



University of Novi Sad  
**INSTITUTE  
OF FOOD  
TECHNOLOGY  
IN NOVI SAD**

**feed**  
congress

16-18 October, 2024  
Novi Sad, Serbia

**5<sup>th</sup>**

**INTERNATIONAL CONGRESS  
FOOD TECHNOLOGY,  
QUALITY AND SAFETY**

**e-ABSTRACT BOOK**

**ISBN 978-86-7994-063-6**

**V INTERNATIONAL CONGRESS “FOOD TECHNOLOGY, QUALITY AND SAFETY”, 16-18 OCTOBER 2024, NOVI SAD, REPUBLIC OF SERBIA**

***Publisher***

University of Novi Sad  
Institute of Food Technology in Novi Sad  
Bulevar cara Lazara 1  
21000 Novi Sad  
Republic of Serbia

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*Food Technology, Processing and Engineering*

## ELEVATING SOURDOUGH QUALITY WITH QUINOA MALTS

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This study evaluated the quality characteristics and health-promoting properties of wholemeal quinoa flours and malts, along with their corresponding sourdoughs. The flours were analyzed for color, moisture, falling number, total protein, ash, fat, dietary fiber (soluble, insoluble, total), and minerals via Atomic Absorption Spectroscopy (AAS). Sourdoughs were prepared using a control sample (no malt) and samples with 5% and 10% malt additions, under consistent conditions (yield: 250, fermentation: 46 hours at 30°C). These sourdoughs were assessed for quality indicators (color, dynamic viscosity, pH, total titratable acidity (TTA), total protein, ash, volatile compounds via GC-MS, lactic and acetic acid content) and health benefits (dietary fiber, antioxidant activity, micro- and macronutrient content). The results revealed significant differences based on quinoa type and malt addition. White quinoa sourdoughs had the highest TTA, acetic acid, and specific minerals (sodium, magnesium, copper), the lightest color, and lower levels of protein, total dietary fiber (TDF), insoluble dietary fiber (IDF), phytic acid, lactic acid, potassium, and manganese. Black quinoa sourdoughs exhibited the darkest color, highest protein, ash, TDF, IDF, lactic acid, potassium, manganese, and lowest viscosity, acetic acid, and certain minerals (sodium, calcium, zinc). Red quinoa sourdoughs had the highest viscosity, soluble dietary fiber (SDF), and zinc content but the lowest ash, magnesium, and iron levels. Sourdoughs from colored quinoa showed higher antioxidant activity compared to white quinoa. GC-MS identified 25 volatile compounds, predominantly alcohols, followed by organic acids, aldehydes, terpenes, phenols, esters, furans, and cyclic hydrocarbons. Malt addition generally increased sourdough acidity, TDF, lactic acid, and decreased total protein. Specifically, white quinoa sourdoughs with malt showed reduced IDF, acetic acid, and mineral content (sodium, calcium, potassium, magnesium, iron), while black quinoa sourdoughs demonstrated increased viscosity and acetic acid. This study highlights the influence of quinoa variety and malt addition on sourdough quality and nutritional profile.

**Keywords:** quinoa, sourdough, malt, quality, health-enhancing properties

**Acknowledgements:** This research was funded by the National Science Centre of Poland, grant number 2022/06/X/NZ9/01403.

## EVALUATION OF THE ADDITION OF QUINCE SEED OIL CAKE ON THE TEXTURE AND SENSORY PROPERTIES OF TOAST BREAD DURING STORAGE

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Bread enriched with fruit seed oil cake represents a novel innovation in the food industry, aligning with the growing trend towards developing functional foods. The use of oil cake as a by-product in food production is a multifunctional approach that contributes to food safety, sustainability and public health. By using these nutrient resources, the composition and quality of food can be significantly influenced, waste reduced and healthier eating habits promoted. The aim of this study was to analyze the chemical composition, technological and functional properties of quince seed oil cake flour and the effects of its addition on the freshness and sensory properties of standard toast bread. The oil was extracted from the quince seeds by cold pressing, and the resulting cake was then ground into flour. Before being added to the dough, the quince seed oil cake flour was hydrothermally processed. 5 % and 10 % quince seed oilcake flour were added to the standard recipe for toast bread. The quince seed oil cake affected the chemical composition of the toast by increasing the protein, oil, mineral and moisture content and decreasing the carbohydrate content. The addition of quince seed oil cake influenced the textural property of hardness and had a positive effect on the freshness of the final product. The change in freshness was lowest in the Q10 sample (22.0 %) and highest in the control sample (63.0 %). The greatest negative change during storage was observed in the crumbliness, while the odor and taste characteristics showed the least negative changes. After seven days of storage, mold appeared in all samples.

**Keywords:** quince, oil cake, toast, technological and functional properties, sensory evaluation

**Acknowledgements:** The authors are grateful to the Ministry of Education, Science and Technological Development, Republic of Serbia, Contract number 451-03-65/2024-03/200116 (University of Belgrade, Faculty of Agriculture).

## COLD PLASMA TREATMENT AS GREEN TECHNOLOGY IN FOOD PROCESSING

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Cold plasma has become an emerging technology in the food industry. It is widely used as a non-thermal sterilization technique due to its antimicrobial activity. Besides direct plasma treatment, plasma-activated water (PAW) also showed potential for increasing fresh food shelf life. This is possible thanks to PAW's ability to inactivate enzymes responsible for food browning. Additionally, cold plasma could modify existing materials or adjust biopolymer properties, making them suitable for functional food packaging. This study aimed to examine the possibility of cold plasma jet utilization both for less studied applications, including isolating lignin from agri-food waste and producing PAW for fresh-cut apple treatment. By combining cold plasma and alkaline treatment on corn stalks, we obtained oxidized lignin micro- and nanoparticles possessing antioxidative activity. These particles were rich in total phenolic content, reaching up to  $140 \pm 20$   $\mu\text{g}$  gallic acid equivalents per mg of lignin. Granny Smith apples were washed, peeled, and diced into  $1 \times 1 \times 1$  cm cubes, and soaked in differently activated PAW for 20 minutes. After PAW treatment, apples were stored in glass bottles with a screw cap at  $4^\circ\text{C}$ , and changes in their appearance were observed. Apples treated with PAW activated for 30 minutes retained a fresh look and bright color even after 7 days. The overall results of this study confirmed that cold plasma could be used in the processing and preservation of fresh-cut products, but its impact on flavor, nutrition, chemical, and textural properties should be further investigated. Cold plasma-based treatments of lignocellulosic biomass open new possibilities for isolating biopolymers, i.e. lignin, cellulose, and hemicellulose, which could be incorporated into active food packaging following circular bioeconomy principles.

**Keywords:** lignin nanoparticles, corn stalks, active packaging, apples, shelf-life extension

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development, and Innovation of the Republic of Serbia (Contract No. 451-03-65/2024-03/200135 and 451-03-66/2024-03/200287), and by the Alliance of International Science Organizations, project SparkGREEN (ANSO-CR-PP-2022-08).

## EFFICIENT RECOVERY OF PHENOLICS AND TERPENOIDS COMPOUNDS FROM *VERBASCUM SINUATUM* FLOWERS THROUGH PRESSURIZED LIQUID EXTRACTION FOR INDUSTRIAL APPLICATION

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*Verbascum sinuatum* is a plant belonging to the Scrophulariaceae family that has been used in traditional medicine to treat several diseases due to its biological properties (against hemorrhoids, diarrhea, liver inflammation, ulcer inflammation, teeth pain, gumboil, hoarse, tonsillitis, cold, cough, asthma, bronchitis, rheumatism, eczema, wounds, conjunctivitis, otitis, and helminthiasis). These properties have been linked with the phytochemical composition of *V. sinuatum* flowers, mainly to their secondary metabolites such as phenolics, terpenoids and alkaloids. Other authors have shown statistical correlations between the biological activities of *V. sinuatum* flower extracts with their secondary metabolites. The aim of this study was to maximize the extraction of the secondary metabolites (analyzed by HPLC-ESI- QqQ-MS/MS) by determining the optimal conditions using pressurized liquid extraction (PLE). For this purpose, response surface methodology (RSM) and a five-level circumscribed central composite design (28 experiments) was used, and the variables studied were temperature (*T*, 50-200°C), time (*t*, 1-25 min), and ethanol (EtOH) concentration (*S*, 0-100%). Briefly, some of the major phytochemical compounds found were phenolics (*p*-coumaric acid; caffeic acid, luteolin and rutin) and terpenoids (plantagonine, harpagoside, buddindeterpene B sinuoside and catalpol) that can be applied for industrial purposes. The most favorable conditions were minimum time, maximum temperature and water as solvent for polyphenols whereas 4 min and 19% of EtOH maximized the extraction of terpenoids. These optimal conditions are characterized by a higher efficiency in breaking down cell wall membranes, increasing chemical solubility, and preserving bioactive compounds from oxidation (by enzymes or other agents). The temperature; time and solvent concentration were very crucial since at the highest temperature, short time and small EtOH proportion, the extractions were higher. In conclusion, RSM was demonstrated to be a helpful and efficient tool for industrial purposes, that allowed the determination of optimal extraction conditions for the recovery of phytochemical compounds from *V. sinuatum*, regardless of the extraction processes. Furthermore, our findings show that this plant is a reliable source of natural ingredients with potential applications for several industrial sectors and that their recovery using PLE is more efficient than other techniques presented in the scientific bibliography.

**Keywords:** *Verbascum sinuatum*, pressurized liquid extraction, optimization, phenolic compounds, terpenoids

**Acknowledgements:** The research leading to these results was supported by Xunta de Galicia for supporting the post-doctoral grant of A.G. Pereira (IN606B-2024/011) and the pre-doctoral grant of P. Barciela (ED481A-2024-230).

## INVESTIGATION OF THE ANTIOXIDANT PROPERTIES OF SPENT COFFEE GROUNDS EXTRACTS

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Coffee is one of the most consumed beverages worldwide. During coffee production, a significant amount of by-products is generated, with coffee grounds being the primary by-product resulting from both beverage preparation and instant coffee production. The yield of bioactive compounds extracted from coffee grounds depends on the type of coffee, storage conditions and extraction technique. The antioxidant activity of coffee grounds is mainly attributed to their high phenolic content. The main goal of this study was to investigate the antioxidant activity of dried coffee grounds remaining in the machine after espresso preparation, with a focus on determining the total phenolic content, flavonoids and antioxidant activity through FRAP, DPPH and ABTS tests. Four solvents were used for the extraction and isolation of antioxidant components: 1. distilled water; 2. 70% ethanol and two mixtures of 70% ethanol and water in different ratios (3. 70% water and 30% ethanol; and 4. 30% water and 70% ethanol). Based on the research results, it was concluded that the type of solvent used significantly influences the phenolic and flavonoid content in the extract. Water was found to be the least effective for extracting phenolic compounds and achieving high antioxidant capacity. The highest flavonoid content and best antioxidant capacity were achieved using a combination of ethanol and water in a 70:30 ratio.

**Keywords:** by-products, coffee grounds, bioactive compounds

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovations of the Republic of Serbia under the programs 451-03-66/2024-03/200134, 451-03-65/2024-03/200134.

## LACTIC ACID FERMENTATION OF OIL PUMPKIN (*CUCURBITA PEPO* L. VAR. *STYRIACA*) FLESH: PHYSICO-CHEMICAL AND MICROBIOLOGICAL CHARACTERISTICS

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This study investigates the potential utilization of oil pumpkin (*Cucurbita pepo* L. var. *Styriaca*) flesh, a by-product of seed harvesting, through lactic acid fermentation. The research aims to evaluate the physico-chemical and microbiological changes occurring during the fermentation process, exploring a novel application for this underutilized resource in human nutrition. Pumpkin flesh samples, with seeds and peel removed, underwent natural fermentation for 72 hours at room temperature in a 1% NaCl brine solution. The study analysed organic acids, sugar profiles, texture, and microbiological characteristics of both raw and fermented samples. Results revealed significant changes in the fermented product: the pH decreased while titratable acidity increased throughout the fermentation period. Lactic acid content rose from 79.79 to 122.15 mg/kg fresh weight. Glucose and fructose levels rapidly declined, from 2.64% to 0.44% and 1.47% to 0.45%, respectively. Texture analysis showed a decrease in firmness and hardness of the fermented pumpkin pieces. Microbiological profiling indicated an increase in lactic acid bacteria ( $10^9$  CFU/g), alongside the presence of yeasts and moulds (300 CFU/g) and *Enterobacteriaceae* ( $10^3$  CFU/g). The study concludes that oil pumpkin flesh can be successfully fermented by lactic acid bacteria, offering potential for value-added applications. However, to ensure the microbiological stability and safety of the final product, appropriate pre-treatment methods need to be developed and implemented. This research contributes to the growing body of knowledge on sustainable food processing and the valorisation of agricultural by-products.

**Keywords:** oil pumpkin, organic acids, sugars, texture, fermentation

**Acknowledgements:** This research was financially supported by Ministry of Science, Technological Development and Innovation, Republic of Serbia, Institute of Food Technology in Novi Sad (No. 451-03-66/2024-03/200222 and 337-00-577/2021-09/14).

## THE IMPACT OF THE PRODUCTION PROCESS ON THE COLOR OF PEACH AND APPLE FRUIT NECTAR

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The aim of this study was to monitor the colour change of peach and apple nectar during the technological production process. Colour is a key quality attribute of fruit juices and nectars, as it directly influences consumer perception and acceptance. During processing, factors such as temperature, pH value, and exposure to oxygen can lead to significant changes in nectar colour, which may affect the market appeal of the final product. In this research, under real industrial conditions, samples of peach and apple puree were collected at the beginning of the production process, and the final products – peach and apple nectar – were obtained. A Minolta Chroma Meter CR-400 colorimeter (Konica Minolta Inc., Osaka, Japan) was used to determine colour parameters. Measurements were performed under D-65 lighting (average mid-morning or mid-afternoon daylight) with a standard observer angle of 2°. The results were expressed in terms of L\* (lightness), a\* (red/green), and b\* (yellow/blue) values. These parameters were analysed to determine the extent of colour change and to identify critical points in the production process where significant changes occur. The results showed that peach and apple juices undergo noticeable colour changes, with the most significant variations observed during pasteurization and storage. The decrease in L\* values indicates a reduction in the lightness of the fruit nectar colour, while a\* and b\* values decrease due to an increase in the green and blue colour components. All of these changes can be attributed to Maillard reactions and the breakdown of pigments during heating. The findings of this study provide valuable insights into the optimization of processing conditions and enhance the understanding of how technological processes impact undesirable colour changes, thereby improving the quality and shelf-life of fruit juices.

**Keywords:** total colour change, peach and apple nectar, technological process

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia under the programs 451-03-66/2024-03/200134, 451-03-65/2024-03/200134, 451-03-66/2024-03/200222.



## ALTERNATIVES FOR NITRITES FROM NATURAL RESOURCES IN COOKED MEAT PRODUCTS

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Nitrites are the most essential synthetic additives and preservatives in the meat processing industry. They are related to the control of different pathogenic bacteria (e.g., *Clostridium botulinum*, *Listeria monocytogenes*), the development of reddish-pink color and cured flavor, and the delaying of oxidative reactions (lipid and protein oxidation) in cured meat products. However, these preservatives have been linked to several health risks, leading to recent regulatory changes. Also, according to "clean label" trends, customer demand for natural meat products has increased. Hence, scientists and meat industry processors focus on developing nitrite alternatives. This paper reviews nitrate-containing vegetable and plant extracts as potential replacements (used entirely or partially) for synthetic nitrites in manufacturing of cooked meat products.

**Keywords:** nitrites, cured meat products, nitrosamines, plant extracts

**Acknowledgements:** This research was funded by the Ministry of Education, Science and Technological Development: Project 451-03-65/2024-03/200134.

## OPTIMIZATION OF PHENOLIC COMPOUNDS REMOVAL FROM RAPESEED CAKE USING MICROWAVE-ASSISTED EXTRACTION TECHNIQUE

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Rapeseed has a balanced amino acid profile, but its application is hindered by undesirable substances such as glucosinolates, phytic acid, tannins, and phenolic compounds. Although phenolics may have beneficial effects, they significantly contribute to the dark color and bitter taste of isolated proteins. These compounds are present in particularly high amounts, up to ten times more than in other oilseeds, further complicating protein isolation. This study focuses on optimizing microwave extraction to reduce the total phenolic content in rapeseed press cake, thereby improving the quality of the material for protein isolation. A Box-Behnken experimental design was used to investigate the effects of four key input parameters: extraction time (2, 11, 20 minutes), ethanol concentration (75%, 82.5%, 90%), solid-to-liquid ratio (30:1, 45:1, 60:1), and microwave power (400 W, 600 W, 800 W). The goal was to identify the optimal conditions that reduce phenolic content, facilitating the extraction of purer protein material. Experimental results showed that parameters such as ethanol concentration and solid-to-liquid ratio significantly influenced the total phenolic content after extraction, with values ranging from 187.82 mg/g to 232.49 mg/g. The most optimal results were achieved with a sample treated with 82.5% ethanol, a microwave treatment duration of 20 minutes, the highest solid-to-liquid ratio of 1:60, and a power of 600 W. This study demonstrates that using microwave extraction can reduce TPC in rapeseed press cake, making the material more suitable for subsequent protein isolation processes. The analysis indicates that varying parameters significantly affect the extraction of phenolic compounds from rapeseed press cake, thus enhancing the production of material suitable for the efficient extraction of high-quality rapeseed proteins.

**Keywords:** Microwave-assisted extraction, phenolic compounds, rapeseed cake

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovations, Republic of Serbia (Contract No. 451-03-66/2024-03/200222), as well as by the Science Fund of the Republic of Serbia, Grant No 6673, PROTein from Rapeseed Oil Processing Waste: Application in Food and Wastewater Treatment – PROTOPOWER.

