredients suitable for incorporation into widely consumed and appreciated food products on an industrial and commercial scale.

Keywords: natural ingredients, food preservatives, colorants, bioactives, phenolic compounds

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KEYNOTE SPEAKERS

INNOVATION AND SUSTAINABILITY AS DRIVERS OF FOOD SYSTEMS TRANSITION

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The agri-food sector is key to economic prosperity and can promote human health and support environmental ecosystems, although it is currently threatening both. The current food system is a substantial driver of global environmental change, contributing to climate change, biodiversity loss, and severe environmental impact also related to the food waste and loss is occurring.
In this scenario, the policy initiatives of the United Nations (Agenda 2030, SDGs) and of the European Commission (e.g. the Green Deal, Farm to Fork strategy) have paved a route to accelerate the transformation of the agri-food systems towards its sustainability and to guarantee safe and healthy food for all at global level.
A paradigm change for the profound transformation the food system needs is required by leveraging a disrupting and systemic innovation connected with future technologies. A number of innovations resulting from scientific and technological advancements, ranging from the agricultural practices to the food production, from land use and emissions to improved waste management, from novel food sources to improved diets, are increasingly available.
The potential of innovation for a transition towards a sustainable food system depends on various factors including their level of readiness, local development, regulations, cultural and a societal recognition of their value. A close interplay of all the stakeholders of the complex agri-food system including policymakers and societal players to enable transformation to occur is needed. Innovation and sustainability are key driving forces to be implemented and integrated in a modern food production system, where quality and safety are pillar objectives and food security has to be guaranteed.
In this framework, academia plays a main role not only as accelerators of innovation but also as training providers of a skills and competences to the next generation of professionals, scientists, managers and leaders meet the challenges of the ecological transition of the food system.

Keywords: innovation, sustainability, food system

CHROMATOGRAPHY AND MASS SPECTROMETRY IN ANALYSES OF PHYTONUTRIENTS IN FOOD SAMPLES

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Plants (e.g. vegetables, fruits, grains, legumes, nuts, spices) are rich sources of many plant secondary metabolites called phytonutrients or phytochemicals. The major groups of phytonutrients are: polyphenols (e.g. flavonoids, phenolic acids), carotenoids, phytosterols, some triterpenoids, isothiocyanates, etc. Due to their diverse chemical structures and bioactivities (antioxidant activity, enhancement of immune response or cell-to-cell communication, lowering blood pressure and/or cholesterol level, etc.) many daily consumed phytonutrients are still unknown. Many phytonutrients have still not been properly investigated, but several phytonutrients are already used as ingredients of food supplements or functional foods. An increased use of phytonutrients in such products created the demand for new sources of these bioactive compounds. Methods based on chromatographic and hyphenated techniques are an indispensable tool in the discovery of new sources of phytonutrients, development of new food products as well as control of food quality and safety. The lecture will focus on targeted and non-targeted analyses of phytonutrients in food samples (e.g., food supplements, food waste, bee pollen, medicinal plants). The methods based on high-performance thin-layer chromatography (HPTLC-densitometry, HPTLC-image analysis, HPTLC-MS/(MS) and high-performance liquid chromatography (HPLC-UV/Vis, (U)HPLC-MS/(MS)), as well as (HP)TLC–effect-directed analysis (EDA) will be discussed. Examples of challenges in development of chromatographic methods will include the lack of chromophores, isomeric structures, stability of the analytes, unknown impurities in standards, lack of commercial standards and standard reference materials. The influence of different parameters (e.g. stationary phase, time, etc.) on (HP)TLC–EDA analyses of antioxidants and enzyme inhibitors in extracts of different samples (e.g. bee pollen, Japanese knotweed, etc.) will also be addressed.

Keywords: phytonutrients, chromatography, mass spectrometry, effect-directed analysis

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BIO-BASED ACTIVE MOLECULES AND INNOVATIVE PROCESSES FOR FOODS AND BEVERAGES

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Besides the recent achievements in obtaining natural agents to act as preservatives, colorants, sweeteners, among others, new formulations including functional foods, energetic drinks, as also ready to eat foods are a hot topic of the current research with a big market demand.
Taking into account the diversity of bioactive molecules/secondary metabolites present in nature, matrices such as plants, mushrooms as also byproducts/bioresidues from the industrial sector are promising sources of these agents. Therefore, the main goal is to obtain these molecules and find viable and sustainable applications. Mushrooms for instance, especially mushroom residues (broken ones, non-commercialized), are interesting matrices to obtain added value molecules such as phenolic acids, statin like molecules and beta-glucans, components with high bioactive potential, namely hypocholesterolemic activity either by inhibiting cholesterol absorption or by decreasing the cholesterol synthesis. Individually or in synergic ratios, these molecules are being exploited as possible natural based hipocholesterolmic formulations under the Mush4Chol project to be included in functional foods. Taking advantage of the high capacity of mushrooms in assimilating components from the growing substrates, ongoing research is also being conducted to elicit these bioactive molecules.
Within the food sector but regarding the processing technologies, an innovative approach has been the development of “ready to eat” formulations, involving important and challenging research in all the processes since dehydration and further hydration capacities of the final products, allowing the easier transportation, availability and maintaining the same properties.
Similar to the food industry, also the beverage industry is investing in innovative drinks, such is the case of functional, energetic/sport beverages. In this field, completely natural isotonic drinks were developed based on thermal waters, apple juice and hibiscus extract, enriched in minerals and in bioactive molecules. Moving to the beer sector, research is also being conducted within the Bio4Drinks project, aiming at preserving the aromatic profile of craft beers, that due to oxidation processes presents a very reduced shelf-life, an issue that can be overcome using natural extracts with preservative capacity, increasing the beer storage time without alteration in the organoleptic and sensorial profiles.

Keywords: functional foods, isotonic drinks, bio-based active agents, craft beers

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APPLICATION OF INNOVATIVE EXTRACTION TECHNIQUES FOR THE ISOLATION OF BIOACTIVE COMPOUNDS FROM FOOD INDUSTRY BY-PRODUCTS

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Generating large quantities of by-products during food production in a large number of cases results in irreparable environmental damage. The aforementioned directly impacts the economic efficiency of the production process due to the underutilisation of the potential of raw material input, as well as the waste-disposal costs. Insufficient utilisation of natural resources is, among other things, a consequence of the application of traditional extraction procedures. Recent trends in the extraction techniques have largely focused on finding solutions that minimize the use of harmful solvents and allow the use of alternative, so called "green" solvents that ensure safe and high quality extracts. This research is focused on food industry by-products valorisation through the application of different green extraction techniques including supercritical CO_2 extraction, subcritical green solvent extraction, ultrasonic-assisted extraction etc. Food industry by-products could be an alternative source of bioactive compounds which possess different biological activities that could provide health benefits. These bioactive compounds can be isolated, encapsulated and incorporated into new products with added value.

In this presentation, the development of high-valuable extracts rich in biologically active compounds from selected food industry by-products will be reviewed. Therefore, the waste generated during food processing could be effectively utilised by applying different green extraction techniques in the development of value-added products, thus ensuring a complete circular economy, which is imperative today.

Keywords: food by-products, green solvents, extraction, bioactive compounds, circular economy

INVITED LECTURERS FROM UNIVERSITY OF BELGRADE

DESIGN OF EXPERIMENTS: CONCEPTS, APPLICATIONS AND EXAMPLES IN THE FOOD INDUSTRY

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The main goal in food processing and production is to find a compromise between maintaining key quality characteristics and increasing system performance without increasing production costs and time. Determining the optimal nutritional and functional properties, processing conditions and combination of ingredients can be achieved using design of experiments concepts. Experimental design is a statistical approach in which mathematical relationships can be established between dependent (responses) and independent (factors) variables that influence system characteristics. In experimental design, all investigated (independent) parameters are varied simultaneously and the mathematical models created are usually validated using analysis of variance. The two main applications of experimental design are screening and optimization. This concept of experimental design enables to determine how individual factors and relationships between variables influence the tested responses. The choice of experimental design depends primarily on the objectives of the experiment and the number of investigated parameters. The usual approach is to start with a screening design in order to select which variables are significant and at which levels. For this purpose, a full factorial, a fractional factorial or a Plackett-Burman experimental design can be used. After the initial screening experiments, further investigation must be carried out in which the most important factors are further optimized. Multivariate statistical methods are employed in this context, with central composite design and Box-Behnken as part of the response surface methodology being the most common used. In food analysis and processing, optimal conditions need to determined by optimizing a number of responses simultaneously, and the most commonly used methods are the Derringer desirability function and mixture design. There are many examples of the application of experimental design methodology in food analysis and processing. Examples of the drying process, the preparation of edible packaging and the preparation of functional products based on milk proteins and mushroom extracts will be presented here.

Keywords: experimental design, factorial designs, response surface methodology, multi-response optimization, food processing

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APPLICATION OFARTIFICIAL INTELLIGENCE IN THE DEVELOPMENT AND CHARACTERIZATION OF NATURAL PRODUCTS

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Artificial intelligence (AI) is changing the landscape of natural product research, providing innovative tools for development and characterization that increase both efficiency and precision. This talk will highlight the central role of AI techniques, including machine learning (ML) algorithms such as artificial neural networks (ANN), decision trees (DT), clustering techniques, dimensionality reduction methods, etc. Several case studies will be explored to highlight the superiority of AI models in predicting outcomes and analyzing data complexity, which can outperform traditional approaches. Focus will be put on using AI in the optimization of compounds extraction from medicinal herbs, analysis of bioactive compounds and their effects in natural products, as well as on analysis of multivariate nature of data obtained from characterization of such products. AI methods can discern intricate patterns and predict product characteristics with remarkable accuracy, and can thus be used for quality control and optimization. This discussion will extend to nanoencapsulation technologies, emphasizing how AI algorithms can model and predict release behaviors of bioactive compounds, thereby guiding the design of advanced delivery systems for nutritional and pharmaceutical applications. This predictive capability is crucial for designing controlled-release formulations that optimize the therapeutic efficacy and/or bioavailability of bioactive compounds. Additionally, the lecture will explore the use of text mining and natural language processing, in conjunction with AI, to aggregate and interpret vast amounts of literature on biomedicine and natural products, showcasing AI's capacity to streamline research and discovery in nutritional and pharmaceutical applications of natural products.

Keywords: artificial intelligence, machine learning, data science, natural products, development, optimization, characterization

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POLYPHENOLS: THE ROLE OF FOOD BIOACTIVE AGENTS IN COMBATING MICROBIAL VIRULENCE

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Polyphenols are well-known group of bioactive molecules distributed in various foods such as citruses and honey. They have been recognized mainly due to their antioxidant properties but recent insight into their antimicrobial spectrum implies potential novel application for this group. Polyphenols are able not only to block growth of pathogenic microorganisms, but they can also interfere with microbial ability to induce disease, microbial virulence. With the abundant studies conducted worldwide we now know about range of virulence factors that are necessary for disease induction and though we have new antimicrobial targets. One of the most studied virulence factors is microbial ability to form biofilms. We have recently studied rutin, flavonoid glycoside found in citruses. This molecule has the ability to block biofilm formation of *Pseudomonas aeruginosa* IBRS P001 and *Staphylococcus aureus* IBRS MRSA 011, strains that are resistant to antimicrobial therapeutics. In addition to reduction of biofilm biomass, rutin antibiofilm mechanisms also included reduction in cell viability, exopolysaccharide, and extracellular DNA levels. Moreover, moderate inhibition of bacterial adhesion to keratinocytes upon rutin treatment was observed. Rutin antivirulence mechanisms involved inhibition of *P. aeruginosa* protease, pyocyanin, rhamnolipid, and elastase production and the downregulation of the *lasI*, *lasR*, *rhlI*, *rhlR*, *pqsA* and *mvfR* genes. This research has proven wide antivirulence potential of rutin. Moreover, we have tested range of flavonoids as inhibitors of fungal virulence and resistance. Rutin, but also apigenin and apigetrin, have shown promising antibiofilm and anti-hyphal properties in several clinical *Candida albicans* strains examined, which was also confirmed on molecular level.
Polyphenols have wide antivirulence capacity employing a range of mechanisms and might be used for the development of novel antimicrobial strategies. Intake of food polyphenols might have some health benefits in the terms of limiting microbial pathogenicity, which is the possibility that should be explored in more detail.

Keywords: polyphenols, antimicrobial, antivirulence, bioactivity

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SILICON AND IRON IN FOOD CROPS: IMPACT ON HUMAN HEALTH

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Silicon (Si) and iron (Fe) are the second and fourth most abundant elements in the Earth's crust, respectively. The essentiality of Fe was recognized in the mid-19th century, yet essentiality of Si for both higher plants and humans is not fully accepted. Silicon serves as a beneficial mineral for plants, enhancing their resilience to biotic (diseases and pests) and abiotic (drought, low pH, salinity, nutrient disbalances, etc.) stresses. The beneficial effects of Si for human health, including contributions to bone and collagen development and the prevention of Alzheimer's disease, have also been well established. Approximately one-third of global agricultural soils are conductive to Fe-deficiency in various crops, leading to anemia in over two billion world population. To improve content of Si and Fe in edible plant parts (biofortification), two primary strategies are proposed: (1) increasing soil bioavailability of both minerals alongside the use of Si/Fe-based fertilizers, and (2) improving the nutritional quality of plant-derived foods through molecular breeding techniques to modify the content of Si and Fe in crops. We revealed that Si supplementation in crops can promote the root acquisition of Fe and enhance its phloem transport to the edible crop parts. Our recent results demonstrate the ability of Fe-deficient crops to increase Si availability in the rhizosphere, which, in turn, enhances the uptake and transport of both minerals. This opens new innovative approaches in crop Si/Fe biofortification practices for improved human health.

Keywords: biofortification, crops, human health, iron, silicon

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ENHANCING GUT HEALTH AND LONGEVITY THROUGH NOVEL STARTER CULTURES IN FERMENTED PRODUCTS

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For nearly 10,000 years, fermented foods have been essential to the human diet, boasting a significant variety today. The health advantages of fermented foods have been recognized for a long time. These benefits may result from direct interactions of ingested live microorganisms with the host, as a probiotic effect. They may also come indirectly as a result of the ingestion of microbial metabolites synthesized during fermentation. Research suggests that probiotics found in dairy products have positive effects on human health. In recent years, there has been extensive research into the use of starter cultures with probiotic characteristics to address various health conditions. A Western-type diet characterized by a high daily intake of saturated fats and refined carbohydrates leads to the accumulation of a wide variety of molecular and cellular damages over time, leading to several age-related diseases. According to the World Health Organization, the number of people worldwide over 60 years of age was estimated to be 1 billion in 2019, with an expected increase to 2.1 billion by 2050. The increasing number of the elderly population will be accelerated in the coming decades, particularly in developing countries, such as Serbia. We have tested several carefully selected natural isolates of lactic acid bacteria, originating from artisanal dairy products from specific geographical locations in the Balkan peninsula for the ability to decelerate the cell-aging process. Our results revealed that these strains possess exciting probiotic features such as strengthening the epithelial intestinal barrier through stimulation of autophagy, upregulating the tight junctions between the epithelial cells of the intestine which prevent the passage of harmful substances from the intestine to other organs, activating the antimicrobial defense, and extending the lifespan of *Caenorhabditis elegans* via autophagy activation, making them great candidates for probiotic starter cultures for functional dairy food.

Keywords: probiotic, starter cultures, functional food, fermented products, age-related diseases

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THE IMPORTANCE OF TRICHINELLA PROFICIENCY TESTS IN FOOD SAFETY

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Public health, consumer health and food safety are very important internationally and are well regulated. Trichinellosis is a serious zoonotic disease (caused by parasitic nematodes of the genus *Trichinella*) and represents public health issue. People acquire trichinellosis by consuming raw or undercooked meat infected with the *Trichinella* larvae. The gold standard in meat inspection for the presence of *Trichinella* larvae is the use of the artificial digestion method. For the prevention of human infection and for international trade purposes there are regulations for the inspection of meat as well as for quality control. European Union Reference Laboratory for Parasites (EURLP), Rome, Italy, organizes once a year quality controls - *Trichinella* Proficiency Tests (PTs) for all National Reference Laboratories for *Trichinella* (NRLTs). The Serbian Reference Laboratory for *Trichinella* successfully participates for many years. According to Regulations National Reference Laboratories for *Trichinella,* in all member states of the European Union, organize *Trichinella* proficiency tests every year and all laboratories performing artificial digestion of meat need to participate. The Accreditation Body of Serbia requires only from accredited laboratories a certificate of participation in *Trichinella* PTs. In Serbia Institute for the Application of Nuclear Energy INEP and Faculty of Veterinary Medicine successfully and independently organize PTs (from 2017 and 2022 respectfully). The results were published in Serbian and international scientific journals. In 2022 Serbian *Trichinella* legislation was changed and now all laboratories which control meat for the presence of *Trichinella* larvae need to participate in PT. It is desirable that all these laboratories have and use reference material (*Trichinella* larvae in alcohol) and, if necessary, request test samples for training purposes. European and our own experience shows that participation in proficiency tests leads to better laboratory results over time and better consumer protection.

Keywords: Trichinella, proficiency test, food safety

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ELECTROCHEMICAL SENSING OF PESTICIDES IN FOOD

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Pesticides are chemical compounds widely used in agricultural activities to prevent, control, regulate, and eliminate various pests (fungi, insects, mites, rodents, weeds) on agricultural crops. Plant crop yields increased significantly after the introduction of pesticides in agricultural cultivation. On the contrary, numerous pesticides have toxic properties. The uncontrolled use of these chemical compounds affected their significant environmental accumulation. As a result, plant crops (vegetables, fruits), water, groundwater, air, and agricultural soil are contaminated, simultaneously representing the pathways of pesticide residue entry into the food chain. The human organism is frequently exposed to pesticide residues through contaminated food, which may lead to acute or chronic effects on consumers' health. Accordingly, control of pesticide residue levels in food (generally in the environment) is of great importance. Electrochemical methods offer a promising avenue for pesticide residue determination. They outshine other analytical methods (gas chromatography, liquid chromatography, fluorescence spectroscopy, colorimetry) due to their distinct advantages, such as simplicity, low-cost equipment, fast analysis, simple sample preparation, and high sensitivity and selectivity during target analyte determination. The development of nanotechnology and the use of nanomaterials/nanocomposites in constructing various electrochemical sensors have significantly improved sensing performances (low limit of detection, high selectivity). In addition, nanomaterials have proven to be suitable matrices for immobilizing biological species (enzymes, antibodies, nucleic acids) during the development of electrochemical biosensors and immunosensors for pesticide residue detection in food samples. The miniaturization and commercialization of these sensors are among industrial production's main challenges and interests. Today, portable miniature devices (touch- or paper-based sensors) connected to smartphones can be found on the market, especially in food quality assessment. Therefore, there is a constant need for the development of electroanalytical platforms that include innovative approaches in preparing electrochemical sensors for food contaminants detection.

Keywords: electrochemical sensors, food safety, food contaminants, nanomaterials, pesticides

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GREEN LEAF BIOMASS AS AN ALTERNATIVE PROTEIN SOURCE

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Ensuring enough protein for growing world population while respecting sustainability goals puts alternative proteins in the spotlight of food science research. Green leaf biomass represents prospective plant-based alternative source of proteins. The use of leafy biomass as protein source has long research history, but recent increase in protein demand highlighted its potential for food applications. This is mostly because of the enzyme ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO), major fraction of leaf water-soluble (white) proteins. RuBisCO is ideal for human consumption due to its low allergenicity, high digestibility and excellent amino acid composition. In addition to the acknowledged nutritionally valuable RuBisCo protein, other leaf proteins, namely membrane (green) leaf proteins, could be used more in human nutrition.

However, leafy biomass remains underutilized due to the complexity of protein extraction and purification. Insufficiently purified proteins compromise their functionality and applicability in food formulations. Therefore, the development of scalable processes remains an imperative for the cost-efficient and sustainable production of leaf-based proteins. One of the key features for the efficient process is the right choice of the starting material. The starting material should be rich in protein, available in excessive amount, and preferably without other applications.

Pumpkin leaves represent an appropriate choice for production of leaf-based proteins. These leaves are rich in proteins and they are obtained as waste (or by-product) during pumpkin production, hence their usage is in accordance with the trend of making agroindustry more sustainable. Our results showed that the highest yield of pumpkin leaf proteins is obtained when twin-screw press was used for tissue disruption with subsequent repressing. Usage of alternative extraction (ultrasound-assisted) and purification (membrane separation) methods positively affected protein functionality.

Keywords: alternative protein, leaf protein, RuBisCO, agro-waste valorization

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NUTRITION AND GUT MICROBIOTA, DIFFERENT GUT-AXES

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Gut microbiota (GM) is a complex, dynamic, and heterogeneous ecosystem comprised of bacteria, viruses, fungi, and protozoa that colonise the gastrointestinal tract. GM has a symbiotic relationship with the host and regulates both local and systemic physiological functions. The GM profile of each individual is unique and is influenced by various factors such as genetics, nutrition, lifestyle, environmental conditions, early microbial exposure and the immune system. Dysbiosis is regarded as any imbalance in composition and/or function of microbial ecology. Considering the influence of GM is extended not only to the gastrointestinal tract, a concept of multiple bidirectional gut-organ axes has been suggested. In that sense, the effect of dysbiosis can be observed in different organ systems. Dysbiosis leads to mucin degradation, disruption of gut barriers, increased permeability and consequently local and systemic infiltration of pathogenic microorganisms and their by-products. This invasion leads to immune cells activation and systemic inflammation affecting the heart, the liver, kidneys and central nervous system. Nutrition plays a critical role in regulating the multiple gut-organ axes. Specific dietary patterns have been shown to affect the abundance and prevalence of certain bacterial strains thus affecting the GM homeostasis. The intake and ratio of macro- and micronutrients highly impacts the susceptible GM balance and can exert both harmful and favourable effects. Additionally, supplementation with certain probiotics, prebiotics and synbiotics promotes local protective mechanisms that lead to antiinflammatory effects, exerting systemic beneficial outcome. Gut modulation through nutrition and therapy-tailoring according to individual health profile is a promising tool for targeting various diseases, emphasizing the pivotal role of nutrition in shaping human health.

Keywords: microbiota, dysbiosis, nutrition, gut-organ axes

SECTION LECTURERS

CROCUS SATIVUS TEPALS EXTRACT AS A PROMISING TREATMENT FOR OBESITY-RELATED METABOLIC DISORDERS

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Obesity and the associated metabolic complications have become a global health problem for which the usual therapeutic strategies are only of limited effectiveness. To date, saffron (*Crocus sativus* stigmas) has been used primarily in the food, cosmetics and pharmaceutical industries. While numerous studies have confirmed the potential of *Crocus sativus* stigmas to combat obesity, the role of *Crocus sativus* tepals, which are usually wasted during saffron production, as a source of bioactive compounds is still unexplored.

To this end, we investigated the effects of oral administration of an extract of *Crocus sativus* tepals in an animal model of diet-induced obesity. We analysed visceral (VAT) and subcutaneous (SAT) adipose tissue and lipid metabolism in mice fed a 60% fat diet for 14 weeks and orally treated with an extract of *Crocus sativus* tepals for the last 5 weeks of the diet. Energy intake, body mass, triglycerides, systemic insulin sensitivity, adipose tissue histology, insulin signalling and lipid metabolism in VAT and SAT were analysed.

We have demonstrated for the first time that oral administration of an extract of *Crocus sativus* tepals in obese animals results in weight loss, improved systemic insulin sensitivity, lower triglyceride levels and improved lipid peroxidation. Treatment with *Crocus sativus* tepals extract had a suppressive effect on SAT hypertrophy, while it was absent in VAT, suggesting that treatment with *Crocus sativus* tepals extract has differential local effects on adipose tissue development and metabolism. The suppressive effect on hypertrophy of subcutaneous adipocytes was accompanied by reduced inflammation and preserved insulin signalling in this tissue, which most likely contributed to the improved systemic insulin sensitivity.

Considering the results obtained, future pharmaceutical/nutraceutical interventions to improve subcutaneous adipocyte function may help to maintain adequate insulin sensitivity and reduce the risk of developing obesity-related complications.

Keywords: Crocus sativus, adipose tissue, obesity, insulin sensitivity, lipid metabolism

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PHENYLAMIDES IN FOOD: UHPLC/QTOF/MS IDENTIFICATION AND STRUCTURAL ELUCIDATION

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Ultra-high-performance liquid chromatography coupled with quadrupole time-of-flight mass spectrometry (UHPLC/Q-ToF/MS) is a powerful technique that has been used for target and untarget analysis of various bioactive compounds in food and plants. The Q-ToF detector (quadrupole and ToF mass analysers) have unique characteristics (high resolution, fast acquisition, wide mass range, and high ion mobility), which enables its application for identification and structure elucidation of biocompounds. In addition to phenolic compounds, various phenylamides are being increasingly analyzed recently, due to their health benefits such as neuroprotective (agents for treatment of Alzheimer's and Parkinson's diseases), anti-aging, anti-cancer, antioxidative, anti-inflammatory and anti- microbial effects. Phenylamides exist as polyamine (putrescine, spermidine and spermine) or arylmonoamine (tyramine, tryptamine, dopamine, serotonin, octopamine, agmatin) conjugates connected with various phenolic acid moieties, and some of them present very important metabolites of plants and pollen grains. This review is primarily focused on the application of UHPLC/Q-ToF/MS technique for identification of various phenolic acid-polyamine derivatives, as well as their recently confirmed glycosylated forms. These phenylamide derivatives have characteristic MS fragmentation in positive ionisation mode, which can be used for prediction of their structures. Typical MS fragments can indicate the type of polyamine core, phenolic acid moieties (coumaroyl, caffeoyl, dihydrocaffeoyl, feruloyl, sinapoyl) and their positions on the polyamine core, as well as degree/position of glycosylation. Identification of phenylamides is additionally explained on several representative examples (derivatives) detected in tomato leaves, pollen samples and red goji berry. The use of this technique in the analysis of food enriched with phenylamides, as well as their behaviour during *in vitro* digestion, is also considered. Finally, this review points out advantages and limitations of UHPLC/Q-ToF/MS technique for identification of phenylamides with the tendency to facilitate the future characterization of these derivatives.

Keywords: phenylamides, UHPLC Q-ToF MS, food, spermidine, spermine

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EFFET OFAPPLE FIBER ON PROBIOTIC *L. CASEI*SURVIVAL DURING YOGURT PRODUCTION, STORAGE AND SIMULATED GASTROINTESTINAL TRANSIT

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Consumer interest in functional foods and probiotics, has increased in recent years. Dairy products, such as yogurts, fermented milks, and beverages, are considered ideal for delivering probiotic bacteria to humans, and have been part of the human diet for many years. However, to provide health benefits, probiotics should survive both in the food matrix during storage and through gastrointestinal transit after consumption. In the present study, probiotic yogurt with *Lacticaseibacillus casei* ATCC 393 was produced via fortification with apple fibers (0, 1, 2, 3% w/v). The number of *L. casei* cells during yogurt production and refrigerated storage (even after 28 days) was above the requirement of 10^6 cfu g^{-1} (10^8 – 10^9 cfu g^{-1}) for microorganisms (other than the starter culture) added to yogurt according to FAO/WHO, and for probiotic food, according to the US FDA and the food industry. Of note, the number of yogurt starters during storage was higher than 10^8 cfu g^{-1} for all formulations, which is higher than the established FAO/WHO requirement of a minimum number of viable cells of yogurt starter bacteria of 10^7 cfu g^{-1} during consumption. Although no significant positive effect of apple fiber on the probiotic viability during storage was detected, the addition of apple fiber seems to have a beneficial effect on *L. casei* viability during simulated gastrointestinal conditions. More specifically, a slight decrease in the numbers of *L. casei* was detected during the simulated oral and gastric phase and a more significant one (10^3 – 10^4 cfu g^{-1}) during the simulated intestinal phase. However, in all yogurts a viability of 10^5 – 10^6 cfu g^{-1} after the simulated gastrointestinal conditions was observed. The results of this study suggested the efficiency of apple fiber to increase the survival of *L. casei* in yogurt during simulated gastrointestinal conditions.

Keywords: functional food, probiotic, L. casei, dairy products, apple fiber

BEE POLLEN AND BEE BREAD AS SOURCE OF PROTEIN AND BIOACTIVE COMPOUNDS

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The need for diets with adequate amounts of proteins and essential amino acids is increasingly demonstrated. For chronic diseases such as diabetes, cardiovascular disease, cancer and neurodegeneration, dietary interventions can sometimes serve as a treatment strategy. This request is important even for other target populations such as malnourished children or the elderly, but also vegetarians and/or athletes. For all these situations, meat, fish and eggs are available on the market as sources of protein, but in vegetarian and/or calorie-restricted eating plans it is always a problem to find sources of protein, so we must consider looking for new options. Both bee pollen and bee bread are, depending on the botanical origin, a possible choice. These products are also a source of minerals and vitamins, providing added value to complement an unbalanced diet and boost the immune system. Many other constituents of these two bee products can also be used in drug discovery to develop new medicines and/or contribute to other therapeutic strategies. The best known are phenolic and polyphenolic compounds with anti-inflammatory and antioxidant bioactivity, among many other applications. More recent research reveals that bee pollen and bee bread are also good sources of spermidines. These compounds have emerged as well-tolerable calorie restriction mimetics targeting several age-associated molecular and physiological adversities. To ensure that all possible applications of these two products obtain the best final result, product quality control must be guaranteed. For Bee Pollen, the ISO standard was already completed in 2023, ISO 24382:2023- Bee pollen — Specifications. Currently, the same group (ISO/TC 34/SC 19/WG 3) has started working on the Project ISO/AWI 25097 - Management standards for the production and packaging of bee pollen. Additionally, they intend to propose a new project related to Bee Bread. Another advance was a compilation of Standard Methods that can be used for analytical control (https://www.tandfonline.com/doi/pdf/10.1080/00218839.2021.1948240), published by the Bee Pollen Working Group of the International Honey Commission in 2021. In conclusion, conditions exist to allow bee pollen and bee bread to be high- quality dietary products.

Keywords: amino acid, flavonoid, ISO, nutrition, spermidine, standard

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METABOLIC REPROGRAMMING IN PROSTATE CANCER: THE SIGNATURE OF CELLULAR TRANSFORMATION WITH CLINICALIMPLICATIONS

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Cancer, a multifaceted and exceptionally complex disease, originates from disrupted biological circuits and control mechanisms, giving rise to a wide range of inter- and intra-type variations, elaborate mutational landscapes, and multifactorial genetic and phenotypic anomalies. The sustainability of malignancies relies heavily on their aptitude to obtain sufficient resources, adapt to changing microenvironment, and strategically rewire their metabolic architecture. The excessive energy demands imposed by uncontrolled proliferation present significant challenges to cellular biochemistry, forcing cancer cells to undergo substantial reorganization of signaling pathways and transcriptional networks. These adjustments cater to the distinctive anabolic preprequisits of neoplastic growth and contribute to the unique rearrangements observed in tumor metabolism. A notable degree of variability exists within and across diverse cancer entities concerning substrate utilization and pathway reprogramming, highlighting the inadequacy of a uniform metabolic blueprint in accuratelly portraying the intricacies of cancer-driven metabolic alternations. Prostate cancer ranks as the second most frequently diagnosed malignancy in the male demographic, with an estimated 1.4 million cases and a consequential toll of 375000 deaths reported globally in 2020. Driven by these statistics, contemporary urologic oncology is experiencing a surge in research interest aimed at comprehensively understanding the metabolic reprogramming inherent in prostate neoplasia. These investigations delve deeply into the regulatory machinery behind cancer-related metabolic alterations and their profound ramifications on disease initiation and progression. Unveiling the unique metabolic circutry governing this malignancy holds vast potential for pioneering advances in both scientific and clinical realms, offering avenues for more precise risk assessment and the discovery of novel biomarkers and therapeutic targets. Such research endeavors offer the prospect of fundamentally reshaping the landscape of diagnostic and management strategies for this intricate and heterogeneous urooncological condition.

Keywords: prostate cancer, urologic oncology, metabolic reprogramming, hallmarks of malignancy

WINE-MAKING SUSTAINABILITY IN THE ERA OF CIRCULAR ECONOMY: THE CASE OF WINE LEES

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Winemaking, as an agro-industrial process, generates substantial quantities of by-products like grape pomace and wine lees. The latter refers to the residue left behind during racking or bottling of wine and is predominantly composed of dead yeast cells, along with ethanol, phenolic compounds, and tartrates. Yeast extract, a widely used nutrient media component, is a powder-like substance derived from commercially grown yeast biomass, serving as a nitrogen source. In the context of by-products' valorization, wine lees, being rich in dead yeast cells, can be utilized through the process of autolysis for the production of an autolysate, acting as a yeast extract substitute. In the present study, the effect of different parameters, e.g. pH, temperature, on wine lees autolysis efficiency was examined, which was calculated based on the free α-amino nitrogen increase in autolysates. In addition, growth of two major winemaking microorganisms, i.e., *Lactiplantibacillus plantarum* and *Saccharomyces cerevisiae*, was monitored in culture media containing wine lees yeast extract and commercial yeast extract. The effect of yeast extracts mixing, concentration and inoculum cell concentration on major kinetic parameters was evaluated. Results indicate that a significant free α-amino nitrogen increase was observed, reaching a maximum of 834%, while the amino acid profile of autolysates was significantly affected by autolysis temperature. Regarding *L. plantarum* growth, results indicate that similar kinetic parameters were reported for media containing either wine lees or commercial yeast extract, while lag phase duration was not significantly affected by inoculum cell concentration. Moreover, regarding *S. cerevisiae* growth, it appears that addition of both yeast extracts resulted in increased maximum specific growth rate values, although negatively affecting lag phase duration and maximum cell concentration. Findings of the present study appear promising towards the holistic valorization of wine lees, promoting the concept of circular economy and sustainable development for wineries.

Keywords: wine lees, yeast extract, L. plantarum, S. cerevisiae

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ASSESSMENT OF MYCOTOXIN ASSOCIATED HEALTH RISK IN PEOPLE ON GLUTEN FREE DIET

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In recent years, an increased variety of gluten-free products intended to people with celiac disease has been observed. These individuals need to avoid gluten in their diet, which in practice means substitution of wheat, barley, rye and oat with corn, rice, buckwheat and millet. However, all these cereals and products thereof could be contaminated with mycotoxins – aflatoxins (AFs), ochratoxin A (OTA), zearalenone (ZEA), deoxynivalenol (DON) and fumonisins (FUM), which are regulated by the law. The objective of the study was to assess mycotoxin associated health risk in people on gluten free diet. For that purpose, a total of 60 samples, including flour, pasta, bread, breakfast cereals, biscuits and crackers, all marked with a crossed grain symbol or "gluten free" wording, was purchased in supermarkets in Novi Sad (Serbia). In order to assess the risk, mycotoxins concentrations, determined using high performance liquid chromatography, were combined with food consumption data. For toddlers and other children, at mean level of consumption of all investigated food categories except flour margin of exposure indicated risk of AFB1, as well as risk of OTA in case of bread, pasta and crackers. In adolescents, the risk was indicated regarding AFB1 in bread, breakfast cereals and biscuits and for vegetarians additionally in pasta (which also caused the risk of OTA); for adults, elderly and pregnant women the risk of AFB1 was related to bread and breakfast cereals. Mean exposure levels of ZEA, DON and FUM were below their respective tolerable daily intakes even when all food groups were combined. On the level of individual samples, 8-10 of them (depending on the population group) were responsible for low margins of exposure in case of AFB1 and 5-8 in case of OTA. The study findings underline the need of a constant surveillance of population exposure to mycotoxins.