## EXPLORING FUNGAL BIOMASS PRODUCED FROM WHEY POWDER WITH *Aspergillus oryzae* AND *Neurospora intermedia* IN SUSTAINABLE FOOD SYSTEMS: FLAVOR COMPOUNDS AND AROMA CHARACTERISTICS

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Whey, a clear, yellowish to greenish liquid obtained by separating coagulated milk, is processed into powder and utilized as a by-product in the dairy industry. Fungal biomass with potential applications in human nutrition can be produced by using various food industry by-products as substrates. *Aspergillus oryzae* var. *oryzae* CBS 819.72 and *Neurospora intermedia* CBS 131.92 are starter cultures responsible for producing specific aroma compounds in some traditional fermented foods (oncom and koji). This study was focused on determining the flavor compounds and characteristic aroma properties of biomasses produced from whey powder through *A. oryzae* and *N. intermedia*. The volatile compounds of fungal biomass were extracted using solid-phase microextraction (SPME) and identified by gas chromatography-mass spectrometry (GC-MS). The sensory properties of the samples were evaluated by six panelists, who had prior experience in evaluating various products using the Spectrum™ procedure. The correlation between volatile compounds and sensory attributes was illustrated through Principal Component Analysis (PCA). Acids, alcohols, aldehydes, phenols, esters, and ketones were major groups of volatile compounds determined in the biomass samples. Specifically, 1-octen-3-ol (mushroom aroma), 2-ethyl hexanol (rose, green), nonanal (fat, citrus, green) and benzaldehyde (almond, burnt sugar) were found at higher concentrations in *A. oryzae* biomass and *N. intermedia* biomass compared to non-fermented whey powder. Characteristic sensory attributes of the biomass samples were described as mushroom, hazelnut, soil/peppery, wet carton, and fruity. Based on the PCA results, the flavor profiles of biomasses produced by *A. oryzae* and *N. intermedia* exhibited distinct characteristics compared to non-fermented whey powder. These findings will provide valuable insights into the flavor and aroma characteristics of biomasses produced through fungal fermentation for human consumption.

**Keywords:** food by-products, descriptive sensory analysis, filamentous fungi, sustainability

**Acknowledgements:** This work is part of the activities of the COST Action FLAVOURsome, CA22161, supported by COST (European Cooperation in Science and Technology). Burcu KAYA would like to acknowledge the direct support by COST (European Cooperation in Science and Technology; [www.cost.eu](http://www.cost.eu)), in the framework of COST Action CA20128 (Promoting Innovation of ferMENTed fOods; <https://fermentedfoods.eu/>).

## **EFFECT OF THE ADDITION OF RICE FLOUR AND CORN STARCH ON THE PHYSICAL QUALITY PARAMETERS OF GLUTEN-FREE BREAD**

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The aim of this study was to determine and analyse the physical quality parameters of gluten-free bread made from rice flour, corn starch and xanthan gum as well as psyllium husk powder. The proportion of rice flour in the mixture with corn starch ranged from 50 to 100% and the proportion of psyllium husk powder in the mixture with xanthan gum ranged from 0 to 100%. The test firings were carried out according to the experimental plan, which was prepared according to the full factorial design. The specific volume and texture profile analysis of the bread samples were determined. The number of cells, their average size and the area of the cells as well as the color parameters in the CIELab system were determined by computer image analysis. The results of the study showed that increasing the proportion of psyllium husk powder increases the firmness and elasticity of the bread as well as the number of cells, while the average cell size decreases. An increase in the proportion of rice flour resulted in a decrease in the firmness of the bread and an increase in the average size of the cells. The amount of rice flour had no significant effect on the number of cells. The recipe for bread production was optimised based on the number of cells and their average size using the response surface methodology. The results showed that the optimal recipe for the production of gluten-free bread (based on the total mass of flour) is as follows: 100% rice flour with the addition of 4.58% xanthan gum and 5.42% psyllium husk powder.

**Keywords:** gluten-free bread, rice flour, psyllium husk powder, porosity, computer image analysis

## KINETIC MODELING OF HYDRODISTILLATION AND MICROWAVE-ASSISTED HYDRODISTILLATION OF DILL SEED ESSENTIAL OIL (*ANETHUM GRAVEOLENS*)

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*Anethum graveolens* is an annual aromatic plant that belongs to the *Apiaceae* family. This plant has been known as a medicinal herb since the times of the Greeks and Romans, and it is now widely used in traditional medicine for treating conditions affecting the digestive and urinary systems, primarily utilizing its fruit. In order to extract high-quality essential oils (EO) from dill, the effects of hydrodistillation (HD) power (205, 410 W) on the EO yields and extraction kinetics were evaluated. Microwave-assisted hydrodistillation (MWHD) at different irradiation power (180, 360, 600, 800 W) was used as a modern extraction technique, while conventional distillation was comparatively analyzed. Four empirical models that include the stages of washing and diffusion during distillation and describe the kinetics of EO isolation, were utilized to determine which model best fits the experimental data. Furthermore, the kinetics of HD and MWHD were compared to estimate the optimal technique for maximizing EO production. The kinetics of the distillation process of EOs are crucial for the optimization of the process itself, as well as for its control and management, both in laboratory and industrial conditions. The yield of the obtained EOs ranged from 2.43 to 3.23%. The highest yield was obtained using MWHD at 600 W. Mean absolute percentage error (MAPE) and the coefficient of determination ( $R^2$ ) are statistical parameters used to describe the capabilities of applied mathematical models to describe experimental results. Model I and Model III provided the best fit, with very similar results and nearly identical quality of fit. Coefficient of determination was the same for Model I and Model III and it was 0.996. The MAPE for Model I was 7.8%, slightly higher than Model III's value of 7.44%. It can be concluded that MWHD presents an excellent alternative to conventional HD for the recovery of EOs, and can be used on an industrial scale.

**Keywords:** Dill essential oil, hydrodistillation, microwave-assisted hydrodistillation, mathematical modeling, kinetics

**Acknowledgements:** This research was funded by the Science Fund of the Republic of Serbia for the funding of the IDEAS project 'Novel extracts and bioactive compounds from under-utilized resources for high-value applications-BioUtilize', number '7750168' and by the Ministry of Science, Technological Development and Innovations of the Republic of Serbia, 451-03-66/2024-03/200134.

## PRINTING OF FUTURE FOOD - CHALLENGES, OPPORTUNITIES AND PERSPECTIVES

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In today's world, marked by global crises, consumers are becoming increasingly aware of the importance of proper nutrition in preventing diseases and maintaining health. As a result, there is growing demand for functional foods that are nutritionally, biologically, and sensorially appealing, while also being easy, quick, and convenient to consume. Additionally, interest in personalized food is rising, as consumers seek tailored ingredients to meet their individual nutritional needs. 3D printing (3DP) technology offers solutions to these demands, providing a sustainable, simple, and highly versatile approach. A key feature of this technology is the layer-by-layer addition of material, which enables the production of complex geometric shapes that would be difficult or impossible to achieve using traditional manufacturing methods. The food industry is particularly focused on developing innovative functional foods by adding functional ingredients to provide health benefits. In this regard, 3DP offers many advantages, including the customization of food textures, shapes, and nutritional content, which is particularly useful for creating personalized diets or functional foods tailored to specific health needs. 3D food printing also opens possibilities for reducing food waste by utilizing alternative ingredients, including food by-products. Furthermore, it presents opportunities for producing more sustainable and eco-friendly food options, while improving efficiency in food production and presentation. However, adapting 3DP technology to the food sector presents challenges, as food materials often consist of various components with differing physicochemical properties. Additionally, 3DP faces challenges related to food safety and regulatory concerns. Recent studies suggest that innovative non-thermal technologies may help extend the shelf life and preserve the nutritional value of 3D-printed food. In conclusion, key areas for future development in 3D food printing include improving its speed and precision, expanding the range of printable ingredients, and making the technology more accessible to both consumers and food manufacturers.

**Keywords:** functional food, 3D printing, sustainability, quality

**Acknowledgements:** PRIMA Project „From Edible Sprouts to Healthy Food – FEED“ (Prima Call 2022, Prima Section 2 – Multi Topic 2022, Topic 2.3.1 (RIA) Enabling the transition to healthy and sustainable dietary behaviour) (HORIZON 2020 Programme).

## OPTIMISATION OF OREGANO ESSENTIAL OIL ENCAPSULATION USING FLUIDIZED BED GRANULATION

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The essential oil (EO) from the aerial parts in the flowering phase of *Origanum heracleoticum* L. (OEO), has been traditionally used in treating urinary tract infections, bronchitis, rheumatic diseases, and gastroenteritis. It also has important applications in the food industry due to its significant antimicrobial potential such as anti-fungal, bactericidal, and antiviral. Beyond antimicrobial properties, OEO, rich in phenols thymol and carvacrol, showed also antioxidant properties. Food industries are looking for natural antimicrobial agents and antioxidants to replace synthetic ones due to health reasons. EOs are natural products that can satisfy that need, but their composition and activity could change during thermal storage conditions. The process of encapsulation protects against the evaporation of volatile components from EO and minimizes unwanted changes in chemical composition. The aim of this study is to optimize the process of OEO encapsulation using fluidized bed granulation (FBG). FBG is a process that involves the conversion of atomized liquids like suspensions and emulsions into free-flowing granular solids including processes such as wetting, drying, particle size enlargement, and homogenization into a single step of processing. Our study monitored the effects of various factors (type of carrier, temperature in the fluid bed, and flow rate of the sprayed o/w emulsion of OEO with carrier solids) on the yield and encapsulation efficiency. In all samples, lactose monohydrate was used to form the fluidized bed. The highest yield and encapsulation efficiency were obtained with carboxymethylcellulose sodium as a carrier, average temperature of less than 35° C, and a 10 ml/min flow rate of OEO emulsion. The number of total phenols in prepared samples was determined using GC-MS and spectrophotometrically at 275 nm after FBG, and in selected samples after nine months of storage at ambient temperature. It was observed that fluidized bed granules had good stability. Our results showed that FBG, as a one-step drying process with gentle treatment of temperature-sensitive substances, is suitable for volatile substances such OEO.

**Keywords:** *Origanum heracleoticum*, essential oil, encapsulation, fluidized bed granulation

**Acknowledgments:** Ministry of Science, Technological Development, and Innovation (451-03-66/2024-03/200003).



## BIOLOGICAL SYNTHESIS OF SELENIUM ENRICHED CELLULOSE FILMS FOR FOOD AND COSMETIC INDUSTRY APPLICATIONS

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Increasing environmental protection requirements in the food and cosmetics industries are leading to a shift in their development towards the production of active biodegradable films based on biopolymers. Biopolymers are seen as an alternative to non-biodegradable single-use plastics, which are widely regarded as pollutants. In addition, biodegradable materials can exhibit biological properties such as antioxidant and antimicrobial activities, which can be enhanced by the incorporation of metal ions such as selenium. The cellulose-producing acetic acid bacterium *Komagataeibacter rhaeticus* was isolated from kombu broth and used to produce bacterial cellulose biopolymers. The liquid medium was enriched with selenium by adding sodium selenite at a concentration of 2.5 mg Se/L - SE-CeL2.5, and 5 mg Se/L - SE-CeL5 and cultivation was carried out under aerobic and static conditions. As a result, control, non-enriched (C-Cel) and selenium-enriched samples of cellulose pellicles were obtained. The significant selenium content in the enriched samples was determined by ICP-OES after microwave digestion. SEM analysis showed that the bacterial cellulose was densely interwoven, while the incorporated selenium was detected by EDS. FTIR analysis showed all peaks characteristic of bacterial cellulose in the C-Cel sample. Se=O vibrations were detected at 887 cm<sup>-1</sup> in both the SE-CeL2.5 and SE-CeL5 samples. According to the results of the XRD analysis, the main peaks of cellulose and Se were identified, which also confirms the incorporation of Se compounds into the cellulose matrix. The ability of the samples to reduce the number of *Staphylococcus aureus* and *Escherichia coli* was tested. SE-CeL2.5 significantly reduced the initial number of *S. aureus* cells, from 5.62 log<sub>10</sub> CFU/mL to 2.26 log<sub>10</sub> CFU/mL, while SE-CeL5 showed a weaker effect. The results obtained indicate a possible application of the process used for the production of selenium-enriched biodegradable materials with antibacterial activity for use in the food and cosmetics industry.

**Keywords:** selenium, cellulose, antibacterial activity, food industry, cosmetic industry

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Contract numbers: 451-03-65/2024-03/200116, 451-03-66/2024-/200017, 451-03-66/2024-03/200175) and the project "Green technologies for obtaining antimicrobial composites for use in cosmetics", "EU for Green Agenda in Serbia", with the technical and financial support of the European Union and in partnership with the Ministry of Environmental Protection, implemented by UNDP in cooperation with the Embassy of Sweden and the European Investment Bank (EIB), with additional funding from the Governments of Sweden, Switzerland, and Serbia (Contract number 00136377/00127312/2023/24).



## EVALUATION OF SURVIVAL IN SIMULATED GASTROINTESTINAL CONDITIONS OF APRICOT LEATHER WITH ENCAPSULATED PROBIOTIC BACTERIA ADDITION

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Probiotic-added fruit leathers are functional foods designed to provide probiotic bacteria in an easily consumable form. The primary purpose of probiotic-supplemented products is to deliver beneficial bacteria directly to the intestines, thereby supporting digestive health. Probiotics also potentially boost the immune system, alleviate digestive issues, and enhance overall gut health. Studies indicate that for a food or probiotic supplement to offer such benefits, it must reach the ileum of the small intestine at a minimum concentration of 6 log cfu (colony forming unit) g<sup>-1</sup>. This study examined the bioaccessibility of encapsulated probiotics in apricot fruit leather. Apricot fruit leather has been selected as a probiotic carrier product due to its dual role as a healthy snack and a suitable matrix for probiotics. Three probiotic strains (*Lactiplantibacillus plantarum* subsp. *plantarum* W2, *Lactiplantibacillus pentosus* XL640, and *Limosilactobacillus fermentum* W8), were added in both encapsulated and non-encapsulated form to apricot leather using maltodextrin as the coating material. Probiotic viability was evaluated using in vitro gastrointestinal conditions simulating the gastric, enteric I, and enteric II phases with minor modifications of several bioavailability methods. Lactic acid bacteria counts were measured by making appropriate dilutions on the aliquots taken from the samples after 0 minutes (before) and after each gastrointestinal phase (gastric – 2 h, enteric I–4 h, and enteric II–6 h). Consistent with similar studies, there was a decrease in the total lactic acid bacteria count during gastrointestinal transit. Encapsulated bacteria demonstrated better survival in each sample compared to their non-encapsulated counterparts of the same strain. Encapsulated *Lactiplantibacillus plantarum* subsp. *plantarum* W2 bacterium showed the highest survival at the end of the enteric II phase (6.52±0.11 log cfu/g). In previous studies, in parallel with this study, it has been found that maltodextrin coating protects samples, especially in the stomach environment. It has been found that the encapsulated forms are more suitable for the probiotic leather, and the encapsulated forms of all three strains have captured the amount that should be present even at the last stage. As a result, leather with encapsulated probiotics stands out as an innovative food supplement that can increase the bioavailability of probiotic bacteria. When the correct formulation and preventive techniques are used, these products can be effective in supporting digestive health.

**Keywords:** Probiotic apricot leather, Microencapsulation, *Lactiplantibacillus plantarum* subsp. *plantarum* strain W2, *Lactiplantibacillus pentosus* strain XL640, *Limosilactobacillus fermentum* strain W8

## EFFECTS OF COLD PLASMA-TREATED WHEAT FLOUR ON THE SENSORY CHARACTERISTICS OF WHEAT, WHOLE GRAIN, AND BRAN-ENRICHED BREADS

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This study investigated the effects of cold atmospheric plasma-treated wheat flour on the sensory quality of bread. Two plasma treatments were applied: OT\* (minimized SRCLa and maximized SRCSu) and OT\*\* (maximized both SRCLa and SRCSu). Three types of bread were examined: wheat bread (T-400 flour), mixed whole grain bread (50% T-400 flour: 50% whole grain flour), and bran-enriched bread (90% T-400 flour: 10% wheat bran). For each bread type, four samples were prepared: a control, using untreated flour, two samples with 10% of the control flour substituted by treated flour, and two samples made entirely from treated flour (T-400 flour). In total, 11 bread samples were produced and evaluated. Sensory evaluation, including crumb structure assessment and scoring method, was performed to evaluate the impact of treated flour on bread quality. Addition of plasma-treated flour improved the sensory quality of wheat and bran-enriched breads but had a negative impact on mixed whole grain bread quality. In wheat bread, 10% substitution with treated flour improved crumb quality and overall sensory scores, particularly this was evident for the second treatment. Bran-enriched bread showed unexpected improvements in crumb elasticity and sensory quality with treated flour addition. Conversely, whole grain bread exhibited poorer crumb structure and lower sensory scores when incorporating treated flour. Substitution of 10% of flour with treated flour showed better effects on bread quality compared to the use of 100% treated flour. The study indicates that cold atmospheric plasma treatment of wheat flour can enhance the sensory qualities of certain bread types, but its effectiveness varies depending on bread composition and treatment conditions.

**Keywords:** Cold atmospheric plasma, wheat flour, bread quality, sensory evaluation

**Acknowledgements:** This work was funded by the Ministry of Science, Technological Development and Innovations, Serbia, contract no. 451-03-66/2024-01/200222.