Keywords: cereals, food safety, risk assessment, public health

LECTURE AND ORAL PRESENTATION WITHIN SECTIONS

COMPARISON OF THE EFFECTS OF ROSMARINIC AND CAFFEIC ACIDS ON KIDNEY REDOX STATUS IN EXPERIMENTAL HYPERTENSION

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Hypertension is associated with increased oxidative stress. Reactive oxygen species play an important role in the physiological regulation of renal function, and oxidative stress occurs because of peroxidative damage to kidney macromolecules and cell membranes. Thiols are effective endogenous antioxidants that can preserve protein structure, protecting cells and tissues from damage induced by oxidative stress. We previously characterized rosmarinic (RA) and caffeic (CA) acid as the main polyphenolics of the *Thymus serpyllum* L. aqueous extract (TE) with strong hypotensive and antioxidative effects when administered acutely to spontaneously hypertensive rats (SHR). Here we compare the effects of chronic consumption of RA and CA on kidney redox balance in SHR. Adult, male rats were divided into three groups: the control SHRC group received vehicle, SHR+RA group received RA (15mg/kg/day) and SHR+CA group received CA (3mg/kg/day), by gavage during 4 weeks period. These doses of RA and CA corresponded to their amount in previously used TE. Quantity of kidney free-protein thiol groups, superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) activities were detected spectrophotometrically, while systolic blood pressure was measured by direct method. Chronic consumption of RA and CA significantly reduced SBP compared to control ($p < 0.05$) and both treatments increased the activities of SOD ($p < 0.001$) and CAT ($p < 0.05$, and $p < 0.001$, respectively), while GPx activity was increased in SHR+RA compared to both SHRC ($p < 0.001$) and SHR+CA groups ($p < 0.01$). The level of thiol groups decreased significantly in SHR+RA ($p < 0.001$) and SHR+CA ($p < 0.01$) compared to the SHRC group. The activities of antioxidant enzymes were in a significant negative correlation with thiol group. Our results promote the advantages of exogenous antioxidants, such as RA and CA, compared to the endogenous, such as thiols, in establishing kidney redox balance in SHR.

Keywords: hypertension, rosmarinic acid, caffeic acid, kidney redox balance, thiols

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THE APPLICATION OF OKARA IN GLUTEN-FREE BAKERY PRODUCTS

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During the processing of soybeans into milk, up to 30% of the soybeans can be lost as waste, in the form of okara - the insoluble residue of the soybeans that remains after the soymilk is pressed. About 1.2 kg of fresh okara is obtained from 1 kg of soybeans processed into milk. The global production of soybean okara is about 1.4 billion tons per year, but the utilization index is very low - 40% for animal consumption and only 10% for human consumption. More than 50% of okara ends up in waste and causes significant environmental pollution. Okara is characterized by a high content of total proteins (35.07-40.36%), a low residual trypsin-inhibitory activity (4.82-7.99%) and a very low content of lectins (0.06-1.73%), while the total content of trypsin-inhibitors in the extracted proteins is high (12.03-14.40%). Okara has a high content of total carbohydrates (34.12-42.96%), which consist of monosaccharides (0.89-19.52%), disaccharides (8.07-22.76%; lactose-free), and dietary fiber (17.90-36.32%). It is also characterized by a very low total energy value (65.76-78.72 kcal/100g). Okara has the highest content of nitrogen and potassium from the group of macroelements and iron and zinc from the group of microelements. Such a composition of biologically active components, as well as good techno-functional properties, enable the use of okara as a nutritionally complete herbal supplement with the aim of obtaining a functional food with a low energy value (in the formulation of: meat products, pasta, beverages, candies, flour, as edible packaging in the food industry, or as an ingredient of fish feed and also in the bakery industry). The formulation of gluten-free bakery products represents a major challenge for food technology in the 21st century. Gluten-free breads have poorer sensory and textural properties compared to gluten-containing breads because they lack gluten, which is the carrier of the product structure. In order to improve the sensory characteristics and compensate for the missing gluten, gluten-free bread is prepared with various ingredients such as sugar and additives. This not only leeds to poorer sensory properties, but also to a poorer nutritional quality of the bread. The addition of okara in the production of gluten-free products is one of the solutions to overcome these problems. For example, a 30% proportion of okara in the formulation of gluten-free bread (70% - mixture of pseudocereal and cereal flours) results in a bread that is characterized by a high fiber content (14%), a rich source of protein (8.8%) and certain minerals (e.g. zinc, iron), low content of saturated fatty acids (0.8%), the absence of sugar, significant content of total phenolic components and favorable antioxidant activity, as well as low energy value1. According to sensory characteristics, this bread belongs to the category of very good and excellent quality with an average rating of 4.59 by consumers)1. It can be concluded that okara is very suitable for the production of eco-innovative low-energy gluten-free bread, enriched with dietary fibers and proteins with a pleasant taste, which is in line with the principles of sustainable waste management in the food industry.

Keywords: sustainable food production, bakery products, by-product of soybean processing

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IS THERE A DIFFERENCE IN THE MINERAL COMPOSITION OF ORGANIC AND CONVENTIONAL HONEY?

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Honey is a nutritionally valuable product in the form of a sweet syrup mixture produced by honey bees. It has been used in human nutrition since ancient times. The production of organic food is experiencing great growth due to the human health and protection of the environment. Organic honey production requires production conditions that are significantly different from the conditions of conventional beekeeping. In conventional beekeeping the use of various pesticides is allowed for which the maximum permissible values in honey are defined. Improper use of these pesticides makes honey a threat to human health. It is a generally accepted opinion that organically produced honey has a higher nutritional value than conventionally produced honey. However, is that so? The aim of this research was to determine the potential difference in the mineral composition between honeys of the same botanical origin produced in organic and conventional beekeeping. Eight conventional and organical honey samples were used with different botanical origin (linden, acacia, chestnut, meadow) produced in the Balkans region. Optical emission spectrometry (ICP-OES) on a Thermo Scientific iCAP 6500 Duo ICP instrument (Thermo Fisher Scientific, Cambridge, UK) with iTEVA operating software was used for analysis of samples mineral content. The most abundant mineral elements in all samples analyzed were potassium, calcium and phosphorus. Significant content of sodium, magnesium and sulphur were also found in all examined samples. From the group of microelements, the presence of: iron, zinc and manganese were registered. Among toxic elements the presence of boron and aluminum, as well as lead and arsenic was not recorded while lithium was practically in traces. The results showed that the mineral composition of organically and conventionally produced honey does not depend significantly on the method of honey production (organic/conventional beekeeping) but much more on the botanical origin.

Keywords: linden/acacia/chestnut/meadow honey, production method, mineral composition

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EFFECTS OF PROTEOLYTICALLY-ACTIVE LACTOBACILLI STRAINS ON SOURDOUGH STARTER FERMENTATION PROCESS

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Gluten-related disorders have surged in recent years, underscoring the necessity for innovative approaches to alleviate symptoms and enhance gluten digestion. In this study, we embarked on a comprehensive exploration of proteolytic activity within a collection of *Lactobacillus* strains to uncover their potential in gluten peptide hydrolysis and sourdough fermentation process. We initiated our investigation by screening 120 *Lactobacillus* strains for proteolytic activity on gluten peptides, with a goal of identifying the most potent candidates. Subsequent probiotic characterization followed, focusing on antimicrobial capabilities against prevalent pathogens. Safety characterization ensued, including testing for antibiotic resistance, ensuring the suitability of selected strains for further investigation, ending in the refinement of our selection to 11 candidates. To assess their applicability in sourdough fermentation, the selected strains were introduced into khorasan wheat sourdough starters, and their impact on growth kinetics and pH modulation was monitored. Among the selected strains, only one strain of *Lactobacillus brevis* (BGZLS30-24) demonstrated a significant effect on the growth kinetics and pH reduction of the sourdough starter. Additionally, protein isolation from the mature sourdough starter facilitated the evaluation of proteolytic activity within the dough inoculated with the selected strain. While a detectable proteolytic activity was observed, its magnitude appeared to be attenuated compared to the initial screening test. Furthermore, metagenomic analysis of sourdough starters was conducted to gain insights into microbial diversity dynamics during the maturation process, revealing that the addition of the BGZLS30-24 significantly shortens the microbiota maturation time. Textural analysis of sourdough breads was conducted to elucidate the impact of BGZLS30-24 on the final product, revealing its contribution to increased bread volume and reduced hardness, indicating improvements in textural properties. Our findings show the versatile role of selected *Lactobacillus brevis* BGZLS30-24 in gluten digestion and sourdough fermentation, hinting at it's potential as an adjunct in developing novel strategies for managing gluten-related disorders and enhancing the quality of bakery products.

Keywords: sourdough, lactobacilli, gluten, microbiota, bread

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INFLUENCE OF VITAMIN E ON THE ADIPOGENIC POTENTIAL OF BONE MARROW AND ADIPOSE TISSUE MESENCHYMAL STEM CELLS ISOLATED FROM OSTEOARTHRITIS PATIENTS

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Osteoarthritis (OA) is a chronic inflammatory degenerative disease which leads to articular cartilage and subchondral bone destruction. A clear risk factor for developing OA is being overweight since population-based studies have confirmed a link between obesity and OA. Also, it has been established that vitamin E, a fat-soluble antioxidant affects the lipolysis of adipose tissue and prevents cholesterol accumulation in obesity, emphasizing its application as a supplement in patients suffering from OA as essential. Comprehensive profiling of mesenchymal stem cells (MSCs), cells exhibiting self-renewal and multilineage differentiation potential is of great importance for defining molecular phenotype of overweight or obese OA patients and their treatment. In this study we investigated the effects of vitamin E on adipogenic differentiation of MSCs derived from hip bone marrow (BM MSCs) and peripheral adipose tissue (PAT MSCs) isolated from matched OA patients undergoing hip arthroplasty. For that purpose, the occurrence of lipid droplets after 3-weeks incubation of BM MSCs and AT MSCs in adipogenic medium with increasing concentrations of vitamin E (0, 20 and 40 μM) was studied by Oil Red staining. Gene and protein expression of markers of adipogenic differentiation (adiponectin and PPAR gamma) were analyzed. Our results revealed statistically significant decrease of adipogenic differentiation upon vitamin E treatment in both sources of MSCs determined by counting differentiated cells with lipid droplets. Protein and gene expression of PPAR gamma was decreased in MSCs from both cell sources. Moreover, vitamin E reduced the protein expression of adiponectin and leptin in MSCs of both sources. All the results represent the potential benefits of vitamin E supplementation in overweight OA patients.

Keywords: osteoarthritis, obesity, vitamin E, bone marrow mesenchymal stem cells

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ENHANCING HONEY QUALITY CONTROL USING VIBRATIONAL SPECTROSCOPY TECHNIQUES

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Ensuring the quality of honey is vital for consumer safety, product authenticity, and maintaining its market value. Given honey's economic importance, developing a rapid and cost-effective method to certify its quality is essential. Fourier-transform infrared spectroscopy with Attenuated Total Reflection (FTIR-ATR) and FT-RAMAN techniques have been extensively used in food analysis, particularly for honey. This study aims to compare the effectiveness of these two techniques in the quality control of honey.

Calibration models were performed using Partial Least Squares Regression models for the parameters of total acidity, reducing sugars, hydroxymethylfurfural (HMF), electrical conductivity, ash content, proline content, diastase index, total flavonoids and phenolic compounds content.

The calibration models for each parameter demonstrated determination coefficients greater than 0.965 for FTIR-ATR and greater than 0.983 for FT-RAMAN. The residual prediction deviation (RPD) values were higher than 5.5 for FTIR-ATR and higher than 7.6 for FT-RAMAN. Although both techniques provided excellent results, it can be concluded that FT-RAMAN is a more accurate technique compared to FTIR-ATR for the evaluation of the physicochemical characteristics of honey.

Keywords: FT-Raman, FTIR-ATR, honey, chemical composition, quality control

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MICROBIOLOGICAL QUALITY OF CHOCOLATE-COATED CONFECTIONERY FROM VARIOUS MANUFACTURERS

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Confectionery has become a part of our daily lives due to its high energy value and the improvement of consumer mood through its consumption. Due to their composition and high moisture content, they provide a suitable environment for the growth and development of microorganisms. The aim of this study was to investigate the microbiological quality of chocolate-coated confectionery from different manufacturers, testing microbiological parameters that may affect the quality and also the safety of the product, according to the valid microbiological criteria guidelines. Eight chocolate-coated confectionery products from different manufacturers, which are on the market in the Republic of Serbia, were tested according to the valid ISO methods (ISO 4833, SRPS ISO 21527-2, ISO 21528-2, ISO 6888-1). In addition to the use of standard culture media prescribed by the ISO methods, Petrifilms were also used in this study. According to the Regulative, sample F showed an acceptable result for total aerobic mesophilic bacterial activity with a value of 3.37 log CFU/mL (limit values 3 log CFU/mL and 4 log CFU/mL). The unsatisfactory results of the microbiological examination showed the following samples: Sample labeled H showed the highest activity of Enterobacteriaceae with a count of 2.11 log CFU/mL (the maximum allowable value is 2 log CFU/mL); samples C and E showed the highest activity for yeasts and molds with a cell count of 2.96 log CFU/mL (maximum value is 2 log CFU/mL); the cell count of *Staphylococcus aureus* in sample E was 3.23 log CFU/mL (maximum value is 2 log CFU/mL), which is the highest activity for this microbiological parameter. In conclusion, the unsatisfactory results obtained with the above-mentioned ISO methods indicate improper performance of the production process. It is therefore necessary to work on improving hygiene practices and creating better conditions for product storage.

Key words: candy products, microbiological analysis, microbiological parameters, petrifilms

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OPTIMIZING PARAMETERS RELATED TO THE PHENOLIC AND FLAVONOID CONTENTS OF STEEPE PEONY LEAVES

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The current study had two objectives: (1) the determination of the total phenolic content (TPC) and total flavonoid content (TFC) of steppe peony leaf extracts prepared in two different extraction methods, maceration using conventional orbital shaker (M_{COS}) and ultrasound-assisted extraction (UAE) and (2) optimization of the extraction process parameters *via* varying solvent type (aqueous, and ethanolic (50% and 70%, *v/v*), time of extraction (5, 15, 30, and 60 min), and solid-to-solvent ratio (1:10, 1:20, 1:30, 1:40, and 1:50). The total phenolics were extracted most effectively by UAE (411,3 mg gallic acid equivalents (GAE)/mL of raw extract). The M_{COS} gave lower values, approximately 112,9 mg GAE/mL of raw extract. Total flavonoids content were ranged from 3,0 to 20,0 mg catechin equivalents (CE)/mL of raw extract for UAE, and from 2,98 to 22,87 mg CE/mL of raw extract for M_{COS}. For all tested analytes, it was observed that a concentration of ethanol of 50% (*v/v*), a time of extraction of 15 min, and the solid-to-solvent ratio of 1:20 were the most effective for extracting bioactive substances (phenolics and flavonoids) from Steppe peony.

Keywords: *phenolics, colorimetric determination, gallic acid equivalents, conventional and „green" extraction*

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ENCAPSULATION OF BROCCOLI MICROGREEN JUICE: PHYTOCHEMICAL COMPOSITION AND ANTIOXIDANT ACTIVITY

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Encapsulation is a process that implies the active compounds are enclosed in a wall material using various techniques, creating a barrier that protects the active ingredients from unfavorable environmental conditions. The most commonly encapsulated active compounds derive from plant extracts and juices. Broccoli microgreen juice as a source of active compounds for encapsulation has not been used so far. The aim of this study is the encapsulation of broccoli microgreen juice (BCJ) in maltodextrin as wall material by spray drying technique and the characterization of the obtained powder in terms of phytochemical composition and antioxidant activity. The spectrophotometric assays were used to determine the content of total phenolics (TPC), flavonoids (TFC) and antioxidant activity (AA) ($ABTS^{\cdot+}$, $DPPH^{\cdot}$ and FRAP). The TPC, TFC, and AA were expressed in mg equivalents (gallic acid, quercetin, and Trolox, respectively) per 100 g of the encapsulates. The values determined for TPC were higher than those for TFC. Regarding antioxidant activity, the results followed the order FRAP>$ABTS^{\cdot+}$> $DPPH^{\cdot}$. It should be noted that the antioxidant potential expressed by the encapsulated BCJ varied due to different mechanisms of the employed antioxidant assays. In summary, broccoli microgreen juice encapsulated in maltodextrin showed a high content of phenolic compounds and good antioxidant activity and can be defined as a novel food ingredient. In addition, future studies should focus on the addition of encapsulated broccoli microgreen juice in food products and the characterization of such products.

Keywords: encapsulation; spray drying; microgreen juice; phytochemical composition; antioxidant activity

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NUTRITIONALAND PHYSICOCHEMICAL PROPERTIES OF TOMATO JUICE ENRICHED WITH BLUE SPIRULINA POWDER

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Tomato (*Solanum lycopersicum* L.) juice is one of the most widely consumed juices in the world. Due to its attractive colour and flavour, it represents a suitable vehicle for nutritional enrichment. Spirulina (*Arthrospira platensis*) is microalga characterized by high protein content and represents a rich source of various macro- and microelements. Spirulina powder is sometimes purified to reduce its unpleasant sensory properties and this powder is characterized by a bright blue colour originating from the high concentration of phycocyanin. The presented study aimed to assess the influence of blue spirulina powder addition on the nutritional composition and physicochemical properties of tomato juice.

Spirulina powder was added to tomato juice in two concentrations (0.8% and 1.6% w/w) to achieve the recommended daily intake (2-4 g) in one glass of enriched juice (250 ml). Standard methods for the juice analysis were used to examine the basic nutritional composition, while atomic absorption spectroscopy (Thermo Fisher, USA) was used to determine the mineral profile. The colour of juices was measured using a Chrome Meter CR-400 (Konica Minolta, Japan), and their textural properties (firmness, consistency, cohesiveness and index of viscosity) were determined by a TA.XT Plus Texture Analyser (Stable Micro Systems, UK).

The amount of proteins and all analysed minerals increased with the addition of blue spirulina powder. Although potassium to sodium ratio decreased, it was still higher than the World Health Organization's daily intake recommendation. Juices enriched with blue spirulina powder had significantly lower lightness (L*), colour saturation (C*), red tone (a*) and yellow tone (b*) intensity than control tomato juice. The addition of spirulina powder significantly changed the textural properties of tomato juice, decreasing its firmness and consistency and increasing its cohesiveness and viscosity. Consumers' acceptance test is necessary to determine the likability of enriched tomato juices.

Keywords: tomato juice, spirulina, nutritional composition, colour, texture

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BETWEEN HOT AND COLD: A FOCUS GROUPS INVESTIGATION OF SLOVENIAN CONSUMERS' FOOD SAFETY THROUGH TEMPERATURE MANAGEMENT

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Consumer food safety is very important as most foodborne outbreaks in the European Union occur in the home environment (EFSA, 2016-2021) [1]. In Slovenia several thousand cases of food poisoning are reported every year and the most frequent intestinal infectious diseases in 2021 included: campylobacteriosis (more than 1080 cases), noroviruses, Clostridium difficile infections, Escherichia coli infections, rotaviruses and salmonellosis [2]. Therefore, a better understanding of consumer food safety is crucial to prepare more efficient interventions to improve food safety in the home environment. The aim of this study was to investigate consumer food safety through a series of focus group discussions about a wide range of food handling activities from shopping to cooking. More than 30 consumers in Slovenia with different demographic characteristics (gender, age and education) participated in the study. The focus groups were conducted in person and online. Each participant took part in one focus group, typically comprising four participants on average. Focus groups discussions were structured - based on 19 questions - and the conversation lasted approximately 1 to 1.5 hours. The presented results will include highlights of temperature management during various food handling activities of the participating consumers from shopping to cooking and leftovers management. Diverse examples of good practices and risk increasing practices will also be presented. These practices can serve as important indicators of where different interventions could be needed to protect and improve public health.

Keywords: consumer food safety, focus groups, temperature management

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ISOLATION OF RUBISCO PROTEIN FROM PUMPKIN LEAVES USING ULTRAFILTRATION: ENHANCHMENT OF YIELD AND PROTEIN CHARACTERIZATION

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RuBisCO (ribulose-1,5-diphosphate carboxylase/oxygenase), an enzyme crucial for plant photosynthesis, is the most widespread protein in the living world. It is predominantly found in the leaves of plants, which mostly remain unused after harvesting. Despite current efforts, the RuBisCO protein isolated from leaves has not yet found significant application as a supplement in human nutrition, due to the complexity and cost-effectiveness of the production process and the low yield. Two ultrafiltration methods were applied for isolation of RuBisCO protein. Initially, cross-flow ultrafiltration was used, employing a membrane module with a membrane's pore size of 10 kDa. The second method involved dead-end ultrafiltration with a feed flow under a nitrogen stream at a defined pressure, using a membrane with a pore size of 100 kDa.

The obtained ultrafiltration fractions were used for the analysis of soluble protein content and protein yield, reducing sugars and polyphenols. The efficiency of the ultrafiltration process and the protein purity were determined by SDS-PAGE electrophoresis. Also, the obtained protein powders were analyzed for the total and free sulfhydryl group contents and surface hydrophobicity. These protein powders were characterized in terms of their technological and functional properties, including solubility, emulsifying properties, and water/oil binding capacities. The results were then compared with those of protein powder obtained from pumpkin leaves using the most common method, isoelectric precipitation. The different techniques for isolating RuBisCO protein showed certain variations in protein yield as well as in its structural and technological-functional properties. Different isoelectric point values were shown for all three protein powders, including variations in the content of free and total sulfhydryl groups. Furthermore, surface hydrophobicity and oil binding capacity showed lower values for the RuBisCO protein obtained by ultrafiltration methods. The solubility of RuBisCO protein obtained by ultrafiltration methods exceeds 90% at pH > 6, indicating that the protein is highly soluble.

Keywords: RuBisCO, proteins, cross-flow ultrafiltration, dead-end ultrafiltration, structural properties, functional properties

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PRESENTING THE RESEARCH ON CHEMICAL FINGERPRINT AND BIOLOGICAL EFFECTS OF CROATIAN PROPOLIS *IN VITRO*

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Due to the scarce data on chemical composition and biological effects of Croatian propolis, the goal of this study is to give an overview of the research on elemental and phenolic profile of propolis, its antioxidant capacity and protective effects at the cellular level. To our knowledge, this is the first study on geno-/cytoprotective effect of ethanolic extract of propolis on human blood lymphocytes *in vitro*. Propolis samples, taken on 60 particularly chosen locations in Istria & Primorje-Gorski Kotar County, were submitted to extraction in 70% ethanol for 40 days, following beekeeping procedures. Chemical profiling was performed using inductively coupled plasma-mass spectrometry (ICP-MS) (for elements) and gas chromatography-tandem mass spectrometry (GC-MS/MS) (for phenolics). Total phenolic and flavonoid content and antioxidant activity of ethanolic extracts of propolis will be determined using spectrophotometry. The protective effect at the cellular level will be assessed using micronucleus-test *in vitro* on human blood lymphocytes treated with cytostatic drug irinotecan, following standard protocol proposed by the Organisation for Economic Co-operation and Development (OECD). Preliminary results showed that the season of sample collection and geolocation of the stationary apiaries significantly affected chemical fingerprint of the propolis, both qualitatively and quantitatively. We expect that this research will fill in gaps in knowledge regarding the specificity of the chemical fingerprint of Croatian propolis, seasonal variation of its chemical content and will provide the first results on its protective role at the cellular level.

Keywords: propolis, phenolics, elements, micronucleus test, cytoprotection

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EFFECT DIRECTED ANALYSIS OF BIOACTIVE COMPOUNDS IN PEELS OF 14 APPLE CULTIVARS

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The individual steps of HPTLC are technically independent among each other and they provide access to versatile methods and flexibility in the detection of targeted compound groups. After the removal of the mobile phase, the bacterial or enzyme assays can be performed directly in a compatible medium. The bioactivity of the sample can be observed from a more detailed perspective than with the strictly defined fractions separated by elution on a column. The subjects of this study were methanol extracts from peels of 14 apple cultivars (6 autochthonous, 4 standard, and 4 resistant cultivars), with the aim to detect and distinguish the extracted compounds by observing their antimicrobial activity. The extraction was performed with a 0.1% hydrochloric acid solution in methanol. The chromatographic separation was carried out on HTPLC silica gel plates with a mobile phase consisting of ethyl acetate: *n*-hexane: formic acid: water (48/33/6/3, *v/v/v/v*). The bioautographic assay was performed with the *Escherichia coli* (ATCC 35218). Visualisation with *p*-anisaldehyde – sulfuric acid reagent (ASA) was used to detect phenolic acids, flavonoids and terpenes. Images of plates (both assays) were taken and processed in ImageJ, while R software was used for further processing of the chromatographic data. Antimicrobial activity was observed in zones between R_F 0.6 and 0.9. The presence of compounds was determined using the ASA reagent, by comparing the R_F values of the active zones and the standards used, and by the colours of the zones after derivatisation. The R_F values of oleanolic, ursolic, ferulic, *p*-coumaric and caffeic acid standards are in the range of the R_F values defined by the zones active against *E. coli*. The prevalence of terpene compounds is striking in the zones with antimicrobial activity. Regarding the antimicrobial activity of the used standard mixture, oleanolic and ursolic acids showed antimicrobial activity.

Keywords: Malus domestica, autochthonous cultivars, oleanolic acid, triterpene, Esecherichia coli

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EFFECTS OF NEXT-GENERATION PROBIOTICS ON NEURAL GENE EXPRESSION IN *Caenorhabditis elegans*

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There is growing body of evidences that the gut microbiota composition is associated with host health making the human gut a unique environment to explore commensal microorganisms with probiotic properties. The emerging subgroup of probiotics named Next-Generation Probiotics represent a novel group of gut bacteria which are strict anaerobes, whose safety status is still not fully determined, but could confer the specific healthy effect on the host. For several microbial strains that are part of the gut ecosystem have been shown to improve behaviour-related functions in the host. Therefore, the aim of this study was to evaluate the neuromodulating properties of gut anaerobic bacteria isolated from healthy individuals in *Caenorhabditis elegans* model system through assessment of the expression of the genes important for proper neural function of the worms. Gut anaerobic bacteria including *Dorea longicatena* NGB204, *Dorea formicigenerans* NGB229, *Bacteroides xylanisolvens* NGB220, *Bacteroides cellulosilyticus* NGB202, *Roseburia intestinalis* NGB215 and *Lancefieldella pravula* NGB231 were isolated from human fecal material in BACTEC media followed by serial dilutions spreading on Columbia Blood Agar supplemented with cysteine and sodium thioglycolate in Whitley Anaerobic Workstation. All of tested gut anaerobes showed capability to differently regulate the expression of the genes involved in serotonin, dopamine and GABA synthesis (*tph-1*, *cat-2*, *unc-25*), neurotransmitters' vesicle release (*unc-64*, *snb-1*, *snt-1*), neuropeptide Y receptor homolog synthesis (*npr-1*), and different classes of neuropeptide Y homologs (*flp-18*, *flp-21*) in *C. elegans* in comparison to worms fed with standard laboratory food *Escherichia coli* OP50. These results imply that selected bacterial species could be considered as potential novel Neurobiotics, the Next-Generation Probiotics with capability to target neural function and modulate different neurodegenerative and psychiatric disorders.

Keywords: next-generation probiotics, Caenorhabditis elegans, gut anaerobic bacteria, neuropeptides, neurotransmitters

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ANTIMICROBIALACTIVITY OF CINNAMALDEHYDE AGAINST COMMON FOOD-BORNE BACTERIA

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Influenced by the shift of consumers' demand toward more natural, sustainable, less processed, and clean-label food, the use of essential oils and their active compounds as antibacterial agents from natural sources in food, especially meat and dairy products, as one of the vehicles most frequently associated with food-borne outbreaks, regain scientific attention. Therefore, the present study aimed to evaluate the antibacterial effects of cinnamaldehyde against some of the most common food-borne bacteria. Susceptibility of the *Salmonella* Typhimurium ATCC 14028, *Salmonella* Enteritidis ATCC 13076, *Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, *Listeria monocytogenes* ATCC 19111 and *Yersinia enterocolitica* ATCC 9610 serovars to cinnamaldehyde (98% purity, Carl Roth, Germany) was investigated by the broth microdilution method. Cinnamaldehyde showed a minimal inhibitory concentration (MIC) of 160 μg/mL against all the tested serotypes, indicating strong antibacterial properties against both, Gram-positive and Gram-negative bacteria. As expected, antibiotic exhibited greater antibacterial activity with MICs ranging from 0.25-1 μg/mL. Namely, tetracycline showed the highest MIC of 0.25 μg/mL against the *S.* Typhimurium and *S. aureus*, while for inhibition of *E. coli*, *L. monocytogenes*, and *Y. enterocolitica* 0.50 μg/mL of antibiotic was required. Finally, *S.* Enteritidis was least susceptible to tetracycline with MIC of 1 μg/mL. In conclusion, despite showing lower antibacterial activity than tetracycline, cinnamaldehyde exhibited antibacterial activity against all tested microorganisms. Since lipid and protein components of the food could interact with active compounds and decrease their antibacterial effect, further studies should investigate the antibacterial activity of cinnamaldehyde in food models.

Keywords: cinnamaldehyde, pathogens, antibacterial properties, minimal inhibitory concentration

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GREEN LEAVES: FROM OIL PROCESSING BY PRODUCTS TO NOVEL PROTEINS NANOPARTICLE STRUCTURES

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Given their high protein content and widespread use, by-products of the oil processing sector, such as pumpkin leaves, are promising alternative protein sources. The most abundant protein on the planet is found in the soluble protein fraction of leaves, commonly referred to as white food protein. This protein is mainly composed of the photosynthetic enzyme RuBisCO (ribulose-1,5-bisphosphate carboxylase/oxygenase). Food proteins are intriguing GRAS (Generally Recognized As Safe) components for nanoparticle delivery systems due to their unique functional properties, enabling the encapsulation of hydrophilic and lipophilic bioactive substances. These traits include the ability to produce gels and emulsions. Furthermore, food proteins can bind bioactive compounds offering opportunities for improved protection until their release. This research serves as proof of concept that the cold gelation protocol, after optimisation, can be effectively employed for the creation of novel protein nanoparticles. The white protein fraction from leaves was isolated using a three-step process, consisting of screw pressing, thermal coagulation, and acid precipitation. Protein nanoparticles (50–150 nm) were prepared by employing a cold gelation protocol with calcium as the cross-linking cation. Nanoparticle characteristics including size, surface charge, and hydrophobicity, were adjusted by changing the cross-linking cation concentration (1–5 mmol/dm^3), environmental pH (7–9), and temperature of the alkaline treatment (70–100 °C at pH 12). The yield of protein nanoparticle and their morphology characteristics, determined via SEM imaging, were the unique parameters for validating and selecting the optimal ultracentrifugation conditions for protein suspensions. For this purpose, time (10–60 min) and rotation speed (40000–80000 rpm) were varied using the Optima^{TM} XPN-100 ultracentrifuge. The obtained nanoparticles exhibited a uniform size distribution and spherical shape. A lower pH value and higher concentration of cross-linking cations found to promote the development of larger and surface-charged nanoparticles. Protein conformation analysis revealed that the calcium ions likely protected the negative charges on the protein polypeptide chains and served as a salt bridge, enabling the polypeptide chains to approach each other. These findings have significant implications for the production of food protein nanoparticles appropriate for the synthesis of novel carriers for bioactive substances.

Keywords: green leaves, white proteins, nanoparticle structures, cold-set gelation protocol

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INSECTS AS ALTERNATIVE SOURCE OFANIMAL PROTEINS IN HUMAN DIET: SAFETY CONCERNS

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With the world's population constantly increasing, edible insects emerge as a very promising, sustainable and alternative food source. Nevertheless, there are a number of potential safety concerns for consumers associated with edible insects and their products, including allergic reactions, pathogenic microorganisms, parasites, pesticide residues and heavy metals. Allergic reactions are the most important safety concern for insect consumers. Most allergens from edible insects are proteins such as hyaluronidase, microtubulin, phospholipase A, arginine kinase and proto-myosin. However, the allergens in edible insects can be mitigated via various methods (heat treatment, fermentation, hydrolysis) to reduce their allergenicity. While many microorganisms specific to edible insects may not pose direct safety concerns for humans, they can still transmit diseases that are potentially harmful. Edible insects harbor numerous pathogenic bacteria that can impact human health, whereby the most important are *Staphylococcus aureus, Clostridium, Bacillus cereus*, *Vibrio* and *Streptococcus*. Additionally, viruses present a notable safety concern for consumers, with over 70 viruses have been detected in edible insects, 36 of which can cause infections in humans. Furthermore, certain parasites found in edible insects, such as *Dicrocoelium dendriticum*, *Entamoeba histolytica*, *Giardia lamblia* and *Toxoplasma* spp., can also pose risks to human health upon consumption. Pesticide residues and heavy metals pose safety concern for wild-caught insects. Most commonly found heavy metals in edible insects are cadmium, mercury, arsenic and lead. Other significant chemical hazards in edible insects include dioxins, residues of veterinary drugs, polychlorinated biphenyls, mycotoxins, plant toxins and polyaromatic hydrocarbons, all of which can pose important safety risks. In conclusion, the primary safety concerns associated with both farmed and wild-caught insects are allergic reactions and pathogenic microorganisms. However, pesticide residues and heavy metals represent particularly significant safety issues in wild-caught insects. Therefore, from the food safety perspective, it is strongly recommended that only edible insects sourced from controlled farm conditions should be consumed.

Keywords: edible insects, pathogenic microorganisms, allergic reactions, heavy metals, pesticides

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DEFINING ENZYMATIC HYDROLYSIS OF BANANA PEELS WITH COMBINATIONS OF ENZYMES (CELLULASE, VISCOZYME LAND PECTINASE)

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The global consumption of bananas (*Musa* spp., Musaceae family) generates significant quantities of banana peels (BP), constituting 30-40% of the fruit's weight, which are typically discarded as waste. However, there is a growing interest in using BP for eco-friendly biotechnological processes. To use BP effectively, pretreatment is crucial to enhance enzyme activity in the hydrolysis of complex carbohydrates. This study is the second stage of BP pre-treatment screening, and it aims to determine the best pretreatment and enzymatic hydrolysis (EH) conditions with different enzymes to produce a fermentation substrate suitable for producing lactic acid. Commercial enzymes were used for the assays, namely cellulase cocktail, viscozyme L and pectinase. A diluted-acid pretreatment was used with 0.25% H_2SO_4 in an autoclave for 10 min at 121 ºC. The pretreated material was subjected to EH tests in which two variables were evaluated for their influence on hydrolysis yield: solid load (% dw/v), and enzyme combinations. The first step was to characterize the biomass. BP showed 11.42±0.09% cellulose, 5.9±0.3% hemicellulose, 17.2±0.5% total lignin and 44.8±0.2% extractives. After this stage, tests were performed on the EH, namely sugars concentration, with results showing that 20% BP (86±12 g/L) yielded more heterogeneous outcomes compared to 15% BP (64±9 g/L). This can be attributed to the higher solid loads and reduced medium mobility. Sugar concentrations increased from 0 h (15%: 66.7±9 g/L; 20%: 53±1 g/L) to 12 h (15%: 91±8 g/L; 20%: 71±4 g/L) and remained constant between 12 h and 24 h, with no significant differences in either group (15%: $p = 0.705$; 20%: $p = 0.664$). Hydrolysis with visccozyme L demonstrated superior performance in terms of concentration and biomass fluidity throughout the process. Practically, at both solid loads, the medium with viscoszyme L was always more liquid than the others. Statistically, cellulase plus viscoszyme L showed a significant difference ($p = 0.037$) compared to the use of cellulase alone. Thus, viscozyme L together with cellulase were chosen to produce the fermentation BP medium for the next stages of research.

Keywords: by-products; sugars; acid pre-treatment; solid load; fermentation medium

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BIOACTIVE COMPOUNDS AND THEIR USE TO PRODUCE ACTIVE EDIBLE COATINGS AS FOOD PACKAGING MATERIALS

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Currently, many foods are wasted because they are highly perishable or have not been properly packaged, and when they reach the final consumer, they do not meet minimum quality requisites. To overcome this problem, the food and packaging industries have developed several active packaging systems to increase the food's shelf life. One of the systems developed to control food spoilage and increase its shelf life are edible active coatings. Edible coatings are thin, protective, edible, and biodegradable layers deposited on a food's surface. The edible coatings are an environmentally friendly technology produced with biodegradable polymers (polysaccharides, proteins, lipids, and composites) and, can also enhance food safety, nutritional and sensory attributes by adding bioactive compounds (antioxidants, antimicrobials, or specific nutrients) to the polymeric matrix. Different studies have shown the effectiveness of edible active coatings in different foods products, particularly in fruits and vegetables. The edible active coatings can control moisture transfer, gas exchange, microbial growth, oxidation processes and other chemical reactions. Within presentation the current state of knowledge on edible active coatings, discussing their different applications as carriers of active compounds, such as antimicrobials or antioxidants, to improve quality and extend food's shelf life will be presented.

Keywords: active packaging, edible coatings, shelf life, antioxidant, antimicrobial, bioactive compounds

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CHEMICAL PROFILING AND ANTIBACTERIALACTIVITY OF *HERICIUM ERINACEUS* EXTRACTS

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Hericium erinaceus (Bull.) Persoon (fam. Hericiaceae) or Lion's mane, widely used in traditional Chinese medicine, represents one of the most promising mushrooms with many biological activities, such as neuroprotective, neuroregenerative and antitumor. The main focus of this study was to investigate the antibacterial potential of four extracts obtained from *H. erinaceus* fruiting bodies using ultrasound-assisted extraction with water, ethanol, ethyl acetate and dichloromethane/methanol 2:1 (v/v) mixture. The ^{1}H NMR profiles revealed the metabolite fingerprint of all four extracts. The highest amount of phenolic compounds, such as caffeic and ferulic acids, was detected in ethyl acetate extract, comparing the total ionic current of the detected compounds between all four extracts obtained by HPLC-Orbitrap-MS/MS. Antibacterial activity of the extracts was evaluated by microdilution method against the following pathogenic bacteria: *Pseudomonas aeruginosa*, resistant *P. aeruginosa*, *Escherichia coli*, *Staphylococcus lugdunensis*, *S. aureus* and methicillin-resistant *S. aureus* (MRSA). Compared to the other two, dichloromethane/methanol and ethyl acetate extracts possessed higher antibacterial activity against *P. aeruginosa* and resistant *P. aeruginosa*, with MIC and MBC values of 2 mg/mL and 4 mg/mL for *P. aeruginosa* and 3 mg/mL and 4 mg/mL for resistant *P. aeruginosa*. In case of *E. coli*, ethyl acetate extract had a MIC value of 3 mg/mL, while all extracts had a MBC value of 8 mg/mL. The obtained MIC values of 2 mg/mL for *S. lugdunensis* and 3 mg/mL for *S. aureus* and MRSA and MBC value of 4 mg/mL for *S. lugdunensis*, *S. aureus* and MRSA indicate that ethyl acetate extract was empirically validated as the most appropriate, among tested extracts, regarding the obtained antibacterial potential, and should be considered for further research. The obtained results emphasize the medicinal potential of *H. erinaceus different extracts,* especially considering the worldwide trend towards seeking supplements of natural origin.

Keywords: Hericium erinaceus, fruiting bodies extracts, antibacterial activity, NMR spectroscopy, LC-MS/MS

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RESVERATROL EFFECTS ON HUMAN DIABETIC BLOOD VESSELS

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Polyphenols are secondary phytometabolites, chemically characterized by a series of phenolic structural units. One of the most studied natural polyphenol is resveratrol (RSV). The studies imply that RSV has broad range of properties, such as antiinflammatory, antioxidant, antithrombogenic, antiproliferative, antiaging, antimicrobial, neuroprotective, cardioprotective, estrogenic and antidiabetic.

It is known that diabetes mellitus increases contractility of blood vessels, impairs endothelial function, which may contribute to cardiovascular (CV) and metabolic complications. Therefore, proven efficacy of RSV in the relaxation and prevention of contraction of blood vessels in animal and human tissue may highlight RSV as potential new agent for the treatment of CV diseases (CVD). In humans with insulin resistance, RSV improves glycemic control. In addition, hypoglycemic effect of RSV has been confirmed in both types of diabetes mellitus (type 1 and 2). Also, RSV stimulates glucose uptake by increasing GLUT4 (glucose transporter type 4) expression in the plasma membrane of peripheral tissues. It has an important role in protecting endothelial cells, smooth muscle cells, and other important cell types in blood vessels. The results of clinical trial showed that treatment with RSV reduced secretion of pro-inflammatory cytokines, like VCAM-1 (vascular cell adhesion molecule 1), ICAM-1 (intercellular adhesion molecule 1), IL-8 (interleukin 8), which could be protective mechanism for CVD complications caused by diabetes. Additionally, RSV has been shown to improve mitochondrial function.

Potential of the use of RSV and mechanisms of its action has also been investigated in the cardiosurgery. Nowadays we know that vasodilation of diabetic blood vessels commonly used as bypass grafts (human saphenous vein and internal mammary artery) by RSV is partly produced through potassium (K) channels in vascular smooth muscle cells.

However, there are several properties of RSV that limits its full bioactivity (antioxidant capacity, low bioavailability and unfavourable pharmacokinetics), which need to be addressed and solved in the future. Although literature offers technical solutions for overcoming these issues, further studies should be done highlighting the proportion of patients with diabetes (type 1 and 2) and CV complications, which are candidates for bypass surgery.

Keywords: polyphenols, resveratrol, diabetes mellitus, blood vessels, potassium channels

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EFFECT OF DIFFERENT SUGAR TYPES ON HMF CONTENT AND COLOR OF WHOLEGRAIN WHEAT-BASED COOKIES

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The Maillard reaction and caramelization are chemical processes that induce the formation of chemical compounds responsible for the desired color and taste of thermally treated food, but also to the formation of potentially hazardous compounds. Among them, acrylamide and 5-hydroxymethylfurfural (HMF) are the most studied. HMF is formed by dehydration of hexose sugars or the Maillard reaction during thermal treatment and has been identified as a contaminant with high concentrations that pose a health risk due to its biotransformation into 5-sulfoxymethylfurfural, a highly genotoxic and mutagenic metabolite. The objective of this study was to evaluate the impact of different sugar types on the formation of HMF and color in wholegrain wheat-based cookies. The cookies were made using AACC method with some modification. The recipes differed in the type of added sugar, while the total amount of sugar was kept constant. Sugars (sucrose, fructose, dextrose, brown cane sugar and coconut sugar), sweeteners (agave syrup and stevia), sugar alcohols (erythritol and xylitol) and natural sweeteners (acacia honey) were used. Baking was performed at 180°C for 7, 10 and 13 minutes in the Memmert UF55 oven. Pure sucrose and dextrose had a lower impact on HMF formation in cookies, as well as coconut sugar and brown cane sugar with 82 and 50% of sucrose in the total sugars. Fructose was the most reactive. After 13 min of baking, the content of HMF in cookies with the addition of pure fructose, agave syrup (rich in fructose) and honey (glucose:fructose=1:4) was 103.2, 42.1 and 104.5 µg/g, respectively. No HMF content was detected in cookies with addition of alcoholic sugars and stevia whose sweetness comes from steviol glycosides. The lightest was the cookies with xylitol, while the darkest was the coconut sugar cookies. Correlation analysis shows high negative correlations (>0.97) between the L values of the color and the content of HMF in cookies.

Keywords: wheat-based cookies, HMF, color, Maillard reaction

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HORIZON 2020 IMPTOX PERSPECTIVE

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Micro- and nanoplastics (MNPs) are pervasive environmental pollutants found in marine water, freshwater, agroecosystems, atmosphere, food, drinking water, and biota. Despite their widespread presence, comprehensive data on MNPs prevalence and impact remain limited due to analytical challenges. Beyond their physical presence, MNPs carry adsorbed contaminants such as organic pollutants, heavy metals, and microbes, raising concerns about their potential health risks.
The European Union's Horizon 2020 Imptox project – "An innovative analytical platform to investigate the effect and toxicity of micro and nanoplastics combined with environmental contaminants on the risk of allergic disease in preclinical and clinical studies" - under the European Research Cluster to understand the health impacts of micro and nanoplastics (CUSP), consists of consortium of 12 partners from 8 European countries, and addresses these concerns by focusing on the indirect hazards of MNPs, specifically through adsorbed contaminants like allergenic proteins, pathogenic bacteria, and combined hazards. Allergies, affecting over 150 million Europeans, are on the rise, with predictions indicating that half the EU population may be affected by 2025. Understanding whether environmental MNPs contribute to this increase is critical. Project key objectives include the production and characterization of high-quality in-house MNPs, development of protocols for biological material digestion, innovative labeling for NPs tracking, and the conduct of large-scale environmental studies and a clinical trial on pediatric exposure assessment. Imptox aims to improve our understanding of MNPs exposure through ingestion and inhalation, provide risk assessment scenarios, inform policy, and enhance communication strategies between scientists and stakeholders. By focusing on vulnerable populations and fostering interdisciplinary collaboration, Imptox will contribute to societal, health, and environmental advancements.

Keywords: microplastics, proteomics, allergies, pollution, health

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EFFECTS OF NOVEL LIGHT-BASED TECHNOLOGIES ON THE INACTIVATION OF *ESCHERICHIA COLI* AND THE QUALITY CHARACTERISTICS OF SPINACH

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In recent years, there has been an increasing trend towards the consumption of fresh or minimally processed foods. Researchers are therefore investigating non-thermal technologies for surface decontamination of fresh fruit and vegetables as an alternative to conventional methods. Pulsed light (PL) and ultraviolet light (UV-C) are two widely used techniques to effectively decontaminate fresh produce and food contact surfaces in food processing lines. The Centers for Disease Control (CDC) and the Food and Drug Administration (FDA) have identified *Escherichia coli* O157:H7 as one of the major microorganisms that can spoil leafy vegetables and pose a risk to human health. To address this issue, this study investigates the effects of light-based techniques on fresh spinach decontamination and quality changes. The study also evaluates the effects of single and combined applications and their sequencing to determine the efficiency of microbial inactivation. Spinach was inoculated with *Escherichia coli* ATCC 26 by spot inoculation and surface decontamination was performed by a combination of UV-C and PL technologies. The process parameters were optimized considering color and inactivation rate. The distance between the samples and the light source was set at 12 cm, and the fluence of 12 J/cm^2 was determined to be optimal. Quality characteristics evaluated included total soluble solids, pH, moisture, water activity, titratable acidity, total phenolic compounds, total chlorophyll content, color, texture, and microbiological analysis.The combined PL and UV-C treatment resulted in a reduction of *E. coli* by 2 log cycles with no significant effect on pH, acidity, water activity, moisture and texture compared to the control group. This study highlights the effectiveness of the combined treatment of spinach with UV-C and PL, which can be used as an alternative to conventional washing methods to achieve microbial inactivation while maintaining the quality of the spinach.

Keywords: light-based technologies, surface decontamination Escherichia.coli, spinach, quality

RAMAN SPECTROSCOPY IN FOOD ADULTERATION: CHALENGES AND PROSPECTS

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Raman spectroscopy has become an important analytical tool in the detection and prevention of food adulteration as it provides a rapid, non-destructive and highly sensitive method for analyzing the molecular composition of various food products. The applications of Raman spectroscopy in food fraud analysis are diverse and include detection of adulteration, authentication of food origin and verification of labelling claims. In dairy products, Raman spectroscopy has been used to detect the addition of non-milk fats and proteins. In honey, it can detect the presence of sugar syrups and other non-natural additives. The technique is also suitable for determining the geographical origin of olive oil and wine, distinguishing between genuine and counterfeit alcoholic beverages and ensuring the authenticity of high-quality spices such as saffron and turmeric. The authenticity of essential plant oils and the purity of seeds can also be verified using Raman spectroscopy. Despite its advantages, the application of Raman spectroscopy in food adulteration faces some challenges. A major obstacle is fluorescence interference, which often obscures the Raman signal and makes analysis difficult. The complexity of interpreting spectra from heterogeneous food matrices can also be an issue. Furthermore, the high cost and need for specialized equipment and expertise limit accessibility, especially in resource-poor areas. The prospects for overcoming these challenges are promising. Advances in instrumentation, such as time-gated Raman spectroscopy, surface-enhanced Raman spectroscopy (SERS), Spatially Offset Raman Spectroscopy (SORS), Tip-Enhanced Raman Spectroscopy (TERS) and hyperspectral imaging are being developed to reduce fluorescence interference and improve signal clarity. The transferability of chemometric models, which is crucial for the broad application of Raman spectroscopy, is another challenge. Variability between different instruments, even of the same model, can lead to inconsistencies in spectral data, making it difficult to create robust and universal predictive models. In addition, differences in sample presentation, environmental conditions and operator handling can further affect the reproducibility and reliability of results in different environments. In summary, while Raman spectroscopy faces significant challenges, its prospects in the field of food adulteration detection are solid. With continued progress, this technique will become an indispensable tool for ensuring food authenticity and safety, ultimately protecting consumer health and preserving market integrity.

Keywords: SERS, transfer model, chemometrics

Acknowledgements: This work was supported by Science Fund of the Republic of Serbia, #GRANT No 7750160, R-SPECT.

EFFECT OF LEMON BALM HYDROSOL ON TWO ISOLATED DISTAL SECTIONS OF THE SMALL INTESTINE OF RATS

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Melissa officinalis has been used for centuries as an herbal remedy because of its antibacterial, antiviral, antispasmodic and sedative properties. According to the FDA list, it is generally considered safe for oral use. The Council of Europe has recommended that a leaf of this plant can be used to flavour food, as it is already listed in the N2 category. In addition, it can be used to flavour tea, beer or wine. Compared to the corresponding essential oils, hydrosol, aromatic liquids produced during the hydrodistillation of essential oils, have the potential to be used as medicinal beverages due to their mild activity.

The aim of this study was to investigate the effect of different volumes (10-500µl) of hydrosol obtained by hydrodistillation of the aerial part of *Melissa officinalis* L. (Lamiaceae) on the spontaneous contractions in the isolated rat jejunum and ileum in a tissue bath *in vitro*. The obtained results demonstrated that the applied hydrosol does not alter contraction patterns of the isolated segments of small intestine since they produced ca. 10% decrease in contraction intensity of both segments.

Having in mind the demonstrated effect of the lemon balm hydrosol on two isolated distal parts of the small intestine (jejunum and ileum) of rats and the previous ones published concerning gastric fundus one can think of this hydrosol as safe for usage. Research into hydrosols may lead to the development of functional and non-alcoholic beverages as a safe way of using essential oils or even the development of new components or therapeutic agents.

Keywords: ileum, jejunum, hydrolate, Melissa officinalis, contractility

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ESSENTIAL OILAND HYDROSOL FOR THE PRESERVATION OF FRESH PRODUCE: QUALITYAND SAFETY ISSUES

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Postharvest losses of fruit and vegetables are considerable, with chemical applications to be of great consumer concerns regarding food safety. Alternative sanitizers are under investigation, with natural compounds such as essential oils (EOs) and hydrosol to achieve scientific and consumer's interest for the preservation of fresh produce. *Origanum dubium* EO and hydrosol (at different concentrations and time of dipping application) examined for the preservation of tomato and cucumber fruits quality, and their effectiveness as sanitizing agents against two foodborne pathogens (*Listeria monocytogenes* and *Salmonella enterica*). The results indicated that increased concentrations of EO combined with longer time of application resulted into less marketable fruit compared to the hydrosol application. Interestingly, EO application at lower concentrations and shorter time of application (i.e., 0.01%-5 min) increased fruit antioxidants, ascorbic acid and carotenoids levels (for tomato fruit), suggesting the increase of the nutritional value of the treated fruit, as to the control. The EO and hydrosol application was able to decrease the bacterial populations (both bacteria) on fruit. In a second study, tomato fruits at breaker and red ripening stage were exposed to sage (*Salvia fruticosa*) EO (50 μL/L or 500 μL/L) for 2, 7 and 14 days, at 11 ^{o}C. Quality-related attributes were examined during (sustain effect-SE) and following (vapour-induced memory effect-ME; 7 days vapours + 7 days storage) vapour treatment. In breaker tomatoes, EO-enrichment (sustained effect) retained fruit firmness, respiration rates and ethylene emission in low EO levels (50 μL/L), while fruit metabolism was speed up in high EO levels of 500 μL/L, with decreased firmness and increased rates of respiration and ethylene and affecting the antioxidant metabolisms. The effects were more pronounced during the storage period of 14 days, comparing to the fruit exposed to common storage-transit practice. In red fruits, the EOs impacts were evidenced earlier, at 2 and 7 days of storage with increased rates of respiration and ethylene, increased *β*-carotene and decreased lycopene content. Considering the pre-exposed fruits to EOs, quality attributes were more affected in green fruits and to a less level of the red fruits. Overall, the results suggest that EO and Hydrosol may be considered as an alternative food preservative treatment, significantly reducing or eliminating decay/pathogens infection during fresh produce storage.

Keywords: vegetables, natural products, quality-related attributes, volatiles, foodborne pathogens

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ADDRESSING HUMAN NUTRITION AND AGRICULTURAL SUSTAINABILITY: THE DIVERSICROP INITIATIVE

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In response to the pressing need for sustainably produced nutritious food in Europe, the DIVERSICROP project focuses on underutilized crops such as rye and legumes. These crops, known for their resilience to stress, possess the potential to enhance diets and contribute to agricultural sustainability. Leveraging an innovative, cross-sectoral, and multidisciplinary approach, DIVERSICROP aims to analyse the historical cultivation, genetic diversity, and adaptation of these crops to climate change. Central to the project's objectives is the nutrition working group, tasked with understanding consumption patterns and nutritional contributions across Europe. Through a comprehensive bottom-up approach, the group will conduct literature reviews to assess the nutritional value, crop quality, and health benefits of target crops. Furthermore, databases will be examined to analyse trends in production, supply, and consumption, utilizing resources such as FAO food balance sheets and EFSA databases. Nutritional case studies will be undertaken to explore the consumption of foods containing the target crops and their representation in food-based dietary guidelines. These activities aim to inform strategies for sustainable cultivation and dietary guidance, aligning with the Farm to Fork and Biodiversity strategies under the European Green Deal and contributing to the UN Sustainable Development Goals. Overall, the DIVERSICROP initiative underscores the importance of interdisciplinary collaboration in addressing agricultural sustainability and human nutrition challenges. DIVERSICROP seeks to leverage the rich historical background of these crops in Europe, in conjunction with modern farming techniques, nutritional research, and policy data. This holistic approach aims to shed light on the reasons behind the underutilization of these crops and offer valuable insights to guide future strategies and choices. By adopting this comprehensive methodology, the project attempts to rejuvenate underutilized crops, encourage diversified crop production, and enhance nutritional outcomes. In doing so, it aims to strengthen the resilience and sustainability of European food systems.

Keywords: underutilized plants, health, nutrition, sustainability, food security

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ZINGIBER OFFICINALE ROSCOE AND BRYONIA DIOICA JACQ: PROMISING ANTI-INFLAMMATORY TUBERS

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The discovery of new, efficient, and safer anti-inflammatory agents is crucial to prevent and treat inflammatory processes. Natural matrices, such as tubers, have been explored and described as having a huge diversity of bioactive molecules with anti-inflammatory capacity and few undesirable effects. Thus, in the present work, the hydroethanolic extracts of two tubers, *Zingiber officinale* Roscoe (Ginger) and *Bryonia dioica* Jacq. were studied as potential anti-inflammatory agents.The main objective of this work was to validate the anti-inflammatory activity of these tubers through two anti-inflammatory assays *in vitro*: i) inhibition of the production of nitric oxide (NO) in a cell model, RAW 264.7 - stimulated rat macrophage cell line by lipopolysaccharide (LPS); ii) inhibition of the activity of the inducible nitric oxide synthase (iNOS) enzyme. To ensure the safe use of these extracts, cytotoxic activity in tumor and non-tumor lines and cellular antioxidant activity were evaluated. Cell cycle analysis and apoptosis were also assessed using flow cytometry techniques. In addition, these extracts were characterized in terms of organic acids and phenolic compounds through chromatographic techniques to establish a structure-activity relationship. According to the results obtained, the two tubers revealed the presence of organic acids, well-described bioactive molecules, and four phenolic compounds were also identified in *Z. officinale*. Both samples are promising anti-inflammatory agents, since they exhibited the ability to inhibit NO production with very low IC_{50} values. It should be noted that *B. dioica* showed an anti-inflammatory and cytotoxic activity more effective than the standards used. The results of antioxidant activity were less promising, as both samples were less effective than quercetin. The cell cycle results show that *B. dioica* suppresses the G0/G1 phase. At the same time, apoptotic analysis indicates that *Z. officinale* has a greater capacity to induce cell death by the apoptotic pathway. These results are very promising and highlight the anti-inflammatory potential of these tubers, however further tests are needed to evaluate other inflammatory mediators and confirm *in vivo* their efficacy and safety.

Keywords: anti-inflammatory agents Natural matrices, Z. officinale, B. dioica

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3D FOOD PRINTING: RECENT DEVELOPMENTS IN THE APPLICATION OF NOVEL FOOD PROCESSING TECHNIQUES

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In recent years, three-dimensional (3D) food printing has been intensively researched in the food industry. 3D printing offers some advantages over conventional processing techniques as it is used in some specific food areas such as the confectionery market, military and space food, personalised food and food for the elderly. With 3D printing technology, we can create customised foods with complex shapes, geometries, textures and nutritional content. This article provides a comprehensive overview of the applications of the various 3D printing techniques for food. There are currently four 3D printing techniques: extrusion–based printing, inkjet printing, selective sintering and binder jetting. The first and second are two of the most popular techniques in the application of 3D printing in food production. The equipment for extrusion-based printing is simple and this technique offers a wider range of materials. Its limitations lie in the inability to produce complex food designs and the difficulty of holding 3D structures in post–processing. The advantages of inkjet printing are fast production, better print quality and a wide range of materials. With this technique, it is possible to print simple food designs that only have a surface filling or image decoration. The main limitations of the other two techniques are the use of limited materials and the production of less nutritious products. Extrusion-based printing technology is the most suitable for food applications. The influence of material pre-treatment methods and post–processing techniques is very important. The quality of the printed food depends on the material properties (thermal properties and rheology), as the composition of the food and its physical properties are very complex. Researchers have made significant progress in the study of printable materials, but many factors that affect the accuracy and quality of 3D food printing pose major challenges for researchers.

Keywords: 3D printing, food printing, printing techniques

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IMPACT OF MICROPLASTICS ORIGINATING FROM FORMULA PREPARATION ON PROTEIN DIGESTION IN INFANT DIGESTION MODELS

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Microplastics (MPs) of various morphologies have been ubiquitously detected in the environment, food, drinking water, and biota and may pose a threat to food safety and human health. Interestingly, the highest reported concentration of MPs comes from the processing of foods in plastic packaging. MPs exposure to humans is more prevalent in infants than in any other age group due to the use of polypropylene (PP)-based products in formula preparation. Although the effect of MPs on human health is still controversial, multiple studies conducted using *in vivo* and *in vitro* animal models have suggested that MPs can lead to a variety of health problems for humans, including gastrointestinal disorders, obesity, inflammation, cardiovascular disease, disruption of the endocrine system, and damage to vital organs including the liver and spleen. In addition, MPs could disrupt the digestion and bioavailability of important nutrients such as proteins, affecting the proper functioning of the human body, or triggering chronic health diseases and allergic reactions. Meanwhile, cow's milk forms an essential part of the diet of infants as a source of protein and other nutrients and a primary component of infant formula. MPs have been found in milk products, including prepared infant formula. Despite the overwhelming evidence of the presence of MPs in infant foods, the literature remains deficient in information relating to the impact of MPs on the digestion and absorption of proteins in infants. This research gap gave birth to the European Union's Horizon Europe-funded Microprot project. The broad aim of the Microprot project is to investigate the impact of polypropylene-based MPs from plastic packaging material on the digestibility of proteins in adults and infants. Therefore, the occurrence of MPs in various foods and their potential effect on health will be presented, highlighting the approach and impact of the Microprot project.

Keywords: microplastics, plastic packaging, health, infant formula, food, and beverages

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POSTER PRESENTATIONS WITHIN SECTIONS

Section
EDUCATION, INNOVATION AND TRANSFER OF KNOWLEDGE

EVALUATING SENSORY ASPECTS OF INNOVATIVE FOOD PRODUCTS INCORPORATING UNDERUTILIZED INGREDIENTS

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Introduction: Since the 1900s, less than 25% of the plant genetic diversity has been used in everyday nutrition. As a result, thousands of cultivated and wild food crops are no longer used despite their high nutritional value. In the frame of BioValue project (GA 101000499) innovative food products containing non and underutilised food species (*Lens culinaris*, *Lathyrus* spp., *Fagopyron* spp., Tomato ideotypes, Eggplant landraces, *Cucumis melo* var. *flexuosus* and *Sonchus oleraceus*) were created in order to bring biodiversity back to the plate in a way consumers desire. **Methods:** Prototypes of food products were prepared in the experimental kitchen. As vital in developing new food products, sensory evaluation is conducted focusing on satisfaction, sensory suitability, and consumer acceptance. Nine created products were evaluated in Serbia and Greece during May 2023. Each product was blind tasted, rated and ranked according to its organoleptic quality by professionals and lay representatives. Based on the feedback, product recipes were adapted to offer their healthier versions in alignment with Nutri-Scores. These adaptations ensured the promotion of agrobiodiversity, while maintaining an enticing and satisfying taste, texture and mouthfeel. **Results:** The results of the sensory evaluation of all samples (Chickpea and Grass Pea Flour Crackers, Buckwheat Pretzels, Gingerbread Cookies, Red Lentil Brownie, Lentil Paté, Buckwheat Pasta, Lentil Pie, Vegan Burger and Eggplant Salad), conducted with the merged sample of both Greek and Serbian participants has shown that the Red Lentil Brownie received the highest scores in all measured attributes. On the other hand, Chickpea and Grass Pea Crackers, received the lowest scores in almost all attributes. **Conclusion:** By conducting sensory evaluations and consumer surveys improved understanding was attained to align with consumer expectations. Gathering feedback from consumers was useful for refining the recipes and ensuring that innovative food products meet their expectations in terms of taste, texture, aroma, and overall sensory experience.

Keywords: non and underutilized food, sensory evaluation, innovative food product

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Section
FOOD NUTRITION AND HEALTH

THYMUS SERPHYLLUM AND *TH. VULGARIS* ESSENTIAL OIL AND HYDROLATE AS A POTENT ANTIOXIDANTS AND *A*-GLUCOSIDASE INHIBITORS

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Thymus serphyllum L. (wild thyme) and *Th. vulgaris* (common thyme) are well-known medicinal and culinary herbs with a significant amount of essential oil, rich in bioactive phenolic terpenes. Both herbs are consumed as tea and syrup, especially for their antimicrobial and anti-inflammatory properties. The aim of this study was to investigate the chemical composition, antioxidant and antidiabetic effects of essential oils (EO) and hydrolates (HYD) of two *Thymus* species grown in the fields of the Institute for Medicinal Plants Research "Dr. Josif Pančić", Pančevo, Serbia. The EO and HYD were obtained by hydrodistillation and subjected to analysis using GC-FID and GC-MS techniques. Wild thyme contained mainly bornyl acetate (almost 50% of EO) and thymol (even 98% of HYD). Common thyme was characterized by high amounts of thymol (over 50% and 90%, respectively). The results of the Folin-Cioacalteu method show that the EO of both species have a total phenolic content of over 350 mg GAE/g, while the HYD displayed values of approximately half of that. The total reduction power test results show that the EO of both species has dose-dependent antioxidant activity, reaching values of about 170 µM AAE/g at the highest concentration applied, while the values for HYD are about 125 µM AAE/g. The EO of both species inhibited α-glucosidase in a dose-dependent manner and achieved an inhibition of about 80% at the highest applied concentration. The inhibition of α-glucosidase by the HYD of wild and common thyme (87.39 and 83.44%, respectively) was similar to that of the positive control acarbose (87.74% at 1 mg/mL). These findings underscore the significant presence of phenolic compounds with potent antioxidant and antidiabetic properties in EO and HYD of both thyme species, thereby highlighting their potential applications in the food and pharmaceutical industries.

Keywords: Thymus, essential oil, hydrolate, antioxidants, antidiabetic

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Vaccinium myrtillus LEAF EXTRACT-LOADED LIPOSOMES: THE INFLUENCE OF UV IRRADIATION

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Due to the present phytochemicals (phenolic acids, flavonoids, and tannins), the extracts of *Vaccinium myrtillus* leaf were shown to scavenge free radicals *in vitro*, to enhance glutamate decarboxylase gene expression in dermal fibroblasts, resulting in the stimulation of cell growth and synthesis of hyaluronic acid and glutathione, to inhibit collagenase and elastase, enzymes responsible for the ragging and wrinkled nature of skin, to decrease the melanin content in B16 melanoma cells, and to suppress the release of histamine from mast cells. With the aim to improve the stability and bioavailability of the extract compounds, *V. myrtillus* leaf extract was encapsulated in the liposomes, and the influence of the UV irradiation on the particle size, polydispersity index (PDI), zeta potential, and mobility of obtained liposomes was investigated. The liposomal particles were prepared using the proliposome method and 4 g of phospholipids, 50 mL of *V. myrtillus* ethanol extract, and 20 mL of ultrapure water. Vesicle size, PDI, zeta potential, and mobility were measured by photon correlation spectroscopy before and after UV irradiation. The size and PDI did not change after UV irradiation and amounted to 5508.67±56.58 nm and 0.249±0.047, respectively. On the other hand, the zeta potential (as a parameter of the stability of the system) and mobility possessed low values (absolute values) at the beginning and additionally significantly changed in the UV-treated sample (-3.93±0.10 mV and -0.315±0.016 μmcm/Vs, respectively). The beneficial effects of biologically active *V. myrtillus* leaf phytochemicals on human and animal health, as well as their sensitivity, particularly under UV irradiation, encourage their encapsulation in liposomes. However, future experiments should be focused on the improvement of the stability of *V. myrtillus* extract-loaded liposomes, *i.e.*, increasing the zeta potential value.

Keywords: encapsulation, liposomal particles, Vaccinium myrtillus, UV irradiation

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EFFECT OF BLACK GOJI BERRY EXTRACT ON SPONTANEOUS SMALL BOWEL CONTRACTIONS

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Goji berries are becoming increasingly popular among the exotic berry fruits in many countries, as they can be used in medicine and pharmacy as well as in the food industry. In a variety of foods such as dairy and baked goods, ice cream, jams, sauces, salads and beer goji berries can be found. Although the goji berry has not been consumed in large quantities in Europe in the past, it is not subject to any regulations under EU novel food legislation and there are no legal requirements or restrictions on its use in other foods. Antioxidant, anti-inflammatory, anti-radiation, immunostimulant, anticancer are just some of the biological activities of black goji berries. These advantageous qualities are ascribed to the separate or combined actions of the components of goji berries. Black goji berry, *Lycium ruthenicum* Murray, has the potential to contribute to the development of food additives and functional foods as well as the treatment of diseases based on various pharmacological activities.

We investigated the effect of different concentrations (0.01-100 mg/mL) of goji extract, obtained from fresh black goji berries by percolation with water in a ratio of 1:5, on the intensity of spontaneous contractions in the isolated rat jejunum and ileum. The application of different concentrations of goji water percolate produced no notable change in the tone of the isolated small intestine segments.

The global popularization of the goji berry and goji berry-based products is supported by scientific evidence of their health-promoting effects. The efficacy and safety of black goji berry-based foods must be proven by well-designed clinical studies. In addition, the possible interactions of goji berries with conventional medicines and natural health products need to be further investigated. Here obtained results indicate that the goji water percolate does not alter the contraction patterns of the isolated rat small intestine segments and thus it could be considered safe for utilization.

Keywords: ileum, jejunum, Lycium ruthenicum, spasmolytic activity

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ANTIMICROBIALACTIVITY OF BIOCHANIN AAND ITS EFFECT ON ENZYMES

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Biochanin A is a natural isoflavone most abundant in red clover, chickpeas and soybeans. It has numerous biological activities, and food rich in biochanin A and the other isoflavones can contribute to the reduction of the symptoms of neurodegenerative, cardiovascular and inflammatory diseases.

The aim of this work is to investigate the microbiological activity of biochanin A and its influence on the inhibition of cholinesterase enzymes (acetylcholinesterase and butyrylcholinesterase), tyrosinase, α-amylase, α-glucosidase. Microbiological activity against Gram-positive and Gram-negative bacteria was tested (*Escherichia coli* 8739, *Pseudomonas aeruginosa* 9027, *Salmonella enteritidis* 13076, *Klebsiella pneumoniae* 10031, *Staphylococcus aureus* 25923 and fungi *Candida albicans* 10231, *Aspergillus niger* 16404) using the microdilution method. The inhibitory activities of BHA were tested on the enzymes acetylcholinesterase and butyrylcholinesterase using the Elmanov method, tyrosinase using the dopachrome method, α-amylase using the iodine/potassium iodide method and α-glucosidase using the p-nitrophenyl-α-glucopyranoside method.

The results showed that the minimum inhibitory concentration (MIC) for biochanin A is in the range of 0.84-1.69 mg/cm^3, except in the case of *A. niger* 16404. The most sensitive of Gram-negative bacteria are *E. coli*, *P. aeruginosa* and *S. enteritidis* (MIC is 0.84 mg/cm^3), and from Gram-positive *S. aureus*, whose growth is inhibited at the same MIC. Gram-negative bacteria *K. pneumoniae* and yeast *C. albicans* showed less and the same sensitivity to biochanin A. The minimum concentration of biochanin A at which the growth of these microorganisms was stopped is 1.69 mg/cm^3. Biochanin A did not show activity on acetylcholinesterase and butyrylcholinesterase, while it has an inhibitory effect on α-glucosidase, α-amylase and tyrosinase.

Keywords: biochanin A, antimicrobial activity, enzyme inhibition

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ANTIOXIDANT ACTIVITY OF ISOLATED AND COMMERCIAL LAUREL LEAF ESSENTIAL OILS (*Laurus nobilis* L.)