## TECHNOLOGICAL CHALLENGES AND UP-TO-DATE SOLUTIONS IN FORMULATING LOW-FODMAP CEREAL-BASED PRODUCTS

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The prevalence of irritable bowel syndrome (IBS) has highlighted the need for low-FODMAP (Fermentable Oligosaccharides, Disaccharides, Monosaccharides, and Polyols) diets, particularly concerning cereal-based products. The aim of the study was to review the technological challenges and contemporary solutions in formulating low-FODMAP cereal-based products, with a focus on low-FODMAP bread production. The primary challenges include the inherent high FODMAP content in grains such as rye and wheat, which are staples in many diets. This research explores various methods to reduce FODMAP levels and the challenges of maintaining the nutritional and sensory qualities of these products. The solutions involve mainly the materials selection and optimizing fermentation processes, particularly through the use of specific bacterial strains. Studies have shown that extended fermentation times, significantly reduce FODMAP content. For instance, a 72-hour fermentation with these strains reduced FODMAPs in wheat bread to less than 0.1 g/100 g while increasing antioxidant levels. Similar results were observed in rye bread, where controlled fermentation with selected bacteria strains decreased FODMAP levels despite slight changes in organoleptic properties. Additionally, the type of flour and fermentation methods significantly affects FODMAP content. For example, rye flour, a common ingredient in sourdough bread, presents high levels of fructans. However, the use of endosperm rye flour combined with sourdough fermentation showed improved baking properties and consumer acceptability, with reduced fructan content. Sourdough inoculation with strains like *Saccharomyces boulardii* also demonstrated potential in lowering fructan levels and enhancing dietary fiber solubility. The integration of targeted fermentation techniques with specific bacterial strains offers a promising approach to developing low-FODMAP cereal-based products. These advancements address dietary restrictions associated with IBS and enhance the nutritional profile and consumer acceptability of cereal-based foods. Further research could lead to wider adoption and innovation in the food industry, benefiting individuals with FODMAP-related disorders.

**Keywords:** low-FODMAP diet, cereal-based products, IBS, sourdough bread, LAB fermentation

## RECOVERY OF PROTEIN FROM POTATO PROCESSING WASTEWATER USING THERMOCOAGULATION TECHNIQUES

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Food waste reduction and industrial side-streams conversion into new functional and nutritional ingredients are crucial today for promoting environmental sustainability and building zero-waste circular bio-refineries. Among these, potato processing wastewater has been a lasting challenge in potato processing due to the high quantity generated during starch extraction, along with significant level of chemical oxygen demand (COD) and biochemical oxygen demand (BOD), which generates sludge. However, potato wastewater also contains a significant amount of valuable proteins with high functionality and nutritional value, making potato proteins highly regarded among plant proteins. These proteins are usually allergen-free soluble proteins, characterised by considerable value if effectively extracted and valorised. Some of these proteins also exhibit beneficial techno-functional as well as bio-active properties, such as anti-tumor and free radical scavenging activities. The methods used for proteins extraction from potato wastewater, including extraction variables, significantly influence the purity, yield and quality of the final product. Common methods include ethanolic or acid precipitation, thermal coagulation, and combinations of these treatments. A protein recovery process from potato wastewater constituting heating at 70 °C before and after iso-electric pH adjustment, resultant in a precipitate having 65% protein content. This study demonstrates that proteins can be recovered from potato wastewater, providing the possibility of converting this waste product into a valuable resource.

**Keywords:** potato, wastewater, protein extraction, side-stream, resource

**Acknowledgements:** This research was funded by the project “Upcycling Side-Streams for Sustainable and Healthy Ingredients and New Food Concepts (UP4Food, 2024-2027)”, supported by the Department of Agriculture, Food and the Marine (DAFM). The funding falls under the EU Joint Programming Initiative “A Healthy Diet for a Healthy Life” (JPI HDHL).

## DETERMINATION OF OPTIMAL QUANTITY TOMATOES BY-PRODUCT IN FERMENTED CRACKERS USING MATLAB – A SUSTAINABLE APPROACH FOR VALORIZATION OF AGRICULTURAL WASTE

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The use of certain waste products in the production of crackers can lead to improved characteristics and increased nutritional content. However, the optimal amount of additives to replace basic raw materials remains uncertain. The present work aims to determine the optimal amount of tomato pulp in crackers. Crackers were developed by using waste from tomato processing and the evaluation of their physical, chemical, and sensory characteristics. The study used a selection method to choose relevant features for regression analysis and a second-order polynomial model. A linear programming algorithm was used to determine the optimal amount of additive. Results of a study found that the addition of 9.27% of raw material to crackers improved their characteristics and remained acceptable to consumers. The study's optimal amount is higher than what is reported in existing literature, indicating an 8% permissible addition of tomato pomace in crackers. The higher optimal amount determined in this work can help replace more of the main raw materials used in the production of crackers.

**Keywords:** Food waste, Sustainability, Statistical analysis, Optimal quantity

**Acknowledgements:** This work was financially supported by the Bulgarian Ministry of Education and Science, National Research Fund under contract number: КП-06-КОСТ/20.

## THE EFFECT OF AMARANTH SEEDS ON THE CHEMICAL, MICROBIOLOGICAL AND SENSORY QUALITY OF YOGURT

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The objective of this study was to investigate the chemical, microbiological, and sensory characteristics of yogurt enriched with amaranth seeds, stored at a temperature range of  $5\pm 1^\circ\text{C}$  for 21 days. The quantity of amaranth incorporated into the yogurt was 5% and 10%. The changes were regularly observed and recorded every 7 days. Statistically significant results ( $p \leq 0.05$ ) show that as storage time increases, pH values decrease, and acidity increases. The fat content remains consistent regardless of the amount of amaranth stored. The inclusion of amaranth seeds in yogurt resulted in a statistically significant increase ( $p \leq 0.05$ ) in the protein content of the final product. All yogurt samples demonstrate the absence of *Enterobacteriaceae*, yeast and molds, Coagulase-positive staphylococci, *Salmonella* spp. and *Listeria monocytogenes* after 21 days of storage, ensuring product safety. Sensory quality was evaluated based on color, taste, aftertaste, texture, and overall acceptability at 1, 7, 14, and 21 days. The results indicate that the sensory quality of yogurt decreases as the concentration of amaranth seeds increases. Except for the control sample (without amaranth seeds), yogurt with 5% amaranth seeds exhibited superior sensory quality compared to yogurt with 10% amaranth seeds.

**Keywords:** yogurt, amaranth, storage, quality

**Acknowledgements:** This work was financially supported by the Bulgarian Ministry of Education and Science, National Research Fund under contract number: KP-06-KOCT/20.

## PREPARATION AND CHARACTERISATION OF $\beta$ -CAROTENE-INULIN PARTICLES

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$\beta$ -carotene (CAR) is a precursor to vitamin A, an essential nutrient in the human diet. As an antioxidant, CAR helps to protect cells from damage caused by free radicals, potentially reducing the risk of chronic diseases. However, due to their fast-paced lifestyles, people do not include enough CAR- or other antioxidant-containing foods in their diets. Also, CAR is very unstable, sensitive to visible and UV light, and is not soluble in water. Therefore, in this study, it was aimed to encapsulate CAR with plant-based polysaccharide inulin (IN) to stabilize and improve CAR properties. Inulin is prebiotic as it is not digested in the human digestive system, does not elevate blood sugar levels, and is soluble in water. Thus, it is a reasonable candidate for the encapsulation of CAR. The preparation of CAR encapsulated in inulin particles (CAR-IN) was carried out by addition of CAR in an organic solvent to hot water dispersion of inulin and following evaporation of the organic solvent. The CAR-IN particles were characterised with DLS, FTIR, DSC and NMR methods. The antioxidant properties were investigated with ABTS, FRAP, and DPPH methods and showed high antioxidant and radical scavenging abilities. The particle size of CAR-IN complexes was 200 – 300 nm and depended on the amount of IN used in the preparation of the particles. Most importantly, the solubility of CAR-IN particles was achieved as high as 45 – 50 mg/mL in water, which is a notable improvement when compared to CAR solubility. Overall, the preparation of CAR-IN particles improved CAR properties and it could be used as a food additive to enhance the human diet with antioxidants.

**Keywords:**  $\beta$ -carotene, inulin, antioxidant, solubility

**Acknowledgements:** This research was funded by Research Council of Lithuania project No. S-PD-24-55.

## EFFECTS OF NATURAL COLORANTS AND APPLICATION OF PULSED ELECTRIC FIELD (PEF) TECHNOLOGY ON QUALITY PARAMETERS OF NITRITE-FREE BACON

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The replacement of nitrites in cured meat products, such as bacon chops, presents challenges in maintaining the characteristic pink colour and other quality attributes. This study evaluates the effectiveness of natural colorants: betanin, red yeast (*Monascus*) rice extract, and roselle (*Hibiscus sabdariffa*) extract, as alternatives to nitrites for colour enhancement in nitrite-free bacon. Pulsed electric field (PEF) pre-treatment was applied to samples prior to immersing in brine with colorant. Bacon loins were then treated with brines containing different concentrations of these colorants, vacuum packaged, tumbled, and stored at 4°C until further testing. Instrumental colour parameters (L\*, a\*, b\*, hue angle, chroma, and cured colour ratio) were measured for both uncooked and cooked samples. Results showed that uncooked bacon treated with 0.05% betanin exhibited similar redness (a\*) and cured colour ratios to nitrite-treated controls, indicating that betanin has potential as a feasible colorant. Red yeast rice extract at 0.10% also enhanced redness but increased yellowness (b\*), affecting the overall colour. Though a\* values were comparable with the control, all samples with colorants displayed significantly (P<0.05) higher b\*, hue angle, and chroma values, and decreased L\* and cured colour ratio values than nitrite-treated samples upon cooking. While PEF treatment applied did not affect the instrumental colour of the uncooked and cooked samples, it was beneficial for reducing lipid oxidation on uncooked bacon with betanin and roselle extracts. These findings suggest that while natural colorants can mimic the colour effects of nitrites in uncooked bacon, additional strategies are required to improve colour stability.

**Keywords:** nitrite-free, meat products, natural colorants

**Acknowledgements:** This research is funded by the Department of Agriculture, Food, and the Marine (DAFM) Ireland, Funding Agency Reference Number 2021R426, Project "Novel Nitrate/Nitrite Reduced and Nitrate/Nitrite Free Meats: Safety, Quality and Shelf-life" and the Teagasc Walsh Scholarships Programme.



## FAT REDUCTION OF ROLL-IN FAT FOR PUFF PASTRY FROM RHEOLOGICAL AND TEXTURAL ASPECTS

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Puff-pastry bakery products are specific by flaky structure made of alternating layers of dough and semi-solid roll-in fat. Adequate quality of puff pastry bakery products relies on rheological properties of the roll-in fat. Significant amount of fat in puff pastry, up to 60%, is considerable factor for these products from health aspect. Thus, new aspects of puff pastry production should include tendency for fat reduction. The possibility to reduce fat content in roll-in fat for puff pastry with cellulose-based fat mimetic was observed in this work from rheological and textural aspect. The cellulose-based fat mimetic is microcrystalline cellulose gel (MCG) that contains fibers of colloidal microcrystalline cellulose with cellulose gum. Sufficiently hydrated particles of colloidal microcrystal cellulose form soft, spherical aggregates, physically similar to the globular structures of fat and simulate the functional and sensory properties of fat. Different concentration of MCG was formed (3, 5 and 7%) and used for fat reduction in mixture with roll-in fat in amount of 15, 30 and 45%. After homogenization the rheological and textural characteristics of the obtained mixtures were analyzed. Rheological determination of flow properties showed that obtained mixtures maintained a partial structure and had a certain degree of plasticity, favorable property of spreadable food components. Increasing amount of MCG decreased the consistency of observed mixtures. Elastic modulus  $G'$  was higher than viscous modulus  $G''$  and pointed to domination of elastic linkages. Textural determination significantly confirmed rheological results and firmness and work of shear of the mixtures decreased at high amount of MCG fat mimetic. According to rheological and textural results, obtained mixture can realize the functional properties of fat comparing to control roll-in fat. Further analysis should be directed to specific application of this mixture in a fat-reduced puff pastry product, in the aim to confirm its functionality.

**Keywords:** MCG fat mimetic, roll-in fat, puff pastry, rheology, texture

**Acknowledgements:** This research was supported by the Program of Ministry of Science, Technological Development and Innovation of Republic of Serbia (number: 451-03-65/2024-03/200134 and 451-03-66/2024-03/200134).

## MUSTARD SEED FLOUR ADDITION IN BREADMAKING – EFFECTS ON DOUGH RHEOLOGY AND BREAD QUALITY

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The incorporation of mustard seed flour in breadmaking has garnered interest due to its potential to enhance nutritional and functional properties of bread. Mustard seeds are rich in proteins, dietary fiber and bioactive compounds, offering potential health benefits and improved sensory attributes when used as a flour substitute in baking. However, its addition affects rheological properties of the dough, which are critical for the baking process and the final product quality. In this study, the rheological properties of dough in which part of the wheat flour was substituted with 1, 5 and 10% mustard seed flour (MF) were examined using farinograph and extensograph. Dough samples with 5 and 10% of MF had 3 times longer development time, and stability of mentioned samples was increased by 33,3% in comparison to the control sample. This could be an effect of the MF composition, especially the high content of mucilage, which acts similarly to polysaccharide gums (glucomannans and pectins). However, regardless of quantity, introduction of MF into the dough resulted in significant increase of degree of softening compared to control, suggesting the weakening of gluten structure due to its dilution. Furthermore, the addition of MF reduced the dough energy by 4 to 37% in regards to control sample. Resistance of dough with MF addition increased by 40-50% and extensibility decreased up to 50-60% compared to the control dough. As a consequence of deteriorated rheological properties, bread with 1-10% of MF had significantly lower specific volume (1,05-1,71 cm<sup>2</sup>/g) in regards to the control (3,03 cm<sup>2</sup>/g). Obtained results indicate that MF addition caused weakening of gluten structure, which prohibited expansion and retention of gases formed during breadmaking. In conclusion, further research should aim to develop strategies to maximize the benefits of MF in bread while mitigating its adverse effects on dough rheology and product quality.

**Keywords:** mustard seed flour, dough rheology, bread quality

**Acknowledgements:** This work was supported by the Program of Ministry of Science, Technological Development and Innovation of Republic of Serbia (number: 451-03-65/2024-03/200134 and 451-03-66/2024-03/200134). The work of M. R. in Serbia was financially supported by the CEEPUS Network CIII-HR-0306-12-1920 For Safe and Healthy Food in Middle-Europe.

## APPLICATION OF PROTEIN POWDERS IN PRODUCTION OF HIGH-PROTEIN PASTRY

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This study aims to provide a comprehensive analysis of how the addition of plant and animal-based protein powders impacts the rheological properties of wheat dough, using farinograph and extensograph for assessment. The novelty of this research is based on comparing well-known protein isolates such as soy and whey with less common ones like hemp, wheat bran (WBPI), brown rice, and hydrolysed collagen. By combining dough analysis with an in-depth examination of the techno-functional properties of these protein isolates, the study seeks to elucidate the mechanisms by which they influence dough structure and functionality. Protein powders had significantly different functional properties. The solubility of tested protein powders varied, with animal-origin powders showing the highest solubility. The lowest solubility occurred at pH 3–5, near the isoelectric point of the most proteins. Soy protein isolate had the best water-holding capacity (10.65 g<sub>H<sub>2</sub>O</sub>/g), foaming capacity (109.6%), and emulsion properties, followed by pea protein with a WHC of 5.32 g<sub>H<sub>2</sub>O</sub>/g. WBPI exhibited the best oil-holding capacity (2.18 g<sub>oil</sub>/g). Foaming properties varied but correlated positively with water-holding capacity. Most protein powders showed good emulsion activity, except for beef, hemp, and rice proteins. Protein powders were used for the fortification of wheat flour to 20% protein content. Farinograph and extensograph analysis revealed significant impact of fortification on rheology of dough. Extensograph measurements showed significant differences in dough energy with protein powder addition. Adding protein to 20% typically reduced extensograph energy, except with wheat gluten and hydrolyzed collagen, which increased the resistance. Longer rest periods improved dough resistance. Gluten, pea, collagen hydrolysate, and rice isolates were suitable for longer processing, while whey, hemp, and WBPI resulted in low energy and reduced extension. Plant-based proteins generally made dough tougher and less extensible, impacting dough expansion, shape stability, and proofing, thus affecting overall product quality.

**Keywords:** high protein pastry, farinograph, extensograph, protein isolates

**Acknowledgements:** This research was supported by project no. FCH–S-23-8330 financed by the Ministry of Education, Youth and Sports of the Czech Republic.

## OPTIMIZING POTENTIAL OF UNDERUTILIZED HOT TRUB AS A BREWING BY-PRODUCT USING GREEN EXTRACTION TECHNIQUES

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Large amounts of by-products are produced in beer production, with hot trub being one of the most valuable brewing streams. The hot trub has an abundant chemical composition that includes insoluble hop materials, condensation product of hop polyphenols and wort proteins, and isomerised hop acids. All of these compounds have biological activity such as antioxidant, antimicrobial, antifungal, antiviral, anti-inflammatory, and anticancer. Hops waste is commonly used as livestock feed, disposed of in the fields or incinerated. The extraction or recovery of high-value bioactive compounds from hot trub as a strategy to solve this environmental and economic issue has recently been emphasized. The aim of this study was to determine the total phenols and flavonoids content and to examine antioxidant activity in hot trub using novel extraction techniques with special emphasis on green and environmental aspects instead of conventional extraction techniques. Ultrasound-assistant extraction was performed and ethanol was used as the solvent in different concentrations (20, 40, 60, 80, and 96%). Total phenols and total flavonoids content was evaluated by HPLC assays, while the antioxidant activity was tested *in vitro* by DPPH, FRAP, and ABTS tests. The highest yields of these bioactive compounds and the highest antioxidant activity were obtained using ethanol concentration of 60%. Then, the impact of extraction temperature on total phenols and flavonoids content and antioxidant activity was investigated at various temperatures. The highest total phenols content and antioxidant activity were obtained by applying extraction temperature of 50 °C (0.5961 g GAE/100 g and 11.1060 µM/g, respectively), while the most optimal extraction temperature for total flavonoids content was 40 °C (0.1572 g CE/100 g). The obtained results indicate that using this extraction technique has a huge potential for the valorization of hot trub as a brewery by-product in the isolation of novel extracts and bioactive compounds.

**Keywords:** beer, hot trub, green extraction technologies, polyphenols, flavonoids

**Acknowledgements:** This research was funded by the Program of the Ministry of Education, Science, and Technological Development of the Republic of Serbia (No. 451-03-47/2023-01/200134), Science Fund of the Republic of Serbia for the funding of the IDEAS project 'Novel extracts and bioactive compounds from under-utilized resources for high-value applications – BioUtilize', number '7750168', and by Provincial Secretariat for Higher Education and Scientific Research, Republic of Serbia, Autonomous Province of Vojvodina (No. 142-451-3498/2023-01/01).

## HYPOGLICEMIC POTENTIAL, POLYPHENOLIC PROFILE, HYGROSCOPIC AND THERMAL PROPERTIES OF ELDERBERRY (*SAMBUCUS NIGRA* L.) MICROCAPSULES

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Elderberry (*Sambucus nigra* L.) is characterized by a high content of nutritionally valuable components, mainly polyphenolics. However, the isolated elderberry polyphenolics are unstable compounds and very susceptible to degradation towards environmental conditions, especially during processing and storage. The high polyphenolic instability limits their incorporation into food and medical products, which encapsulation can overcome. In this research, a microwave-assisted extraction was used to prepare extracts from lyophilized elderberries. Demineralized water was used as a solvent for extraction. The obtained extracts were encapsulated by using maltodextrin (MD), gum arabic (GA), whey protein concentrate (WPC), sodium alginate (ALG), soy protein (SP) and inulin (IN) as carriers by lyophilization for 48 hours. Two different ratios (1:1 and 6:1) of carrier/core material to the total solid content of elderberry extract (EE) were used. The thermal properties of obtained elderberry powders were analyzed by thermogravimetric analysis. The hygroscopicity of powders was determined at 75.29% relative humidity at 25 °C. Quantitative analysis of 40 polyphenolic compounds by HPLC-UV/VIS technique was applied to elucidate differences in samples' phenolic profile. The hypoglycemic potential of samples was evaluated *in vitro*, as an inhibitory effect on  $\alpha$ -amylase, a digestive enzyme involved in carbohydrate metabolism. The best thermal stability of EE was obtained by using WPC or GA (carrier/core ratio of 6:1). The usage of WPC, MD, and IN with carrier/core ratio 6:1 can be selected as carriers with the lowest hygroscopicity. However, regardless of hygroscopicity value, most elderberry powders suffered aggregation and swelling at investigated humidity, where only WPC and IN (at IN: EE= 6:1) powders did not physically change. The polyphenolic profile evaluation results in quantification of 13 compounds, with qualitative and quantitative differences between samples. Also, differences in amylase inhibitory potential were noticed. The obtained results suggest that elderberry encapsulation could enhance its application in the food, cosmetic, and pharmaceutical industries.

**Keywords:** elderberry extracts, freeze drying, encapsulation, thermal stability, polyphenolic profile, hypoglycemic effect

**Acknowledgments:** This research study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Project No. 451-03-65/2024-03/ 200134).

## SUPERCritical EXTRACTION OF LADY'S MANTLE HERB (*ALCHEMILLA SUBCRENATA* BUSER) - PHYTOCHEMICAL CHARACTERIZATION AND PHARMACOLOGICAL POTENTIAL

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*Alchemilla subcrenata* Buser belongs to the Rosaceae family and is characterized by nutritionally valuable components that are especially distributed in the herbs of the plant. Plant material was collected in the vicinity of Lake Vlasina in the territory of Serbia. The dried herb of the plant was used to obtain Lady's mantle extract using the supercritical fluid extraction (SFE) technique. During the SFE the temperature (40 °C, 50 °C and 60 °C) and pressure (100 bar, 250 bar, and 400 bar) were varied, while the gas flow was unchanged. The obtained extracts were analyzed for the content of total phenols and flavonoids, and the content of terpenes determined by GC-MS technique. The raffinate was collected from the extract with the highest yield (T=50 °C, p=400 bar) and its chemical composition (LC-MS/MS technique) and pharmacological activity were examined. The pharmacological potential was investigated using several *in vitro* antioxidants (DPPH, ABTS, FRAP, CUPRAC, metal chelation) and enzyme inhibition assays (cholinesterase, tyrosinase,  $\alpha$ -amylase, and  $\alpha$ -glucosidase). The results of the investigation showed that phytol was dominant in all examined extracts. The highest phytol content was determined in the extract obtained at the following process parameters: T= 40 °C and p=250 bar (30.34%). Apart from phytol, the content of linolenic acid was significant, which was found in the highest percentage in the extract obtained at T=40 °C and p= 400 bar (17.38%). The best antioxidant potential was shown by the extract obtained at the process parameters: T= 50 °C and p= 100 bar (87.49 mg TE/g), while the extract obtained at T= 40 °C and p=100 bar was the most effective enzyme inhibitory agent against cholinesterases and  $\alpha$ -glucosidase. Namely, it could be assumed that the terpene compounds present in the investigated extracts in different percentages are the result of the influence of the selected process parameters.

**Keywords:** *Alchemilla subcrenata*, chemical composition, pharmacological potential

**Acknowledgments:** This research study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Project No. 451- 03-65/2024-03/200134).

## NUTRITIONAL CHARACTERISTICS AND BIOACCESSIBILITY OF GLUTEN-FREE CRACKERS CONTAINING PUMPKIN SEED PRESS CAKE FLOUR

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Utilizing chickpea and pumpkin seed press cake flours as nutritionally valuable ingredients in the production of gluten-free crackers offers a promising approach to addressing nutritional gaps while catering to the increasing demand for healthier gluten-free alternatives. However, pumpkin seed press cake, a by-product of the oil extraction process, is often discarded despite being a good source of valuable compounds with multitudes of positive health effects. Therefore, the objective of the present work was to evaluate the potential of chickpea and pumpkin seed press-cake flours as base ingredients for gluten-free cracker production, considering nutritional quality as well as to study the changes in the total phenolic content, antioxidant capacity, protein composition and mineral content caused by the digestion processes. The crackers were produced by substituting 20% and 35% of chickpea flour by using two types of pumpkin seed press cake flour, one from virgin oil pressing and the other from cold-pressed oil pressing. The proximate composition of the crackers is influenced by the gradual replacement of chickpea flour with pumpkin seed press cake flour. The inclusion of pumpkin seed press cake flour led to higher levels of protein, fat, and ash in the crackers, while reducing the total carbohydrate content. Given the high dietary fiber and protein content of these raw materials, the crackers could be labelled as "high in fiber" and a "source of protein." In most cases, the substitution of chickpea flour with press cake flour improved the functional properties of crackers. Specifically, all samples after digestion demonstrated a significant increase in total phenolic content and antioxidant activity, with a greater increase in crackers with press cake flour obtained from the cold-pressed oil production. The protein digestibility of crackers with cold-pressed cake flour was similar to that of the control sample, while other crackers showed lower values. Mineral content analysis indicated good bioaccessibility for zinc, iron, and magnesium. These results highlight the potential of pumpkin seed press cake as a rich source of bioactive compounds. Moreover, this by-product, along with chickpea flour, offers a valuable alternative to traditional gluten-free flours for producing nutrient-rich, health-promoting gluten-free crackers.

**Keywords:** gluten-free crackers, pumpkin seed oil processing by-products, in vitro digestion, antioxidant activity, mineral content

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).



## COMPARISON OF POLYPHENOLIC COMPOSITION, ANTIOXIDANT ACTIVITY, AND SENSORY CHARACTERISTICS OF MERLOT WINE AGED IN OAK (*QUERCUS PETRAEA*) BARRELS FROM SERBIA AND FRANCE

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The influence of wood on wine maturation is complex. The effect of oak on wine is determined by the type, origin, and production technology of the barrels. *Quercus petraea* is commonly used for wine barrels due to its favorable mechanical properties and high aromatic potential. This study examined the phytochemical profile of Merlot wine after one month of aging in four oak (*Quercus petraea*) barrels—two from Serbia (Fruška gora, Vrdnik area, and Batot forest of Kopaonik) and two from France. The analysis included the determination of the content of total polyphenols, flavan-3-ols, anthocyanins, monomeric anthocyanins, and tannoid substances using spectrophotometry, while HPLC was used to determine qualitative and quantitative content of phenolic acids. Antioxidant activity was assessed through DPPH, ABTS, and reducing power assays. Sensory evaluation was conducted by five evaluators following the standardized procedure (ISO 3591, 2016). The results showed high polyphenolic content in all wine samples. A statistically significant difference ( $p < 0.05$ ) in polyphenolic content was found between wine aged in barrel from Batot forest and wine aged in French barrel. However, no significant differences were found in total anthocyanins, monomeric anthocyanins, or tannoid compounds between wines aged in Serbian and French oak barrels. Flavan-3-ols and monomeric anthocyanins decreased in wine samples from Serbian and French oak barrels compared to the control ( $p < 0.01$ ), likely due to polymerization and condensation reactions. HPLC analysis identified gallic acid as the most abundant, with a 21% higher level in wines aged in French oak barrels. All samples showed high antioxidant activity, with the wine aged in the barrel from Batot forest standing out. After 30 days of aging, the scores for overall appreciation were similar to the control. The research will continue to monitor the wine's chemical composition and sensory characteristics, as significant differences are expected to become more noticeable over time.

**Keywords:** wine, oak barrels, antioxidant activity, chemical characterization, sensory analysis

**Acknowledgements:** This work was supported by The Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant No. 451-03- 66/2024-03/200358, 451-03-65/2024-03/200134 and 451-03-66/2024-03/200222).



## **FREEZE-DRIED HORSERADISH LEAF POMACE AS A NOVEL VALUABLE SOURCE OF ANTIOXIDANTS**

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Cold-pressed horseradish leaf juice processing results in large quantities of pomace as a by-product, which is usually disposed of as waste. However, the pomace contains various antioxidant compounds that can be recovered and potentially used in the food industry. To reduce enzymatic and microbiological activity and achieve a more favorable form for addition to food, it is preferable to dry the fresh pomace. One of the most suitable drying techniques for the preservation of bioactive compounds is freeze-drying, which is performed under mild, non-thermal conditions. Therefore, this study aimed to obtain freeze-dried horseradish leaf pomace and to investigate the physicochemical properties of this by-product along with its potential as a source of phenolic compounds by spectrophotometric and chromatographic analysis. The freeze-drying process was performed under the following conditions: at a temperature of -40 °C and a pressure of 0.12 mbar for 48 h. In addition to the significantly reduced moisture content, low values for water activity and hygroscopicity were determined for the freeze-dried horseradish leaf pomace using standard analytical methods. Spectrophotometric methods were used to characterize the ethanolic (80% v/v) pomace extract, and high values were obtained for total phenolic, flavonoid, and phenolic acid contents as well as antioxidant activity (determined by DPPH, ABTS, and FRAP methods). Using an ultra-high-performance liquid chromatography system, coupled with a quadrupole time-of-flight mass spectrometry, a total of ten phenolic compounds were quantified after extraction from the freeze-dried powder. Phenolic compounds from the classes of flavonoids and phenolic acids were detected, with the kaempferol derivatives dominating quantitatively. The obtained results highlight freeze-dried horseradish leaf pomace as an underexplored source of antioxidant compounds whose addition to food products could reduce the amount of oxidation-caused waste.

**Keywords:** horseradish leaf pomace, freeze-drying, phenolic compounds, antioxidant activity, waste reduction

**Acknowledgments:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant numbers 451-03-65/2024-03/200116 and 451-03-66/2024-03/200007) and the Science Fund of the Republic of Serbia (grant number 7744714, FUNPRO).

## HORSERADISH LEAF BY-PRODUCT: A NATURAL ANTIOXIDANT IN MAYONNAISE PRODUCTION

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Due to the high biological activity of fresh horseradish leaves, it is desirable to press them to obtain juice that can be used in the food industry. In addition, a pomace rich in phenolic compounds with a pronounced antioxidant activity remains as a by-product of pressing. 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 analyze the effect of adding freeze-dried horseradish leaf pomace on the mayonnaise's oxidative stability and quality during an eight-week storage period. The mayonnaises were produced using the following ingredients: sunflower oil (75%), egg yolk (3%), vinegar (3%), sugar (3%), and salt (1%). The water content (15%) was reduced by adding freeze-dried pomace (in an amount giving a total phenolic content of 400 mg gallic acid equivalents/kg mayonnaise). The control mayonnaise was prepared without adding horseradish pomace. The total oxidation value, calculated as the sum of the primary and secondary oxidation products, was higher in the control sample after eight weeks of storage than in the mayonnaise containing horseradish pomace. Based on an accelerated oxidative stability test, the horseradish pomace proved to be very effective in delaying mayonnaise oxidation by prolonging the induction period compared to the control sample. The horseradish pomace also improved the product quality with a higher pH value after storage compared to the control. In conclusion, freeze-dried horseradish leaf pomace positively affected the oxidative stability and quality of mayonnaise, indicating the great potential of this natural antioxidant as a substitute for synthetic ones in the food industry. Furthermore, this study promotes the circular economy by providing insights into the assessment valorization of horseradish leaf pomace regarded as agricultural biowaste.

**Keywords:** horseradish leaf pomace, freeze-drying, antioxidant activity, mayonnaise, oxidative stability

**Acknowledgments:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant number 451-03-65/2024-03/200116).

## RESEARCH PROGRESS AND FUTURE TREND OF GINGER PROCESSING IN CHINA

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Ginger (*Zingiber officinale*) belongs to the Zingiberaceae family, which has been widely used in culinary recipes as an edible spice either in fresh or dried powder form due to its powerful refreshing and pungent aroma. Records including *Analecets*, *Compendium of Materia Medica* show that ginger has been used as one kind of edible and medicinal vegetable in ancient China for over 3000 years, attributing to its beneficial nutrients and pharmacological activity against cough, cold, nausea, vomiting, food poisoning, arthritic pain and radiation-induced toxicities and diseases. FAO reported that the total output of ginger around the world has reached nearly 15 million tons in recent years, and the top countries including China, Nigeria, India, Indonesia, Thailand, Bangladesh and Brazil, accounted for more than 90% of the total cultivation area all over the world. Among them, the cultivation area and total output in China reached more than 300 000 hectares and 10 million tons, accounting for about 1/2 and 2/3 of the world, respectively. Due to its large output and great popularity among consumers, we studied the agronomic characters and chemical compositions of gingers derived from 25 varieties to evaluate their processability and potential applications, such as fresh consumption, drying, curing and extraction of functional components. It would provide theoretical basis on the yield, structure and physicochemical properties of essential oil, resistant starch and dietary fibre in ginger. However, the full utilization of ginger's benefits and ginger agro-residues as well as the relative large-scale processing equipment remain largely unexplored. Therefore, further studies are necessary to confront and resolve these challenges.

**Keywords:** ginger, bioactivities, production status, processing methods, outlook

**Acknowledgements:** This research was funded by the Natural Key Research and Development Plan, grant number 2023YFD1600203.

## MICROSTRUCTURE AND MECHANICAL PROPERTIES OF YOGURT PRODUCTS

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Homogenization is an important and necessary pretreatment for processing milk and dairy products, such as yogurt, as it prevents creaming during incubation and storage. This process enhances the stability, consistency, and body of yogurt, resulting in superior product quality. Ultrasonic processing or sonication is a promising alternative technology in the food industry for homogenizing milk, resulting in a decrease in the size of milk fat globules. Therefore, this process can significantly modify the final technological and functional properties of yogurts, influencing the texture, microstructure and water holding capacity. The aim of the research was to evaluate the impact of ultrasonication for sheep milk homogenization on the mechanical properties and microstructure of yogurt products. The study investigated the effect of ultrasonication on both whole and skimmed milk, with varying parameters such as amplitude and time interval. Yogurt acidification was achieved using a starter culture of lactic acid bacteria. Light scattering and confocal microscopy were used for the determination of droplet size and microstructure of the sonicated emulsions, respectively. The rheological properties, water holding capacity and acidification kinetics of the obtained yogurt products were also evaluated. Ultrasonication significantly decreased the droplet size of milk fat globules, resulting in alterations in the microstructure of the yogurt and a reduction in syneresis. Also, the utilization of ultrasound in milk homogenization affected the rheological properties, resulting in the formation of stronger gels, while improving the water holding capacity in yogurts. Overall, ultrasonication contributes to the formation of yogurts with superior texture, consistency and functional properties.

**Keywords:** yogurt, milk fat globules, ultrasonication

**Acknowledgements:** This project was co-funded by Greece and European Union (Project title: Utilization of genetic material of breeds and farming systems to create products with increased added value from sheep's milk, M16SYN2-00069).

## PRODUCTION OF PROTEIN HYDROLYSATES BY ENZYMATIC HYDROLYSIS OF RAPESEED PRESS CAKE

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The worldwide increasing demand in proteins for human nutrition and animal feeding leads to a growing interest in novel protein sources. Therefore, rapeseed as an established raw material for the production of edible oils could be a promising alternative, as large amounts of press cakes are available. At present, rapeseed cake is primarily directed into the animal feed market. However, there is an opportunity for its use as a high-quality food protein source. Despite decades of research, several technologies being developed, and products being brought to large scale production, there are still not many commercially available rapeseed protein products. The conventional way of obtaining protein - alkaline extraction with isoelectric precipitation, in this work was replaced by direct enzymatic hydrolysis of rapeseed cake in order to increase the protein yield and obtain hydrolysate with improved properties. Hydrolysis was performed using two enzymes, alcalase and pepsin, the degree of hydrolysis was determined and the obtained hydrolysates were characterized using electrophoresis. In addition, the antioxidant activity of the obtained hydrolysates was determined. The results showed that protein hydrolysates obtained with the application of both used enzymes have high antioxidant activity, which makes them potentially good for use in different formulations of food products.