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Laurel (*Laurus nobilis* L.) is an aromatic evergreen plant from the Lauraceae family. Laurel leaf is widely used as a spice and a flavoring agent in food preparation. It is also used in the pharmaceutical and cosmetic industry, due to its antibacterial, antifungal, antioxidant, antidiabetic and cytotoxic activities. The content of essential oil in the laurel leaf is about 1.5%. The most abundant components in the laurel leaf essential oil are oxidized monoterpenes (1,8-cineole and α-terpinyl-acetate), then monoterpene hydrocarbons and phenylpropanoid compounds. This work aimed to compare antioxidant activity of commercial (Probotanik, RS) and isolated laurel leaf essential oils (Vitamin Horgoš, RS) by applying DPPH and ABTS assays. The laurel essential oil is isolated by using Clevenger type hydrodistillation (hydromodule 1:10, m/V, distillation time 120 min). The yield of the isolated essential oil was 1.77 ± 0.02 cm^3/100 g of plant material. Both essential oils showed good, concentration-dependent antioxidant activity. The isolated laurel leaf essential oil showed stronger antioxidant activity ($EC_{50(DPPH)}$ = 0.391 ± 0.023 mg/cm^3 and $EC_{50(ABTS)}$ = 9.631 ± 0.042 mg/cm^3) than commercial laurel leaf essential oil ($EC_{50(DPPH)}$ = 0.909 ± 0.012 mg/cm^3 and $EC_{50(ABTS)}$ = 20.168 ± 0.173 mg/cm^3). The results indicate that both laurel essential oils are good antioxidant agents and have potential to be applied in industry as a safer alternative to the synthetic antioxidants.

Keywords: laurel, essential oil, DPPH assay, ABTS assay, antioxidant activity

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ANTIMICROBIALACTIVITY OF DIFFERENT HYDROLATES AGAINST *CANDIDAALBICANS*

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In recent years, there has been a growing interest in natural products obtained from the distillation of aromatic plants: essential oils and hydrolates. There are many scientific articles on the efficacy of essential oils in various fields: antimicrobial, immunomodulatory, antioxidant, anti-inflammatory, analgesic, etc., but there is little evidence on the activities of hydrolates. In 2012, the French Pharmacopoeia defined hydrolates as a product obtained by distilling various parts of aromatic plants and separating them from the essential oil during distillation. Previous studies indicate that hydrolates are also suitable for applications in the food industry to inhibit the development of pathological microorganisms in food and to remove biofilms that pose a threat to public health in food, pharmaceuticals and cosmetic products. In order to improve the knowledge about the potential applications of hydrolates, the antimicrobial activity of six hydrolates was tested and compared in this study against the fungal strain *Candida albicans*. Four dilutions of each sample were tested in the microdilution assay and the results were expressed as minimum inhibitory concentration (v/v%). The best results were obtained with thyme hydrolate and Roman chamomile hydrolate (MIC values were 50 (v/v%). Lavender hydrolate showed activity only undiluted (MIC value was 100 (v/v%)), while lemon balm, hyssop and wild carrot showed no activity even undiluted. As hydrolates are considered potential antimicrobial agents, further studies are needed to define which hydrolates to focus on when considering potential antimicrobial agents.

Keywords: microdilution assay, Thyme hydrolate, Roman chamomile hydrolate, minimal inhibitory concentration

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ASSESSMENT OF THE ANTIOXIDANT AND ANTIDIABETIC ACTIVITY OF THE EXTRACTS OF HOUSELEEK LEAVES AND HONEY MIXTURE BASED ON THE PHYTOCHEMICAL PROFILE

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Exploration of the health benefits of plants and different natural products has revealed that they possess diverse biological effects. Houseleek (*Sempervivum tectorum*) leaves and honey are rich in phenolic compounds, which are responsible for their medicinal properties, including antioxidant, antidiabetic, anti-inflammatory, and antimicrobial activities. The combination of houseleek and honey is noted for its various benefits in traditional medicine, yet it has not been extensively studied. Fresh houseleek leaves were mixed with acacia and meadow honey to create a mixture whose extracts were prepared with acidified water (pH 2) over five weeks. The aim of this research was to characterize the phytochemical profile of the mixture extracts and to evaluate their antioxidant and antidiabetic activities using high-performance thin-layer chromatography. Chromatograms were developed using a mobile phase consisting of ethyl acetate-toluene-formic acid-water (16:4:3:2, *v/v/v/v*) and derivatized with Naturstoff reagent, 2,2-Diphenyl-1-picrylhydrazyl radical solution and α-amylase solution to obtain phytochemical, antioxidant and antidiabetic profiles, respectively. The phenolic profile of the honey extract shows light blue zones, while the houseleek leaves extracts contain more polar phenolics of different colours. The mixture extracts display a rich phenolic profile with compounds originating from both components. The presence of neochlorogenic acid, isoquercetin, astragalin, gallic and caffeic acids, kaempferol, aesculetin, and galangin was identified using standards. The antioxidant profile of the mixture extracts prepared from both types of honey is rich in active zones, with aesculetin, caffeic acid and galangin showing the strongest activity. The antidiabetic profile of the mixture extracts shows zones of active compounds with low intensity. Mixture extracts with acacia honey exhibit more active zones compared to the mixture extracts with meadow honey. The assessment of biological activity using high-performance thin-layer chromatography is a rapid and straightforward method for bioprofiling of different complex matrices.

Keywords: Houseleek, Honey, Phytochemical profile, Antioxidant activity, Antidiabetic activity

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FATTY ACIDS COMPOSITION OF KIDNEY AFTER TREATMENT WITH DIFFERENT KINDS OF OIL

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The kidneys are organs that require energy from the metabolism of fatty acids and glucose. Lipid metabolism in the kidneys is important because of the exchange of free fatty acids and apolipoproteins from the peripheral circulation.
The composition of fatty acids (FA) depends on the external intake of FA as well as endogenous synthesis. Literature data showed that there is a tissue specifity concerning activity of desaturase and elongase activity.
Our study aimed to determine the effects of treatment of three different oils linseed oil, palm oil, and high oleic sunflower oil on the phospholipid fatty acid composition of C57BL/6 mice kidneys. Treatment lasted for 4 weeks, under controlled experimental conditions, water ad libitum. Group one (n=10) was control group and mices were fed with standard food for experimental animals, group 2 mices were treted with standard food plus linseed oil (n=10), group 3 were treated standard laboratory food plus palm oil (n=10) and group 4 mices were treated with standard food plus sunflower oil (n=10). Phospholipid fatty acids of mice kidneys were analysed by gas-liquid (GC) chromatography while samples were prepared with standard procedure.
Linseed oil treated group (2) significantly increased kidney linolenic acid (18:3, n-3), eicosapentaenoic acid (20:5, n-3) compared to control and other 3 groups. Consequently, for the competition for the same set of enzymes for desaturation and elongation arachidonic acid (20:4n-6) significantly decreased in kidney related to other treatments. Treatment with palm oil increased percentage of kidney linoleic acid, but treatment with high oleic acid sunflower oil, significantly increased kidney oleic acid compared to control.
Our results confirmed that kidney phospholipids FA composition depends on the type of fat intake and endogenous synthesis as well.

Keywords: fatty acids, kidney, oil treatment, C57BL/6 mi

STRAWBERRY CULTIVAR 'APRIKA' INCREASES INSULIN SENSITIVITY IN HIGH-FAT DIET OBESITY MICE MODEL

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The main culprits in the increasing prevalence of obesity worldwide are considered to be the frequent consumption of energy-dense, processed food, in combination with a sedentary lifestyle. Energy misbalance in the obese state disrupts lipid and glucose metabolism, leading to insulin resistance. Obesity is further associated with health problems, causing cardiovascular diseases and cancer. The new approach to the prevention and management of obesity and associated diseases is the use of functional foods and their bioactive components. Polyphenols as naturally-occurring phytochemicals, can modulate physiological and molecular pathways involved in energy metabolism, by reducing oxidative stress, increasing β-oxidation, etc. 'Aprika' is a new cultivar with relatively high phenolic content compared to other strawberry varieties (*Fragaria x ananassa*, Duch.). Here, we investigated the effect of lyophilized 'Aprika' 5-days-a-week supplementation on pathology induced by the high-fat diet. Male mice C57BL/6J were on a high-fat diet (HFD) for 9 weeks; by the end of the experiment, the HFD mice developed obesity (30% increase in body mass relative to control), followed by insulin resistance and increased liver fat accumulation. The food intake in energy (kcal) did not significantly differ among HFD groups, w/wo strawberries. However, relative to the HFD group, mice on HFD simultaneously supplemented with 'Aprika' had lower body mass. Importantly, strawberry supplementation resulted in increased insulin sensitivity, assessed by intraperitoneal glucose tolerance test and insulin level in serum, as well as a lower liver lipid accumulation, investigated with histological stains Oil Red O and hematoxylin&eosin. The exact mechanisms by which these physiological phenomena occurred are under investigation. Previously, we showed 'Aprika' methanol extract causes activation of the AMPK-dependent mechanism of GLUT4 trafficking *in vitro*, and presumed increased glucose uptake. The in vivo model presented here further supports the hypothesis that introducing 'Aprika' strawberry cultivar into everyday diet can significantly reduce the HFD-obesity-induced pathological changes.

Keywords: high-fat diet, obesity, functional foods, polyphenols, strawberry

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ANTIOXIDANT CAPACITY OF SILIBININ-LOADED LIPOSOMES

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Silibinin is the major active constituent of silymarin, a standardized extract of milk thistle seeds. Silibinin has been used traditionally as a chemopreventive and therapeutic agent in human lung cancer. It also possesses hepatoprotective, antioxidant, hypocholesterolemic, antitumor, cardioprotective, neuroprotective, and antiviral activities. However, its application is limited due to poor water solubility, intestinal resorption, and low bioavailability. Liposomes, as non-toxic, biodegradable, and biocompatible lipid carriers, can provide controlled delivery of bioactive components and their protection from degradation caused by light, oxygen, UV irradiation, different pH values, and enzymes. Additionally, phospholipids from the liposomes do not provoke a reaction with taste receptors, and, therefore, the liposomal bilayer is an appropriate carrier for covering the unpleasant taste of numerous polyphenols. With the aim of investigating the antioxidant potential of silibinin-loaded liposomes after different technological procedures (UV irradiation and lyophilization), two antioxidant assays were employed (ABTS and DPPH tests). Liposomes with silibinin were prepared using the proliposome method and phospholipids. According to the results of the ABTS test, the antioxidant activity of pure silibinin was 0.769 µmol Trolox equivalent (TE)/mL, while the antioxidant potential was lower after the encapsulation in liposomes; 0.548 µmol TE/mL after preparation, 0.549 µmol TE/mL after UV irradiation, and 0.436 µmol TE/mL after lyophilization. Furthermore, the DPPH radical scavenging activity was 20.97% for pure silibinin, 22.48% immediately after the liposomal preparation, 22.24% after UV irradiation, and 18.73% after lyophilization. As can be seen, UV irradiation did not cause significant changes in the overall antioxidant potential of silibinin-loaded liposomes. Nevertheless, lyophilization significantly decreased the radical scavenging activity of the liposomes. Considering that the two used antioxidant assays are based on different principles and reactions, the obtained results provide good insight into the overall antioxidant activity of silibinin-loaded liposomes.

Keywords: antioxidant capacity, encapsulation, liposomes, proliposome method, silibinin

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THE INFLUENCE OF LYOPHILIZATION ON LIPOSOMAL PARTICLES WITH SILYMARIN

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Silymarin is the group of biologically active polyphenols from *Silybum marianum* (milk thistle) that contains silibinin, isosilybin, silydianin, and silychristand. The mentioned components show numerous pharmacological activities promoting human health and well-being, including antioxidant, antimicrobial, anti-inflammatory, antiviral, immunomodulatory, and antitumor effects. Nevertheless, silymarin is quite sensitive to temperature, light, and oxidation and has poor water solubility and low bioavailability. Therefore, their application in food, functional food, dietetic supplements, and pharmaceutics is limited. The encapsulation of silymarin in liposomes represents a technique that can be widely used to strengthen and supplement formulations by enhancing stability and bioavailability and controlling the delivery of the active compound. Lyophilization is a widely employed procedure for drying thermosensitive components to obtain freeze-dried products with active compounds that are stable over a long period, due to the prevention of hydrolytic and oxidative degradation which can occur in water surrounding. Hence, lyophilization can result in significant modifications of the liposomal vesicles, thus its effect should be examined. The liposomes were prepared using 0.5 g of silymarin, 5 g of phospholipids, 10 mL of ethanol, and 40 mL of water in the proliposome procedure. After the preparation, the liposomes were freeze-dried for 24 h. The characterization is performed using photon correlation spectroscopy. Vesicle size and polydispersity index (PDI) of lyophilized silymarin-loaded liposomes were changed from 4080.0±24.0 nm and 0.346±0.044 to 4628.1±45.2 nm 0.426±0.038, respectively. Zeta potential was -20.55±1.34 mV, mobility was -1.55±0.13 μmcm/Vs, and conductivity was 20.15±1.06 μS/cm. In comparison to non-treated liposomes, lyophilization caused an increase in vesicle size and the absolute value of zeta potential, and a decrease in the conductivity value. On the other hand, freeze-drying did not have a significant influence on PDI values and mobility of silymarin-loaded liposomes.

Keywords: lyophilization, silymarin, size, zeta potential

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HARNESSING THE POTENTIAL OF UNDERUTILIZED PLANTS IN PROMOTING HUMAN HEALTH AND NUTRITION

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Introduction: Amidst rising concerns surrounding sedentary lifestyles, obesity, global food insecurity, and malnutrition, there is a growing imperative to promote the consumption of underutilized plants in diets. These plants possess unique nutritional and health-promoting characteristics that could mitigate various diseases prevalent in modern life. **Methods:** The Web of Science, Scopus and PubMed databases were searched using relevant keywords such as 'underutilised,' 'plants,' 'health,' and 'health benefits.' Among the 350 research articles and review papers identified, only the most relevant ones were included, n=112. The selected sources provided appropriate data for a narrative review, with a primary focus on papers published within the last decade. **Results:** The shift to Western diets has led to more non-communicable diseases, emphasizing the need to address persistent nutritional deficiencies despite adequate calorie intake. Underutilized crops show great promise in combating hidden hunger, filling crucial nutritional gaps often overlooked, especially in diets heavy on refined carbohydrates and fats. These plants are rich in essential nutrients and bioactive compounds, such as anti-inflammatory and antioxidant agents, which have beneficial effects on human health. Moreover, their combined effects are anticipated to surpass those of individual constituents, suggesting their potential as holistic dietary interventions. Incorporating underutilized plants into diets has been associated with a reduced risk of major diseases, including diabetes, cardiovascular disease, and various cancers, particularly in industrialized nations. Additionally, their consumption enhances plant and food diversity, thereby fostering overall human health and well-being. Strategic interventions focused on underutilized plants could serve as cost-effective investments in developing secure and healthy food systems on a global scale. **Conclusion:** Identifying barriers to the reintroduction of underutilized plants into the food system is crucial for creating actionable solutions to address nutrition-related challenges. Given their multifaceted contributions to nutritional and food security, underutilized plants should be integrated into broader strategies promoting balanced and healthy diets.

Keywords: underutilized plants, non-communicable diseases, malnutrition, health, nutrition, food security

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HYPOGLYCEMIC POTENTIAL OF CORNELIAN CHERRY

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The latest epidemiological estimation suggests that the worldwide prevalence of metabolic disorders related to diabetes is rising. Among numerous harmful factors, hyperglycemia and oxidative stress are proposed as crucial in the progression of this disease and the development of micro- and macro-vascular complications. Herbal products are recognized as useful therapeutic agents in diabetes mellitus management. Pharmacological research has found that cornelian cherry fruit (*Corni fructus*, *Cornus mas* L., Cornaceae) exhibits a variety of activities such as anti-inflammatory, antioxidative, antidiabetic, anticancer, antimicrobial, and due to these attributes take part in the prevention or treatment of diarrhea, inflammatory bowel disease, kidney disorders and atherosclerosis associated with dysregulated blood glucose and lipid profiles. In line with this evidence, phenolic compounds composition, *in vitro* antioxidant and hypoglycemic activities of dry hydroethanolic extract, prepared from fresh cornelian cherry fruits collected in Montenegro, were analyzed. Valuable total phenolics (17.18 mg GAE/g), total flavonoids (29.69 mg QE/g) and total anthocyanins (2.22 mg CGE/g) content were determined spectrophotometrically. Based on HPLC analysis of the extract, the main secondary metabolites were anthocyanins (cyanidin-3-galactoside 0.29 mg/g), phenolic acids (gallic acid 1.92 mg/g, protocatechuic acid 0.25 mg/g, chlorogenic acid 0.21 mg/g, *p*-hydroxybenzoic acid 0.08 mg/g, ellagic acid 0.14 mg/g) and flavonols (quercetin-3-glucopyranoside 0.28 mg/g, kaempferol-3-glucoside 0.07 mg/g, quercetin 0.02 mg/g). Furthermore, notable antioxidant abilities, evaluated by DPPH (IC_{50} 155.70 μg/mL), ABTS (IC_{50} 53.73 μg/mL) and FRAP (0.82 mmol Fe^{2+}/g) assays, were revealed. The investigated extract exhibited significant inhibition of *α*-amylase (IC_{50} 260 μg/mL) and particularly strong inhibition of *α*-glucosidase (IC_{50} 2.80 μg/mL), gastrointestinal enzymes involved in carbohydrates digestion and control of blood glucose levels. Obtained results of chemical analysis and demonstrated antioxidant and hypoglycemic activities showed that cornelian cherry fruit extract represents a herbal preparation with potential use in diet therapy or phytotherapy for a mild form of diabetes.

Keywords: Cornus mas, phenolics, antioxidant, α-glucosidase, diabetes

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WOUND HEALING POTENTIAL OF BLACK RASPBERRY SEED OIL

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The skin is subject to various influences from the external environment that lead to minor or serious problems and diseases. Among the most prevalent is trauma, an injury characterized by disrupting the normal anatomical structure and function of the skin caused by a cut, a blow or a burn, which consequently makes the body become more susceptible to infection. On the basis of numerous animal experiments and human studies, plant oils are recognized to play an important role in regenerative processes, whether they are used as an integral part of the diet or an active ingredient in pharmaceutical preparation intended for local application. This study aimed to investigate the wound-healing potential of black raspberry (*Rubus occidentalis* L., Rosaceae) seed oil, as well as its chemical characterization and antimicrobial activity against the most common skin pathogens. GC-MS analysis was performed to identify the chemical composition of fatty acids in the black raspberry seed oil and revealed that polyunsaturated fatty acids (PUFA) dominated – the most abundant were linoleic (C18:2n-6; 39.30%) and α-linolenic (C18:3n-3; 30.49%) acids, followed by monounsaturated oleic acid (C18:1n-9; 18.94%). The amount of saturated fatty acids (SFA) was notably lower, including palmitic (C16:0; 2.91%), arachidic (C20:0; 1.37%) and behenic (C22:0; 1.01%) acids as the main representatives. The black raspberry seed oil demonstrated significant antimicrobial activity against the bacterial strains *Salmonella* Typhimurium, *Escherichia coli* and *Bacillus cereus*, in addition to antifungal activity against micromycetes *Aspergillus fumigatus*, *A. niger*, *A. versicolor* and *Trichoderma viride*. After examination of the relative growth rate (%) of HaCaT cells treated with different concentrations of black raspberry seed oil (20–400 µg/mL), by cell viability assay, no cytotoxicity was found (IC_{50} > 401 µg/mL). The wound healing activity, in terms of migratory ability, was evaluated by scratch assay in the same cell line, whereby cell migration into the wounded area was monitored using an Inverted Microscope. The closure of the gap distance (wound closure) was found to be approximately 2-fold higher in cells treated with 200 µg/mL of black raspberry seed oil (41.77%) than in non-treated cells (17.34%). The results showed that black raspberry seed oil is a rich source of PUFA, with promising potential in the treatment of skin wounds associated with infections.

Keywords: Rubus occidentalis, seed, oil, fatty acids, skin wounds

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THE POTENTIAL OF POPPY SEEDS AS AN INGREDIENT IN FUNCTIONAL FOODS

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The poppy plant has great economic value as a source of opium latex, edible seeds and oil. Rich in essential nutrients, such as proteins, dietary fiber, polyunsaturated fatty acids and other bioactive compounds, vitamins and minerals, poppy seeds can contribute to the nutritional profile of food and its health benefits, while providing unique sensory attributes, such as nutty flavour and chrunchy texture. Although poppy seeds have a long tradition of use, especially in the Slavic countries, they were mainly used as a sprinkle or auxiliary ingredient in the formulations of various confectionery and bakery products. The main obstacle to the wider use of poppy seeds was related to their safety due to the possibility of contamination with opium alkaloids such as morphine and codeine during harvesting. However, various seed treatment methods which precede direct consumption, have been shown to be effective in reducing the risk, e.g. washing (40%), grinding (25-34%), heat treatment (90%) or their combination (100%). Furthermore, recently, it has been shown that the use of seed constituents, such as cold pressed oil and residual cake, and their incorporation into novel products can represent a promising avenue of expansion the use of poppy. Notwithstanding the use of poppy seeds is limited and related to legal issues, Serbia has very good conditions for poppy cultivation. In addition, the ongoing legalization of three new domestic varieties of poppy could also contribute to the expansion of poppy cultivation and uses in the diet. The aim of this study was to provide an overview of the nutritional and sensory properties of poppy seeds, as well as their safety in terms of alkaloid residues content in order to indicate new possibilities of using poppy seeds in food production.

Keywords: poppy seed, poppy seed oil, poppy seed cake, opium alkaloids, functional food

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IN VITRO ACETYLHOLINESTERASE INHIBITORY POTENTIAL OF *ROSA* LEAFAND FRUIT EXTRACTS

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Neurodegenerative diseases are gradually growing, irreversible brain illnesses that affect almost every fifth person worldwide. Close to 24 million people suffer from Alzheimer's disease, the most common form of dementia. Natural acetylcholinesterase inhibitors (galantamine and rivastigmine) are used to treat this disease. Studies have shown the synergistic action between the chemical composition of plant extracts and their enzyme-inhibiting potential. Phenolic compounds can affect the active site of enzymes such as acetylcholinesterase by reacting with their amino acid residues. This study aimed to analyze the acetylcholinesterase inhibitory potential of ethanolic extracts of leaves and fruits (hypanthium and achenes) obtained from selected wild-growing *Rosa* species. Fully developed leaves and mature fruits of *Rosa pendulina* L., *R. spinosissima* L., and *R. tomentosa* Sm. were collected on natural sites across Serbia. The extracts were prepared using ultrasound-assisted extraction with 70% ethanol (v/v) as a solvent. The enzyme inhibitory potential was measured by the Ellman method using acetylthiocholine iodide and 5,5'-dithio-bis-2-nitrobenzoic acid (DTNB) as substrate. The results were expressed as IC_{50} value (mg/mL). The tested extracts showed moderate enzyme inhibitory potential compared with galantamine as a reference substance (0.12 mg/mL). The inhibitory activity of leaf extracts ranged from 2.115 mg/mL (*R. pendulina*) to 3.977 mg/mL (*R. spinosissima*). The highest inhibitory potential of hypanthium extracts was observed for *R. pendulina* samples (15.723 mg/mL), whereas the lowest inhibitory activity was noted for *R. spinosissima* (22.407 mg/mL) achenes. The results show that the extracts obtained from *Rosa* leaves have several times higher *in vitro* inhibitory activity of acetylcholinesterase in contrast to the widely used fruit extracts. Since *Rosa* leaves present a rich source of phenolic compounds, further investigation of wild roses is needed.

Keywords: acetylholineesterase, Rosa, leaf, fruit

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WILD FOOD PLANTS USED FOR DIGESTIVE DISEASES IN EAST AND SOUTHEAST SERBIA

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According to the report of the Institute of Public Health in East and Southeast Serbia, the most common diseases that the population suffers from are respiratory, circulatory, musculoskeletal, and genitourinary diseases. The limited capacity of the health care system indicates that herbal medicines could be used more intensively in the treatment of various diseases.
The aim of the ethnobotanical and ethnopharmacological study was to collect the knowledge of the local population in the villages of the Timok and Svriljg region, ten communal villages each. Data on the traditional use of medicinal plants were collected through semi-structured anonymous ethnopharmacological interviews with 161 informants.
The most commonly used preparations were fresh (68.22%, 161 use reports), eat (23.31%, 55 use reports) and fresh juice (7.63%, 18 use reports). The most treated affections among the digestive diseases were: abdominal pain (48.31%, 114 use reports), stomach pain (30.93%, 73 use reports) and constipation (11.86%, 28 use reports). A total of 236 pharmaceutical uses were reported belonging to 25 plants which belong to 9 families, mainly represented by Rosaceae (26.69%, 63 use reports), Asteraceae (21.19%, 50 use reports) and Juglandaceae (19.92%, 47 use reports). The results of this study show that the most commonly used plants were *Artemisia absinthium* (21.18%, 50 use reports), *Juglans regia* (19.92%, 47 use reports), and *Viola tricolor* (11.86%, 28 use reports). The use of plants as food is present in the local population, as our study also showed. As reported by the European Medicines Agency of the most commonly used plants, only *Artemisia absinthium* is therapeutic for gastrointestinal disorders, while the other two are used therapeutically for skin disorders and minor wounds.

Keywords: abdominal pain, Artemisia absinthium, ethnopharmacology, Southeast Serbia

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WILD GROWING PLANTS OF EAST AND SOUTHEAST SERBIA BENEFICIAL FOR CARDIOVASCULAR DISEASES

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Although the treatment of many disorders is guided by current medical practice, ethnopharmacological approaches are being investigated to provide long-term symptom relief. Plants are still widely used because they are inexpensive, easy to use and have fewer side effects than modern pharmaceuticals. According to the World Health Organization, cardiovascular disease is one of the leading causes of death worldwide.

This study was conducted to collect and preserve the ethnopharmacological knowledge of local people about cardiovascular diseases in the ten municipality villages in the Timok and Svrljig regions. Semi-structured anonymous ethnopharmacological interviews with 161 informants were used to collect data on the traditional use of medicinal plants.

Medicinal plants are reportedly prepared in various pharmaceutical forms, such as fresh (48.03%, 61 use reports), boiled (30.71%, 39 use reports), juice (11.03%, 14 use reports), and for eat (10.23%, 13 use reports). The most treated affections among the cardiovascular diseases were: cardiac insufficiency (65.35%, 83 use reports), blood vessels (11.81%, 15 use reports), reduction of blood fat (7.9%, 10 use reports) and anemia (7.09%, 9 use reports). The seven families belong to the 16 plants for the treatment of cardiovascular diseases, mainly represented by Rosaceae (42.52%, 54 use reports), Urticaceae (30.71%, 39 use reports) and Amaryllidaceae (12.59%, 16 use reports). According to the results, the most reported use is that of *Urtica dioica* (30.71%, 39 out of total 127 uses), *Crataegus monogyna* (22.83%, 29 out of 127 use reports) and *Allium ursinum* (12.59%, 16 out of 127 use reports). The European Union herbal monograph on *Crataegi folium cum flore* listed the use for circulatory disorders, but for the radix, leaves, and herba of *U. dioica* do not mention indications for circulatory diseases.

The results show that the use of plants as food and medicine is widespread in east and southeast Serbia, partly due to the fact that health capacities in rural areas are limited.

Keywords: cardiovascular, ethnopharmacology, Svrljig, Timok

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NUTRITIONAL PROPERTIES OF NETTLE (*URTICA DIOICA* L.) SEEDS AND THEIR USE AS A POTENTIAL INGREDIENT IN THE DIET

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Accelerated depletion of plant resources due to population growth requires a greater commitment to research new plant resources that have nutritional and health benefits to meet the growing needs of human society. For a plant to be used as a food raw material, it is necessary to know its composition, which is related to its nutritional value. Food products must have a certain nutritional value to meet certain physiological needs of people. In this regard, this paper determined the chemical composition (carbohydrates, starch, soluble sugars, proteins, oils, fibers, ash, moisture) and the mineral composition (essential minerals, essential trace minerals, and non-essential minerals) of nettle seeds and wheat flour-based products obtained by the application of nettle seeds. The work aimed to determine the nutritional value of nettle seeds and the obtained food products and to evaluate the potential possibility of their application in food and other industries. The results show that nettle seeds are a rich source of proteins and lipids, with a content exceeding 20 g/100g, and this species is expected to provide higher concentrations of essential amino acids and essential fatty acids. Fiber (7.92 g/100g) and ash (13.06 g/100g) content also play a significant part in the overall chemical composition of nettle seeds. The results also show that using nettle seeds in the production of food products based on common cereals could potentially increase the content of protein, ash, fiber and lipids. In addition, the high calcium and iron content indicates that the nettle seeds would be useful in osteoarthritis and osteoporosis and make it recommendable for the treatment of anemia. After the incorporation of nettle seeds into a product based on wheat flour, it was determined that the content of these components increased statistically significantly, indicating that nettle seeds are an adequate incorporative component for the formulation of food products with improved nutritional potential and such products could be used in the diet, especially due to the high content of fibers and minerals such as calcium and iron.

Keywords: nettle seeds, food products, nutritional value, chemical composition, minerals

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A PALLADIUM(II) COUMARIN COMPLEX WITH POTENT *IN VITRO* DNA PROTECTIVE ACTIVITY

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Coumarins are compounds widespread in the entire plant world, found in fruits, vegetables, trees, seeds, and vines and are used in the food industry. They are known for their distinct sweet smell and application as flavouring agents in various products. These compounds have been studied worldwide for their potential health benefits, including anti-inflammatory, antioxidant, antimicrobial and anticancer properties. Research has shown promising results in using these compounds to treat various diseases and improve overall health. Due to the fact that some coumarin derivatives can be harmful to health, it is important to evaluate their effects on DNA. The potential DNA-protective activity of coumarin bis(3-(1-((3-hydroxyphenyl)amino)ethylidene)chroman-2,4-dione-palladium(II)) complex at various concentrations (6.25, 12.5, 25, 50, and 100 μg/mL) against hydroxyl and peroxyl radicals-induced oxidative DNA damage was evaluated by two *in vitro* assays. To assess the level of DNA integrity of the tested compound at the same concentrations the acridine orange assay was performed. The evaluated compound had the same ability to protect DNA against oxidative damage caused by hydroxyl as well as from peroxyl radicals in all tested concentrations. According to the acridine orange assay, the test compound significantly reduced denaturation and degradation of DNA molecules caused by both radicals. These findings suggest that the **tested compound** has great potential as a new DNA protective agent.

Keywords: palladium(II)coumarin derivative, DNA protection, acridine orange, hydroxyl radical, peroxyl radical

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ASSESING THE EFFICACY OF PYRAZOL-CHROMENO[2,3-D]PYRIMIDINE DERIVATIVES AS ANTIOXIDATIVE AGENTS FOR DNA PROTECTION

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Pyrazol-chromeno[2,3-*d*]pyrimidine derivatives, 5-(5-methyl-3-oxo-2,3-dihydro-1*H*-pyrazol-4-yl)-1,5-dihydro-2*H*-chromeno[2,3-d]pyrimidine-2,4(3*H*)-dione (**DS-214**) and 9-methoxy-5-(5-methyl-3-oxo-2,3-dihydro-1*H*-pyrazol-4-yl)-1,5-dihydro-2*H*-chromeno[2,3-*d*]pyrimidine-2,4(3*H*)-dione (**DS-210**), are obtained in the three-component reaction of pyrazolone, barbituric acid, and corresponding salicylaldehydes. The structures of these compounds are confirmed using NMR and IR spectroscopy. The most important structure fragment of these compounds is the chromeno part, which is also present in coumarins. Coumarins have been found in different plant sources such as vegetables, fruit, seeds, spices, nuts, and other diet sources, while the greatest contribution of coumarin intake is certainly through the consumption of cinnamon and cinnamon-containing foods. On the other hand, the pyrimidine part originating from barbituric acid is an important building block of riboflavin (vitamin B_2) which is found in food and sold as a dietary supplement. The protective effects of **DS-210** and **DS-214** against DNA damage induced by hydroxyl and peroxyl radicals were tested *in vitro* at different concentrations (25, 50, 100, 200, and 400 μg/mL). Using the acridine orange assay at the same concentrations, DNA integrity was assessed to distinguish between single-stranded (ssDNA) and double-stranded DNA (dsDNA) by measuring the red-to-green fluorescence ratio (R/GFR). Antioxidative DNA protection against peroxyl and hydroxyl radicals has been confirmed by both evaluated compounds at all tested concentrations. According to the acridine orange assay, **DS-210** induced higher green fluorescence, which indicates protection of DNA from degradation against both radicals. On the other hand, **DS-214** induced slightly higher red fluorescence against peroxyl radical, indicating it has lower DNA protectivity against this type of radical compared to **DS-210**. These results suggest that tested compounds possess potential as protective agents in therapeutic strategies aimed at combating oxidative stress-related DNA damage. The study not only confirms the antioxidant capacity of **DS-210** and **DS-214** but also underscores the utility of acridine orange as a sensitive probe for assessing DNA integrity in oxidative environments.

Keywords: pyrazol-chromeno[2,3-d]pyrimidine derivatives, hydroxyl radical, peroxyl radical, acridine orange, antioxidative DNA protection

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CHRONIC ADMINISTRATION OF CAFFEIC BUT NOT ROSMARINIC ACID DOWNREGULATES HO-1 AND HO-2 IN SPONTANEOUSLY HYPERTENSIVE RATS

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Reactive oxygen species play an important role in the development of hypertension. We found the inverse correlation between vascular resistance and plasma heme oxygenase-1 in response to acute wild thyme (a spice plant, rich in polyphenolic compounds, namely rosmarinic (RA) and caffeic (CA) acids) treatment in spontaneously hypertensive rats SHR. Heme oxygenase-1 (HO-1) is an inducible form of the rate-limiting enzyme in the degradation of heme, and heme oxygenase-2 (HO-2) is a constitutive isoform of this enzyme. Bilirubin (a chain-breaking antioxidant) formed by heme oxidation, acts as a lipid peroxyl radical scavenger, becomes significantly increased after wild thyme treatment, and induces the reduction of plasma lipid peroxidation in SHR. The aim of this study was to evaluate whether chronic application of RA and CA affects HO-1/HO-2 enzymes expression and activity in the kidney of SHR. Adult, male rats were divided into three groups: control SHRC group received vehicle, SHR+RA group received RA (15mg/kg/day) and SHR+CA group received CA (3mg/kg/day), by gavage during 4 weeks period. Kidney tissue homogenates were analyzed for WB HO-1 and HO-2 protein expression, as well as oxidative stress marker TBARS and prooxidant/antioxidant balance (PAB). Plasma bilirubin (BIL) and iron concentrations (markers of HO activity) were also determined. Chronic administration of RA and CA significantly reduced PAB compared to control ($p < 0.05$) even though CA significantly reduced the expression of both isoforms of HO ($p < 0.05$). The kidney TBARS level was reduced only in the SHR+RA group. RA and CA reduced plasma iron levels compared to control ($p < 0.05$, and $p < 0.001$), while in SHR+CA BIL was significantly lower compared to SHR+RA group ($p < 0.05$). Our results indicate that mechanisms different from the HO pathway contribute to the antioxidant effects of RA and CA in these experimental conditions.