**Keywords:** protein hydrolysates, rapeseed press cake, bioactivity

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovations, Republic of Serbia (Contract No. 451-03-66/2024-03/200222), as well as by the Science Fund of the Republic of Serbia, Grant No 6673, PROTein from Rapeseed Oil Processing Waste: Application in Food and Wastewater Treatment – PROTOPOWER.

## OPTIMIZING FRUIT JUICE PROCESSING: THE BIOTECHNOLOGICAL IMPACT OF A NOVEL PECTIN LYASE

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The food processing industry significantly utilizes pectinolytic enzymes. These enzymes are crucial for cleaving the  $\alpha$ -1,4 linkages between galacturonosyl residues, which breaks down the pectic structure. Through  $\beta$ -elimination processes, they form unsaturated C4–C5 bonds at the nonreducing end of polysaccharide fragments. Importantly, these reactions occur without the production of methanol, making them particularly advantageous for use in the beverage industry. This study dives into the unique pectin lyase, BvPelB, which is produced from *Bacillus velezensis* 16B and aims to improve fruit juice processing. BvPelB was successfully expressed in *Escherichia coli* and purified, exhibiting robust activity under alkaline conditions and thermal stability. Additionally, BvPelB's molar mass was determined, and its characterization and structure were predicted. The conditions for orange and apple juice treatment were optimized to identify the most effective parameters. Furthermore, BvPelB is beneficial for processing neutral juices that are naturally acidic but are neutralized to serve as a stable base for various applications. This neutralisation further improves the quality of the finished product by enabling the addition of proteins and other necessary nutrients without affecting the beverage's texture or capacity to hold its shape over time.

**Keywords:** fruit juice processing, optimization, pectin lyase, *Bacillus velezensis*, pectin

**Acknowledgements:** This research was supported by the Ministry of Science, Technological Development and Innovation of Republic of Serbia (Contract numbers: 451-03-66/2024-03/200026 and 451-03-65/2024-03/200168).

## FIBRE-RICH INGREDIENT OBTAINED FROM DEFATTED COLD-PRESSED RAPESEED CAKE AFTER ALKALINE PROTEIN EXTRACTION

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Rapeseed is an important oil crop in many countries, particularly due to the increasing demand for rapeseed/canola oil production. During the cold-pressed rapeseed oil production process, rapeseed cake is generated as a by-product, which can be utilized as feedstuff or as raw material for reutilization to prevent food loss. After defatting the cold-pressed rapeseed cake, a protein-rich rapeseed meal is produced, which can be used for protein isolate production by alkali extraction. Nutrient profiling and safety evaluation of the fiber-rich by-product obtained after protein alkali extraction are essential to define its potential for further application. Therefore, the aim of this work was to characterize this fiber-rich residue in terms of: 1) basic chemical profile, 2) mineral content, 3) amino acid profile, and 4) safety characteristics. The rapeseed cake used in this study was obtained from Suncokret d.o.o., Hajdukovo, Serbia. It was defatted and treated with 84% ethanol to minimize phenolic content, assisted by ultrasonic treatment (45 Hz at room temperature for 15 minutes). Alkaline extraction (pH 12, 60 min) was performed using an ultrasonic bath (45 Hz at room temperature for 30 minutes). Following extraction, the insoluble material, or fiber-rich residue, was analyzed. The basic chemical composition of the fiber-rich product includes proteins and carbohydrates. Total fiber accounts for 63.1%, comprising 2.73% soluble fiber and 60.3% insoluble fiber. Mineral analysis revealed the presence of potassium (13.5 g/kg), sodium (0.14 g/kg), calcium (3.22 g/kg), magnesium (6.55 g/kg), manganese (78.2 mg/kg), zinc (104 mg/kg), copper (8.76 mg/kg), iron (56.7 mg/kg), lead (0.22 mg/kg), and cadmium (< 0.003 mg/kg). The amino acid profile indicated a total amino acids content of 20.2 g/100 g, with lysine accounting for 1.22 g/100 g. Safety parameters of the fiber-rich product were also analysed, including antinutrients, mycotoxins, and pesticides. Among the antinutritional factors, glucosinolates were not detected. Tannins were present at 0.72 mg catechin/g, and polyphenols at 0.01 mg GAE/g, indicating that the ethanol washing step was successful. Phytic acid content was measured at 1.37 mg/100 g, suggesting the potential application of phytases at the beginning of the process. Mycotoxins and pesticides were not detected in the fiber-rich residue obtained after alkali extraction. Based on the investigated nutritional and safety parameters, it can be concluded that further research is necessary to explore its potential applications. It appears that this product can be utilized as a feedstuff.

**Keywords:** rapeseed cake, protein isolate, fiber-rich ingredient, characterization

**Acknowledgements:** This work was financially supported by the Science Fund of the Republic of Serbia, Grant No 6673, PROTein from Rapeseed Oil Processing Waste: Application in Food and Wastewater Treatment – PROTOPOWER, and the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## UTILIZATION OF BIGEL STRUCTURES AS FAT REPLACERS IN TURKISH FERMENTED SAUSAGE: EFFECTS ON QUALITY AND PROCESSING

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Turkish fermented sausage (sucuk) is a traditional meat product made from beef, beef fat, sodium chloride, spices, sodium nitrate, ascorbic acid, starter culture, and sucrose. It is stuffed into natural casings and then cured, fermented, and dried for a long shelf life. With growing consumer health consciousness, there is a need to reformulate these high-fat products without compromising quality. Bigel structures, combining oleogel and hydrogel, offer potential as fat replacers, but research on their feasibility in food products is still limited. In this study, 50:50 oleogel:hydrogel ratio bigel structures consisting of 5% carnauba wax and 0.5% beef gelatine were prepared to replace the beef fat content of sausages by 25 and 50%. All groups were cured for 4h at 20-25°C and 60% RH, followed by a fermentation process at 20-22 °C and 80-85% RH and finally dried at 18-20°C at 70-75% RH until pH 5.4 and stored under vacuum at +4°C until analysis. Sausage dough and final product were analysed for colour, pH, peroxide number, TBARS, moisture and fat content, water activity, cooking yield, process yield, water holding capacity, texture profile analysis and total titratable acidity. While replaced groups exhibited significantly lower fat content (35,9 and 33,6 for control and 50% (B50) replacement respectively), the replaced groups suffered in terms of cooking yield (83,2 and 77,1 for control and B50 respectively) and oxidative stability (Peroxide number: 37,9 and 47,3; TBARS: 0,893, and 1,05 for control and 50% (B50) respectively). Additionally, moisture content, process yield, and total titratable acidity analyses yielded similar results among all groups. This study suggests that bigels are an excellent alternative to other fat replacing solutions with minor adjustments.

**Keywords:** fermented sausage, bigel, fat substitution, gelled emulsion



## NUTRITIVE PROFILE EVALUATION OF THE COMBINEDLY DEHYDRATED CELERY

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Celery (*Apium graveolens* L.) is an aromatic vegetable, a member of the Apiaceae family. Celery is a globally cultivated vegetable. Celery root is rich in dietary fibre, mineral elements, vitamins and essential oils and, as such, has health benefits. Celery root has few calories, it can be consumed raw or treated. There are different drying methods used to preserve plant material, such as convective, lyophilisation and combined drying methods. Convective drying is characterized by a high temperature which influences the sensory and nutritional characteristics of the product. The lyophilisation method preserves the nutritional and sensory characteristics of products, but it involves increased process costs and longer drying times. The combined dehydration method consists of osmotic dehydration in molasses and lyophilisation as a successive dehydration phase. This study aims to investigate the effect of different drying methods on the chemical and mineral composition of the celery root. The significant differences between different methods of drying of celery root are confirmed by the application of post-hoc Tukey's HSD test at a 95% confidence limit. The calculation of Z-Score Analysis, based on chemical and mineral parameters, points out the best score of celery root dehydration by the combined method, obtaining 91.67%, while celery lyophilization and convective drying obtained only 15.67% and 7.60% of maximal score values, respectively.

**Keywords:** dehydration methods, chemical composition, mineral composition, celery root

## COMPARISON OF DARK AND MILK COCOA TOPPINGS PRODUCED BY FIVE ROLL MILL AND BALL MILL

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Chocolate toppings are confectionery products comprised of non-fat dark cocoa solids and sugar particles dispersed in cocoa butter, as a continual matrix. On the other hand, dark cocoa toppings contain cocoa powder and sugar dispersed in vegetable fat that is cheaper than cocoa butter, but with adequate characteristics. Also, with the addition of a 7% non-fat milk fraction, this product is regarded as a milk cocoa topping. This kind of product, dark and milk cocoa toppings, does not demand long-term conching or tempering. This study aimed to determine and compare the impact of the production process on rheology, particle size distribution and content of moisture, fats, sucrose and lactose. Based on the rheology properties, it was found that the values of the Casson and linear viscosity of the samples produced using a five-roll mill and conching were lower compared to those produced using a ball mill. Regarding particle size distribution, the results showed that the volume-weighted mean parameter D (4,3) was lower for the milk and dark cocoa confectionery topping produced in the ball mill. Additionally, the values of the yield stress in samples produced in the ball mill were 2-fold higher for the milk cocoa topping and 4.5-fold higher for the dark cocoa topping compared with the values of the samples produced using the five-roll mill and conching. The NIR-spectroscopy analysis did not show any significant difference among the samples. It can be concluded that the difference between the samples produced in the ball mill and the five-roll mill and conching exists and that the ball mill can be used for the production of high-quality cocoa toppings thus providing a shorter time of production.

**Keywords:** cocoa toppings, five roll mill, ball mill, rheology, particle size distribution

**Acknowledgements:** This research was supported by the Ministry of Science, Technological Development and Innovations, Serbia, program (451-03-47/2023- 01/200134).

## THE POWER OF SOUR CHERRY POMACE AS AN INGREDIENT: COLOUR, NUTRITIONAL PROFILE AND ANTIOXIDANT ACTIVITY AFFECTED BY DIFFERENT DRYING METHODS

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A viable solution in striking the balance between nutritionally enriched products, resource utilization, and sustainable raw material processing is certainly valorisation of nutrient-rich side-streams originating from the food industry. The juice industry side-streams owing to their polyphenols and dietary fibres content are taking the lead in the innovative ingredient development race. Accounting to a production of 164 446 t in Serbia for 2022, and with a prediction of 136 820 t in 2024, the sour cherry after juicing delivers 15–28% of waste consisting of skin, flesh, and seeds regarded as sour cherry pomace (SCP). Further handling of the corresponding residue in terms of drying and milling, impacts its appearance as well as its original nutritional value. Accordingly, the aim of this research was estimation of the SCP proximate composition, total phenolic content (TPC), antioxidant activity (DPPH and FRAP), and colour in relation to the applied drying method: lyophilisation (0.05 mbar, 72 h) (SCPL) and air drying by condensation (58°C, 32 h) (SCPC). The most visible difference between the samples was in their visual appearance. Namely, SCPC was characterized by lighter colour ( $L^*$  36.96) with less red ( $a^*$  16.78) and more yellow ( $b^*$  12.54) coloration compared to SCPL. Distinction among samples was evident for moisture and fat content. Additionally, minimal variation in acid insoluble ash, total sugars, and proteins was noticed between the samples implying minor influence of the drying type on proximate composition of SCP. Nevertheless, significant differences between the samples were observed concerning antioxidant activity primarily in terms of DPPH where for SCPL ( $IC_{50}$  117.38  $\mu\text{g/ml}$ ) higher radical scavenging potential was detected compared to SCPC ( $IC_{50}$  195.00  $\mu\text{g/ml}$ ). Additionally, increased values of TPC and FRAP were recorded in SCPL.

**Keywords:** sour cherry, side-stream valorization, proximate composition, bioactive compounds, antioxidant activity, colour

**Acknowledgements:** This research was supported by the Science Fund of the Republic of Serbia, #12928, Advanced formulation of sour cherry pomace-based tea cookies: Integrating nutritional enhancement and 3D printing - CheTea3D.

## OPTIMISATION OF ULTRASOUND-ASSISTED SOLID-LIQUID EXTRACTION OF BIOACTIVE COMPOUNDS FROM STINGING NETTLE LEAVES

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Stinging nettle (*Urtica dioica* L.) is a perennial plant that has been used by humans since ancient times for nutrition, treatment and the manufacture of textiles. This plant, with its rich chemical composition, has found its way into industry and is once again at the focus of scientific research due to the bioactive effect of its constituents. The aim of this study was to determine the influence of the parameters of ultrasound-assisted solid-liquid extraction on the content of total phenols and total flavonoids as well as chlorophyll and total carotenoids in the extract of dry nettle leaves and its antioxidant activity. The extraction was carried out in an ultrasonic bath under different conditions: extraction temperature (60–80 °C), extraction time (5–35 min) and the ethanol content in the water-ethanol solvent (0–100%). The Response Surface Method and the Box-Behnken design were used to optimise the process. It was found that the ethanol concentration in the solvent was the process parameter that had the greatest influence on the properties of the analysed extract. Furthermore, different optimal conditions for the extraction of pigments and for the extraction of phenolic compounds were determined. The extraction of chlorophylls and carotenoids was favoured by a lower temperature, a longer extraction time with a higher concentration of ethanol (40 °C, 35 min, 100% ethanol), while extraction at a higher temperature with a lower concentration of ethanol and at short time (80 °C, 5 min, 43% ethanol) resulted in an extract with the highest content of total phenolic substances and maximum antioxidant activity. It can be concluded that the studied extraction method is effective in isolating bioactive compounds from nettle leaves and the obtained extract has the potential of a functional supplement.

**Keywords:** stinging nettle, ultrasound-assisted extraction, antioxidant activity, pigments

## HYBRID DRYING: THE PLANT FOOD WASTE AND BY-PRODUCTS CASE STUDY

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The food industry in the EU member states annually produces about 100 million tonnes of food waste and by-products, and more than 35% of them are processed. Significant quantities of these by-products contain high-value ingredients that could be used for the production of new food products or that they are defined as final products (noodles or other forms) in drying processes or whether they are powdered and used in other products as functional ingredients (egg replacements, natural colorants, nutraceuticals). Current focus on research of fruit and vegetable drying methods as well as food waste and by-products is based on the application of traditional and commonly used convection drying in hot air flow and also conduction drying as well. Any improvements in such an energy and time-consuming process can have a significant economic impact. The application of new technologies in pre-treatments such as microwaves, ultrasound or high pressures, but also in the drying process itself, can also lead to better quality of the product. Hybrid drying along with innovative non-thermal raw material processing techniques are promising technologies that can produce semi-finished products and products with significantly improved nutritional properties, texture and color, while saving time and energy.

**Keywords:** food waste, by-products, hybrid drying, non-thermal technologies

**Acknowledgements:** The work was supported by the Croatian Science Foundation (research project "Hybrid drying and valorization of plant food waste and by-products" IP-2019-04-9750) - HYDRYBY.

## THE PHYSICOCHEMICAL DETERMINATION OF POMEGRANATE PEEL ENCAPSULATES AS A POTENTIAL FUNCTIONAL FOOD PRODUCTS

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Pomegranate (*Punica granatum* L., Punicaceae) is a widely known and used fruit. Besides the edible part of the pomegranate, the inedible part of the fruit is the peel, which represents 40% of the total weight and is usually considered a waste product. Pomegranate peel is rich in bioactive compounds, containing medically and nutritionally important phytochemicals. Almost 48 phenolic components, with numerous health benefits, have been detected in the peel. Implementing the encapsulation process, using biocompatible carriers represents a promising approach for preserving extracted pomegranate bioactive compounds. This study aims to extract the bioactive components from pomegranate peel by-products and convert them into innovative products, which could serve as functional food products. The ripe wild pomegranate fruits were harvested in November 2019 in Bosnia and Herzegovina, municipality of Stolac. Peel was extracted (80% ethanol in a ratio of 1:10), then encapsulated using the freeze-drying technique in the presence and absence of coating materials (maltodextrin, whey protein, and innovative carrier, polydextrose). The obtained powders were analyzed on physicochemical properties. The yields of encapsulates ranged from 88.35-92.3%. Comparing the FTIR spectra of wild pomegranate peel extract and encapsulates, no significant difference can be observed, from which it can be concluded that the drying (encapsulation) process did not affect the change in the composition of the extract. By analyzing the thermal stability of encapsulates, it was confirmed that carriers protected the encapsulates against temperature degradation. All encapsulates showed good thermal stability at temperatures often used during food processing. DSC test also showed that the peak degradation temperature reached the highest value for pomegranate peel extract embedded in a whey protein carrier at 179 °C. Also, exposure to temperatures of 140-216 °C did not affect the chemical composition of the extract, which is significant for the food industry in which food products are exposed to high-temperature temperatures during processing. The obtained results promised that waste by-product reuse would contribute to the circular food economy and reduce the environmental degradation associated with waste disposal.

**Keywords:** pomegranate, encapsulation, carriers, thermal degradation, FTIR

**Acknowledgments:** Ministry of Science, Technological Development, and Innovation (451-03-66/2024-03/200003).

## COMPREHENSIVE ANALYSIS OF DAMAGED STARCH FORMATION IN INDUSTRIAL WHEAT FLOUR MILLING

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Damaged starch, primarily caused by mechanical action from roller mills, plays a critical role in influencing the functional properties of flour. Therefore, monitoring and controlling its formation is essential for producing flours of desired quality. This research involved a comprehensive analysis of passage flours from two commercial mills, where granulation of the flour, flour yields, ash content, and damaged starch content were systematically examined. Detailed passage analysis revealed that neither the generation of damaged starch nor ash content was uniform across the milling process. Damaged starch content progressively increased from the initial to the final passages, particularly within the break, sizing, and reduction systems. Also, cumulative ash and damaged starch curves were formed, analysed and compared. In both mills, the cumulative ash curve had a standard shape and did not significantly increase in the area of low flour extraction. Curves for damaged starch were almost linear in the first half of the curves, with much more direct increase in starch damage compared to ash content. It was noted that similarity between these two types of curves lies in the particular regions, which represents flour streams from the end passages of the reduction system. This insight underscores the importance of careful monitoring and control during these critical phases to optimize damaged starch and ash content in the flour. The results of this study offer valuable guidance for optimizing milling operations and provide insight into how different milling passages affect wheat flour quality, helping to produce flours with specific characteristics.