Keywords: HO-1/HO-2 expression, rosmarinic acid, caffeic acid, hypertension

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TAXIFOLIN MODULATES mRNA EXPRESSION OF INVASION-ASSOCIATED MARKERS AND STIMULATES MIGRATION OF HUMAN TROPHOBLAST CELLS

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During first trimester of pregnancy, the specific regulation of extravillous trophoblast migration and invasion into the maternal uterine wall is essential for successful placental development. Taxifolin (TAX) is a flavonoid of strong antioxidant activity that recently showed cytoprotective effects in trophoblast cells. The aim was to investigate if TAX can affect cell adhesion and migration in HTR-8/SVneo extravillous trophoblast cells as well as the expression of molecules that mediate these processes. Concentrations of TAX 10 and 100 μM were selected according to the previously reported cytoprotective effects.

Analysis of cell adhesion by crystal violet staining on 3 different substrates (plastic, collagen, Matrigel) showed that HTR-8/SVneo cells pre-treated with 100 μM TAX displayed increased adherence to Matrigel, substrate that imitate extracellular matrix, while adhesion to plastic and collagen was unaltered. TAX 10 μM did not lead to a significant change in cell adhesion. Further, the influence of 24h incubation with TAX on the mRNA expression of matrix metalloproteinases 2 and 9 (*MMP2*, *MMP9*) and integrin α1, α5 and β1 (*ITGA1*, *ITGA5* and *ITGB1*) was examined by qPCR. It was shown that TAX at 10 μM can elevate the expression of all integrin subunits, as well as *MMP2* and *MMP9*, while TAX 100 μM showed significant inhibitory effect on *MMP2* and stimulated the expression of *MMP9* and *ITGB1*, compared to the levels in untreated cells. A functional test - scratch assay, showed that TAX 10 μM significantly promotes cell migration after the 24h incubation.

It can be concluded that TAX can influence the cell adhesion and migration and mRNA expression of proteolytic factors crucial for trophoblast invasion. These are preliminary findings of TAX effect on functional role of human trophoblast cells *in vitro* and provide rationale for further studies of its potential application in pregnancy pathologies associated with disturbed trophoblast invasion.

Keywords: taxifolin, trophoblast, integrins, matrix metalloproteinases, cell migration

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ANTIOXIDANT CAPACITY OF APIUM GRAVEOLENS, APIUM GRAVEOLENS VAR. RAPACEUM, AND DAUCUS CAROTA EXTRACTS

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Celery (*Apium graveolens*) represents an edible plant used in traditional medicine due to its numerous health benefits: prevention of cardiovascular disease, lowering blood glucose and blood pressure, antifungal anti-inflammatory, anticoagulant, antioxidant, and antitumor properties. Celeriac (*Apium graveolens* var. *rapaceum*) is commonly used for its edible fleshy tap root and stalk. It contains flavonoids, violate oil, vitamins, and minerals, showing anticancer effects. Carrot (*Daucus carota*) is recognized for its nutraceutical and health benefits and due to the presence of phenolic compounds, carotenoids, and ascorbic acid possess antioxidant, anti-aging, anti-inflammatory, and anti-proliferative activities. Ultrasound-assisted extraction represents a modern technique for the extraction of various antioxidant compounds from plant material. Thus, in the present study, the extracts were prepared using celery root, celeriac stalk and leaves, or carrot root, and water or 30% ethanol in an ultrasound bath. The antioxidant potential was determined by analyzing total reducing (Folin-Ciocalteu assay) and DPPH radical scavenging activities. The total reducing capacity varied in a range of 0.27 to 2.10 mg gallic acid equivalents (GAE)/g of fresh plant material, achieving the highest values in the following samples: ethanol celeriac leaf extract > water celeriac leaf extract > water celery root and celeriac stalk extracts. On the other hand, the lowest total reducing capacity was obtained in the samples: water and ethanol carrot root extracts < ethanol celery root and celeriac stalk extracts. The DPPH radical scavenging potential, expressed as the concentration required for neutralization of 50% of radicals, follows the trend: ethanol celeriac leaf extract (0.10±0.01 g/mL) < water celeriac stalk extract (0.13±0.00 g/mL) < water celery and carrot extracts (0.18 g/mL) < ethanol celeriac stalk extract (0.26±0.03 g/mL) < water celeriac leaf extract (0.43±0.07 g/mL) < ethanol carrot root extract (0.62±0.05 g/mL) < ethanol celery root extract (1.32±0.06 g/mL) (the lowes IC_{50} = the highest antioxidant capacity). As can be seen, the highest antioxidant capacity was determined in ethanol celeriac leaf extract in both assays. Thus future experiments should be focused on the chemical characterization of the mentioned extract and individual compounds responsible for the antioxidant activity.

Keywords: antioxidant activity, carrot, celery, celeriac, extraction

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ANTIOXIDANT PROPERTIES OF GOAT MILK ENRICHED WITH GRAPE POMACE SEED EXTRACT AND MUSHROOM EXTRACT

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Goat's milk has a higher calcium, phosphorus, and potassium content than cow's and human milk, and also contains several bioactive peptides that enhance its antioxidant effect. It is of great importance because of its potential as a functional food. Various health institutions have recognized the nutritional problem and introduced the concept of functional foods, which, in addition to their basic nutritional content, can be enriched with additional components that have a positive effect on human health. Previous research has shown that enrichment of skimmed, thermally treated goat's milk powder (TM) with grape pomace seeds extract (GPE) could be a potential new functional food ingredient with good antioxidant properties of phenolic compounds. On the other hand, *Agrocybe aegerita* (V. Brig.) Singer is a popular edible mushroom known for its anti-inflammatory, antifungal, antibiotic and anti-tumor properties. It possesses functional compounds related to the identified organic acids, fatty acids and tocopherols. The aim of this research was to evaluate the antioxidant properties of TM enriched with 0.1% GPE and 0.1% mushroom extract (ME) using ferrous ion-chelating capacity (FCC) and ferric reducing power (FRP) tests. The results showed that goat milk has good FCC, but after addition of grape pomace seed and mushrom extracts, FCC increased by 30.30%, compared to TM samples. On the other hand, FRP increased about 30 times (2286%) compared to TM sample after GPE and ME enrichment. The addition of bioactive compounds extracted from grape pomace seeds and edible mushrooms can improve the antioxidant properties of goat milk and could be a promising ingredient for the formulation of functional foods.

Keywords: skimmed goat's milk; grape pomace seed; antioxidant properties; Agrocybe aegerita

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ENCAPSULATION OF *FUMARIA OFFICINALIS* EXTRACT IN THE LIPOSOMAL VESICLES

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Fumaria officinalis (fumitory) extracts have shown antimicrobial, antioxidative, antispasmodic, laxative, anthelmintic, cholagogue, cytotoxic, and sedative effects. The mentioned health benefits can be attributed to the high content of polyphenol and alkaloid compounds. However, the mentioned compounds possess low solubility, stability, and bioavailability. Therefore, their encapsulation in different carriers is necessary. Liposomes are widely used as carriers for the encapsulation, preservation, and controlled release of numerous hydrophilic and lipophilic bioactive principles from different plant sources. Thus, the aim of the presented study is the development and physicochemical characterization of *F. officinalis* extract-loaded liposomes in terms of encapsulation efficiency, vesicle size, polydispersity index (PDI), zeta potential, conductivity, and mobility. The extract-loaded liposomes were obtained in the proliposome method using 1 g of phospholipids, 1 mL of fumitory ethanol extract, and 10 mL of ultrapure water. Encapsulation efficiency was indirectly calculated by the polyphenol concentration determined in the supernatant. Particle size, PDI, zeta potential, conductivity, and mobility were measured by photon correlation spectroscopy. The encapsulation efficiency of polyphenols was >73%. The vesicle size and PDI were 274.0±0.7 nm and 0.307±0.020, respectively. The zeta potential, conductivity, and mobility were -6.34±0.16 mV, 0.465 mS/cm, and -0.491±0.011 μmcm/Vs, respectively. The beneficial effects of biologically active compounds from *F. officinalis* herba on human health, as well as their sensitivity, highlight the application of small, uniform, and stable phospholipid liposomal vesicles as their carrier.

Keywords: encapsulation, fumitory, liposomes, polyphenols

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TOTAL POLYPHENOL AND PROTEIN CONTENT IN DIFFERENT *FUMARIA OFFICINALIS* EXTRACTS

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Fumaria officinalis L. (Fumariaceae) is a component of various phytotherapeutic formulations in the European ethnobotany used in hepatobiliary dysfunction, illnesses of gastrointestinal and urogenital tracts, cancer, rheumatism, high blood pressure, and skin disorders. Various extraction techniques can isolate bioactive compounds from plant material; however, they differ in terms of extraction speed and efficiency, the yield of target molecules, solvent and energy consumption. Therefore, in the present study, microwave- and ultrasound-assisted extractions (MAE and UAE, respectively) and two different solid-to-solvent ratios (1:20 and 1:30) were used for polyphenol and protein extractions from *F. officinalis* herba. MAE process was performed in a microwave reactor for 2 min, while UAE was done in an ultrasound bath for 15 min. The total polyphenol content (TPC) was determined in the Folin-Ciocalteu assay, while the total protein yield was measured in the Bradford protein assay. The TPC varied in a range of 15.2 to 23.7 mg gallic acid equivalents (GAE)/g of dried plant material (dw), achieving the highest value in the extract prepared using MAE and a 1:30 ratio, followed by MAE and a 1:20 ratio and UAE and a 1:30 ratio, while the lowest polyphenol yield was obtained using UAE and a 1:20 ratio. The concentration of proteins in *F. officinalis* extracts follows the trend: MAE and a 1:30 ratio (68.3 mg/g of dw) > MAE at a 1:20 ratio (67.2 mg/g of dw) and UAE at a 1:30 ratio (67.5 mg/g of dw) > UAE and a 1:20 ratio (64.3 mg/g of dw). Due to significantly higher polyphenol and protein yields, *F. officinalis* extract prepared using MAE (as a more rapid technique) and a higher employed level of solid-to-solvent ratio (1:30) can be potentially implemented in different food, functional food, dietetic supplement, or pharmaceutical formulations.

Keywords: Fumaria officinalis, extraction, polyphenols, proteins

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QUERCETIN IN GENETIC CAUSE OF LUNG DISEASE

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Quercetin (QRC) belongs to flavanoid family of natural polyphenols and is present in variety of foods such as onions, apples, eldberries, coriander and broccoli. The molecular mechanisms underneath its potent antioxidant and anti-inflammatory effects are well established and have been in the basis of notable studies of inflammatory and with-oxidative stress related pulmonary diseases (asthma, chronic obstructive pulmonary disease and pulmonary emphysema/fibrosis). However, may QRC contribute to genetic causes of lung disease, such as cystic fibrosis (CF)?

CF is an autosomal recessive disorder caused by a mutation on chromosome 7 at position q31.2. This specific region contains the cystic fibrosis transmembrane conductance regulator (CFTR) gene which encodes CFTR protein. It acts as a transmembrane anion channel, transporting the anions (specifically bicarbonate and chloride) from the intracellular to the extracellular space. Any dysfunction of CFTR will impair this transport and the disease will occur. Although CF is multi-organ disorder, the main cause of morbidity and mortality is lung disease. Namely, CFTR mutations will highly affect airways and lungs, where overly secreted and viscous mucus impairs mucociliary clearance and increases the risk for recurrent respiratory infections (*S. aureus*, *P. aeruginosa*). The natural course of the disease is characterised with episodic exacerbations and progressive worsening of a lung function, resulting in chronic hypoxia. Over the time, pulmonary hypertension and right-sided heart failure occur. Due to this, CF is associated with shorter life expectancy and unfortunately, some patients will eventually need lung transplant.

After finding that QRC is able to improve CFTR-mediated transepithelial transport of chloride anions in nasal epithelium, its role in the pathogenesis of CF has been extensively investigated. So far, QRC has been studied in several clinical trials and, from recently, we know that QRC is able to activate defective F508del CFTR protein and increases its availability at the cell membrane. Ongoing Clinical trials (https://apps.cff.org/trials/finder) are very optimistic.

Keywords: quercetin, cystic fibrosis, mutations, lung disease, clinical trials

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RESVERATROL AND VASCULAR HEALTH – FROM ION CHANNELS TO GUT MICROBIOTA

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Resveratrol (RSV) has been subject to extensive research regarding cardiovascular diseases (CVD), diabetes mellitus, malignancies, neurodegenerative disorders, metabolic syndrome and obesity. Preclinical studies have shown that RSV has the affinity for numerous intracellular molecules and cell structures, which may vividly depict RSV as "*one molecule - many targets*". Considering high global prevalence and long-term complications, combating CVD (endothelial dysfunction, hypertension, peripheral arterial disease, heart failure, etc.) by introducing RSV to clinical practice has been, for some time now, of the utmost public health interest. After extensive results, it seems that the ability to potently relax different blood vessels is in the centre of its cardio-protection.

RSV relaxes vascular smooth muscles cells (VSMC) regardless of the presence of functional endothelium, for which two different mechanisms are proposed. First (endothelium-dependent) mechanism includes increased production of nitric oxide (NO) and activation of signal pathways involving SIRT1 (sirtuin 1), AMPK (5' adenosine monophosphate-activated protein kinase) and ROS (reactive oxygen species). Second (endothelium-independent) mechanism includes modulation of VSMC ion channels, decreasing serum concentrations of vasoconstrictor (e.g., angiotensin II, endothelin-1) and inhibition of VSMC contractility.

Novel mechanistic studies go a step further, introducing a biological model that contributes to RSV's bioavailability and effects, referring to its synergy with the human gut microbiota. On one hand, gut bacteria (e.g *Lactobacillus*) may synthesise RSV from its food form, piceid, while some other (*Bacillus cereus*) may form RSV-derivatives (dihydroRSV, dihydropiceid), with relaxant and anti-atherosclerotic properties. On the other hand, RSV positively changes the gut composition towards 'good' bacteria such as *Bacteroides, Bifidobacterium, Verrucomicrobia*, etc. Indeed, some conflicting results from the clinical trials may be explained exactly with disproportion in the concentration of RSV metabolites, due to interindividual variation in qualitative aspect of gut microbiota. However, this broadens scientist's knowledge of RSV complexities and helps understanding the enormous value of Mediterranean diet.

Keywords: resveratrol, cardiovascular diseases, ion channels, gut microbiota

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RUBUS GLANDULOSUS – ANTIOXIDANT ACTIVITY OF ETHANOL EXTRACTS

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Blackberries (*Rubus* spp.) are known for a variety of health benefits attributed to their phytochemical composition. They have the potential to mitigate metabolic disorders and chronic diseases such as cancer, diabetes, hypertension, cardiovascular disease, gastrointestinal diseases, atherosclerosis, aging, Parkinson's disease, and Alzheimer's disease. Blackberries have a powerful antioxidant capacity due to their high levels of anthocyanins and other phenolics.

In general, fruits are rich in dietary fiber, low in calories and lipids, and contain high levels of simple sugars such as glucose and fructose. Organic acids are present in low concentrations and contribute to the fruit's distinct flavor. The presence of certain minerals, vitamins, and phenolic compounds gives *Rubus* fruits the status of functional food.

Do the leaves, stem, or root of the plant also contain phenolics and flavonoids, whose primary function is to protect cells from UV radiation? Do these compounds accumulate in specific plant parts?

This study examines the total phenolic (TPC) and flavonoid (TFC) content and the antioxidant potential of ethanol extracts from the root, stem, and leaf of *Rubus glandulosus*.

The antioxidant capacity of blackberry samples was assessed using three spectrophotometric assays: $DPPH^{\bullet}$ (2.2, diphenyl-picryl-hydrazyl radical), $ABTS^{\bullet+}$ (2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonate radical cation) and FRAP (ferric reducing antioxidant power). Synthetic antioxidant butylated hydroxyanisole (BHA) and ascorbic acid (vitamin C) were used as controls.

The highest total phenolic and flavonoid content were found in root ethanol extracts (666.99 mg GAE/g . d.e. and 91.63 mg QE/g d.e.. In contrast to the root, TPC and TFC were much lower in the ethanol extract of the fruit (32.74 mg GAE/g d.e.).The fruit extract exhibited the lowest antioxidant activity in $DPPH^{\bullet}$ test (IC_{50} 27.49 µg/mL). The root ethanol extract showed the strongest inhibitory effects, achieving IC_{50} at the lowest concentration (4.92 µg/mL). The values of ABTS radical cation scavenging activity ranged between 3.38 and 26.32 mg AAE/g. d.e. The root extract possessed superior scavenging activity (26.32 mg AAE/g). The FRAP value ranged from 1.05 to 2.83 µmol Fe/mg d.e.. The phenolic and flavonoid content in leaf extracts were half that found in root extracts, with antioxidant activity two times lower than that of the root extract.

Considering the traditional use of *Rubus* spp. in our folk medicine (leaf decoctions), current objective was to explore the broader biological potential of *R. glandulosus*, with a view towards its potential commercialization and the feasibility of producing herbal mixtures or tinctures utilizing all parts of *R. glandulosus.*

Keywords: Rubus glandulosus, phenols, flavonoids, antioxidant activity

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CHARACTERIZATION OF THE TOTAL LACTOSE CONTENT IN RAW MILK SAMPLES

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Mammals are known for their distinctive product milk, which is nutritionally rich and white product. It is generated by their mammary secretory cells during a process known as lactation. Milk quality parameters are of main importance, thus, it needs to be delivered and preserved in the best possible ways. As a high nutritionally fluid, it contains large amounts of lipids, proteins, such as casein, which supplies human body with necessary amino acids for muscle building, and also carbohydrates and energy components. In trace amount are presented vitamins, salts (phosphates and chlorides of calcium and potassium) and enzymes. When it comes to food industry, milk is actually a primary source of lactose. It is a disaccharide sugar composed of glucose and galactosemolecules. Wide usage, beside food industry, is found in pharmaceutical industry, when it comes to production of some medicines. The importance of lactose is manifold because lactose components are also constituent ingredients of some macromolecules (e.g. glycoproteins), and is the least cariogenic of all fermentable sugars (glycemic index is low; GI = 46). To determine the lactose content in raw milk samples, high performance liquid chromatography (HPLC) was used. An HPLC system Waters (Milford, USA), with a Waters 2414 refractive index detector (RID) wasemployed.The injection volume of each sample was 10.0 µl at a flow rate of 1.3 ml/min, with a run time of 12 minutes per sample. Retention time of lactose was 8.87 min. Each sample was performed in triplicate. The final results are expressed in g/100 g raw milk and vary between 3.0 – 5.65 g/100 g. The aim of the determination was to establish a method based on the precipitation method using Carrez I and II reagents. Of the 27 samples analyzed, two were samples of standardized sterilized milk that were used to monitor the validation and setup of the method. The other 25 tested samples of raw milk and lactose concentration were the initial stage of setting up the experiment and monitoring the amount of lactation and the amount of lactose depending on feeding.

Keywords: raw milk, lactose, chromatography

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BIO-ACTIVITY OF CHAGA MUSHROOM: DETERMINING CYTOTOXICITY AND ANTIOXIDANT POTENTIAL IN HUMAN CELL LINES *IN VITRO*

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The Chaga mushroom (*Inonotus obliquus* (Ach. ex Pers.) Pilat), a parasitic mushroom with broad spectrum of metabolites, such as polysaccharides, minerals, carbohydrates and polyphenols, has previously exhibited anti-inflammatory, antioxidant, antigenotoxic and immunomodulatory properties. Although Chaga is a promising natural resource for various applications, there is insufficient data about its biological activity and safety. Thus, it is necessary to investigate how its properties affect the functions of human cells to determine the safety of its use. The cytotoxic effect of Chaga at range of concentrations 25, 50, 75, 100 and 200 μg/mL was investigated in human extravillous trophoblasts HTR-8/SVneo, lung fibroblasts MRC-5 and cervical cancer HeLa cell lines using the MTT and cristal violet (CV) assays. The ability of Chaga to decrease oxidative stress-induced ROS production in MRC-5 cells at 25, 50 and 75 μg/mL was investigated with H2DCFDA assay. The MTT results showed an significant increase in percentage of viable cells at concentrations of 25, 50 and 75 μg/mL in HTR-8/SVneo and of MRC-5 cells, while a significant reduction in cell viability was observed for cancer cells HeLa at concentrations of 100 and 200 μg/mL following the 24h incubation with Chaga. The CV method showed no significant change in the number of adherent cells of HTR-8/SVneo and HeLa cells. The number of adherent MRC-5 cells increased at a Chaga concentration 25 μg/mL, and decreased after 200 μg/mL treatment. Analysis of ROS levels in MRC-5 cells co-incubated with Chaga and 100 μM H_2O_2 showed that Chaga reduces ROS production at 50 and 75 μg/mL. Overall, the exhibited antioxidant ability of Chaga together with its stimulatory effect on cell viability in normal cells, while having a moderate cytotoxic effect in cancer cells, provides evidence of its biological potential.

Keywords: chaga; medicinal mushrooms; cytotoxicity; reactive oxygen species

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HERBAL FOOD SUPPLEMENTS LABELLING: HEALTH CLAIMS, PRECAUTIONS AND WARNINGS

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For years, we have been witnessing an extremely rapid increase in the market offer of food supplements and exceptional creativity in terms of both supplements composition and information provided on their labels. Although it is understood that voluntary informationprovided to facilitate consumers choices must be true and trustworthy, the volume and severity of misleading claims is also on the rise. This study's aim was to challenge regulatory compliance of voluntary health claims and presence of *mandatory statements* on the labels of herbal food supplements commercialized in Novi Sad (Serbia). Health claims were listed on as much as 86.2% of a total of 87 herbal supplements, but only 10.7% of them, all associated with vitamin and mineral ingredients, were in compliance with the EU List of authorised health claims. An additional 38.7% of supplements carried 'on-hold' claims from the EFSA Register of questions for botanicals, continuously used under *transitional regulatory measures. The remaining supplements comprised those* attributed with strictly prohibited properties of disease prevention, treatment or cure (9.3%), and those containing at least one botanical -related health claim out of the scope of the Register of Questions. Among mandatory warnings intended to *safeguard consumer well-being, the one* that recommended daily doses cannot be exceeded was absent from 16.1% of labels. A recommendation to consult a healthcare professional was not included in 72.0% of the products bearing claims. The recently issued resolution of the European Parliament warns that applied practices *"could mislead consumers and constitute a health risk for them, as they may falsely assume that the 'on-hold' claims have been scientifically assessed and risk managed"*. Consumers′ understanding of health promoting claims is considered questionable, but they certainly tend to overconsume supplements bearing claims. Consumers need to be empowered through education and awareness campaigns to guide them through the future food environment.

Keywords: botanicals; on-hold claims; consumer protection; public health

BIBLIOMETRIC ANALYSIS ON NUTRITIONAL PROFILE AND HEALTH BENEFITS OF PEA

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We performed a bibliometric analysis on the scientific literature related to the nutritional composition and health benefits of pea. The bibliographic data was downloaded from Scopus database, which resulted in 715 English written research articles published before June 2024. The bibliographic data was analyzed using Bibliometrix package in R studio. The first literature in this area was published in 1975, and there was an increasing trend of publications over time with an annual growth of about 9%. The core group consisted of 23 out of 376 journals with Nutrients (39 documents), followed by Food Hydrocolloids (19 documents), and Food Chemistry (16 documents) publishing the highest number of documents on the field. The USA made the highest contribution on the topic followed by China and Canada. The top countries performing research on the nutritional composition and health benefits of peas did not show a very high international collaboration. The papers published in the field were mostly single-country publications. The most frequent author keywords projected with the word cloud after pea, legumes and pulses were protein, antioxidant, diet, nutrition, dietary fiber, inflammation, and obesity, which also reflects the current research direction of the field. Other significant keywords were phenolic compounds, carotenoids, extrusion, palmitoylethanolamide, proteomics, etc. Collaboration network analysis was run to show associations between top countries, institutions, and authors. Two collaboration clusters of countries, three sub-clusters of institutions, and ten subnetworks among authors were detected with at least two collaborative papers. The aim of this study is to explore the conceptual, intellectual and social aspects of the nutritional composition and health benefits of peas and to identify current research topics in this area through descriptive and retrospective bibliometric methods.

Keywords: text mining, performance analysis, science mapping, network analysis

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ADVANCEMENTS IN ZINC STATUS ASSESSMENT - A SYSTEMATIC REVIEW AND FUTURE DIRECTIONS

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The discovery of zinc's role in human health occurred six decades ago, yet the search for a sufficiently sensitive and specific biomarker remains ongoing. Plasma/serum Zn, presently considered the most accepted population indicator, is known to respond well to severe Zn deficiency, although mild to moderate deficiency states often go unnoticed. Early-stage detection of Zn deficiency necessitates the exploration of additional robust biomarkers. The systematic review was conducted to assess changes in FADS1 and FADS2 (fatty acid desaturase 1 and 2) activities in response to dietary Zn intake. PubMed, Web of Science, Scopus, Web of Knowledge, and Central were searched using predefined search criteria. Twenty-one studies, including both animal and human trials, were analysed following PRISMA guidelines. Following data extraction, a random-effects meta-analysis was performed. Descriptive analysis and qualitative synthesis were completed to assess variations in desaturase activities and identify potential confounders and covariates, respectively. The relationship between dietary Zn intake and FADS1 and FADS2 activities was examined. While there was no significant correlation between delta 6 desaturase and Zn status, delta 6 desaturase responded more strongly to Zn-containing interventions. However, the magnitude of the correlation between LA:DGLA (linoleic acid: dihomo-gamma-linolenic acid) and Zn intake and between Zn status and Zn intake remained consistent. Desaturase activities varied inconsistently in response to Zn intake among studies, with factors such as study population and dietary interventions contributing to this inconsistency. Further research is needed to clarify the potential of using desaturase activities as biomarkers of Zn status. Further research efforts are recommended to explore the potential of desaturases as reliable indicators of variations in bioavailable zinc intake over time. These efforts should include investigations into periods of both high and low zinc intake, as well as short and long provision times. Additionally, there is a need for refined marker specificity across diverse populations, which warrants continued investigation. Understanding the factors influencing desaturase activities and the interaction of zinc with other minerals will be crucial moving forward. Future studies should consider the insights obtained from qualitative synthesis regarding the various factors that affect the activity of fatty acid desaturases in response to interventions involving zinc. This provided framework should guide the design of upcoming clinical trials aimed at further elucidating the proposed relationships and confirming the effectiveness of FADS1 and FADS2 activity as indicators of zinc status, either alone or in combination with other biochemical markers.

Keywords: zinc, biomarker, desaturase, zinc deficiency, LA:DGLA ratio

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ANTIMICROBIALACTIVITY OF SWEET CHERRY BY-PRODUCT

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Due to its pleasant taste, aroma, color and smell, sweet cherry (*Prunus avium*) is one of the fruits whose production is in growing demand. During its processing, a significant amount of waste by-products is created, dominated by kernels. Cherry kernels are characterized by a high content of bioactive compounds, primarily lipophilic, dominated by unsaturated fatty acids, but also by a significant amount of hydrophilic components, among which polyphenolic compounds are particularly important. In this work, the antimicrobial activity of lipophilic extracts of cherry kernels obtained by supercritical fluid (SFE) and Soxhlet extraction was examined. SFE was performed at 70°C, pressure of 350 bar and with a CO_2 flow of 0.4 kg/h for 4 hours. Soxhlet extraction was performed in a laboratory apparatus with hexane and methylene chloride for 6 hours. The obtained extracts were characterized in terms of chemical composition (GC-FID). The antimicrobial potential was evaluated on a panel of 8 different microbial strains: *Staphylococcus aureus, Klebsiella pneumoniae, Escherichia coli, Proteus vulgaris, Proteus mirabilis, Bacillus subtilis, Candida albicans, Aspergillus niger*. In total 8 fatty acids were identified whereby linoleic (41.10-41.54%) and oleic acid (31.17-31.22%) were the most abundant. The obtained results showed the highest antimicrobial effect of methylene chloride extracts, where the MIC values were in the range of 4.88-78.125 µg/mL, while the lowest sensitivity of the used microorganisms was towards the SFE extract (MIC= 39.1-156.25 µg/ml). The effect on *Bacillus* was remarkable where the MIC value was equal to that obtained with commercial antibiotic (amracin). Also, notible effect was observed against *P.mirabilis* and *S.aureus* where the MIC values were very close to that of amracin. Methylene chloride extracts showed the same effect against *Candida* as the Nystatin standard. High activity of the obtained extracts is of great importance and opens new perspectives in the utilization of this bio-waste.

Keywords: extraction, bio-waste, GC-FID, natural products, antimicrobial activity

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ANTIOXIDATIVE AND ANTIMICROBIALACTIVITY OF CURCUMIN AND ITS INCLUSION COMPLEX

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Curcumin, a phyto-constituent of the rhizome of the *Curcuma longa* plant, has an important application as a food coloring agent and in medicine. It exhibits numerous biological effects: anti-inflammatory, hypoglycemic, antioxidant, antimicrobial and antiviral. The main obstacle for the effective manifestation of these effects is insufficient aqueous solubility of curcumin, and therefore low bioavailability. The aim of this work was the preparation of the inclusion complex curcumin:2-hydroxypropyl-β-cyclodextrin and the investigation of the antioxidant and antimicrobial activity of curcumin and the inclusion complex curcumin:2-hydroxypropyl-β-cyclodextrin. The curcumin:2-hydroxypropyl-β-cyclodextrin inclusion complex was prepared by the coprecipitation method in a molar ratio of 1:1. The antioxidant activity of curcumin and the inclusion complex was determined using DPPH, ABTS and FRAP methods. The *in vitro* antimicrobial activity of curcumin and the inclusion complex was tested using the micro-dilution method. The results of DPPH ($EC_{50(curcumin)}$=6.77 μg/cm^3, $EC_{50(complex)}$=4.4 μg/cm^3), ABTS ($EC_{50(curcumin)}$=98 μg/cm^3, $EC_{50(complex)}$=83 μg/cm^3) and FRAP (9.297 mmol $Fe^{2+}/g_{curcumin}$, 9.509 mmol $Fe^{2+}/g_{curcumin\ in\ the\ complex}$) methods show that curcumin in the inclusion complex exhibits better antioxidant activity than curcumin. Curcumin and the inclusion complex show antibacterial activity against all tested Gram (+) (*Staphylococcus aureus*, *Enterococcus faecalis*, *Streptococcus pneumoniae*, *Streptococcus pyogenes*, *Bacillus cereus*) and Gram (–) (*Pseudomonas aeruginosa*, *Salmonella enteritidis*, *Escherichia coli*, *Enterobacter aerogenes*) bacterial strains and antifungal activity on the tested *Candida albicans* strain. The results of antimicrobial tests show increased effectiveness of curcumin in the inclusion complex. The lowest MIC value (5 μg/cm^3) for curcumin in the inclusion complex was achieved for the *E. faecalis* strain, while the activity of curcumin on the same tested microbe strain is less pronounced (MIC 780 μg/cm^3). Based on the presented results, it can be concluded that the complexation of curcumin with 2-hydroxypropyl-β-cyclodextrin achieves better antioxidant and antimicrobial activity as a consequence of the increased solubility of curcumin in the complex.

Keywords: curcumin, inclusion complex, 2-hydroxypropyl-β-cyclodextrin, antioxidant activity, antimicrobial activity

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QUALITATIVE CHEMICAL COMPOSITION OF THE FRANKINCENSE (*Boswellia serrata*) RESIN METHANOLIC EXTRACTS

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Frankincense (*Boswellia serrata*, Burseraceae) is an important medicinal plant due to high content of bioactive pentacyclic triterpenoid acids (α-boswellic, β-boswellic and lupeolic acids) and their derivatives. They are intensively tested clinically for use in the treatment of asthma, arthritis, diabetes, inflammatory bowel diseases, skin diseases, central nervous system disorders and cancers. This work aimed to examine the influence of the extraction technique on qualitative chemical composition of the frankincense resin methanolic extracts. Extracts were prepared using maceration, Soxhlet extraction and ultrasound-assisted extraction. The macerate was prepared by macerating 1 g of frankincense resin with 10 ml of methanol for six days, with occasional shaking at room temperature. Soxhlet extraction was performed at the boiling temperature of the solvent for 4 hours, with the ratio of plant material to solvent being 1:10, m/V. Ultrasound-assisted extraction was performed at 45 °C for 2 hours, at the same ratio of plant material and solvent (1:10, m/V). The chemical composition of the extracts was determined by applying ultra-high-performance liquid chromatography-tandem mass spectrometry (UHPLC-DAD-MS/MS) method. Twenty-seven, twenty-five and twenty-three different compounds were detected in methanolic extracts obtained by maceration, Soxhlet extraction and ultrasound-assisted extraction, respectively. Sixteen components were identified: α- and β-boswellic acid, their acetyl-, keto- and acetyl-keto derivatives, then lupeolic acid and oleanolic, tirucallic and urosolic acids derivatives. Based on the obtained results, it can be concluded that maceration is the method of choice for the extraction of bioactive principles from frankincense resin, as it enables the isolation of the largest number of bioactive components. However, further quantitative analysis should be performed in order to determine the content of the individual components in the frankincense methanolic extracts.

Keywords: frankincense resin, maceration, Soxhlet extraction, ultrasound-assisted extraction, boswellic acids

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CAFFEIC ACID MODULATES EXPRESSION OF PROINFLAMMATORY CYTOKINES IN HUMAN TROPHOBLAST CELLS

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One of the crucial processes for human pregnancy establishment is invasion of the extravillous trophoblast cells (EVTs), specific cells of the placenta, into uterus wall. This process is tightly regulated by numerous factors including proinflammatory cytokines secreted by different cells and tissues at feto-maternal interface. Caffeic acid (CA), polyphenolic compound abundantly present in fruits, vegetables, spices and beverages used in everyday diet, exhibits numerous physiological effects including antioxidant and anti-inflammatory activities. Our previous *in vitro* studies on human EVT HTR-8/SVneo cell line showed that CA has a potential to modulate processes important for early pregnancy.

The aim of this study was to further investigate CA effects on trophoblast cells by evaluating expression of proinflammatory cytokines involved in regulation of EVT invasion in HTR-8/SVneo cells. Our results assessed by real-time PCR showed that treatment with both 10 µM and 100 µM CA downregulated expression of *TGFB1* and *IL1B* in treated HTR-8/SVneo cells. However, CA upregulated mRNA levels of IL-8 (*CXCL8*) while *CXCR1*, gene coding for IL-8 receptor, was downregulated, concentration-dependently in both cases.

Based on these preliminary results it can be concluded that CA could affect cytokine and chemokine expression in trophoblast cells. Also, it could modulate IL-8 signalling pathway not just by affecting *CXCL8* expression but also expression of its receptor. These results could contribute to revealing molecular mechanisms involved in CA-induced modulation of trophoblast invasion and migration indicated in our previous studies.

Keywords: caffeic acid, trophoblast, cytokines

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Section
FOOD PRODUCTION, PROCESSING, SUSTAINABILITY, ADDED-VALUE FOOD

VALORISATION OF APPLE PEEL THROUGH MODERN EXTRACTION TECHNIQUES

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The apple (Malus domestica) is one of the most consumed fruits in the world, and its utilization is high. A large proportion of the peel remains as waste during processing. Considering the richness of the apple peel in bioactive compounds, especially phenolics, fibers, and minerals, this by-product is of great importance. In order to extract the bioactive compounds from the apple peel, the influence of different modern extraction techniques and solvents on the extraction of phenolics was investigated.