**Keywords:** commercial milling, passage analysis, damaged starch content, ash content, flour yield

**Acknowledgements:** This work was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (451-03-66/2024–03/200134 and 451-03-65/2024-03/200134).

## THE EFFECT OF COLD PRESSING METHODS ON THE MINERALS DISTRIBUTION DURING BLACK MULBERRY JUICE PRODUCTION

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Black mulberry fruits are recognized for the well-being of human beings due to their high nutritional significance and potential health benefits. It is a good source of carbohydrates, lipids, proteins, vitamins, fibers and polyphenols (phenolic acids, flavanol derivatives, anthocyanins). Mulberry is also an excellent source of some important minerals, particularly calcium, phosphorus, potassium, magnesium, and sodium. Mulberry fruit is commonly consumed fresh but can also be consumed as shelf-stable processed products, such as juices, molasses and marmalades. Washing, mechanical milling into a pomace, mashing, cold pressing, pasteurization and filling-packing are the major steps in mulberry juice production. Some of them, such as pressing affect the distribution of nutrients, including minerals, into juice. In this study we investigated the effect of pressing on the distribution of macro- and microelements into mulberry juice and pomace. Black mulberries were pressed using laboratory hydraulic press, as well as using vertical cold press juicer. The content of macro elements (Na, K, Mg, Ca, S and P) of fresh fruit, juices and pomaces was determined by ICP-OES whereas the content of 13 micro and trace elements, including Fe, Zn, Cu, Mn, Mo, Co, Cr, Se, B, Al, Ni, Pb and Cd were determined by ICP-MS. Juice obtained by vertical cold press juicer had almost two times higher content of Ca, Mg and Zn, six times higher content of Fe compared to juice prepared using hydraulic press. The results clearly indicated that the way of pressing significantly determined the distributions of minerals.

**Keywords:** Mulberry juice, pomace, minerals

**Acknowledgements:** This research was funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia, University of Belgrade—Faculty of Agriculture (Agreements No. 451-03-47/2023-01/200134, 451-03-47/2023-01/200116, and 451-03-1202/2021-09).



## RHEOLOGICAL BEHAVIOR OF DOUGHS FROM ANCIENT WHEAT FLOURS: THE IMPACT OF SPONTANEOUSLY FERMENTED SOURDOUGHS

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Ancient wheat varieties have attracted significant interest due to their nutritional and functional attributes. They are often comparable to or even superior to modern wheat varieties, particularly in terms of minor components like proteins, dietary fiber, resistant starch, minerals, vitamins, and phenolic compounds. Typically used as whole grain, the higher fiber content of ancient wheat can adversely affect dough rheology, texture, and sensory qualities of the final products. To address these challenges, sourdough fermentation is recognized as an appropriate technological process for producing bakery products based on ancient wheat grains. The aim of this study was to evaluate the rheological properties during mixing and heating of dough consisting of wholewheat flour and sourdough, both originating from modern and ancient wheats (emmer, spelt and Khorasan). Two forms of sourdough (native and lyophilized; pH ranged from 4.22 to 4.55), obtained through spontaneous fermentation of whole grain flours from modern and ancient wheats, were used. In order to achieve appropriate flour substitution level, special attention was given to maintaining an equal proportion of dry matter from fermented dough (approximately 25%) derived from both types of sourdough samples. According to the obtained Mixolab profiles, the addition of sourdough significantly influenced dough behaviour in most parameters. In the initial phase of Mixolab mixing, native sourdough addition results in a more pronounced decrease in dough development time and stability, indicating even less stable and weaker dough. The C2 value, which is related to protein weakening, decreased with more pronounced lower values in the case of native sourdough addition. Upon heating, only the wheat control sample without sourdough addition had pronounced maximum starch paste consistency (C3), followed by a slight decrease in the torque for stability of hot starch paste (C4). However, in the rest of the tested samples there was a constant increase in measured torque, so the values of the C4 parameter were higher compared to the C3 values. During further heating, the presence of sourdough influenced the increase in the final baked dough consistency. The results indicate that sourdough significantly impacted dough behaviour. Native sourdough contributed to a softer and more adhesive texture, enhancing dough elasticity and reducing firmness. In contrast, samples with lyophilized sourdough exhibited characteristics more similar to the controls and were less challenging to process.

**Keywords:** ancient wheat, sourdough, rheology, Mixolab

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## OPTIMIZATION OF ULTRASONIC EXTRACTION FOR ENHANCED PURITY OF PHYTOCHEMICALS FROM *CITRUS AURANTIUM* L. FLOWERS

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*Citrus aurantium* L., commonly known as bitter orange, is a versatile medicinal plant well-known for its diverse therapeutic properties, including anti-inflammatory, antioxidant, and antimicrobial effects. These benefits are attributed to its rich phytochemical profile, especially its diverse array of secondary metabolites. Strong correlations between these compounds and the biological activities of *C. aurantium* extracts, highlighted the need for optimized extraction techniques to enhance their yield and purity for industrial applications. This study aimed to optimize the extraction of secondary metabolites (analyzed by HPLC-ESI- QqQ-MS/MS) from *C. aurantium* flowers using an ultrasonic-assisted extraction (UAE). For this purpose, response surface methodology (RSM) and a five-level circumscribed central composite design (28 experiments) was used, using the following variables as the critical parameters that affected extraction: *time* (*t*, 5-45 min), *amplitude* (*A*, 30-80%), and *ethanol concentration* (*S*, 0-100%). The HPLC-ESI- QqQ-MS/MS identified high amounts of phenolic acids such as caffeic acid, terpenoids like genipin, and flavonoids such as gardenin, that were further selected for individual optimization of the extraction conditions. A key finding was that most compounds exhibit maximum values at experimental runs. This trend corresponded to solvent concentrations of 100% ethanol, showing the affinity of the compounds. Under these circumstances, the extraction yield was reduced to 6-13% but the purity of the compounds was improved. This finding suggests that while higher ethanol concentrations may decrease the yield, they contribute to obtaining extracts with higher phytochemical purity. Such conditions could be advantageous for applications where the purity of bioactive compounds is more critical, such as in pharmaceutical or high-end cosmetic formulations. These insights underline the importance of fine-tuning extraction parameters to balance yield and purity according to the specific requirements of industrial applications. The use of UAE combined with RSM presents a robust method for maximizing the recovery of high-purity phytochemicals from *C. aurantium* L. flowers.

**Keywords:** *Citrus aurantium* L., ultrasonic technique, optimization, phenolic acids, flavonoids, high-performance liquid chromatography (HPLC)

**Acknowledgements:** The research leading to these results was supported by Xunta de Galicia for supporting the pre-doctoral grant of P. Barciela (ED481A-2024-230). The authors are grateful to the National funding by FCT, Foundation for Science and Technology, through the individual research grants of J. Echave (2023.04987.BD).

## THE IMPACT OF THERMAL PROCESSING ON MECHANICAL CHARACTERISTICS OF GLUTEN IN MODEL SYSTEMS

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During the heating phases of fermentation and baking, the gluten network undergoes significant alterations, leading to changes in its viscoelastic properties, which greatly influence the final bread loaf volume. Understanding gluten structural changes for each wheat variety during the initial stages of breadmaking, with mild heat treatment below 90 °C, is critical for regulating end product quality. Six commercial wheat flour samples from the U.S. hard red winter class, varying in protein content, were analyzed, along with one sample each of soft red winter, hard red spring, and durum wheat for reference. The viscoelastic properties of isolated gluten were measured at various temperatures (25, 35, 45, 55, and 65 °C) under a constant shear stress of 100 Pa. The experimental data were fitted to a Burgers model to understand the viscous flow and elastic components of gluten. The results revealed two major transitions in viscoelastic behavior at 45 °C and 65 °C. At 45 °C, there was an increase in creep compliance (flowability, with maximum strain reaching 36.8% from 25 to 45 °C in hard red winter wheat) and a decrease in recovery compliance (elasticity, with recoverability up to 10.8% from 25 to 45 °C in hard red winter wheat), indicating that gluten started to denature and become more deformable. At 65 °C, the trend in flowability reversed (maximum strain decreased to 22.3% from 45 to 65 °C in hard red winter wheat), suggesting that gluten aggregation predominated at this higher temperature. Principal Component Analysis (PCA) distinguished two groups of hard red winter wheat gluten samples based on their associations with strength/elasticity and deformation/flowability. This study demonstrates the efficacy of mathematical modeling in capturing and differentiating the viscoelastic properties of gluten, providing valuable insights for optimizing bread quality through controlled heat treatment.

**Keywords:** Gluten, Heat treatment, Processing

## INVESTIGATING THE INCORPORATION OF LOTUS ROOT STARCH IN THE DEVELOPMENT OF NUTRITIOUS SNACK

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This research aimed to study the process of producing starch from lotus roots and to develop a snack mixed with lotus root starch. Three drying methods (tray drying, drum drying, and freeze drying) were studied. The selected lotus root starch was then used to develop a snack. The result showed that tray drying affected the lotus root starch to have total phenolic compound and total flavonoid compound as 297.80 mg Trolox/100g sample and 79.45 mg Catechin/100g sample and antioxidant activity using DPPH, FRAP, and ABTS assays as 68.38, 92.54 and 120.97mg Trolox/100g sample, respectively. The study on the direction of product improvement using the Just-About- Right (JAR) method suggested that color value and overall aroma should be increased, whereas sweetness and crispiness should be decreased. Those results were further used to develop the optimized formulation using mixture design (D-optimal) using lotus root starch (20-34%), cake flour (10-17%) and sugar (10-17%). The result showed total phenolic compound and antioxidant activities (DPPH, FRAP, and ABTS) of snack mixed with lotus root starch were in the range of 111.54-186.69, 43.01-57.26, 89.78-261.56, 57.49-63.29 mg Trolox/100g sample respectively. The sensory preference rating was in the range of slightly like to moderately like (6.1-7.8). The physical, chemical, and sensory properties analyzed the relationship between independent variables (lotus root starch, cake flour, and sugar) and dependent variables (physical, chemical and sensory properties). The results showed that moisture content, hardness, total phenolic compound, overall aroma and overall liking can create regression equation and response surface which can be used to create overlay plot that predict the optimum amount of lotus root starch (29%), cake flour (10%), and sugar (15%) with the product desirability at 0.718.

**Keywords:** Drying, Lotus root starch, Snack

**Acknowledgements:** The authors wish to express their profound gratitude to the esteemed Faculty of Agro- Industry at Prince of Songkla University, Thailand. Their invaluable support through the provision of essential laboratory equipment played a pivotal role in facilitating the successful completion of this research endeavor, underscoring the significance of their contribution to advancing scientific knowledge in this field.

## VISUALISING PULSED ELECTRIC FIELD EFFECTS IN PLANT TISSUES USING THE REDOX INDICATOR TTC

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Electroporation or pulsed electric field treatment (PEF) involves applying short-duration electric pulses (nanoseconds to milliseconds), and of electric field amplitudes of 0.1 to 80 kV·cm<sup>-1</sup>. This external electric field, if of sufficient strength, can increase cell membrane permeability and biological tissue conductivity, which is commonly attributed to the formation of hydrophilic pores in the cell membrane. PEF technology is gaining relevance and popularity in the food industry due to its relative simplicity, compatibility with industrial parameters, and low energy requirements. It can be easily integrated with other food processing steps or used independently. A common use as (pre)treatment of plant tissues is to achieve either better mass transfer from/into tissue, to soften the tissue for facilitation of cutting, or for structural modification of the tissue for more efficient processing steps (e.g. peeling). One of the challenges in applying PEF to food products is the electrical inhomogeneity stemming from the biological diversity of treated products and their diverse morphology. In simpler terms, tissues of different origins respond differently to PEF, e.g. apple fruit responds differently than potato tubers, and differently than asparagus. Treatment parameters must thus be optimized for each product separately. During optimization, it helps quantifying how extensively we affected the tissue using PEF using reliable detection methods. Some of these methods either in use or under study are impedance measurements; magnetic resonance imaging; texture analysis; and dyeing cells with redox indicators such as triphenyl tetrazolium chloride, a metabolic activity indicator that colours living but not dead cells. Our study explores how inhomogeneities in tissue cause a supposedly homogeneous treatment using parallel plate electrodes to result in inhomogeneously treated tissue when field strength is insufficient. We highlight the importance of sufficient field strength coverage of entire tissue for the success of the treatment and achieving the intended effect.

**Keywords:** pulsed electric fields, electroporation, triphenyl tetrazolium chloride, plant tissue, electric field strength

**Acknowledgements:** The authors acknowledge the financial support through research programs and projects granted by the Slovenian Research Agency (ARIS), namely the research programme P2-0249, granted to and led by Damijan Miklavčič. This study was conducted within the Infrastructure Programme: Network of research infrastructure centres at the University of Ljubljana (MRIC UL IP-0510).

## KINETIC MODELING OF THE CARAWAY (*Carum carvi*) ESSENTIAL OILS ISOLATED UNDER DIFFERENT CONDITIONS

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*Carum carvi*, known as caraway, is a biennial plant which belongs to the *Apiaceae* family. Caraway seed represents the target part of the plant that is used as seasoning to flavor dishes, and for essential oil isolation. Caraway essential oil (CEO) reaches 3–7% usually composed of d-carvone and d-limonene among volatile monoterpenes. Due to its unique aroma, caraway's application is widespread in culinary, alternative medicine, cosmetic, beverage and pharmaceutical industries. The two main bioactive compounds (carvone and limonene) are directly associated with antispasmodic, carminative, antioxidant and antimicrobial properties. Hence, this research was focused on obtaining applicable high-quality CEO isolated by hydrodistillation (HD) and microwave-assisted hydrodistillation (MWHD). Moreover, distillation kinetics was determined to compare the quality of caraway HD and MWHD EO. CEOs from seeds were obtained by HD at 205 and 410 W and MWHD at 180, 360, 600 and 800 W. CEO yield was sequentially measured after 2.5, 5, 7.5, 10, 15, 30, 45, 60, 90 and 120 min and distillation kinetic was monitored. Four kinetic models were applied to closely explain the experimentally obtained results. Model I included simultaneous washing and diffusion, model II included washing followed by diffusion, model III included diffusion without washing and the second-order model was model IV. Based on the statistical parameters, the model with the best fit of the experimental results can be chosen. Isolated CEOs were characterized in terms of extraction yield. For CEO obtained by HD, yield ranged from 3.61% at 205 W to 3.78% at 410 W. MWHD CEO yield was lowest at lower power (2.81% at 180 W) and increased by increasing the microwave power (3.81% at 800 W). Through the kinetic model it is possible to optimize and better explain the phenomena of the distillation process. By choosing optimal conditions, highly valuable CEO may be obtained with certain application in many fields.

**Keywords:** caraway, essential oil, hydrodistillation, microwave-assisted hydrodistillation, kinetic models

**Acknowledgements:** This research was supported by the Science Fund of the Republic of Serbia, 7750168, Novel extracts and bioactive compounds from under-utilized resources for high-value applications—BioUtilize and by the Ministry of Science, Technological Development and Innovations of the Republic of Serbia, 451-03-65/2024-03/ 200134.

## EFFECT OF SHADING ON THE PHYTOCHEMICAL BURDEN OF *MORINGA OLEIFERA* LAM. PRODUCTION

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*Moringa oleifera* Lam. is a multipurpose plant, and its demand is increasing in close association with the derived health benefits of its phytochemicals, among which highlight glucosinolates (GSL) and phenolics. This study aims to explore the combination of soil amendments (application of combinations of poultry manure, biochar, and enzyme) and the effects of shade on the phytochemical content of moringa leaves. Moringa leaves were assessed on their profile of glucosinolates that allowed describing the presence of 4-( $\alpha$ -L-rhamnopyranosyloxy)benzyl-glucosinolate and 4'-O-acetyl-4-( $\alpha$ -L-rhamnopyranosyloxy)benzyl-glucosinolate, as well as the phenolics apigenin di-C-glucoside (vicenin-2) y quercetin acetyl-hexoside. The results obtained evidenced a high concentration of 4-( $\alpha$ -L-rhamnopyranosyloxy)benzyl-glucosinolate under no shading conditions (1516.77  $\mu$ g/g dry weight (dw)), while 4'-O-acetyl-4-( $\alpha$ -L-rhamnopyranosyloxy)benzyl-glucosinolate was found in concentrations lower than the limit of quantification (<LOQ). The analysis of moringa leaves of trees grown under shading conditions evidenced concentrations <LOQ for both organosulfur compounds. This evidenced the light incidence's relevance for the moringa tree's secondary metabolism and the synthesis of these bioactive compounds. Concerning phenolics, vicenin-2 was found in higher concentration in leaves protected from light (365.17  $\mu$ g/g dw). Still, the total phenolic concentration suggested that the highest burden for this phytochemical class corresponded to leaves of trees under no shading growing conditions. In conclusion, light incidence is key for phytochemicals' metabolism and concentration in moringa, thus contributing to an enhanced content.

**Keywords:** *Moringa oleifera* L., biomass yield, biochar, enzyme, poultry manure

**Acknowledgements:** The present work has been funded by the Spanish Ministry of Science and Technology through the CSIC Programme for Scientific Cooperation for Development iCOOP (iCOOP/COOPA/22040).