The water extracts were prepared by ultrasound (UAE) and microwave (MAE), while NADES extracts were prepared by maceration using fructose-glycerol 1:4 (MAC1) and choline chloride - glycerol 1:2 mixtures (MAC2). In the extracts obtained phenolics were analyzed and their *in vitro* antioxidant activity was tested.

The total phenolic content (TPC) of the extracts was determined by Follin-Ciocalteu method and expressed as chlorogenic acid equivalents (mg CAE/g). The UAE yielded the highest TPC (0.038 mg CAE/g dw) while MAC1 had the lowest phenolic content (0.010 mg CAE/g dw). The same situation was observed for total flavonoids contents (TFC) which were determined by a spectrophotometric method based on the $AlCl_3$ reaction and expressed as rutin equivalents (mg RE/g dw). In the UAE extracts TFC was 0.008 mg RE/g, while in MAC2 it had a value of 0.002 mg RE/g. LC-MS analysis confirmed the presence of phenolic acids, flavonoids and phloretin derivatives.

UAE, MAE, and MAC showed significant anti-radical DPPH ability and the IC_{50} values were in the range of 0.69-1.187 mg/g. The reducing power test showed remarkable activity, with EC_{50} values ranging from 5.02 to 8.93 mg /g. The results were highly variable depending on the extraction method, underlining the importance of extract preparation for the overall nutritional profile of apple peel.

Keywords: extractions, antioxidants, apple peel, LC-MS/MS

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ENHANCING STRAWBERRY TREE FRUIT SPIRIT: EFFECTS OF OAK WOOD AGEING

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The present study focuses on the impact of wood ageing on strawberry tree fruit spirit, a high-quality beverage in Mediterranean regions, particularly Portugal. It explores the chemical and sensory changes resulting from ageing the spirit for two periods of time with oak wood of varying toasting levels. Analysis included acidity, colour, pH, dry extract, volatile compounds (methanol, acetaldehyde, ethyl acetate and fusel alcohols), identified and quantified through GC-MS and GC-FID, and low molecular weight compounds, quantified by HPLC (gallic acid, ellagic acids, syringic acid, vanillin, syringaldehyde, coniferaldehyde, sinapaldehyde, furfural, 5-hydroxymethylfurfural and 5-methylfurfural). Sensory evaluations were made by a trained panel.
Results showed enhanced colour and enrichment with volatile and low molecular weight compounds due to oak wood contact, with slight differences among toasting levels. Almost all parameters increased with ageing time, as confirmed by principal component analysis, which effectively discriminated between aged spirits and toasting level. These findings contribute to understanding and optimizing the ageing process of strawberry tree fruit spirit for commercialization. Medium toasted oak yielded the best results.

Keywords: strawberry tree fruit, spirit, volatile compounds, low molecular weight compounds, sensory analysis

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DIVERSITYAND PROPERTIES OF CULTIVABLE YEASTS ISOLATED FROM GRAPE BERRIES OF PROKUPAC

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The aim of this work was to study the biodiversity of yeasts isolated from grape berries of Prokupac (*Vitis vinifera* L.; proles pontica, subproles balcanica), the autochthonous Serbian red wine variety. The grapes were harvested from vineyards in two geographically different wine regions in Serbia (Šumadija and Toplica), which differ in both soil and climatic characteristics. Epiphytes and endophytes were isolated from the grape berries and additionally from the must during one-week spontaneous alcoholic fermentations. Different restriction profiles of the ITS-5.8S rDNA region corresponding to *Rhodotorula glutinis, Hanseniaspora uvarum, Aureobasidium pullulans,* and *Saccharomyces cerevisiae* were observed. The presence of yeasts varied considerably in the grape berries and in the grape must, where we could only detect *S. cerevisiae*. The glucose concentration (0.5, 1, 1.5, and 2%) and the influence of the pH (4, 6.4, 8, and 10) of the YPD medium on the non-Saccharomyces yeasts were calculated. The results showed that the sugar concentration influenced *A. pullulans* the most, while the pH of the medium of 10 completely interrupted the growth of *A. pullulans* as well as that of *R. glutinis*. *H. uvarum* was the most resistant species to pH changes and glucose availability. No geographical variability in yeast diversity was detected in our study, suggesting the influence of grape variety rather than environment. This is the first report on the population dynamics of "culturable" microbiota diversity of Prokupac (the most important Serbian red wine), which allows us to provide the basis for improved management of wine yeasts (both with non-Saccharomyces and Saccharomyces) for the production of typical wines with pronounced terroir impact.

Keywords: autochthonous wine variety, yeast diversity, epiphytes, endophytes

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PROTEIN COMPOSITION, TECNO-FUCTIONALAND ANTIOXIDANT PROPERTIES OF DIFFERENT TYPES OF COMMERCIAL SOY FLOUR

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The aim of this study is to characterized protein composition, techno-functional and antioxidant properties of different types of commercial soy flour. For this purpose, three types of soy flour were used, fully toasted, lightly toasted and enzymatically active soy flour. Protein fraction was characterized by the level of soluble proteins and their composition. Soluble protein composition as well as the composition of total proteins was determined using SDS-PAGE under denaturing and reducing conditions and by densitometric analysis of the obtained gels. Foaming properties of soy flours are characterized by foam capacity (FC%) and foam stability index (FS%) whereas emulsifying properties are determined as emulsion ability index (EAI) and emulsion stability index (ESI). Also, water- and oil holding capacities and swelling properties were determined. In vitro antioxidant properties are characterized using TAC-, CUPRAC- and DPPH-assay. In addition, the level of total phenols and flavonoids were detected. As could be expected the investigated flours significantly differs in water soluble protein content and composition but had similar total protein content and composition. The best solubility at pH 8.0 expressed lightly toasted flour. The investigated flours had significantly different techno-functional properties. EAI and ESI values were in the range of 8.90-25.42 m2/g and 14.17-38.33 min. The highest ability to form emulsion had enzymatically active soy flour whereas the most stable emulsion formed lightly toasted samples. These samples also had the best foaming properties (37.00 FC% and 90.10 FS%). Full toasted samples were the most powerful as water holding and swelling agent. The highest antioxidant power expressed lightly toasted samples. Furthermore, these samples had the highest content of total phenols and flavonoids.

Keyword: soy flour, protein composition, techno-functional properties, antioxidant properties

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THE IMPACT OF DIFFERENT SOLID-TO-SOLVENT RATIOS ON *SATUREJA MONTANA* L. POLYPHENOL AND FLAVONOID CONTENT AND ANTIOXIDANT POTENTIAL

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The presented study aimed to optimize the extraction of polyphenolic compounds from *Satureja montana* L. cultivated at the experimental site of the Institute for Medicinal Plants Research "Dr Josif Pančić", Serbia, by varying on of the most important parameter for maceration, solid-to-solvent ratio. The obtained extracts were characterized on the basis of the total polyphenol content (TPC), and total flavonoid content (TFC). The TPC varied from 35.01 ± 10.41 mg GAE (gallic acid equivalent)/g to 87.96 ± 4.73 mg GAE/g. On the other hand, the TFC values were in the range from 1.8 ± 0.09 to 5.6 ± 0.01 mg CE (catechin equivalent)/g. The highest TFC was obtained at a solid-to-solvent ratio of 1:50, whereas in the case of TFC the optimal ratio was 1:40. The highest DPPH neutralization level (86.12 %) as well as the highest anti-ABTS antioxidant activity (90.35 %) were obtained for the extracts prepared using a solid-to-solvent ratio of 1 g:50 cm^3. Our study shows the initial steps in obtaining polyphenol-rich extract of *S. montana* with good satisfactory antioxidant potential, which could be used in the pharmaceutical, food, or cosmetic industry.

Keywords: polyphenols, winter savory, maceration, flavonoids

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IMPROVING THE FUNCTIONALAND NUTRITIONAL CHARACTERISTICS OF YOGURTS BY INCORPORATING APPLE FIBER

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There is a rising awareness among consumers about healthy and high quality foods, which has led to an increased demand for functional food production. The addition of bioactive ingredients to food not only improves the product's characteristics but also has a positive impact on human health. Dairy products, specifically yogurt, are commonly used for the production of functional foods by incorporating other functional ingredients into them. In the present study, yogurt was fortified with different concentrations of apple fiber (0-3% w/w). Apple fiber is obtained from apple pomace and is known for its high content of antioxidant compounds and dietary fiber. Yogurt fortified with apple fiber showed higher antioxidant activity (measured using DPPH, CUPRAC, and FRAP methods), and displayed a higher total phenolic content compared to the control, without affecting the main physicochemical and microbiological properties. Furthermore, correlation analysis showed a strong positive linear relationship between total phenolic content and antioxidant activity. Additionally, the incorporation of apple fiber in yogurt resulted in a significant increase in water holding capacity and a decrease in syneresis. Syneresis is one of the most common defects in yogurts that may limit the shelf life of the product and affect its acceptability by consumers due to its undesirable appearance. The results of the present study show that apple fiber is a suitable fortification material for the production of functional yogurts. Fortifying yogurt with apple fiber can increase its phenolic content, antioxidant activity, and dietary fiber, while improving its technological characteristics like syneresis.

Keywords: antioxidant activity, total phenolic content, syneresis, water holding capacity

INTRODUCING ALFALFA SEED TO BREAD PRODUCTION – EVALUATION OF TECHNOLOGICAL QUALITYAND SENSORY ACCEPTANCE

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The increasing efforts to use legumes in bread production triggered the search for a novel ingredient that would meet consumers' needs not only in terms of macronutrients such as proteins, but also dietary fibres, minerals, vitamins and phytochemicals. It is precisely these compounds that are found in alfalfa seeds, which are an underutilised food source with great potential for use in staple foods such as bread. Nevertheless, the disadvantages of using alfalfa seeds are mainly the presence of anti-nutrients and the grassy flavour which can be successfully reduced by using bioprocessing method such as germination.

Therefore, this study investigates the substitution of white wheat flour with 5 and 10% flours of ungerminated and germinated alfalfa seeds subjected to convective drying in the conventional bread formulation and the resulting effects on the specific volume, crumb texture and sensory properties of the bread.

The addition of alfalfa seed flour significantly increased the specific volume of the bread, regardless of the level of addition and the type of flour (2.96±0.02 to 3.16±0.38 cm^3/g). At the same time, a significant decrease in crumb hardness was observed compared to the control (from 3.82±0.67 to 2.10±0.42 N), especially with a 10% substitution with alfalfa flour, while minor variations were observed for cohesiveness and springiness.

Sensory acceptability, as assessed by 12 respondents using the 5-point hedonic scale, was highest for bread with 5% ungerminated alfalfa flour (4.3 points) and was characterised by good flavour, soft crumb texture with uniform medium sized air voids, and high specific volume. A 10% substitution with germinated alfalfa flour resulted in the lowest bread acceptability score (3.7 points). The reported results showed improvements in specific volume, softness of the crumb and good sensory acceptability of the breads suggesting that both ungerminated and germinated alfalfa flour are promising baking ingredients.

Keywords: legumes, germination, bread, sensory assessment, technological quality

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FROM FARM TO FUNCTIONAL FOOD: INFLUENCE OF ORGANIC BIOACTIVE COMPOUNDS ON THE POLYPHENOLIC PROFILE AND ANTIOXIDANT ACTIVITY OF RASPBERRY FRUIT

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Raspberry (*Rubus idaeus* L.), also called European raspberry or red raspberry, is a red berry species native to Europe and northern Asia and is commonly cultivated in temperate regions. This fruit is renowned for its abundance of vitamins and minerals, as well as its high content of total dietary fibers and phenolic compounds. With such a diverse nutrient profile, raspberry is recognized as a natural functional food with multiple beneficial properties, including potent antioxidant capacity, anti-inflammatory, and antimicrobial effects. The aim of the research was to investigate the impact of biostimulants on the chemical composition of raspberry fruit extract. The 'Vilamet' variety of raspberry was treated with three types of biostimulants: Epin-Extra®, a brassinosteroid phytohormone; Zircon®, a plant extract; and chitosan, an extract from plants and shells. Following the harvest, the ripe raspberry fruit was dried, powdered, and subjected to hydroethanolic extraction. The polyphenolic profile was determined by HPLC-DAD, and antiradical activity was analysed against DPPH and hydroxyl radical species using Electron Paramagnetic Resonance (EPR) spectroscopy. The results revealed that in all investigated samples, the most abundant were ellagic and syringic acids. The Zircon® treatment exhibits the highest concentration of polyphenols, especially in the case of ellagic and syringic acids, while the Epin-Extra® has the lowest concentrations of these two acids. Extracts of fruits treated with the chitosan biostimulant exhibit the highest antiradical activity, both towards DPPH (94.39%) and $^{\bullet}$OH (73.72%) radicals. Our research shows that chitosan biostimulant treatment can boost the antioxidant qualities of raspberry fruit extract, emphasising the role of farming methods in the bioactive composition of fruits and positioning chitosan as a valuable tool for enhancing raspberries' functional properties.

Keywords: raspberry, biostimulants, natural functional food

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PHYSICOCHEMICAL PROPERTIES AND FUNCTIONAL CHARACTERISTICS OF GREEK CRAFT BEERS

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Beer is the most popular alcoholic beverage in the world (when measured by volume of consumption). There has been a worldwide rise in the number of microbreweries or craft breweries in recent years. The term "craft brewery" typically refers to a brewery that produces small amounts of beer, usually made with traditional ingredients, but also with non-traditional ingredients to provide a unique flavor. In addition to being a lower-alcohol alternative to wine and spirits, beer contains several essential nutrients, making it a nutritious beverage. Craft beer is often unfiltered and unpasteurized, which preserves more functional compounds but reduces its shelf life compared to industrial beers. The present study aimed to examine the validity of this hypothesis. Craft beers from different brands, produced in Northern Greece were analyzed for their physicochemical characteristics, phenolic content, and antioxidant capacity (with DPPH and FRAP methodology) and compared with conventional/industrial beers from Greece. The results showed no significant differences in the main physicochemical characteristics of beers, apart from color, which was correlated with the production method of each beer. However, there were relevant differences in the content of phenolic compounds and antioxidant capacity of the analyzed beers. The results of total phenolic content ranged from 329 ± 9 to 702 ± 33 mg gallic acid/L, while in the case of antioxidant capacity, the results ranged from 0.63 ± 0.14 to 1.42 ± 0.14 mmol Trolox/L, and from 1.58 ± 0.01 to 3.03 ± 0.07 mmol Trolox/L, for DPPH and FRAP methodology, respectively. All craft beers presented higher values of total phenolic content of up to 83% and antioxidant capacity of up to 30-137% in FRAP and 13-155% in DPPH methodology compared to conventional/industrial beers. The results of the present study highlight the superiority in functional characteristics of craft beers compared to conventional/industrial beers.

Keywords: craft beer, total phenolic content, antioxidant capacity, color

THE INFLUENCE OF DIFFERENT CORN SYRUPS ON THE QUALITY PARAMETERS OF JELLY CANDIES

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Corn syrups are products obtained by acidic, enzymatic or combined acid-enzyme hydrolysis of corn starch. They are usually identified by their reducing power, expressed as dextrose equivalent (DE), which represents the total content of reducing sugar, calculated as D-glucose, based on the dry weight of the syrup. Corn syrups are known under different trade names, e.g. glucose-, malt-, high-maltose syrup etc. In the present study, three samples were prepared that were sweetened with different corn syrups (two glucose syrups and one maltose syrup) in combination with erythritol and stevia, as well as a control sample in which sucrose was used instead of corn syrup. The dry matter content of dry matter, DE and pH value were determined in the syrups used. The dry matter content and pH of the syrups were within a narrow range, i.e. between 82.0 and 82.7 and between 4.8 and 4.9 respectively. The DE of glucose syrups was 36.6 and 40.0 and for malt syrup 62.1. The dry matter content of the jelly candies produced was between 77.7 and 78.4, and the pH was between 3.90 and 4.01. The instrumentally examined textural properties of the jelly candies, i.e. hardness and gumminess, were lowest in the control sample and highest in the sample with the addition of glucose syrup at DE 36.6. The results of the sensory evaluation using the scoring method showed that overall quality of the control sample of jelly candies and the sample with glucose syrup with DE 40.0 was classified as very good, while samples of candies with glucose syrup with DE 36.6 and malt syrup belonged to the excellent quality category. It was shown that corn syrups can be successfully used as a substitute for sucrose in the production of jelly candies, without significantly changing the chemical parameters and sensory properties of the end products.

Keywords: corn syrup, jelly candies, sensory properties, textural properties

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EFFECTS OF THERMAL PROCESSING ON THE INFANT FOOD FUNCTIONALITY

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Human milk is widely considered the most beneficial source of nutrition for newborns. As compared to infant formula, human milk has more advantages. It contains essential nutrients and bioactive components that are important for the growth and development of the gut microbiota and immune system. To ensure that preterm babies receive adequate nutrients and healthy doses of bioactive components, human milk banks collect mothers' own or donor human milk, which undergoes thermal processing. Freeze storage and pasteurization are the thermal treatments that contribute to the microbiologically safe human milk, but they alter the nutritional value. The aim was to compare the effects of thermal processing on the functionality of human milk and different infant formulas. Thermal processes such as pasteurization and freeze storage can alter the lipid and protein properties and antioxidant capacity of preterm human milk. Moreover, the lipid content is further reduced when the milk is pasteurized after freezing. Thermal treatments decreased the activity of superoxide dismutase and glutathione peroxidase and influenced the lipid status and energy content of human milk. Although freezing followed by pasteurization is common in milk banks, it has a negative impact on the quality of milk. Therefore, to provide adequate infant nutrition and ensure greater lipid content and higher concentrations of antioxidant components, individual milk supplementation with quality fortifiers should be used in milk banks.

Keywords: human milk, thermal processing, pasteurization, functionality

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EFFECT OF VARIOUS ALTERNATIVE DIETS ON GROWTH PARAMETERS AND BIOMETRIC INDECES OF COMMON CARP (Cyprinus carpio)

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Fishmeal is used as one of the main protein sources in intensive common carp production. The use of this component makes the production of fish food more expensive, due to the decline of the population of wild fish used for its production, so there is an urgent need to find new sources of protein. Worms and other invertebrates are easy to grow and have minimal negative impact on the environment and as such they are candidates for replacing fishmeal in the fish diet. The experiment was carried out under controlled conditions in the fish nutrition laboratory at the Faculty of Agriculture, University of Belgrade. This study aimed to investigate the effects of a complete fishmeal based diet (IFM) replacement with total inclusion of earthworm diet (IEF), mealworm diet (ITM) and zooplankton diet (IZO). At the beginning and end of the feeding period, fish were weighed to calculate growth performance such as specific growth rate (SGR), body weight gain (BWG) and condition factor (CF), while the liver was weighed to determine hepatosomatic (HSI) index. Values for SGR and BWG had the same trend for all four experimental groups, with the highest values achieved in the group whose diet included mealworms, while the lowest value was achieved in the group in which the diet included fishmeal. Common carp fed ITM achieved similar CF values to the control diet, suggesting that *Tenebrio molitor* based diets can replace fishmeal (IFM) based diets. Also, values for HSI range from 1 to 2%. Finally, based on the results obtained, it can be concluded that alternative protein sources can be used in the common carp diet instead of fishmeal without a potentially negative impact on fish growth and health.

Keywords: fishmeal replacement; hepatosomatic index, condition factor

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FATTY ACID COMPOSITION OF RAINBOW TROUT (ONCORHYNCHUS MYKISS) FED MEALWORM, EARTHWORM, AND ZOOPLANKTON DIETS

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Feeding in aquaculture plays a pivotal role in the successful rearing of fish, with particular emphasis on diets that have a positive effect on the chemical composition of fish fillets. This study aimed to assess the effect of the total replacement of fishmeal with alternative animal protein sources on the fatty acid composition of rainbow trout fillets. The fishmeal diet (FD) was substituted with diets based on earthworms (ED), mealworms (MD), and zooplankton (ZD). The gas chromatography technique with a flame ionization detector was used to determine the fatty acid composition of rainbow trout fillets. The lipids from the fillets were extracted with a mixture of chloroform and methanol (2:1) and prepared for GC analysis according to the EN ISO method. The results of the fatty acid composition of rainbow trout fillets fed with ZD showed a higher content of saturated fatty acids (SFA) compared to the control fillets. However, replacing fishmeal in the rainbow trout diet with mealworm and zooplankton resulted in the increase of monounsaturated fatty acids (MUFA) compared to rainbow trout fed with FD, with the rise in MUFA attributed to elevated oleic acid levels. Fish fillets fed with ED and MD exhibited higher levels of n-6 polyunsaturated fatty acids (PUFA), due to higher linoleic acid (LA) content, but lower levels of n-3 PUFA, which was reflected in the lower n3/n6 ratio compared to the control fillet. In contrast, the replacement of fishmeal with zooplankton had lower n-6 PUFA and n-3 PUFA, but a higher n3/n6 ratio. Despite the change in fatty acid profile, the ratio of n-3 to n-6 PUFA in all fillets remained within the optimal range recommended for human health. In conclusion, rainbow trout fillets fed with alternative protein sources had favorable nutritional characteristics in terms of fatty acids.

Keywords: rainbow trout, polyunsaturated fatty acids, mealworm, earthworm, zooplankton

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ENCAPSULATED HORSERADISH LEAF JUICE: A POTENTIAL ALTERNATIVE TO SYNTHETIC ANTIOXIDANTS IN MAYONNAISE PRODUCTION

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Horseradish leaf is an under-researched source of phenolics with pronounced antioxidant potential. Due to the high biological activity of fresh horseradish juices, it is desirable to encapsulate them by spray-drying, a cost-effective one-step process suitable for scaling up production. The potentially harmful effects of synthetic antioxidants have led to an increasing demand for antioxidants from natural sources to maintain the oxidative stability of lipid-rich products. Therefore, this study aimed to compare the effect of encapsulated, spray-dried horseradish leaf juice within maltodextrin/alginate (MD/AL) and maltodextrin/gum Arabic (MD/GA) with the effect of a conventionally used synthetic antioxidant ethylenediaminetetraacetic acid (EDTA) on the mayonnaise oxidative stability, quality, and sensory properties.

Sunflower oil (75%), egg yolk (3%), vinegar (3%), sugar (3%), and salt (1%) were used for mayonnaise production. The water content (15%) was reduced by adding encapsulates (in an amount to achieve a total phenolic content of 400 mg gallic acid equivalents/kg mayonnaise). The mayonnaise containing EDTA was used as a positive control. Based on an accelerated oxidative stability test, MD/AL and MD/GA were found to be more effective than EDTA in delaying the mayonnaise oxidation, by prolonging the induction period (by 39 and 32%, respectively). The mayonnaise quality during the eight-week storage period was determined by measuring the pH and acid values. The horseradish encapsulates also improved the product quality with a higher pH (by 0.5-1.5%) and lower acidity (by 21.4%) after storage compared to the positive control. A nine-point hedonic scale was used for the sensory analysis of the mayonnaises. The overall acceptability of the mayonnaises followed the order: MD/AL>EDTA>MD/GA, with scores above 7 ("like moderately").

Finally, horseradish leaf juice encapsulates positively affected the oxidative stability, quality, and sensory properties of the mayonnaise, indicating the great potential of these natural antioxidants as a substitute for synthetic ones in the food industry.

Keywords: horseradish leaf juice, encapsulation, antioxidant activity, mayonnaise, oxidative stability

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EFFECT OF REPLACING WHEAT WITH PORCINO (*Boletus edulis*) FLOUR ON CONTENT OF FREE AND BOUND PHENOLIC ACIDS

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The porcino mushroom (*Boletus edulis*) has nutritional and medicinal importance. The free phenolics are solvent extractable, while the bound can be extracted only after hydrolysis. For better estimating the phenolics content and antioxidant activities, the bound phenolics (mainly bond to cell wall components by ester linkages) have to be considered too in particular in case of cereals and wheat. In this research the content and composition of free and bound phenolic acids were investigated in the mixture where 30% of wheat (type 500) was replaced by porcino flour. A porcino mushroom was from eko-region Piskupovo, Leskovac, Serbia. The free phenolics were extracted by methanol, and the bound phenolics were liberated after alkali hydrolysis, and then extracted by mixture of diethyl ether and ethyl acetate (1:1, v/v). Solvent was evaporated and dry residue dissolved in methanol. The analyse was performed on an Agilent 1100 Series HPLC system consisted of micro vacuum degasser, binary pump, thermostatted column compartment and variable wavelength detector. Column was Agilent Eclipse XDB-C18 4.6 mm IDx 150 mm (5 μm) 80 Å, the mobile phase, 5 mmol/l potassium dihydrogen phosphate solution (pH 2.5) and acetonytrile (41:9, v/v), elution was isocratic by flow rate of 1.0 ml min^{-1}, at temperature of 40 ^{0}C. The dosing volume was 30 μl, and acids were detected at 280 nm. In investigated extracts chlorogenic, gallic, protocatechuic, caffeic, genistic and *trans*-ferulic acid were detected, and their content was estimated from the calibration curves built for each acid separately. The total content of detected acids in the extract of free phenolics was 257.17, and the bound, 529.60 μg/g. Among the free phenolics, protocatechuic acid had the highest content (166.69 μg/g dry mass), while *trans*-ferulic acid (456.57 μg/g dry mass) was the most abundant in the bound fraction. Replacing the wheat with porcino flour increased the content of detected phenolics acids by about 10%.

Keywords: content, HPLC, mushroom, phenolic acids

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VALORIZATION OF SELECTED PLANT EXTRACTS AS FUNCTIONAL INGREDIENTS IN FERMENTED BEVERAGES

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For millennia, people have incorporated botanical ingredients in fermented beverages to enhance flavor and increase their health-beneficial effects. Thus, a global market has developed that encompasses a wide range of fermented products and is worth a total of 2.02 trillion USD in 2022. This expanding market is driven by the rising popularity of probiotic food and beverages among consumers. However, modern technology, offers new opportunities with their sensorial enhancements and medicinal effects. Herein, we evaluated antioxidant and antimicrobial properties of *Crataegus monogyna* (Jacq.) and *Rosa canina* L. aqueous and ethanolic extracts. This is quite important as estimates suggest that neurodegenerative and cardiovascular diseases may be triggered by increased oxidative stress, while certain bacterial skin infections are associated with vascular complications in diabetes mellitus. Incorporation of plant extracts as a mean to enhance sensory properties of fermented beverages is expected to provide health-beneficial properties as well. Our results showed that the tested extracts have very good antimicrobial activity towards pathogenic microorganisms including *Bacillus cereus*, *Aspergillus flavus* and *Candida albicans* as the most susceptible to the activity of the tested extracts. Aqueous extracts was more active thant the ethanolic, with MIC value in range of 0.50-1.00 mg/mL, and MBC/MFC in range of 1.00-2.00 mg/mL. As for the antioxidant activity, both aqueous and ethanolic extracts showed quite promising potential. Ethanolic extract of *C. monogyna* showed better antioxidant potential than the aqueous (EC_{50} 0.47 mg/mL and 5.1 mg/mL, respectively). In *R. canina*, the aqueous extract neutralized free radicals more efficiently than the ethanolic extract. This activity may be ascribed to high content of phenolic compounds, with the highest amount detected in the aqueous extract of *R. canina* – 412.25 mg/mL. A more in depth chemical analysis showed that both samples had high levels of benzoic and hydroxybenzoic acid in *C. monogyna* (7.409 and 19.781 mg/100g lyophilized extracts, respectively), while dihidroxybenzoic acid, ellagic acid and methylellagic acid were the most abundant in *R. canina* (187.849, 37.070 and 26.850 mg/100g lyophilized extracts, respectively). Given the specific flavor of the extracts and their proven bioactive properties, we believe they would be excellent ingredients for fermented beverages.

Keywords: plant extracts, food enhancers, bioactivity

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ULTRASOUND AND NATURAL DEEP EUTECTIC SOLVENTS AS TOOLS FOR IMPROVING EXTRACTION YIELD AND ANTIOXIDANT POTENTIAL OF WILD THYME EXTRACTS

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The aerial part of wild thyme has widespread use in traditional medicine for treating gastrointestinal and respiratory disorders, rheumatism, menstrual pain, wounds, and eczema. Its extracts possess antioxidative, anti-inflammatory, antihypertensive, antibacterial, and antifungal properties. Ultrasound extraction is a novel procedure used to isolate various plant bioactive compounds, due to the increased extraction yield, fast kinetics, and simple operation. Furthermore, natural deep eutectic solvents can increase phenolic recovery from plant matrix. In the present research, the extracts were prepared using dried wild thyme herb and water, 35% ethanol, or two types of natural deep eutectic solvents with 50 % water - betaine+citric acid and citric acid+saccharose in an ultrasound bath. The obtained extracts were characterized *via* analyzing the total phenolic and flavonoid contents (TPC and TFC, respectively) and antioxidant capacity ($ABTS^{\bullet+}$ and $DPPH^{\bullet}$ assays). The TPC of water, ethanol, betaine+citric acid, and citric acid+saccharose extracts were 25.08±2.3, 27.5±1.2, 30.2±2.9, and 28.1±3.3 mg gallic acid equivalents (GAE)/g of dried plant material, respectively. At the same time, the TFC values amounted to 10.5±0.8, 13.1±1.3, 1.0±0.5, and 2.8±0.1 mg catechin equivalents (CE)/g of dried plant material, respectively. In the $ABTS^{\bullet+}$ assay, antioxidant activity was 7.7±1.0 µmol Trolox equivalent (TE)/g of dried plant material (water extract), 8.5±0.9 µmol TE/g (ethanol extract), 10.8±0.11 µmol TE/g (betaine+citric acid extract), and 17.0±0.7 µmol TE/g (citric acid+saccharose extract). DPPH radical scavenging capacity was expressed as IC_{50} (concentration of the extract required to neutralize 50% of free DPPH radicals); IC_{50} was 4.3±0.2 mg/mL for water extract, 4.0±0.5 mg/mL for ethanol extract, 3.5±0.2 mg/mL for betaine+citric acid extract, and 2.9±0.1 mg/mL for citric acid+saccharose extract. pH values for ethanol, betaine+citric acid, and citric acid+saccharose extracts were 6.97, 6.18, 2.47, and 1.60, respectively. Due to higher $ABTS^{\bullet+}$ and $DPPH^{\bullet}$ scavenging activity, wild thyme extract obtained using citric acid and saccharose with 50% water compared to water, ethanol, and betaine+citric acid extracts was favored as an ingredient in food and pharmaceutical products.

Keywords: antioxidants, natural deep eutectic solvents, phenolics, ultrasound, wild thyme

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OPTIMIZATION OF PHENOLS EXTRACTION FROM GRAPE POMACE BASED ON NON-IONIC SURFACTANTS

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The winemaking process generates large amounts of by-products, whereas grape pomace residue might account for approximately 20-30% of the mass of total processed grapes. Grape pomace is considered both an environmental issue and a rich source of biologically active compounds. There is a growing scientific and industrial interest in the valorisation of winemaking waste and the effective phenolics recovery. The modern approach to the phenolics extraction has led to the development of efficient and environmentally-friendly methods. The methodology suggesting the use of aqueous solutions of surfactants represents a novel approach to the conventional solid-liquid extraction. The factors affecting the efficiency of micelle-mediated extraction of phenolic compounds from red grape pomace were investigated and the optimal conditions for the extraction were determined using Box-Behnken experimental design. Three independent variables (pH value, time and solvent-to-material ratio) to enhance the phenolics recovery from grape pomace by aqueous solution of non-ionic surfactant Brij S20 (3% (w/V)) were examined. The response variable was total phenolic content, determined by the Folin-Ciocalteu method. Applying response surface methodology, the second order polynomial regression equation was derived. The developed model showed a good accuracy. It was concluded that linear coefficients representing time and solvent-to-material ratio are significant and these individual process variables showed the highest level of contribution. It was shown that optimal conditions for micelle-mediated phenolics extraction were pH 3, an extraction time of 90 min, and solvent-to-material ratio of 150 ml/g. Under the optimal conditions, the predicted total phenolic content was 38.01 mg GAE/g dry weight, while the experimentally obtained value was 40.42 ± 2.83 mg GAE/g. Therefore, the proposed extraction technology by using Brij S20 aqueous solution as a solvent allows obtaining the extracts with high phenolics content which are suitable for the applications in food, cosmetic, and pharmaceutical products.

Keywords: extraction, polyphenols, grape pomace, optimization

EFFECTS OF HEN FEED ENRICHED WITH OLIVE EXTRACT ON FATTY ACID PROFILES EGGS -PILOT STUDY

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Olive oil has beneficial effects on health, lowering blood glucose level, blood pressure, decreasing inflammation and adiposity, and it was confirmed in many studies. Bioactive compounds in olive oil are phytosterols, triterpenoids, phenolic compounds, tocopherols, and fatty acids. In our experiment olive pomace oil extracted from a dried pomace was used in enriching the han food. Eggs present essential food some classified it as "superfood" and are widespread used in everyday life.

Fatty acid profiles in animal models using plasma, erythrocytes, organs are influenced by animals food intake as well as animal endogenous metabolism status.

In our study we analysed the effects of feed enriched with olive extract (dried oil pomace extracted from dried pomace and pulp), Lucta, SA, Spain. That food was used for feeding Hans on the farm (Panovo, Srbija) in well-controlled conditions. We followed the experiment for one month feeding and analysed hans eggs fatty acids profiles, comparing them to those eggs of hans fed with standard food.

Fatty acids profile were determined using gas-liquid chromatography (GC), with our standard laboratory procedure.

Our results showed that oleic acid was increased in food enriched with olive extract compared to usual han food (standard). We also examined the fatty acids profile in eggs of those hans and results showed an increase in oleic acid compared to eggs from hans using standard food.

Concerning the fact that oleic acid has beneficial effects on human health and that eggs as a essential food are very commonly used in Serbian nutrition, our aim is to further examine (more examined eggs) their health effects. One day, those eggs could be commonly used in everyday diet and bought in markets. Further research would be vital to unlock its full benefits.

Keywords: hans, eggs, fatty acid profile, olive extract

Acknowledgements: our study were conducted at Panovo farm and in colaboration with Lucta, SA, Spain.

CHEMICALAND SENSORY CARACTERISTICS OF WINE OBTAINED FROM SELECTED GRAPEVINE GENOTYPES OF THE PROKUPAC VARIETY

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The aim of this study was to compare the chemical composition and sensory characteristics of wines derived from different clones of the Serbian grape variety Prokupac. The wines were produced from ten selected genotypes of the Prokupac variety (1T, 2T, 3T, 4T, 6T, 8T, 9T, 15T, 17T and 18T) originating from the Tri Morave wine region, Serbia, using the microvinification method. After destemming and crushing the grapes, $K_2S_2O_5$ was added at a rate of 5 g per 100 kg as well as the enzyme preparation EXV (Lallemand, Canada) (2 g/100 kg). Alcoholic fermentation began with inoculation of the *Saccharomyces cerevisiae* yeast strain (Lalvin V1116, Lallemand, Canada) in amount of 30 g/100 kg and maceration lasted nine days. To evaluate the chemical parameters of the wines obtained, total acidity, pH, alcohol content, hue and colour intensity, total phenolic content, Folin-Ciocalteu index and total anthocyanin content were analysed according to the Compendium of International Methods of Wine and Must Analysis. The total acidity ranged from 3.40 to 5.70 g/L tartaric acid and the pH values were between 3.70 and 4.21. The lowest alcohol content was 12.3 vol% for the 17T wine and the highest was 15.0 vol% for the 2T wine. The highest total phenolic content (1480 mg GAE/L) and Folin Ciocalteu Index (19.20) were found in 4T wine. The highest total anthocyanin content was found in 2T wine at 101.50 mg/L. In terms of hue and colour intensity, the hue value was 0.70 in most wines and the colour intensity varied between 1.30 and 2.10, which is typical of young wines. In terms of sensory characteristics, wine 2T showed the best sensory characteristics. The colour of the wine was dark ruby red. The aroma was the most complex with a pronounced fruitiness characteristic of Prokupac. Rounded taste with a nice balance between alcoholic sweetness, tannins and acids.

Keywords: Prokupac, wine, grapevine genotypes, phenolics, sensory properties

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EXPLORING NADES AS CO-SOLVENT FOR ENHANCED EXTRACTION OF ANTIDIABETIC MOLECULES FROM CORN SILK

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Diabetes mellitus affects over 463 million people globally with increasing prevalence. Research now focuses more on natural molecules with antidiabetic properties, and one source of such biomolecules is corn silk (*Zea mays* L.), whose effects have been recognized by the traditional medicine in many cultures. The presented research focuses on developing corn silk extraction process with the aim of obtaining extracts with a high effectiveness in treating diabetes. Corn silk was extracted using subcritical water extraction with/without a co-solvent. A natural eutectic solvent (NADES) composed of L-proline and lactic acid was used as the co-solvent. The obtained extracts were characterized in terms of total phenolics (TPC) and flavonoids (TFC) content, antioxidant activity, and their ability to inhibit α-amylase and α-glucosidase. The results showed that the use of NADES resulted in a significant increase in TPC (from 124.43 to 255.83 mg GAE/g DW) and TFC (from 25.37 to 38.29 mg RE/g DW). Additionally, in terms of antioxidant activity, extracts obtained with co-solvent showed a higer ability to neutralize DPPH radicals (IC_{50}=0.02 mg/mL) compared to those obtained without co-solvent (IC_{50}=0.03 mg/mL). Regarding reduction capability, extracts obtained with the co-solvent had a 53% better effect on $Fe^{3+}\rightarrow Fe^{2+}$ ions reduction. The use of co-solvent significantly enhanced the ability of extarct to inhibit α-amylase and α-glucosidase. Specifically, for amylase, extracts with co-solvent showed a 100% better effect compared to extracts obtained without co-solvent (7.05 mmol AE/g and 14.98 mmol AE/g DW, respectively). In the case of glucosidase, the effect of extracts with the co-solvent was 64% higher. The combination of L-proline and lactic acid has the ability to form complexes that interact with enzymes, slowing down the process of carbohydrate degradation and reducing glucose release into the bloodstream. These results opens up promising avenues for the application of corn silk in the formulation of official preparations for diabetes treatment.

Keywords: corn silk, antidiabetic effect, extraction, subcritical water, NADES

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VIRGINIA TOBACCO: HOW DRYING AFFECTS QUALITY

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As the first technological step in the tobacco processing procedure, drying determines the direction of the transformation of substances in the tobacco leaf. It is therefore the most important step, as it also has a direct effect on the quality of the final product. The paper discusses the possibilities of different methods of drying for Virginia tobacco, from standard drying (flue cured - FC) to atypical drying (air cured - AC, sun cured - SC, freeze-dried) as well the digitization of individual drying stages and the use of new energy sources. It was found that atypical drying methods change the chemical properties of tobacco, which would make them unfavorable for the production of Virginia cigarettes with a pleasant taste and average strength of tobacco smoke. The chemicals in the freeze-dried tobaccos are basically not converted, as most enzymes lose their activity at temperatures below 0°C. Virginia dried using the AC method comes close to dark tobaccos in terms of its properties. The content of nitrogen substances, ash and sand increases significantly, while the content of soluble sugar decreases. SC Virginia contains larger quantities of undecomposed substances with a high molecular weight (protein and starch). The FC method provides the best results, especially when improving and digitizing the process. In addition, the atypical drying methods last longer than the usual 6 days when the process is carried out in special dryers with controlled conditions.

The greening of Virginia's drying process through the use of biomass briquettes as an energy source has led to good results in terms of energy and environmental efficiency. The growing awareness of sustainability and environmental protection will also influence approaches to tobacco drying. The combination of traditional processes with new alternatives can offer opportunities for the production of diverse and high-quality tobacco products.

Keywords: Virginia tobacco, digitization, greening, drying methods, chemical properties

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Section
FOOD QUALITY AND SAFETY

ANTIDIABETIC AND CYTOTOXIC POTENTIAL OF THE SPONGES FROM THE ADRIATIC SEA

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A balanced diet can promote health and prevent disease. Modern-day diet is based on excessive consumption of processed foods, sugary drinks and, unhealthy fats, that may increase the risk of cancer and diabetes. Natural products can be used as support to the treatment of diabetes, particularly Type 2, by acting as inhibitors of carbohydrate-hydrolysing enzymes, such as α-glucosidase. Inhibition of this enzyme is important in order to slow down the absorption of carbohydrates from the gut after a meal, decreasing postprandial hyperglycemia. Additionally, people with diabetes mainly Type 2 are at increased risk for cancer, especially liver, pancreatic, colorectal, endometrial, breast and bladder cancer.

Extracts from marine sponges collected from the Adriatic Sea, Boka Kotorska bay (*Ircinia variabilis*, *Axinella cannabina*, *Aplysina aerophoba*, *Agelas oroides*, *Sarcotragus spinosulus*, *Chondrilla nucula*, *Petrosia ficiformis*) were analyzed for antidiabetic and cytotoxic potential. Extracts were obtained by supercritical CO_2 extraction. Antidiabetic potential was examined using α-glucosidase inhibition assay. Cytotoxic potential was tested using MTT test against three human malignant cell lines: HeLa (cervical adenocarcinoma), LS174T (colorectal adenocarcinoma), and A549 (lung adenocarcinoma), as well as against human normal lung fibroblasts (MRC-5).

The results of sponge extracts on the inhibition of the α-glucosidase enzyme showed that five out of seven sponge extracts showed better anti-α-glucosidase inhibition than the standard drug acarbose. The highest α-glucosidase inhibition exhibited *Sarcotragus spinosulus* extract with IC_{50}: 84.89±0.71μg/mL. The obtained data of cytotoxic evaluation indicate that the most of the tested extracts of sponges demonstrated considerable to moderate cytotoxic activity against the examined malignant cell lines. The highest cytotoxic activity had *Sarcotragus spinosulus* extract (IC_{50}: 46.88±7.29μg/mL) against HeLa cells. The obtained data did not show selectivity towards normal MRC5 cells, so future toxicity studies are required.

Overall extract of tested sponges showed promising antidiabetic and cytotoxic potential.

Keywords: diabetes, a-glucosidase, marine sponges, anticancer activity, cytotoxic

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PRELIMINARY STUDY ON SENSORY ACCEPTANCE OF VARIOUS FRUIT AND VEGETABLE JUICES ENRICHED WITH GREEN AND BLUE SPIRULINA POWDER

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Spirulina (*Arthrospira platensis*) has become increasingly popular as a dietary supplement in recent decades due to its remarkable nutritional properties and sustainable cultivation. Spirulina is a blue-green microalgae that is usually sold as a dietary supplement in the form of powder, tablets or capsules. The natural colour of spirulina powder is green, but it can be purified to concentrate phycocyanin, which is characterised by vibrant blue colour. However, its consumption as a food ingredient is far less common due to its specific sensory properties. On the other hand, fruit and vegetable juices can serve as carriers for various food ingredients due to their mass consumption and pleasant sensory properties. Therefore, the aim of this study was to perform a preliminary sensory evaluation of different fruit and vegetable juices enriched with green and blue spirulina powder.

Two fruit juices (apple and sour cherry) and two vegetable juices (tomato and celery) were selected for fortification with spirulina due to their popularity and health benefits, while juices without added spirulina powder were used as control samples. Green and blue spirulina powder was added to all juices at two concentrations (0.8% and 1.6% w/w) per serving (250 ml). Overall sensory acceptability was assessed by eight trained panellists on a seven-point hedonic scale (from 1=extremely dislike to 7=extremely like). Twenty juice samples were coded with random three-digit numbers and the evaluation was carried out under controlled sensory laboratory conditions.

Juice samples with a hedonic score of more than 4 were considered acceptable in terms of their sensory characteristics. These samples were selected to be included in further experiments, together with the control juice. Sour cherry juice with the addition of 0.8% and 1.6% blue spirulina powder and tomato juice with the addition of 0.8% and 1.6% blue spirulina powder were selected for further testing.

Keywords: spirulina, juice, sensory analysis, acceptance test, hedonic scale

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OCHRATOXIN A IN CHILDREN'S FOOD: IMPLICATIONS ON CHILDREN'S HEALTH

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Ochratoxin A is a mycotoxin produced by certain molds in cereals. Its presence causes numerous side effects, including nephrotoxicity, immunotoxicity, and carcinogenicity. Infants, with their developing immune and renal systems, are particularly susceptible to these adverse effects. Consequently, the assessment of ochratoxin A levels in cereal products for infants and young children is very important in order to protect children's health. Because of the above, the legal legislation prescribes a maximum level of ochratoxin A in cereal-based baby food, which is 0.50 mg/kg. High-performance liquid chromatography (HPLC/FLD) was used as an analytical method for determining the content of ochratoxin A. A total of 14 samples of baby food containing cereals were examined, of which 9 samples were fruit and vegetable porridge with cereals, and 5 were salty porridge with cereals (porridge with vegetables, mushrooms, fish, milk). The results of the analysis for all tested samples were below the detection limit of 0.5 mg/kg, which is in accordance with the legislation prohibiting the presence of ochratoxin A in cereals. Assessment of ochratoxin A content in cereal products for infants and young children is essential to preserve pediatric health and mitigate risks associated with mycotoxin exposure. Continued research, surveillance, and regulatory efforts are imperative to effectively save the population's public health. The conducted research showed that products intended for infants and small children on the market of the Republic of Srpska are safe in terms of presence of ochratoxin A.

Keywords: ochratoxin A, baby food, cereals, contamination

UNIFORMITY OF MASS OF SINGLE-DOSE HERBAL FOOD SUPPLEMENTS - QUALITY CONTROL

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In order to ensure the availability of products of satisfactory quality for every consumer on the market, it is necessary that food supplements meet all the quality and safety requirements prescribed by current legislation and standards, including pharmacopoeia. Food supplements in the form of tablets, capsules and sachets must meet the pharmacopoeia requirements related to uniformity of mass of single-dose preparations. Also, herbal raw materials often vary in moisture content and also can be insufficiently homogenized during the formulation of the final product, so uniformity of mass can be one of the first indicators of inadequate quality of single-dose products. Therefore, the aim of this research was to examine the uniformity of the mass of herbal food supplements in the form of tablets, capsules and sachets. The gravimetric method for determination of the percentage deviation from the average mass was used (Ph.Eur.10.0_01/2008:20905). For capsules and sachets, it is prescribed that only two individual masses may deviate more than 7.5% from the average mass of 20 capsules/sachets, and none deviates more than 15% from the average mass (average mass of 300 mg and more). For tablets, the condition applies that only two individual masses may deviate more than 5% from the average mass of 20 tablets, and none may deviate more than 10% from the average mass (average mass of 250 mg and more). 15 samples of herbal food supplements were tested, of which 11 samples were in the form of capsules, 3 samples in the form of tablets and 1 sample in the form of sachets. All tested samples correspond to the requirements prescribed by the Pharmacopoeia. The conducted research showed that examined herbal food supplements present on the market of the Republic of Srpska are of satisfactory quality.

Keywords: herbal food supplements, uniformity of mass

ANALYSIS OF TOTAL SULFUR DIOXIDE CONTENT IN FOOD AND BEVERAGES BY MODIFIED MONIER-WILLIAMS METHOD

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Sulfur dioxide (SO_2) and sulfite salts, known as sulfiting agents, are food additives. Due to antimicrobial, antioxidant, and anti-browning properties, these compounds are used as preservative and for color retention in various food and beverage products, including wine, beer, dried fruits, and potato-based products. However, dietary sulfite intake presents safety concerns due to possible allergy-like reactions in certain sensitive people. Therefore, they presence must be declared as allergens on the food label if their level exceeds 10 mg/kg or 10 mg/L (expressed as SO_2). This study aimed to analyze the total SO_2 content of commercial products from different food categories by modified Monier-Williams methods. The total SO_2 content in wines, beers and fruit and vegetable-based products were 80.4-192 mg/L, 9.4-46.5 mg/L, and 14.5-83.7 mg/kg, respectively. Among these food product categories, there were no products exceeding national maximum permitted levels for sulfites. However, although European and national legislation restrict usage of sulfites in meat, there were large number of non-compliant samples of minced meat and meat products with high-sulfite concentrations, in the range 44.8-530.7 mg/kg. These results indicate need for regular control sulfites in foods, especially in meat products for ensure food safety for consumers.

Keywords: additives, preservatives, antioxidants, sulfites, foods

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ASSESSMENT OF THE GLUTEN CONTENT IN FRUIT AND VEGETABLE FOODS FOR CHILDREN

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Recent studies have shown that exposure to gluten during childhood, especially in infants with a genetic predisposition, may increase the risk of developing gluten-related disorders such as celiac disease. As the introduction of baby food is permitted for infants and young children from 4 months of age, it is extremely important to monitor the safety and quality of this product group, including testing for gluten content in order to provide appropriate nutritional recommendations and reduce the risk of adverse health effects. Also, the possibility of cross-contamination during production highlights the importance of testing the gluten content of baby foods, especially those labeled “gluten-free”. The aim of this work was to investigate the gluten content in fruit and vegetable baby food for infants and young children labeled "gluten-free". The ELISA method was used to determine the gluten content. 19 samples of baby food for infants on the market of the Republic of Srpska were analyzed, of which 12 samples were fruit porridges, 3 porridges from a combination of fruit and vegetables and 4 fruit porridges with the addition of rice. All samples were labeled as "gluten-free", which implies that the gluten content in the product must be less than 20 mg/kg, which was verified. Considering that the presence of gluten can have potentially negative effects on the health of infants and young children, especially if there is a hypersensitivity to gluten or a predisposition to develop one, it is very important to regularly check the safety of food for children. The tests carried out have shown that the products intended for infants and young children on the Republic of Srpska market are safe to consume.

Keywords: gluten, children's porridge, Republic of Srpska

HEAVY METAL CONTAMINATION OF HERBAL FOOD SUPPLEMENTS

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Heavy metal contamination in herbal food supplements is one of the major public health risk factors worldwide. Heavy metals, including lead, cadmium, mercury and arsenic, can accumulate in herbal raw materials and dietary supplements through various sources such as soil, water, manufacturing processes, etc. Chronic exposure to these metals is associated with a number of adverse health effects, including neurotoxicity, nephrotoxicity, carcinogenicity, etc. In the Republic of Srpska, as well as in EU countries, legislation prescribes maximum levels of contamination of food supplements with lead, cadmium and mercury (3.0, 1.0 and 0.1 mg/kg, respectively). Considering the possibility of arsenic contamination in herbal raw materials, the guidelines and standards suggest that the arsenic content in plant-based products should also be investigated. The aim of this study was to investigate the potential heavy metal contamination of herbal food supplements. An atomic absorption spectrophotometer (AAS) was used to determine the lead, cadmium and arsenic content and a direct mercury analyzer was used to determine the mercury content. 28 samples of herbal food supplements were tested, of which 15 samples were in liquid form and 13 samples were in the form of tablets, capsules and sachets. All samples tested showed that the lead, cadmium and arsenic content was below the detection limits. The mercury content was below the detection limit in most samples, while an extremely small amount of mercury was detected in some samples (0.005 to 0.016 mg/kg), which is still far below the permissible contamination limits. The tests carried out have shown that the products tested are safe for consumption. However, continuous monitoring of the safety this product group is necessary to protect the health of consumers.

Keywords: heavy metals, contamination, herbal food supplements

DETERMINATION OF FLUORIDE IN INFANT JUICES USING FLUORIDE ION-SELECTIVE ELECTRODE

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Infant juices (concentrated liquids) and baby food in the form of porridge are the main sources of fluoride intake during infancy, alongside breast milk. In addition, fruits and vegetables used to make infant juices play an important role in the fluoride content of infant formula supplements. Monitoring the fluoride content in such formulations is crucial to ensure the safety, quality, and optimal health of the youngest consumers. An unbalanced intake of this ion can lead to diseases such as dental fluorosis and skeletal bone disease, and high fluoride concentrations can have a negative impact on brain development and cognitive abilities in newborns.

The aim of this study was to determine fluoride in 17 samples of selected infant juices available on the Serbian market from four different manufacturers using fluoride ion-selective electrodes, as a rapid, reliable, robust, economical, and environmentally friendly method.

The method was applied without significant prior sample preparation, using the standard addition method, and the fluoride concentrations ranged from 0.0845 to 0.2810 mg/L.

The results obtained were below the values recommended by the European Food Safety Authority (EFSA) and the Scientific Committee on Food (SCF) for fluoride ion concentrations.

Considering that children, especially infants, are the most vulnerable to the harmful effects of fluoride, more attention must be paid to the consumption and contribution of this element from other food sources. From this point of view, health authorities should emphasize fluoride as an additional toxic element and carry out more controls on infant feeding.

Keywords: fluoride, baby formula, food safety, ion-selective electrode

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NUTRITIONAL COMPOSITION OF PLANT-BASED MEAT SUBSTITUTES AVAILABLE ON SERBIAN MARKET

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The consumption of food of animal origin carries the risk of chronic, non-communicable diseases. In addition, the production of food of animal origin, especially red meat, contributes significantly to greenhouse gas emissions. For all these reasons, there are recommendations to replace meat with suitable plant-based substitutes that are rich in proteins. This study aimed to analyze nutritional composition of 39 plant-based meat substitutes on the market of the Republic of Serbia was analyzed. The information on the ingredients of the products and the available nutritional information were taken from the packaging labels, whereupon the nutritional composition was compared with the nutritional composition of the corresponding meat category. The protein sources in the plant-based meat substitutes were legumes, grains, seeds and nuts, with the nutritional composition varying significantly within the same category. In all product categories analysed, meat substitutes have a higher energy value (1203.10 ± 542.11 vs. 932.15 ± 426.13 kJ/100 g) and lower values for total fat (12.00 ± 6.23 versus 16.45 ± 6.46 g/100 g) and saturated fat (1.85 ± 0.45 vs. 4.06 ± 4.00 g/ 100 g). For certain product categories, a higher average protein content was found in plant-based meat substitutes compared to animal-based products, such as sausages (12.63 ± 2.30 vs. 18.15 ± 7.65 g/100 g), steaks (19.69 ± 3.56 vs. 17.03 ± 2.26 g/100 g) and meatballs (15.66 ± 1.60 vs. 13.05 ± 1.80 g/100 g). From the nutritional information on the food labels, it can be concluded that plant-based meat substitutes have a more favorable nutritional composition. However, what is missing to obtain a complete picture of their nutritional potential as meat substitutes is data on micronutrients, such as vitamin B12 and iron content, as well as data on the bioavailability of nutrients considering the presence of antinutritive factors in plant meat substitutes.

Keywords: plant meat substitutes, nutritional information, proteins, energy value, fat

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UNRIPE WALNUT FRIUT EXTRACTS – PHENOLIC CONTENT AND ANTIOXIDATIVE ACTIVITY

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Walnut (*Juglans regia* L., Juglandaceae) is a deciduous tree mainly known for its delicious and nutritious nuts, a source for dermatocosmetic and phytoterapeutic products as well as a raw material in wood industry. It grows wild or semi-wild from the Southeast Europe to the Japan, but is also an important plantation-grown species. Walnut leaves are also used in folk medicine as an astringent, antiseptic or to stimulate the secretion of bile and for preparation of traditional herbal medicines for wound treatment. Unripe walnut fruits are used to prepare liqueur, which is also attributed with medicinal properties.

In this study, six samples of unripe walnut fruits were randomly collected from different region of Serbia in period May-June 2022. The samples were extracted with 70% ethanol following traditional procedure for liqueur preparation: maceration for 40 days on direct sunlight. After this period, extracts were filtered in dark bottles and kept at 4 °C. The content of total phenolics, flavonoids and juglon, as well as their anti-DPPH• activity, were determined spectrophotometrically. The content of total phenolics and flavonoids in investigated samples was ranged 0.78-2.08 mg gallic acid equivalents/ml and 1.10-1.62 mg catechin equivalents/ml, respectively. Both parameters showed clear decrease with fruit maturity: the highest values were in samples of unripe fruits collected in May. In all samples juglon was determined in very low quantities (up to 18.45 μg/ml). Antioxidant activity of analysed samples was also very pronounced (over 80% of DPPH• inhibition).

Obtained results pointed out ethanolic extracts of unripe walnut fruits as a rich and valuable sources of phenolics, with high antioxidative and thus potentially high biological activity.

Keywords: Juglans regia, walnut, polyphenols, DPPH

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HERBS AND SPICES SPICED WITH ALKALOIDS – OVERVIEW OF RASFF NOTIFICATIONS

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Herbs and spices have a long tradition of use in both home and industrial food preparation, aimed at enhancing the sensory characteristics of food. The aim of this study was to observe the occurrence of natural toxins (pyrrolizidine and tropane alkaloids) in herbs and spices. The data originate from the RASFF (**Rapid Alert System for Food and Feed**) database, whose primary objective is to facilitate the exchange of information, thereby enabling timely responses by relevant authorities in case of risks to public health associated with food. The period from 2010 to 2023 was monitored for the type "food", category "herbs and spices" during which 132 notifications were identified. It is noteworthy that no notifications were reported until 2018, attributed to the insufficient capacity of laboratory testing for natural toxins before (the announcement of) their inclusion in Regulation on maximum levels of contaminants in food. The majority of notifications result from official controls on the market (in 67 cases), while a smaller number originate from border controls (36 consignments) and the company's own check (29 cases). Only 6 notifications identified tropane alkaloids, while the remaining 126 were related to pyrrolizidine ones, present in exceptionally variable concentrations, reaching up to 150 mg/kg. In most cases, the raw materials originate from Turkey, and the predominant spices contaminated with pyrrolizidine alkaloids were oregano (approximately 42%) and cumin (approximately 40%) while the remaining 8% of notifications were related to more than 15 types of herbs and spices, including raspberry leaves, parsley and peppermint contaminated with tropane alkaloids. One-half of notifications has been classified as alerts. The risk decision for as much as 120 notifications has been assessed as serious. Considering that pyrrolizidine alkaloids are hepatotoxic and several of them (*lasiocarpine, monocrotaline and riddelliine)* are classified as possible human carcinogens, their presence in food is highly undesirable.

Keywords: pyrrolizidine alkaloids, tropane alkaloids, food safety, public health

MYCOTOXINS IN HERBS AND SPICES – OVERVIEW OF RASFF NOTIFICATIONS

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The primary task of the European Union Rapid Alert System for Food and Feed (RASFF) is to ensure rapid and efficient exchange of information about health risks associated with food, with the aim of reducing potential negative impacts on public health. Herbs and spices are a category of food commonly contaminated with mycotoxins, specifically aflatoxins, characterised as potent hepatocarcinogens, and nephrotoxin ochratoxin A. Bearing in mind the wide use of herbs and spices in culinary practices, the aim of the present study was to monitor such contamination, based on data from the RASFF database for the period 2010 – 2023. A total of 566 notifications were recorded for the food category „herbs and spices“ and hazard category „mycotoxins“. There were significantly more notifications concerning the presence of aflatoxins – 453 (80.0%), while the presence of ochratoxin A was noted in 124 samples. The majority of notifications resulted from border controls, with 413 cases (73.0%), followed by official control on the market (22.4%) and the company's own check (4.6%). In 227 cases (40.0%), the raw material originated from India, followed by a smaller number of notifications from Indonesia, Pakistan, Sri Lanka, Ethiopia, China, etc. The predominant spices contaminated with mycotoxins were chili, chili peppers and paprika (taken together, responsible for 47.2% of all notifications) and nutmeg (24.6%). Concentrations of aflatoxins were highly variable, in several cases exceeding 0.2 mg/kg (nutmeg) or even 0.3 mg/kg (peppers) of total aflatoxins (maximum allowed 0.010 mg/kg), The highest ochratoxin A concentration was recorded in organic dandelion root (0.570 mg/kg; 0.020 mg/kg allowed). The risk decision for as many as 468 notifications (82.7%) was considered as serious, resulting in 372 cases of border rejection. Considering the widespread view that RASFF reveals only the tip of the iceberg, the presented findings should be regarded as clear warning for public health professionals.

Keywords: aflatoxins, ochratoxin A, food safety, public health

CONTEMPORARY METHODS OF TESTING THE QUALITY OF *ROSA CANINA* EXTRACTS

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Rosa canina extracts have traditionally been used in nutrition due to their high vitamin C content and other bioactive substances such as polyphenols. The total antioxidant activity (TAC) of these bioactive components is an important indicator of the product's biological value. While standard spectrophotometric methods have been used to quantify TAC, the use of electrochemical methods offers significant advantages in terms of speed, cost, and sample preparation. This study aims to compare electrochemical and spectrophotometric methods for determining the total antioxidant capacity before and after *in vitro* digestion of *Rosa canina* samples. The TAC of dried fruit extract, the liquid phase after digestion, and the solid phase after digestion were determined using cyclic voltammetry, differential pulse voltammetry, and the ABTS spectrophotometric method. All samples for TAC detection were prepared by dissolving in water. The study found that the TAC of the fruit extract was significantly higher compared to samples after digestion, with the liquid phase having higher TAC compared to the solid phase. The results for TAC obtained by cyclic voltammetry and differential pulse voltammetry were in good agreement, while the results obtained by the ABTS method showed slightly lower values for samples after digestion. The time required to obtain the TAC by voltammetry is much shorter, as the analyses only take a few seconds and do not require additional sample treatments as with spectrophotometric methods. Our results provide valuable data guiding us into further research on electrochemical methods for rapid, routine determination of TAC in extracts before and after digestion. Electrochemical techniques can exceed the limits of spectrophotometric analysis, such as lower sensitivity and slower response, and are successfully applied as a replacement for long-running spectrophotometric methods.

Keywords: Rosa canina, total antioxidant activity, ABTS, cyclic voltammetry, differential pulse voltammetry

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HUMAN HEALTH RISK ASSESSMENT OF PTES IN CAPSELLA BURSA-PASTORIS L. MEDIK AND ITS EXTRACTS

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Owing to mild properties, wide availability, and low reported side effects, medicinal herbs have been traditionally utilized for thousands of years as a folk remedy, while almost 80% of the world population consumes it as a medical supplement of primary healthcare (WHO). Despite widespread distribution and a broad spectrum of benefits for human health, *Capsella bursa-pastoris* L. Medik is an underexplored medicinal species with a recorded significant accumulation capacity for potentially toxic elements (PTEs). In the present study, we investigated the abundance and mobility of PTEs (Ba, Co, Cr, Cu, Fe, Mn, Ni, Sr, and Zn) in the urban soil - *C. bursa-pastoris* system and comprehensively evaluated potential posing human health risks. Samples were collected in four Serbian cities – Belgrade, Bor, Vršac, and Sremska Mitrovica. PTEs concentrations in samples were determined using inductively coupled plasma optical emission spectroscopy (ICP-OES). The pseudo-total concentrations in soils surpassed the world mean levels. Cu, Zn, Sr, and Mn were the most abundant, whereby the calculated *Igeo* index suggested moderate to heavy soil contamination. Non-carcinogenic (HI) and carcinogenic risks (CR) associated with the ingestion, dermal, and inhalation exposure to the examined soils were within the acceptable ranges ($HI < 1$, $CR < 10^{-6}$). Although Sr, Cu, and Ni contents were elevated in the plant tissues (roots - up to 57.04 mg kg^{-1}, 23.36 mg kg^{-1}, and 7.90 mg kg^{-1}, and shoots – up to 60.78 mg kg^{-1}, 20.21 mg kg^{-1} and 6.26 mg kg^{-1}), estimated daily intake (EDI), target hazard quotient (THQ), and lifetime carcinogenic risk (LCR) demonstrated non-significant health risks by consuming *C. bursa-pastoris*, as a raw herb or herbal extracts. This study aimed to underscore the urgency of the PTEs' continual monitoring in soils and medicinal plants and highlight the importance of conducting comprehensive health risk assessments in providing safe medicinal plant utilization.

Keywords: safety, urban soil, contamination, medicinal plant, herbal extracts

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BIOACTIVITY PROFILING OF BERMET WINES: *IN VITRO* HYPOGLYCEMIC POTENTIAL

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Bermet is a specially flavored wine, which is made from grapes from vineyards on the slopes of Fruška Gora (Vojvodina, Serbia) with the addition of up to 26 different aromatic medicinal plants. This wine has been produced since the 15th century according to the traditional procedure, but until now data on chemical composition and biological activities of these wines are scarce. The aim of this study was to evaluated *in vitro* effect of 7 red and 5 white Bermet wine on α-amylase and α-glucosidase, digestive enzymes involved in carbohydrate metabolism. It is well known that polyphenols have certain biological activity and therefore can contribute to the health benefits of medicinal plants and wine. Therefore, quantitative analysis of 7 phenolic acids, 6 flavonoids, 2 stilbens, 15 anthocyanine glucosides, galactosides and arabinosides by HPLC-UV/VIS technique was applied to elucidate differences in samples phenolic profile. Also, the content of total phenols, tanins, flavonoids and monomeric anthocyanins were evaluated by spectrophotometric methods. Analysed Bermet wines had moderate hypoglycemic potential: activity ranged from 100 to 936 μq acarbose eq/mL of wine (α-amylase) and 1.79 to 46.6 mg acarbose eq/mL of wine (α-glucosidase). The most abundant polyphenol compounds in examined samples were gallic acid (2.3–51.3 mg/L) and catechin (to 19.5 mg/L). Stilben piceid was detected in some samples in higher content (0.3-9.9 mg/L) than resveratol (0.1-9.9 mg/L). Unexpectedly, anthocyanins were detected in only four samples. Presented results are just a part of intensive research on biochemical and health-safety aspect of the quality of Bermet wine. Overall results should elucidate possibility of health benefits of moderate consumption of Bermet wines, but also to contribute, at least partially, to the increase of recognition of Serbian Bermet wines on domestic and world market.

Keywords: Bermet wine, polyphenols, α-amylase, α-glucosidase

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ELEMENTAL PROFILES OF BERMET WINES AND ASSOCIATED HEALTH RISK

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Bermet is a sweet wine enriched with medicinal plants and spices, originally intended for medicinal purposes, but later produced as a regular dessert wine. It is a specialty of Fruška Gora wine region in northern Serbia. The importance of the wine elemental composition is multifaceted: certain chemical elements affect sensory properties, multi-element profiling could contribute to geographical origin differentiation and toxic elements have potential to endanger consumers' safety. The goal of the current study was to investigate elemental profiles and to assess health risk associated with Bermet wines. Elemental profiling (Be, B, Al, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Se, Sr, Mo, Cd, Sn, Sb, Te, Ba, Hg, Tl, Pb) of 12 bottled Bermet wines was conducted by ICP-MS analysis. Health risk was estimated for men and women taking into account several wine consumption scenarios: population average, regular drinkers only and chronic heavy drinkers according to the World Health Organization, and the Serbian national food consumption survey data. The study revealed great differences in elements concentrations. However, all samples exhibited hazard indices well below 1, corresponding to the negligible non-carcinogenic risk. The minimum level of margin of exposure (MOE) associated with lead nephrotoxicity and with carcinogenic risk of arsenic exposure indicated no risk (74 and 9, respectively, in both cases for men drinking wine at volumes reported in the national consumption survey). On the other hand, lifetime cancer risk approach applied to arsenic showed that at the mean level of exposure the tolerable risk of 1 extra lifetime cancer case per 100,000 persons was slightly exceeded in heavy drinking and national wine consumption scenarios, for both men and women. Careful approach and monitoring of the influencing factors (endogenous sources, conditions of grape growth and winemakers' interventions) can contribute to the optimisation of the wine elemental composition.

Keywords: wine, food safety, public health, Fruška Gora

INFLUENCE OF DIFFERENT LIGHT SOURCES ON ANTIOXIDANT STATUS OF CAULIFLOWER MICROGREENS

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Plants use light as the main source of energy for photosynthesis, which regulates numerous other processes related to plant growth and morphology. The influence of light of different spectrum depends on the plant species, so it must be optimized for each plant species and working conditions. The aim of the research was to examine the effect of quality of different light sources (LED red, blue, red+blue 1:1 and cold white) on germination rate, photosynthetic pigments content and antioxidants of cauliflower microgreens. The light intensity of 49 PPFD (Photosynthetic Photon Flux Density) was provided in all light treatments. Plants were grown in a growth chamber at a temperature of 23 ± 2 ºC and under a light regime of a long day (16 h day, 8 h night). The highest germination rate after 8 days was recorded under red+blue light treatment (61.7%), while the lowest was under blue light (9.2%). After 2 weeks of growing in growth chamber the photosynthetic pigments, phenolics and flavonoids content as well as antoxidant capacity (DPPH assay) were determined. The highest concentration of pigments (chlorophylls 4.12 mg and carotenoids 1.13 mg per gram of fresh sample weight) as well as content of phenolics (0.81 mg of gallic acid equivalents per g of fresh sample weight) and flavonoids (2.82 mg rutin equivalents per gram of fresh sample weight) was recorded under combination of red and blue lights, while the lowest was recorded under red light (1.49, 0.42, 0.63 and 1.90, respectively). Content of phenolics and flavonoids had positive and significant correlation with DPPH• antioxidative capacity (r=0.83** and r=0.87** respectively). The results showed that combination of red and blue lights was the best for microgreen antoxidant status.

Keywords: microgreens, LED lights, photosynthetic pigments, phenolics, flavonoids

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READY-TO-EAT SANDWICHES AS ASOURCE OF PATHOGENICBACTERIA

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Ready-to-eat (RTE) foods can be consumed without significant preparation and could be one of the main routes for the spread of foodborne pathogens: *Enterobacteriaceae, Staphylococcus aureus, Salmonella* and *Listeria monocytogenes*. The aim of this study was to determine the frequency of occurrence of foodborne pathogens and to investigate the relationship between the seasons and the number of bacteria. The study included 372 sandwich samples that were analyzed in the period 2021-2022. The sandwiches were sampled in catering establishments in the territory of Belgrade, according to the ISO 6887 standard. The microbiological analyzes were performed according to the valid ISO methods (ISO 21528-2:2009, ISO 6888-1:2009, ISO 6579:2008, ISO 11290-2:2017). API tests were also used in this study. The presence of bacteria from the *Enterobacteriacae* family was confirmed in 30.37% of samples from 2021, most frequently in samples in July and September, while in 2022 it was confirmed in 21.96%, in July and April. The presence of *S. aureus* was confirmed in 6.73% of 2021 samples, most frequently in December and July, and in 9.54% of 2022 samples, most frequently in March and April. *Salmonella* spp. and *L. monocytogenes* were not detected in any of the samples. *Enterococcus faecalis, Enterobacter cloacae, Enterobacter aerogenes, Citrobacter* spp., *Proteus vulgaris, Proteus mirabilis* and *Hafnia alvei* were identified with API kits. After summarizing all the collected results, it can be concluded that RTE foods are suitable for increased occurrence of pathogenic bacteria, and also their occurrence in different months of the year. To reduce the number of bacteria in RTE foods, certain measures should be taken, such as improving hygiene and disinfection of surfaces, proper storage of food and creating suitable environmental conditions.