## EVALUATION OF A NOVEL DISPOSABLE AMPEROMETRIC GLYCEROL BIOSENSOR BASED ON A MELDOLAS BLUE-MODIFIED SCREEN-PRINTED CARBON ELECTRODE FOR JUICE BEVERAGE ANALYSIS

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This study details the design and development of an electrochemical biosensor for measuring glycerol, an important compound in food and beverages, due to its role as a naturally occurring triose sugar and food additive (E 422). Monitoring glycerol concentration is essential for ensuring food quality and safety. The biosensor operates by enzymatically oxidising glycerol using glycerol dehydrogenase (GLDH) in the presence of NAD<sup>+</sup>, resulting in the production of NADH and dihydroxyacetone. The NADH then undergoes electrocatalytic oxidation at a Meldolas Blue-modified Screen-printed Carbon Electrode (MB-SPCE), with the resulting current serving as the analytical response. This response is directly proportional to the glycerol concentration. The biosensor's operation was optimised through initial studies that determined the optimal conditions. The hydrodynamic voltammetric studies were performed using NADH at pH 9 with MB-SPCEs. The electrocatalytic current ranging from -0.30 to +0.20 V was investigated, and the voltammograms indicated a maximum electrocatalytic current at 0 V vs Ag/AgCl, which was used for subsequent studies. Standard addition calibration studies using chronoamperometry were performed with glycerol concentrations between 1.0 to 3.0 mM, utilising only 100 µL of diluted (0.1 M phosphate buffer) grape juice (GJ) directly on the biosensor surface. Calibration plots were constructed by taking current measurements at 100 s after application of the applied potential; this demonstrated that the glycerol biosensor produced a linear response across the concentration range studied. The amperometric biosensor was successfully applied to the measurement of glycerol in commercially available GJ, representing non-alcoholic beverages. These analyses have the ability to be conducted outside the laboratory using commercially available, portable potentiostats. Overall, this approach shows promise to form a platform for the development of novel rapid technology for point-of-test evaluation of glycerol in the production and quality control of non-alcoholic beverages.

**Keywords:** Food safety and quality, amperometric glycerol biosensor, chronoamperometry, screen-printed carbon electrode, Meldolas Blue, grape juice



## MORPHOLOGY AND PHYSICAL PROPERTIES OF BROCCOLI MICROGREEN JUICE ENCAPSULATED WITHIN MALTODEXTRIN AND INULIN

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Encapsulation is a process that is used in a variety of industries, including the food industry. There are many reasons for using encapsulation in the food industry, such as to protect an active compound from unfavorable environmental conditions, mask undesirable sensory properties, improve the stability of the encapsulated compound, facilitate handling, and improve physicochemical properties. Broccoli microgreen juice requires an encapsulation process because it contains various bioactive compounds, including phenolic compounds and sulfur compounds, which act as antioxidants. The aim of this study was to determine the morphology and physical properties of spray-dried, non-encapsulated broccoli microgreen juice (control powder) and encapsulated broccoli microgreen juice within maltodextrin (BMD) and inulin (BIN). The bulk and tapped density, moisture content, and color of the obtained powders were determined using the standard methods, while the morphological properties were determined using scanning electron microscopy (SEM). The control powder had a higher moisture content and a lower powder yield compared to the encapsulated powders. On the other hand, the obtained encapsulated powders had a higher bulk density than the control powder without any carrier, indicating better physical properties. In contrast to the BIN powder, the BMD powder exhibited higher values for brightness ( $L^*$ ). In terms of morphological properties, the control powder exhibited irregular particles in the form of agglomerates, indicating the stickiness of the powder. Unlike the control powder, all obtained encapsulated powders with carriers (BMD and BIN) had no agglomerates, resulting in lower stickiness. In addition, the BMD powder had spherical particles with a concave and rough surface, while the BIN powder had a smaller particle size with an irregular shape and wrinkled surface. In summary, the obtained encapsulated powders have good morphological and physical properties and can be further investigated for application in the food industry.

**Keywords:** encapsulation, broccoli microgreen juice, scanning electron microscopy, bulk density, tapped density

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant number 451-03-65/2024-03/200116).

## CHARACTERISTICS OF TRADITIONAL ROLLED PASTA FILATA CHEESE OF VOJVODINA

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Rolled cheeses are traditionally produced in the area of Vojvodina and represent *pasta filata* cheeses, which are mostly made from raw cow's milk, raw sheep's milk, or a combination of these two types of milk. The process of producing rolled cheeses is specific; it requires certain knowledge and skills, which are passed down from generation to generation. The aims of this research were to determine the microbiological (*Enterobacteriaceae*, *Escherichia coli*, coagulase-positive staphylococci, *Campylobacter* spp., *Salmonella* spp., and *Listeria monocytogenes*), physico-chemical (moisture, dry matter, milk fat, fat in dry matter, moisture on a fat-free basis, protein, ash, carbohydrates, salt, energy value, pH value, and water activity), and sensory characteristics of rolled *pasta filata* cheese traditionally produced from raw cow's milk. Pathogenic bacteria, such as *Listeria monocytogenes* and *Salmonella* spp., were not detected in the rolled cheese, while the microorganisms used as an indicator of hygiene during the manufacture of the cheeses, coagulase-positive staphylococci ( $1.40 \pm 0.61 \log_{10}$  CFU/g) and *Escherichia coli* ( $3.12 \pm 0.47 \log_{10}$  CFU/g), showed good hygienic quality. This rolled *pasta filata* cheese belonged to the full-fat ( $47.11 \pm 0.88\%$  milk fat in dry matter) semi-hard ( $62.06 \pm 0.64\%$  moisture on a fat-free basis) cheeses. Rolled cheese had a regular shape and dimension, without deformations or damage, with a clearly defined, compact leafy structure on the cross-section. The colour of the rolled cheese was white with yellowish shades. The consistency of the rolled cheese was semi-hard. Rolled cheese had a slight milky-sour smell. The basic taste of rolled cheese was pleasant, milky, not too salty and sour. To our knowledge, this is the first report of the microbiological, physico-chemical, and sensory characteristics of traditionally made rolled *pasta filata* cheese.

**Keywords:** *pasta filata* cheese, rolled cheese, microbiological status, physico-chemical properties, sensory characteristics

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia by the contract of implementation and funding of research work of NIV-NS in 2024, Contract No: 451-03-66/2024-03/200031.

## APPLICATION OF OENOLOGICAL TANNINS IN WINE TECHNOLOGY

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Optimal conditions for viticulture and production of quality wines are given when the climate factors such as solar radiation, temperature, rainfall and wind are in harmony. The usage of fossil fuels leads to an increase in CO<sub>2</sub> and other gases that are responsible for the greenhouse effect and global warming. Concentration of CO<sub>2</sub> at the beginning of the 20th century was 260 ppm, today it is 380 ppm and is expected to reach 450-1000 ppm by the end of the 21st century. Increased temperatures will shorten the ripening period of the grapes by 10 to 15 days, during which time not all parts of the berry will reach full ripeness. As a result, grapes are harvested before fully ripe skins and seeds. During production of red wines degree of ripeness of skin is very important, if not decisive on the quality of wine. Seeds are often only ripe at harvest. Climate changes are leading to an increasing imbalance between technological (industrial) and phenolic maturity. This imbalance means that astringency develops differently in the different parts of the berry, which may explain why wines from unripe grapes contain many astringent tannins from the seeds and relatively few tannins from the berry skin. In both cases, these are condensed tannins, known as procyanindins, which are polymers of monomeric subunits: (+)-catechin, (-)-epicatechin, (-)-epigallocatechin and (-)-epicatechin gallate. Modern oenology recommends the addition of both condensed and hydrolysable tannins during alcoholic fermentation or later during wine aging and maturation. The aim of this work was to investigate chemical composition and sensory properties of Cabernet Sauvignon wines in which were added different commercial tannins. The results showed that the treated wines had a higher content of phenolic compounds, a better colour stability and a better structure and texture of the wine.

**Keywords:** oenological tannins, red wine, global warming

**Acknowledgements:** This research was funded by grants from the Ministry of Science, Technological Development and Innovation of the Republic of Serbia and the University of Belgrade-Faculty of Agriculture (No. 451-03-65/2024-03/200116) and grant between the Ministry of Science, Technological Development and Innovation of the Republic of Serbia and the University of Belgrade-Faculty of Pharmacy (No. 451-03-66/2024-03/200161).

## EFFECTS OF FOOD PRE-PROCESSING METHODS ON THE STRUCTURE OF BETA-LACTOGLOBULIN AND ITS BINDING MECHANISM TO COMMON POLYPHENOLS

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Plant-derived polyphenols are known for their diverse bioactive properties, including antioxidant, anti-inflammatory, and metabolic disease-modulating effects. However, their limited hydrophilicity often necessitates advanced delivery systems to improve bioavailability.  $\beta$ -Lactoglobulin (BLG), a protein with a hydrophobic core, has been identified as an effective carrier for polyphenols, but the interaction between BLG and polyphenols is highly dependent on the protein’s structural integrity. The influence of heat and high pressure processing (HPP) on the structure of BLG and its binding affinity with polyphenols remains an area of ongoing research. In this study, circular dichroism spectroscopy and molecular dynamics simulations were employed to examine BLG’s structural changes under thermal and high-pressure conditions, along with particle size analysis to assess aggregation behavior. A range of polyphenols—quercetin, hesperetin, dihydromyricetin, gallic acid, (-)-epicatechin, resveratrol, and secoisolariciresinol diglucoside—were evaluated for their binding interactions with BLG. Fluorescence spectroscopy and molecular docking were used to explore binding mechanisms. Results showed that heating and HPP altered BLG’s structure, leading to changes in aggregation. Preheating at 358 K enhanced binding interactions between BLG and dihydromyricetin, while under HPP, optimal binding occurred at 400 MPa for most polyphenols, though secoisolariciresinol diglucoside and (-)-epicatechin exhibited maximum affinity at atmospheric pressure and 600 MPa, respectively. These findings highlight that both heat and high pressure modify BLG’s binding sites and interaction dynamics with polyphenols, with van der Waals forces playing a significant role in the binding free energy.

**Keywords:**  $\beta$ -Lactoglobulin, polyphenols, heating, high pressure processing, molecular simulation

**Acknowledgements:** This work was supported by the National Natural Science Foundation of China under Grant 31972070 and Key Research & Development Project of Shandong Province (2021TZXD007).

## SPRAY DRYING PRESERVATION: ENHANCING STABILITY OF BLACK ELDERBERRY POMACE POLYPHENOLS-RICH EXTRACTS

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Recycling waste from elderberry juice production is an important practice due to the high content of valuable compounds such as anthocyanins, flavonols, and phenolic acids, which are known for their health benefits and distinctive taste and color. This approach not only helps to reduce environmental impact by minimizing food waste but also creates economic value by transforming waste into high-quality products. Following an optimized extraction of elderberry pomace using ultrasound-assisted technology, a spray drying process was carried out. In this process, carriers, including maltodextrin (MD) and gum arabic (GA), were used in different ratios relative to the dry mass of the extract (100% MD; 80% MD and 20% GA; 60% MD and 40% GA; 40% MD and 60% GA; 20% MD and 80% GA; 100% GA), while the other process parameters were kept constant. The resulting powders were analyzed both physically and chemically. Among the different formulations, the powder produced with 100% maltodextrin showed the highest encapsulation efficiency. This formulation successfully retained 93.54% of cyanidin 3-O-glucoside (14.20 mg/g powder) and 91.60% of cyanidin 3-O-sambubioside (17.55 mg/g powder), which are the predominant compounds in elderberry fruit. Scanning electron microscopy showed that the spray-dried powders consisted of spherical particles on a microscopic scale. Microencapsulation significantly improved the stability of the extract and protected the polyphenolic compounds from degradation. The encapsulated elderberry pomace extract has the potential to serve as a natural flavor and color enhancer for finished products while contributing to a high polyphenol content. This innovation not only maximizes the utilization of elderberry waste but also increases the value of the product.

**Keywords:** elderberry fruit pomace, spray drying, polyphenols, anthocyanins, SEM

**Acknowledgments:** Ministry of Science, Technological Development, and Innovation (451-03-66/2024-03/200003).

## OPTIMIZATION OF THE ESSENTIAL OIL HYDRODISTILLATION FROM LAVENDER FLOWERS PRETREATED WITH TRIETHANOLAMINE: LACTIC ACID DEEP EUTECTIC SOLVENT

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The growing demand for high-quality lavender essential oil has intensified research efforts to improve its recovery techniques. Optimizing the distillation process is crucial for maximizing essential oil yield. In the present paper the essential oil hydrodistillation from lavender flowers pretreated with the aqueous solution of triethanolamine:lactic acid deep eutectic solvent (TEOA:LA DES) is studied. The objective is to optimize the pretreatment conditions to achieve the maximum essential oil yield. The influence of three pretreatment factors (time, aqueous solution-to-plant material ratio, and TEOA:LA DES amount in the aqueous solution), on the essential oil yield was evaluated using a Box-Behnken design. The essential oil yield and pretreatment process factors were correlated using a quadratic model. Analysis of variance revealed that pretreatment time had the most significant impact on the essential oil yield, followed by the aqueous solution-to-plant material ratio, and TEOA:LA DES amount in the aqueous solution. The results further indicated that extending the pretreatment time and increasing the TEOA:LA DES amount positively affected the essential oil yield, while increasing the aqueous solution-to-plant material ratio had a negative effect on the oil yield. The reliability of the applied model in predicting the essential oil yield was confirmed by a very low mean relative deviation. The maximum essential oil yield of 2.25 mL/100 g of dry plant material was achieved under the following optimal conditions: a pretreatment time of 64 h, an aqueous solution-to-plant material ratio of 14:1 mL/g, and a TEOA:LA DES amount of 17%. Using TEOA:LA DES in the pretreatment of plant material can contribute to reducing the amount of water used for essential oil hydrodistillation.

**Keywords:** deep eutectic solvent, hydrodistillation, lavender essential oil, optimization

**Acknowledgements:** This work was supported by the Republic of Serbia – Ministry of Science, Technological Development and Innovation, Programs for Financing Scientific Research Work, No. 451-03-65/2024-03/200133 (Project assigned to the Faculty of Technology, Leskovac, University of Niš).

## THE INFLUENCE OF TRIETHANOLAMINE-BASED DEEP EUTECTIC SOLVENT ON THE YIELD AND CHEMICAL COMPOSITION OF LAVENDER (*Lavandula angustifolia*) ESSENTIAL OIL

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Lavender essential oil is recognized for its numerous health benefits and its widespread application in herbal medicine, cosmetics, and the food industry. This study investigates the recovery of essential oil from lavender (*Lavandula angustifolia*) flowers pretreated with triethanolamine-based deep eutectic solvents (DESs). Ground lavender flowers were pretreated with aqueous solutions of two DESs (20 vol%), triethanolamine:lactic acid (TEOA:LA, molar ratio 1:1) and triethanolamine:oxalic acid (TEOA:OA, molar ratio 1:1) for 48 h at room temperature, and at a solvent-to-plant material ratio of 12:1 ml/g. After pretreatment, the essential oil was recovered by Clevenger distillation for 30 minutes, and the process efficiency was compared with hydrodistillation from untreated flowers at the same water-to-plant material ratio (10:1). The yields of essential oil from lavender flowers pretreated with TEOA:LA and TEOA:OA DESs aqueous solutions were 2.14 ml/100 g and 2.03 ml/100 g of dry plant material, respectively, representing an increase of 12.6% and 6.8% compared to the oil yield from untreated flowers (1.9 ml/100 g of dry plant material). The chemical composition of the oils, analyzed by GC/MS, showed that all oils contained 1,8-cineole, linalool, camphor, and linalool acetate. The oils obtained from TEOA:LA and TEOA:OA DESs treated plant material had linalool as the most abundant component, whereas camphor was the most abundant in the oil from untreated plant material. Using TEOA:LA DES in pretreatment almost doubled the linalool acetate content in the essential oil compared to oil from untreated plant material. The application of DESs in the pretreatment of plant material can enhance the efficiency of essential oil recovery and alter the chemical composition of the obtained oils.

**Keywords:** deep eutectic solvents, essential oil, hydrodistillation, lavender, triethanolamine

**Acknowledgement:** This work was supported by the Republic of Serbia - Ministry of Science, Technological Development and Innovations of Serbia, Programs for Financing Scientific Research Work, No. 451-03-65/2024-03/200133 (Project assigned to the Faculty of Technology, Leskovac, University of Niš).



## OPTIMIZATION OF THE PROPERTIES OF CHEESE PRODUCTS WITH TECHNOLOGICALLY ADVANCED AND INNOVATIVE DRYING METHODS

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Cheese drying is a critical step in the production process and has a significant impact on the texture, flavor, color and shelf-life of the final product. Freeze-drying and microwave drying are two innovative methods to dry cheese while maintaining its sensory and textural properties. This research investigates the optimization of cheese drying by comparing two methods: freeze-drying and microwave drying. In this study, three different treatments of cheese (Gouda, Cheddar, and Vegan) were subjected to both freeze-drying and microwave drying processes. Water activity ( $a_w$ ) and humidity levels were measured to assess the stability and shelf-life potential of the dried cheeses. Texture analysis was performed to measure hardness, fracturability, and gel strength. Additionally, color parameters were evaluated using a colorimeter to determine the effects of each drying method on the visual appearance of the cheese. Sensory analysis was conducted by a trained panel to assess attributes such as flavor, aroma, and overall acceptability. The results indicated significant differences between the two drying methods. Microwave drying resulted in a substantially reduced drying time compared to freeze-drying. Water activity and humidity measurements showed that both methods effectively reduced moisture content, but freeze-dried samples had slightly lower  $a_w$ , indicating better long-term stability. Texture profile analysis showed that freeze-dried cheese had higher hardness than microwave-dried cheese. Color parameter evaluation showed minimal differences, though microwave drying resulted in slightly darker (lower  $L^*$ ) cheese. The sensory analysis indicated that both drying methods produced cheese with acceptable taste, flavor, and aroma. In conclusion, while both freeze-drying and microwave drying methods are effective for drying cheese, each has unique advantages. Freeze-drying offers superior sensory qualities and slightly better moisture control, whereas microwave drying provides faster processing times. Despite minor differences in texture and color, the overall quality remains high, making both drying processes viable options for cheese drying.