Key words: RTE food, foodborne pathogens, sandwiches, ISO methods, API

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CENTRAL COMPOSITE DESIGN-BASED INVESTIGATION OFANTIOXIDANT PROPERTIES OF GOAT MILK / *LAETIPORUS SULPHUREUS* EXTRACT MIXTURES

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Functional foods and their health aspects have become one of the main topics in food science. Milk and dairy products have been enriched with various substances in order to increase its potential health benefits. These studies have mainly been performed on bovine milk, but data about goat milk are scarce. In this study, the antioxidant properties of mixtures of thermally treated skimmed goat milk (TM) / *Laetiporus sulphureus* aqueous extract (LA) were evaluated by a response surface methodology using a central composite design (CCD). The concentrations of TM and LA were selected as variable parameters in CCD. The mixtures were prepared according to a matrix based on 14 experimental runs obtained with Minitab software. Antioxidant properties were estimated via three methods based on different mechanisms: $ABTS^{\bullet+}$ scavenging activity, Ferrous ion chelating capacity, Ferric reducing power. For all tested antioxidant properties, a very good correlation was observed between them and investigated parameters ($R^2 \approx$ 94%; 98%; 97.5%, respectively). The $ABTS^{\bullet+}$ scavenging activity of mixtures was high (3.63 – 5.88 mg Trolox/ mL), due to the high casein activity, as well as the non-purified fungal extract probably owing its capacity to synergistic antioxidant activity between proteins and polysaccharides. The results were highest within the LA range 0.6 – 1 % and 6 % TM, with significant effect of LA concentration and the interaction of TM and LA concentrations. Chelating capacity of the mixtures depended only of TM concentration because of chelating properties of goat milk protein and peptides with the highest values for TM ≥ 10%. Conversely, the LA concentration was the only significant factor for the ferric reducing power, mainly due to the fungal polysaccharide content. The results presented indicate the use of different antioxidant methods for the examination of complex systems such as food. These mixtures have great potential for the formulation of functional food ingredients, using goat milk as a base and fungal extracts to enrich and improve functionality.

Keywords: thermally treated goat milk, Laetiporus sulphureus, central composite design, antioxidant properties

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PHYTOCHEMICALANALYSIS OF BALKAN JUNIPERS ESSENTIAL OILS

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Genus *Juniperus* L. (Cupressaceae) is the second largest conifer genus, with over 60 taxa distributed mostly in the northern hemisphere. These evergreen shrubs thrive in dry hills or mountainous tracts. The seed-cones ("berries"), leaves, wood are used for distillation and essential oils (EO) production. Juniper EO has a myriad of different activities. It is used as spices in food industry as coregent of flavour and smell substances in foods and beverages. Juniper berries stimulate digestion, enhance the body's resistance and expel uric acid and salt from the body. In this study, we analyzed EO composition of three species of junipers that grow wild in the Balkans, but have a very distinct distribution range - *Juniperus sabina, J. pheonicea,* and *J. foetidissima*. EO from leaves and berries were obtained using Clevenger type apparatus and their chemical composition was analyzed using GC/MS and GC/FID. Hydrodistillation yielded from 0.95 to 3.7%. Obtained oils had different relative density and organoleptic characteristics. The chemical composition varied based on the species and the plant organ used. In total, 108 compounds were identified, representing 96.9-98.7% of the total oil composition. The main components of the EOs were: sabinene in the leaves of *J. sabina* from both populations (44.23% and 37.5%), α-pinene in the berries of *J. foetidissima* (67.8%) and in all organs of *J. phoenicea* (63.28%-86.48%), and *allo*-cedrol (33.13%) in the leaves of *J. foetidissima*. Antioxidative capacity of essential oils was determined using DPPH assay and TLC bioautography. TLC bioautography showed several monoterpene and sesquiterpene fractions that were responsible for antioxidant activity. Unlike fractions, total oils had moderate to low antioxidant capacity, with *J. sabina* having the highest activity (IC_{50}=22.8 mg/ml). Based on the TLC bioautography and multivariate statistical tests, component responsible for antioxidative capacity were determined, enabling further investigations into isolation and application of these EO.

Keywords: Juniperus sabina, Juniperus phoenicea, Juniperus foetidissima, essential oil, antioxidant activity

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FATTY ACIDS PROFILES OF RAW AND ROASTED ALMOND, HAZELNUT AND WALNUT

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Nuts are commonly promoted as healthy and the possibility that nut intake may defend human health has been investigated worldwide. There is the recognition that nuts are a good source of many nutrients, including monounsaturated and polyunsaturated fatty acid (PUFA), vitamins E and K, selected minerals such as magnesium, copper, potassium, and selenium, dietary fibers, carotenoids, and phytosterols with potential antioxidant action. This study aimed to evaluate fatty acids profiles of raw and roasted almond, hazelnut and walnut. Analysis of total lipids was performed from about 1 g (1.077±0.009) of nut samples. Total lipids were extracted according to the method of Folch using 2:1 chloroform:methanol mixture containing butylated hydroxytoluene (0.05% BHT weight/volume). Fatty acids profiles were determined by gas- chromatography (GC). The content of individual fatty acids was expressed as a percentage of the total fatty acids. The main fatty acids of almond were oleic (66.12-64.86%), linoleic (22.02-22.03%), palmitic (8.62-8.73%) and stearic (2.17-2.95%) acids, while the dominant fatty acids of hazelnut were oleic acid (82.63-81.60%), followed by linoleic (7.25-8.76%) and palmitic acids (7.43-6.71%). The fat content of walnuts is very high. Additionally, walnuts represent nuts rich in α-linolenic acid (ALA). Walnut fat is mostly composed of linoleic (62.51-56.97%), linolenic (12.21-10.06%), oleic (12.85-15.20%), palmitic (8.68-12.93%) and stearic (2.82-3.19%) acids. There are noticeable changes in fatty acid compositions of nuts between groups. The most noticeable change is in roasted walnuts, which have significantly lower PUFAs compared to raw walnuts.

Keywords: nuts, amond, hazelnut, walnut, fatty acid profiles

A LINK BETWEEN DRY ROSELLE CALYCES CONTENT, PH VALUE AND PERCEPTION AND ACCEPTANCE OF THE COLOUR OF HERBAL TEAS

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The global food market is facing increasing consumer demand for health-promoting foods and beverages, which has led researchers to develop products rich in a range of bioactive and nutraceutical compounds. In this regard, the bioactive properties and health benefits of roselle, (*Hibiscus sabdariff*a L.), a plant from the hibiscus or mallow family (Malvaceae) contribute significantly to its growing popularity. Anthocyanins are among the most important phytochemicals responsible for the characteristic colour of roselle calyx. In the present study, blends of dried fruits and different plants, including the dried calyces of roselle flower (RC), were used for the preparation of 30 herbal tea samples. The content of RC in the blends was 20, 30, 35, 40 and 45% (w/w). The colour of the tea samples was sensory characterised by combining a free sorting task with verbal descriptions of the obtained sample groups. The sensory panel consisted of 30 students and staff, whereas acceptance tests were performed by 92 students from the University of Belgrade – Faculty of Agriculture.

The pH of the prepared herbal teas ranged from 2.82 to 3.31. The lower pH values of the tea samples were associated with higher proportion of RC in the herbal blends. Five clusters of tea samples, with colour descriptions ranging from 'dark red/burgundy' to 'yellow-orange/yellow', were formed by hierarchical cluster analysis applied to the sorting data previously analysed using the appropriate dimensional reduction techniques. The lowest proportion of red colour was found in the samples with the highest pH and lowest RC content (20%), where the colour was described as 'yellow-orange/yellow'. The average pH of these samples (3.30±0.02, n=3) was significantly higher ($p<0.05$) than the average pH of the samples in the other four clusters (3.01±0.10, n=27), which were not significantly different from each other in terms of pH. The colour of the samples with the higher RC content (40 and 45%) was described as 'dark red/magenta/burgundy' as well as 'orange-red' and 'intense amber', while the samples with RC content of 30 and 35% were characterised by 'red-orange/amber', 'orange' and 'brown-orange/topaz'. The acceptability tests showed that the colours of the tea samples with an RC content of 35-45% were the most acceptable ('dark red/burgundy' to 'red-orange/amber'), while the colour of the samples with the lowest RC content (20%) was the least acceptable ('yellow-orange/yellow'), at a statistical significance level of 0.05.

Keywords: Hibiscus sabdariffa, pH, color characterisation, labeled sorting, acceptance testing

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PHENOLIC COMPOSITION AND ANTIOXIDANT POTENTIAL OF DRY SWEET CHERRY FRUITS

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Three sweet cherry cultivars, 'Carmen' (early-medium ripening cultivar), 'Kordia' (late ripening cultivar), and 'Regina' (very late ripening cultivar) were studied for total phenolic content (TPC), total flavonoid content (TFC), total phenolic acids content (TPAC) and antioxidant potential. All cultivars have large to very large fruits with an average weight of 8 to 10 g. The fruits were picked at the full maturity stage, in the cherry orchard at the experimental farm "Radmilovac" of the Faculty of Agriculture in Belgrade. Drying was performed at the experimental dryer using the convective drying method (two constant air temperatures, 70 and 80 °C, air flow speed 4 m/s) until a constant mass was reached. The extracts of the dried fruits were prepared with methanol (1:5 ratio) in an ultrasonic bath (240 W, 35 kHz) at 25°C for 30 minutes. The results of TPC, TFC and TPAC were expressed as corresponding equivalents per gram of dry extract (gallic acid GAE/g, quercetin QE/g, and caffeic acid CAE/g, respectively). The antioxidant potential of the extracts was evaluated by total antioxidant capacity (TAC), DPPH radical and ABTS radical-cation scavenging potential assays and expressed either as equivalents of ascorbic acid (AA) or as IC_{50} values for $DPPH^{\bullet}$ and $ABTS^{\bullet+}$ assays.

The highest TPC was determined in the extract of 'Kordia' fruits dried at both temperatures (70 °C - 12.59 and 80 °C - 10.89 mg GAE/g), but without significant differences ($p>0.05$). The 'Kordia' cultivars dried at 70 °C had the highest TFC content (1.10 mg QE/g), which was significantly different from that dried at 80 °C ($p<0.05$), while no flavonoids were detected in 'Regina' and 'Carmen' (dried at 70 °C). On the other hand, cultivars 'Carmen' and 'Regina' dried at 70 °C had the highest TPAC, 3.49 and 3.96 mg CAE/g, respectively. In all three assays, the extract of 'Kordia' cherry fruits dried at 70°C stood out with the highest antioxidant potential, and differed significantly from the extract of the sample dried at 80 °C ($p<0.05$). The opposite trend was observed for 'Carmen' and 'Regina' cultivars, however with no significant differences between extracts of samples dried at 70 and 80 °C ($p>0.05$). The increased contents of polyphenols, flavonoids and phenolic acids in the extract of 'Kordia' cherry fruits were associated with the lower drying temperature of the fruits.

Keywords: Prunus avium L., drying, flavonoids, phenolic acids, antioxidant activity

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LIPID PROFILE OF ORGANIC AND CONVENTIONAL SPELT GRAIN

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Spelt (*Triticum spelta* L., family Poaceae) is one of the oldest grains which is recognized for good nutritional composition (spelt has a higher content of proteins, vitamins, and minerals compared to wheat grain). Spelt has a higher lipid content and also a higher unsaturated fatty acid/palmitic acid ratio than wheat, resulting in the level of oleic acid doubled compared to wheat. The aim of this work was to determine lipids, fatty acids (FAs) and triacylglycerols (TAGs) profile, in the grains of spelt cultivar 'Nirvana'. Spelt was grown under two cultivation systems (conventional and organic) in Maize Research Institute "Zemun Polje" during 2017. The lipid content was determined by the AOAS standard method 963.15, the fatty acid content was determined by the GC-FID method, while HPLC - RI method was used for the determination of the TAG profile. The lipid content is expressed as g/100 g dry weight (DW) while results for FAs and TAGs are expressed as % of total fatty acids/triacylglycerols determined. Lipid content was not significantly different between both types of seeds (organic and conventional grain 1.77 and 1.75 g/100 g DW, respectively). Ten different FAs were identified in examined grains with a predominant presence of linoleic acid (C18:2n-6), while the lowest content was myristic acid (C14:0). Conventional grain was greater amount of arachidonic acid (C20:0) compared to organic grain (0.25 and 0.19%, respectively), while organic grain was greater amount of gondoic acid (C20:1) compared to conventional (0.83 and 0.69%, respectively). When it comes to the TAG content, in spelt grain was predominant presence of ECN44 (38.56-38.86%). Organic grain content greater amount of ECN42 (34.72%) compared to conventional (32.41%), while conventional grain was greater amount of ECN50 (2.65%) compared to organic grain (1.87%).

Keywords: spelt, grain, lipids, fatty acids, triacylglycerols

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CONTENT OF SELECTED TOXIC ELEMENTS IN ORGANIC AND CONVENTIONAL SOYBEAN SEED

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Plants unable to develop high selectivity for biogenic elements intake. Therefore, apart from intake of benefitial elements they will absorb also some potential toxic elements (PTEs) such as Cd, Pb, Ni and Cr. The primary effects of the elemental toxicity depend on the properties of their ions leading to numerous physiological, anatomical and morphological disorders in the plant. In the system of organic agriculture, special attention is paid to the maintaining „healthy“ and fertile soil, without the use of mineral fertilizers, in order to produce food without residues of pesticides and PTEs. The aim of this study was to examine the content of Cd, Pb, Ni and Cr in soybean seed (variety Kaća) produced by conventional and organic farming methods, at the Institute of Field and Vegetable Crops Novi Sad, during 2017. Also, the vigour of the soybean seed obtained through the accelerated aging test was examined. Determination of selected toxic elements was conducted by using Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES) with previous digestion of samples. The obtained results were expressed as mg/kg of dry weight (DW). Conventional soybean seed contained higher amount of Ni (3.604 mg/kg DW), while content of Cd (0.041 mg/kg DW), Cr (0.054 mg/kg DW) and Pb (0.081 mg/kg DW) was higher in the organic seed. After applied the accelerated aging test, the content of Ni, Cd and Pb were higher at the root of the conventional soybean seedling (8.561 mg/kg DW, 0.087 mg/kg DW, 1.027 mg/kg DW, respectively), while the content of Cr (2.618 mg/kg DW) was the highest in above-ground part of the organic soybean seedling. Based on obtained results there are no clear regularity about production system influence on the content of selected PTEs.

Keywords: organic production, conventional production, ICP-OES, heavy metals, soybean

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Section
NOVEL ASPECTS OF FOOD SAFETY AND QUALITY

OPTIMIZATION OF PROTOCOLS FOR SELECTION OF ENDOPHYTIC PLANT GROWTH-PROMOTING BACTERIA FROM TOMATO (*SOLANUM LYCOPERSICUM* L.) SEEDS

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The increasing demand for sustainable agricultural practices highlights the growing need to reduce the use of synthetic chemicals such as fertilizers and pesticides, which pose a serious threat to the environment through soil degradation, water pollution, and biodiversity loss. To overcome these challenges, the use of bioinoculants offers a promising solution to increase crop productivity while maintaining ecological balance. This study aimed to explore a novel approach to improve the resilience and growth of tomato (*Solanum lycopersicum* L.) plants by exploiting the relationship between plants and their endophytic bacteria. We focused on the optimization of protocols for the selection of endophytic bacteria from tomato seeds that exhibit plant growth-promoting (PGP) properties. A screening of 91 bacterial isolates from two different tomato varieties, Narvik and Jabučar, was performed. First, the potential of these isolates to promote the germination and growth of tomato seedlings was tested. Based on the observed plant growth-promoting effects, 9 isolates belonging to the genera *Priestia, Sphingomonas, Exiguobacterium, Paenibacillus, Paracoccus, Bacillus,* and *Brevundimonas* were selected for further characterization of PGP properties. All tested isolates exhibited nitrogen fixation and indole-3-acetic acid production. Phosphate solubilization was observed in 5 isolates, exopolysaccharide production in 4 isolates, and ACC deaminase synthesis in 3 isolates. Numerous isolates showed motility as well as the production of at least one enzyme. One isolate from the collection showed the ability to produce siderophores, while another showed the synthesis of acetoin. All isolates in the collection showed several PGP characteristics. Investigating the properties and effects of endophytic PGP bacteria holds significant potential for addressing numerous challenges in agriculture. We believe that using a method where the beneficial effects are tested first, followed by characterization of the bacteria, is a much more robust approach.

Keywords: PGP bacteria, endophytes, tomato

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FUNCTIONALITY OF ENZYMATICALLY MODIFIED FULL TOASTED SOY FLOUR

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Enzymatic hydrolysis is a powerful and safe method of enhancement of nutritive, techno-functional and functional properties of soy protein products. Its success is determined by a number of factors including the choice of enzyme and hydrolysis conditions. In this study the effect of different hydrolases on techno-functional and antioxidant properties of commercial full toasted soy flour was investigated. The commercial full toasted soy flour was modified with three different hydrolases, commercial multi-enzyme complex (Viscozyme L), commercial α-galactosidase, Flavourzyme and their combinations under the same conditions. Modified samples were thermally inactivated and spray-dried. The effect of hydrolysis was monitored by the change of protein and carbohydrate solubility and by the SDS-PAGE of soluble and total proteins under reducing conditions. The change of emulsifying properties was followed using emulsion stability index (ESI) and emulsion activity index (EAI) whereas foaming properties were determined as foam stability (FS) and foam capacity (FC). Also, the effect of hydrolysis on water holding-, oil holding capacity and swelling ability of modified samples was determined. In addition, the effect of hydrolysis on *in vitro* antioxidant properties of full toasted soy flour was determined (Total antioxidant capacity, CUPRAC and DPPH assay). Hydrolysis increased the level of soluble carbohydrates of al samples by 32.09-46.99% whereas protein solubility increased in the case of flours treated with Flavourzyme. The used enzymes and their combination differently affected techno-functional properties of toasted flour. Hydrolysis improved foam capacity and stability, emulsion stability and oil holding capacity whereas water holding and swelling capacity of modified samples were slightly lower than initial flour. Hydrolysis significantly improved all of antioxidant properties. Based on these results it is obvious that hydrolysis with carbohydrases and proteases as well as with their combination, can be a useful method for improving nutritive, techno-functional and functional properties of full toasted soy flour.

Keyword: soy flour, modification, techno-functional properties, antioxidant properties

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THE INFLUENCE OF CARBOHYDRATE CARRIERS ON THE MORPHOLOGY AND PHYSICAL PROPERTIES OF RED BEET MICROGREEN JUICE ENCAPSULATES

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Red beet microgreen juice is considered a novel functional beverage due to its content of bioactive compounds, including the color pigment betalain and flavonoids. Considering the sensitivity of these bioactive compounds, it is necessary to protect them by encapsulation within carriers to extend their shelf life. The aim of this study was to apply the spray drying technique for the encapsulation of red beet microgreen juice in inulin (RIN) and maltodextrin (RMD) carriers and to determine the effects of spray drying on the morphology and physical properties of obtained encapsulates. The morphological properties of the obtained encapsulates were examined by scanning electron microscopy (SEM), while the moisture content, tapped and bulk density and color were determined by standard methods. The RMD was characterized by a small particle size with the presence of typical spherical particles and pseudo-spherical particles with irregular surfaces due to rapid evaporation at high temperatures in the spray drying chamber. In contrast to RMD, RIN had larger particles with a high degree of agglomeration as inulin is a larger molecule and more hygroscopic than maltodextrin. The moisture content of the encapsulates was below 10 %, with the higher bulk density of RIN compared to RMD. Regarding the color of the encapsulates, those in maltodextrin showed a higher brightness and saturation than the encapsulates within inulin. In summary, inulin and maltodextrin provide good morphological and physical properties and can be used for the encapsulation of red beet microgreen juice. However, further studies should include a detailed phytochemical characterization of the encapsulates.

Keywords: red beet microgreen juice, inulin, maltodextrin, scanning electron microscopy, moisture content

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THE POSSIBILITY OF REDUCING THE ZEARALENONE CONTENT IN THE WHEAT FLOUR BY ATMOSPHERIC COLD PLASMA TREATMENTS

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Zearalenone (ZEA) is an estrogenic mycotoxin, a secondary metabolite of the genus *Fusarium*, especially produced by *F. graminearum,* and *F. culmorum* affected by environmental factors. It is associated mainly with maize but it occurs also in small grain cereals (wheat, barley and sorghum) and represents a significant hazard to the food and feed processing chain, causing health issues and economic losses. Therefore, the aim of this study was to explore the potency of cold atmospheric pressure plasma treatments, as a new non-thermal approach, for reduction of ZEA content in spiked white wheat flour samples (75 μg/kg of ZEA in flour). The flour samples were treated with non-equilibrium (cold) atmospheric pressure plasma generated in the air by using a surface dielectric barrier source. In the experiment, the treatment durations (30 s, 60 s, 90 s, 120 s, 150 s and 180 s) and sample distances from the cold plasma source (6 mm, 21 mm, 36 mm and 51 mm) were varied. The reduction of the ZEA content in the samples after treatment was monitored by the high-performance liquid chromatography with fluorescence detection at excitation and emission wavelengths of 274 nm and 450 nm, respectively. The maximum reduction of the ZEA content was obtained after 120 s treatment performed at 36 mm distance from the plasma source, resulting in reduction of 97.8%.

Keywords: wheat flour, zearalenone, atmospheric cold plasma, HPLC-FLD

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ISOLATION OF DIETARY FIBERS FROM SOY HULLAND INCORPORATION IN NOVEL GLUTEN FREE COOKIES

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Gluten-free baked products often present quality defects, low nutritional value and low sensorial quality. The supplementation of non-wheat flour and dough with healthy dietary fibres (DFs) and prebiotic is a usual practice for increasing the nutritional value of gluten-free products while improving their texture, shelf-life and mouthfeel. However, the effectiveness of DFs to mimic the properties of gluten to overcome problems of gluten-free systems or promote health benefits depends not only on extraction process but also on their source, structural and chemical composition. Pectin and their oligosaccharides are valuable compounds providing the known associated health benefits.

In this paper, we examined two methods for extracting DFs from soy hulls, which are waste streams generated during the production of soy proteins. Specifically, these methods include acid extraction using a 0.5 M HCl solution for 30 min and ultrasonic extraction performed in cycles under the following conditions: 22-25 kHz, duty cycle 0.67, power 490 W (90%). Extracted DFs was characterized regarding its galacturonic acid (GalA) content, degree of methoxylation (DM), degree of acetylation (DA) and functional properties like water (WHC) and oil (OHC) holding capacities, as well as solubility. Significantly higher yield of DFs was achieved with acid extraction, and the functional properties of the extracted fibers were satisfactory. Content of GalA amounted 40% (m/m), DM was determined to be 30% and DA 0.9%. Average MW determined by HPSEC was 1815 kDa, WHC was amounted to 12 g/g while OHC was 2.44 g/g. Solubility was determined to be 50%.

The obtained fibers were incorporated into rice cookies: a control batch with rice flour and two batches with 10% (10CDF) and 20% (20CDF) fiber substitution. The addition of pectic fibers resulted in darker, smaller cookies with decreased hardness (29% for 10CDF and 14% for 20CDF). The cookies also exhibited better nutritional, textural and sensory properties compared to the control. Physico-chemical analysis revealed that 10CDF cookies are a "source of dietary fiber" (4.1% fiber) and 20CDF cookies are "rich in dietary fiber" (7.9% fiber), as per Regulation no. 1924/2006.

Keywords: acid extraction, ultrasound extraction, pectic fibers, soy hull, gluten free cookies

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HORSERADISH LEAF JUICE ENCAPSULATES: PHYSICOCHEMICAL, SPECTROPHOTOMETRIC, AND CHROMATOGRAPHIC CHARACTERIZATION

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Horseradish is mainly used in the food industry for its succulent and spicy roots, while the horseradish leaves are usually disposed of with the above-ground biomass. The cold pressing of horseradish leaves produces a juice that is rich in bioactive compounds with pronounced antioxidant potential. However, due to its high water content and biochemical reactivity, it is prone to degradation. The bioactive compounds of horseradish leaf could be protected from undesirable external factors by spray drying encapsulation within maltodextrin/alginate (MD/AL) and maltodextrin/gum Arabic (MD/GA) carriers. Therefore, the aim of this study was to encapsulate horseradish leaf juice within these carriers and to evaluate the physicochemical, spectrophotometric, and chromatographic properties of the obtained encapsulates. Using standard analytical methods, lower values for moisture content, water activity, solubility, oil holding capacity, and encapsulation yield were obtained for MD/AL than for MD/GA encapsulates. Standard spectrophotometric methods were used to characterize MD/AL and MD/GA encapsulates in terms of total phenolic content (5241.25 and 4849.00 mg gallic acid equivalents/100 g), total flavonoid content (4640.67 and 4159.99 mg catechin equivalents/100 g), total phenolic acid content (5008.34 and 7013.20 mg caffeic acid equivalents/100 g), and antioxidant activity (mmol Trolox equivalents/100 g: 0.88 and 0.44 (DPPH); 58.67 and 63.20 (ABTS); and 15.78 and 13.71 (FRAP), respectively). Using an ultra-high-performance liquid chromatography system, coupled with a quadrupole time-of-flight mass spectrometry, a total of fourteen phenolic compounds were quantified after extraction from the encapsulates. The chromatographic analysis also confirmed the higher total content of all identified phenolic compounds in MD/AL (1896.87 mg/kg) than in MD/GA (1761.27 mg/kg). The obtained results highlight encapsulated horseradish leaf juice as an underestimated and underexplored source of phenolic compounds with high antioxidant potential, whose application in the food industry could valorize crop side streams and reduce synthetic antioxidant usage.

Keywords: horseradish leaf, encapsulation, spray drying, phenolic compounds, antioxidant activity

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DETERMINATION OF OXIDATIVE INDUCTION TIME OF SESAME SEEDS OIL BY DSC ANALYSIS AND THE KISSINGER-AKAHIRA-SUNOSE METHOD

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Oils are important in human nutritio, in particular because of their content of polyunsaturated fatty acids. Sesame seeds (*Sesamum indicum* L.) contain 42-60% oil, which has aromatic properties, rich in unsaturated fatty acids (UFAs), vitamin E and natural antioxidants. Spoilage of oils due to the UFAs oxidation is accompanied by the appearance of "rancidity", and oils then acquire an unsavoury taste and their shelf life is shortened. There are a various methods for determining the oxidative induction time (OIT) which is directly related to oil shelf life. In this work, DSC analysis, under non-isothermal conditions, and the Kissinger-Akahira-Sunose method, based on the oxidation onset temperature and two equations, were used to determine oxidative induction time and kinetic parameters during oxidation of sesame seed oil purchased at a local store. The oil was heated in the temperature range from 25 °C to 450 °C, at rates of 5, 10, 15 and 20 °C min^{-1}, with an empty closed aluminium pan used as a reference. Thermal effects were recorded on a differential scanning calorimeter TA Instruments, Q20, USA. For a temperature of 25 °C (which can be considered as a storage temperature) the activation energy was 118.12 kJ, pre-exponential factor (A), $1.78x10^{13}$ min^{-1} and constant of reaction rate, $3.51x10^{-8}$ min^{-1}. Based on the equations $OIT=Ae^{-DT}$ and $OIT=(T_\infty-T)a[\ln(T_\infty/T_\infty-T)]^{(a-1)/a}$ (D and a are the variable kinetic parameters without physical meaning, T∞ is the is conversion temperature for an infinite heating rate, T is the selected temperature and β is the heating rate), the oxidation induction time of 3.70 and 6.03 months, respectively, was obtained. The obtained oxidation induction time based on the second equation shows better agreement with the shelf life of oil marked on the consumer's specification, specified as 7 months.

Keywords: DSC, oil, oxidation, sesame, shelf life

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"GREEN" EXTRACTION AND NANOENCAPSULATION OF BIOACTIVE COMPOUNDS FROM PLANT BY-PRODUCTS TO PRODUCE SENSITIVE SKIN COSMETICS

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NanoCosmos (www.nanocosmos.eu) is a collective international approach from the research community and industry to participate in the transformation of bioeconomy through bioscience and nanotechnology. The main aim of the NanoCosmos project is to create a multidisciplinary and intersectoral network to harness the recovery of valuable bioactive compounds from post-harvest by-products, such as Saffron petals (*Crocus sativus*), Chamomile (*Matricaria chamomilla*) and Lavender (*Lavandula angustifolia vera*) post distillation biomass and *Rhodiola rosea* leaves including flowers. The application of green extraction techniques and the production of nanocosmetic products for sensitive skin applications will be the major innovative outcomes of NanoCosmos following the circular economy rules. To achieve the ambitious goals of the project plant biodiversity, cropping technologies and different sources of by products will be evaluated and explored by application chemical analysis and of green extraction technology. Recovered bioactive compounds/extracts will be analyzed by state-of-the art metabolomics tools and supported by bioactivity and efficiency testings. The impact of drying process of original plant material but also the by-products on chemical profile will be analyzed. Finally, bioactive extracts/compounds will be implemented by nano-technology resulting in the development of novel formulations for skin cosmetics. Beside the scientific part NanoCosmos will maximize research value by fostering long-term network relationships between academic institutions and industry. Best-practice technologies will be disseminated, with research focusing on emerging developments in recovery techniques as well on the nanotechnological application of the recovered compounds or mixtures.

Keywords: green extraction, bioactive compounds, post-harvest by-products, nanocosmetics

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A PROSPECTIVE CROSS-SECTION SURVEY ON OMEGA-3 FATTY ACID SUPPLEMENT CONSUMPTION IN THE REPUBLIC OF SERBIAAND THE REPUBLIC OF SRPSKA

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Modern diet is often depleted of the nutritionally important omega-3 fatty acids (ω-3-FA), such as alpha-linolenic (ALA; primarily found in seeds, nuts and vegetable oils) and especially eicosapentaenoic (EPA) and docosahexaenoic (DHA) (found in algae and fish). The use of ω-3-FA supplements can be seen as a simple yet effective way to obtain these nutrients, crucial for the health of heart, brain and immune system. In order to obtain insight into the use of ω-3-FA rich foods and supplements, knowledge and opinions of populations of the Republic of Serbia and the Republic of Srpska, following the ethical commission approval, an online questionnaire was made available during May-October 2023. The multiple correspondence analysis was applied to identify specific patterns of the variability of the dataset of 895 respondents (including socio-demographic data, basic anthropometric features and important lifestyle data of respondents). It was found that the respondents use ω-3-FA supplements because of inadequate dietary intake, but also for the purpose of therapy or prevention of certain diseases and conditions. The users take supplements on a regular basis, although for a short period of time. The decision on consumption is made by themselves or by following the advice of healthcare professionals. More than half of parents give ω-3-FA supplements to their children under the age of 3, with the similar purposes, but in such case more frequently, and for longer period of time. The use of ω-3-FA via supplements was reported by nearly half of pregnant (45.2%) and breastfeeding (41.6%) women, although these physiological conditions were not necessarily the single most important reason for the use decision. Regardless of a number of reports of the intake of food rich in ω-3-FA, the consumption pattern indicates that for the majority of population, consumption occasions are not frequent enough to provide recommended amounts of ω-3-FA.

Keywords: food supplements, EPA, DHA, public health

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COMPOSITE HYDROGELS OBTAINED FROM EXTRACTS OF *PORPHYRIDIUM PURPUREUM* AND ALGINATE

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Hydrogels play a significant role in the food industry as three-dimensional polymer networks. Hydrogels have found extensive use not only in the production of traditional food products but also in designing carriers for bioactive components. Developing hydrogels for food applications prioritizes the use of natural ingredients and simple production methods. Extracts from the red microalgae *Porphyridium purpureum* contain sulphated polysaccharides and vibrantly coloured phycobiliproteins (PBPs), notably B-Phycoerythrin. This makes *Porphyridium purpureum* an excellent starting point for developing food-based hydrogels with strong bioactive properties. This work aims to develop and characterize the hydrogels from water extracts of *Porphyridium purpureum*. In order to achieve that, we used the gelling property of the extract under acidic conditions at pH 2.0. Two types of hydrogels based on this algal extract were compared. The first is formed solely in an acidic environment, while the second is formed by adding alginate at the same pH in the presence of calcium ions. The mechanical properties of both hydrogels were determined by frequency sweep measurements using a rheometer with applied plane/plane geometry. Rheological measurements showed that adding alginate significantly increases the mechanical properties and elasticity of the hydrogel. Confocal microscopy demonstrated stronger fluorescence of PBPs in the gel without alginate. Furthermore, the distribution of different PBPs within the gel network is more uniform without alginate. The digestibility of the hydrogels was evaluated using an *in vitro* static digestion model. Although both hydrogels were confirmed to be digestible, the hydrogel without adding alginate showed higher digestibility. Antioxidant assays, ABTS and a reducing power test showed that bioaccessible peptides after digestion of both hydrogels possess antioxidant activity, with those obtained without alginate having a higher activity. Overall, this research provides a simple and effective approach for developing coloured hydrogels with attractive appearance, good bioaccessibility and notable bioactive properties.

Keywords: hydrogel, Porphyridium purpureum, Phycobiliproteins, B-Phycoerythrin, polysaccharides

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LABBANK – THE FIRST BIOBANK OF AUTOCHTHONOUS PROBIOTIC LACTIC ACID BACTERIA

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Lactic acid bacteria (LAB) are a group of bacteria that are generally recognized as safe (GRAS) and are widely used in fermented foods. They are widespread in nature mainly in raw milk and dairy products. The indigenous microbiota of raw milk directly affects the sensory properties of the raw milk products. The WHO recommends ~~to~~ the use of fermented dairy, meat and vegetable~~s~~ products in the daily diet, as LAB have a great impact on human health.

Group for Probiotics and Microbiota – Host Interaction within the Institute of Molecular Genetics and Genetic Engineering, University of Belgrade has a large collection of LAB (about 5000 various strains) isolated from artisanal autochthonous dairy products produced in a traditional way without the addition starter cultures. The characterization of isolated LAB strains was done by classic microbiological methods and identification by 16S rDNA sequencing. These LAB strains are collected in previously 30 years and stored in refrigerator at -80°C.

Until now 23 distinctive LAB strains from LABbank collection have been deposited in the Belgian Coordinated Collection of Microorganisms, University of Ghent, Belgium. The 13 strains have been licensed through license agreements, as the subjects of innovations:

-HiraVet probiotic for prevention and treatment of intestinal infections in humans and animals.

-Diasolution probiotic for diabetes management.

-Lagendairy starter cultures for innovative dairy products for the production of soft white cheese, cheese in brine, yogurt and sour cream.

-Sixteen LAB strains are currently the subject of innovations submitted as 2 PoC projects.

Our findings illustrate the importance of the research on natural isolates of LAB as a valuable source of strains with novel properties, since they can provide a deeper and more complete insight into the functioning and organization of the comprehensive metabolic system in these bacteria and their impact on human and animal health.

Keywords: BIObank, Lactic acid bacteria, Autochthonous dairy products, probiotic fermented foods

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