**Keywords:** cheese drying, freeze-drying, microwave drying, sensory evaluation

**Acknowledgements:** This project was co-funded by Greece and European Union (Project title: Optimizing the properties of dairy products using technologically advanced and innovative drying methods, M16SYN2-00079).



*Feed Technology, Quality and Safety*

## CLIMATE CHANGE AND IRRIGATION STRATEGIES: EFFECTS ON FUNGAL PATHOGENS IN MAIZE CULTIVATION

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Maize is a primary crop for agricultural production and export in the Republic of Serbia. However, despite the region's suitability for maize cultivation, production is often hindered by the presence of mycotoxigenic fungi, genera *Fusarium*, *Aspergillus*, and *Penicillium* are the most common. The occurrence of pathogens leads to reduced yield, quality of seed, and can also cause indirect damage in livestock production. After pathogen infection, maize kernels can become contaminated with mycotoxins. These compounds are harmful to humans and animals. In recent years, there has been particular attention to the presence of mycotoxins in food. Climate change and conditions that favor the emergence of pathogens require the implementation of various measures to create more favorable conditions for plant growth. Consequently, by enhancing plant resistance under adverse conditions, the pressure from pathogens can be reduced. The aim of this experiment was to determine whether the application of irrigation affects the reduction of ear rot. The study is conducted at two locations: Ravno Selo and Kovilj during the year 2024. Field disease assessment for ear rot involved the evaluation of a total of 60 plants in each treatment at the harvest phenological stage. Within each treatment replication, ears from inner rows were visually inspected using the seven-class disease severity scale established by Reid et al. (1996): 1 - 0%, 2 – 1-3%, 3 - 4-10%, 4 – 11-25%, 5 – 26-50%, 6 – 51-75%, and 7 – 76-100% of kernels exhibiting rot symptoms. The Disease severity index (DSI) in the field was determined using the Townsend-Heuberger formula. Characteristic symptoms of ear rot were observed, and disease rating was conducted. At the Ravno Selo locality recorded DSI was 8.15% for the irrigated plot and 16.67% for the non-irrigated plot. At the Kovilj locality, DSI was 8.89% for the irrigated plot and 15.19% for the non-irrigated plot.

**Keywords:** maize, irrigation, mycotoxigenic fungi, mycotoxins

**Acknowledgements:** The authors express their appreciation to the "ANTARES" project, that received funding from the European Union's Horizon 2020 research and innovation program under grant agreement SGA-CSA. No.739570 under FPA No. 664387 (<https://doi.org/10.3030/739570>).

## SUSTAINABLE EXTRACTION AND CHARACTERIZATION OF BIOACTIVE COMPOUNDS FROM *HELICHRYSUM ITALICUM* USING OPTIMIZED ACCELERATED SOLVENT EXTRACTION

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The growing awareness of the link between diet and health has fuelled demand for products that offer more than basic nutrition, driving interest in plant-based compounds. Consequently, the incorporation of herbal extracts and bioactive phytochemicals into food, nutraceuticals, and pharmaceuticals has gained significant attention. *Helichrysum italicum*, known as the "Immortelle" plant, is particularly valued for its rich phytochemical profile, including polyphenols, flavonoids, and terpenoids, which are linked to various health benefits. Capitalizing on the potential of this valuable plant, the present study employed Accelerated Solvent Extraction (ASE) to optimize the extraction of bioactive compounds from *H. italicum*. The research systematically explored the effects of key extraction parameters, including ethanol concentration as a solvent and extraction temperature, and time, to achieve the highest total polyphenol (TP) content, total flavonoid (TF) content, and antioxidant potential of the obtained extracts. The ASE process utilized an ethanol-water mixture as the extraction solvent, with the ethanol concentration varied from 60% to 90%, the extraction temperature set between 120°C and 180°C, and the extraction time tested in the range of 10 to 30 minutes. The results revealed that the optimal extraction conditions for maximizing TP, TF, and antioxidant activity were an ethanol concentration of 60%, an extraction temperature of 123°C, and an extraction time of 21 minutes. Under these optimized conditions, the *H. italicum* extract exhibited a high TP content, a substantial TF content, and a potent DPPH radical scavenging activity, indicative of a rich antioxidant potential. Obtained results highlight the effectiveness of ASE in selectively extracting bioactive compounds from *H. italicum*, with the optimized parameters leading to the highest recovery of polyphenols, flavonoids, and other antioxidants. The results offer valuable insights into the potential of *H. italicum* as a source of natural antioxidants and bioactive compounds for food, pharmaceutical, and cosmetic applications, while the optimized ASE protocol provides a foundation for sustainable extraction and further research, potentially unlocking new avenues for its commercial and therapeutic development.

**Keywords:** *Helichrysum italicum*, Accelerated Solvent Extraction (ASE), bioactive compounds, antioxidant activity, sustainable extraction

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222) and through the bilateral collaboration between Serbia and Slovenia for the period 2023-2025.

## **NUTRITIONAL CHARACTERIZATION OF GOAT MILK AFTER SUPPLEMENTING THEIR DIETS WITH MILK THISTLE MEAL IN THE PRESENCE OF OXIDIZED FAT**

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Our study evaluated the impact of incorporating oxidized flaxseed oil and milk thistle meal (MTM), into goat diets on milk nutritional quality. Thirty goats were assigned to three groups: a control group fed 7% fresh flaxseed oil (C), a group fed 7% oxidized flaxseed oil (E1), and another group fed 7% oxidized flaxseed oil and 10% MTM (E2). The proximate chemical composition was not significantly influenced by the experimental diets, with the exception of urea content, which decreased in the case of E2 (23.20 mg/100g), compared with C (30.9 mg/100 g) and E1 (28.8 mg/100 g) groups. The trace mineral profile of the milk was significantly affected by the experimental diets. In the E1 group, a notable reduction in iron (Fe) concentration was observed compared to the C group, (13.2 mg/kg, vs. 14.06 mg/kg). However, the inclusion of MTM in the E2 group mitigated the negative impact of oxidized oil, resulting in an increased Fe content compared to E1 (14.93 mg/kg),  $p = 0.025$ . A similar pattern was observed for milk phosphorus (P) concentrations, which were reduced in the E1 group compared to the C (1.08%, vs. 1.06%), but the inclusion of MTM in the E2 group counteracted these effects by increasing its content (1.08%),  $p = 0.031$ . Regarding vitamin E isomers, the inclusion of MTM significantly increased the content of gamma-tocopherol ( $p = 0.005$ ) compared to both the C and E1 groups. Additionally, the MTM balanced the negative effects of oxidized flaxseed oil on alpha-tocopherol and the total vitamin E content, which had been reduced by the E1 diet. In the E2 group, both alpha-tocopherol and total vitamin E levels increased compared with E1, but with no significant differences compared to the C. Also, milk cholesterol levels increased in E1 (0.02 0g/100 g raw milk), but E3 reversed this pattern by reducing its concentration to 0.015 g/100 g raw milk.

**Keywords:** flaxseed oil, milk thistle, goat milk, milk vitamin E, milk cholesterol

**Acknowledgements:** This research was funded by the Romanian Ministry of Research, Innovation and Digitalization, Project PN 2320 0301.

## INSECT-BASED FEED BLENDS: EFFECTS OF EXTRUSION ON SELECTED TECHNOLOGICAL AND NUTRITIONAL FEATURES

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The aims of this study were to examine the effect of extrusion process on *Hermetia illucens* (BSFL), *Tenebrio molitor* (YMWL) and *Zophobas morio* (SWL) blends with or without soybean meal (SBM) addition and to evaluate the impact of extrusion on the *in vitro* organic matter digestibility (IVD-DM) and *in vitro* crude protein digestibility (IVD-CP) in insect-based blends. Two blends, for each insect, have been formulated as follows: (i) 50% of corn meal, 25% of insect, 25% of dried carrots meal; (ii) 25% of corn meal; 25% of SBM, 25% of insects, 25% of dried carrots meal. The nutritional content of blends prepared for extrusion with and without SBM were 80 - 81% and 79 - 81% for dry matter; 4.7 - 6% and 3.2 - 4.2% for ash; while 20.8 - 22% and 9 - 11.6% for crude protein, respectively. All these results had no significant difference between the insect's species. All insect-based blends were characterized by a “good” extrudability and torque value remained always under 2 Nm in all samples. Of note, torque value results seem to be higher when SBM was added to the blend, although this difference was not significant. In this study, at 100°C the highest water removal was in the sample YMWL-based and without SBM, while the lowest was obtained in the sample BSFL-based and with SBM, because as reported in other study, inclusion of plant protein, seems to increased the water-holding capacity. IVD-DM and IVD-CP digestibility had no significant difference before and after the extrusion process and also considering the addition of SBM. In conclusion, it's possible to use extrusion technology with different insect's species. The addition of SBM doesn't have any effects on IVD-DM and IVD-CP, but seems to affect torque value and water removal, but further investigations are needed.

**Keywords:** feed technology, extrusion, insects, animal feed

**Acknowledgements:** This study was carried out within the Agritech National Research Center and received funding from the European Union Next-GenerationEU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.4 – D.D. 1032 17/06/2022, CN00000022). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

## MYCOTOXIN RISKS UNDER A CLIMATE CHANGE SCENARIO IN EUROPE: CHALLENGES AND PERSPECTIVES

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As determined by the Intergovernmental Panel on Climate Change, warming of the climate system is unequivocal and has been associated with rising sea levels, diminished amounts of ice and snow and increasing oceanic and atmospheric temperatures. Such climate changes have a significant impact on stages and rates of toxigenic fungi development and can modify host-resistance and host-pathogen interactions, influencing deeply also the conditions for mycotoxin production that vary for each individual pathogen. Moreover, the new combinations mycotoxins/host plants/geographical areas are arising to the attention of the scientific community and require new diagnostic tools and deeper knowledge of both biology and genetics of toxigenic fungi. In this presentation, it is underlined that an extension of the aflatoxin contamination risk in maize in South and Central-Europe is currently higher than predicted by models developed in the recent past, due to favourable climatic conditions to the growth of *Aspergillus flavus*. Moreover, the mycotoxigenic *Fusarium* species profile on wheat in Europe is in continuous change in Northern, Central and Southern-Europe with, in particular, a worrisome growing contamination of *F. graminearum* in the Central and Northern Europe.

**Keywords:** Food safety, *Aspergillus flavus*, Aflatoxin B1, *Fusarium graminearum*, Deoxynivalenol

## THE EFFECTS OF DIETARY ALLICIN (GARLIC EXTRACT AND GARLIC LEAVES) AND OXIDIZED OIL ON PRODUCTIVE PERFORMANCES AND BLOOD PARAMETERS OF BROILERS

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The objective of this study was to evaluate the effects of dietary oxidized oil and allicin (two different dietary sources) as natural antioxidants on productive and blood parameters of broilers. A total of one-day-old 200 Ross 308 broilers were randomly divided into 4 dietary groups (50 animals/group). The experimental groups differ from the control one in terms of the presence of oxidized oil in their structure (peroxide value 5.2 (E groups) vs. 1.7 (C group) meq active oxygen/kg). The experimental groups differ from each other by the presence of allicin (100 mg/kg inclusion rate as extract and 0.5% as garlic leaf powder). At the end of the experiment, six animals per group were sacrificed, and blood samples were collected. The final body weight, feed intake, and weight gain, were not influenced by the dietary treatments nor the presence of oxidized oil in feed. The feed-to-gain ratio was positively influenced by the presence of allicin extract (1.517 g/g vs. 1.655 g/g for the control group). Regarding the health parameters determined in blood, the cholesterol level was significantly decreased for both allicin supplemented groups compared to others. LDH level in blood was also decreased for experimental groups (allicin groups) compared with the group that received oxidized oil without allicin as an antioxidant, proving the beneficial effect of dietary supplements on counteracting the effects of stress induced by the presence of oxidized oil in broiler diets.

**Keywords:** poultry, allicin, performance, blood parameters

**Acknowledgments:** This research was funded by the Romanian Ministry of Research, Innovation and Digitalization, Project PN 23.20.03.01.

## EDIBLE INSECTS: A KEY COMPONENT OF THE FOOD AND FEED REVOLUTION

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The food and feed industries are dealing with numerous challenges, including climate change, population growth, and limited arable land. As we enter a food/feed revolution, there is an ongoing quest for new and sustainable sources of nutrition. Insects have emerged as a valuable protein source in some parts of the world. However, in western countries, the processing and commercialization of food and feed products derived from insects are significantly limited due to low consumer acceptance of these products. This research aimed to investigate the nutritional composition of different insect meals and the techno-functional characteristics of their protein isolates. In this study, three insect species (*Tenebrio molitor*, *Acheta domestica*, and *Zophobas morio*) were reared under various conditions to produce high-quality insect meals for feed. The second phase of the research involved isolating proteins through alkali extraction to create high-value protein ingredients for food applications. The findings revealed that the optimal nutritional composition of insect meals was achieved using a diet of cabbage, carrot, and flaxseed. The inclusion of a small amount of flaxseed increased the levels of omega-3 fatty acids in all insect oil samples. Furthermore, the proteins extracted from these insects exhibited favourable techno-functional properties and biological functionality. All samples demonstrated high antioxidative and antimicrobial properties. Based on these results, insect meals from *Tenebrio molitor*, *Acheta domestica*, and *Zophobas morio* are valuable nutritional ingredients comparable to fish meal and superior to soybean meal in terms of nutritional value in animal feed production. Additionally, the techno-functional and biological properties of the protein isolates suggest their potential use in various food, pharmaceutical, and cosmetic industries.

**Keywords:** insects, proteins, feed, food

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development, and Innovations, the Republic of Serbia (contracts no. 451-03-66/2024-03/200222 and 451-03-65/2024-03/200134).



## EFFECTIVENESS OF ESSENTIAL PLANTS IN THE PREVENTION OF KETOSIS IN DAIRY COWS

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Ketosis, a metabolic disorder often present in dairy cows during the peripartal period, is characterized by low glucose levels and high ketone body levels in the blood. This disorder can significantly reduce milk production and affect the overall health of the cows. Traditional prevention methods include proper nutrition and dietary supplements, but increasing research is focusing on the use of essential plants as an alternative or complementary solution. Essential oils, known for their antioxidant, anti-inflammatory, and antimicrobial properties, can play a significant role in the prevention of ketosis. Plants such as oregano, cinnamon, garlic, and thyme contain bioactive compounds that can improve the metabolism of glucose and fats, thereby reducing the risk of developing ketosis. Studies have shown that adding essential oils to the diet of dairy cows can positively affect metabolism and reduce the level of ketone bodies in the blood. For example, oregano oil, rich in carvacrol, can improve gluconeogenesis, the process by which non-carbohydrate sources are converted into glucose. Additionally, cinnamon oil can improve insulin sensitivity, which is crucial for maintaining stable glucose levels in the blood. The combination of different essential oils can have a synergistic effect, providing more comprehensive protection against ketosis. However, the correct dosage and method of application are important to achieve optimal effectiveness without adverse consequences. Although the research results are promising, further studies are needed to determine the optimal conditions for application and the long-term effects of essential plants on the health and productivity of dairy cows. In any case, essential plants represent an interesting and potentially effective option for improving the health and well-being of dairy cows and can become an important part of the strategy for preventing ketosis.

**Keywords:** essential oils, plant extract, phytotherapy prevention of ketosis, dairy cows

## ONION SKIN WASTE AS A POTENTIAL SOURCE OF FUNCTIONAL INGREDIENTS FOR ANIMAL FEEDS

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Onion is a widely cultivated vegetable and ranks as the second most significant crop after tomatoes. Consequently, large quantities of onion waste, particularly onion skin, are generated, leading to environmental concerns. However, due to the presence of bioactive compounds, onion skin holds the potential for creating value-added products when processed appropriately. The feed industry is increasingly utilizing by-products because of their affordability and beneficial properties. With the substantial amount of onion waste produced, it is crucial to explore its potential as a natural source of valuable functional ingredients. This study aimed to compare the nutritional profile of yellow and red skin onions as potential functional ingredients for animal nutrition. Yellow and red onion skins were characterized by a similar content of crude protein (2.67-2.85%) and crude fat (0.62-0.75%). A higher content of crude fiber (27.24 vs. 25.28%) and ash (15.26 vs. 12.50%) was observed in yellow skin onions. Iron and manganese were the most abundant microelements analyzed. Iron concentrations in yellow skin onions were 2855.34 mg/kg and 1911.92 mg/kg in red -skin onions. Similarly, a greater content of manganese was observed in yellow skin onions when compared to the red variety (141.50 mg/kg vs. 104.46 mg/kg). The antioxidant potential of the onion skin waste was evaluated by assessing total polyphenols and DPPH. It was found that red onion skin had higher values for total polyphenol content (114.85 mg/g GAE) and DPPH (849.82 mM Trolox). The lipid quality assessed by fatty acid profiling revealed that yellow skin onions had the highest percentage of linoleic acid (29.63%) and PUFA content (34.9%). The nutritional profile highlights that both types of skin onions are valuable agricultural by-products, due to the content of biologically active compounds, with potential applicability in animal nutrition.

**Keywords:** nutrients, yellow and red skin onions, by-products, animal nutrition, functional ingredients

**Acknowledgments:** This research was funded by the Romanian Ministry of Agriculture and Rural Development, Project ADER 8.2.2/2023.

## THE POTENTIAL OF CRAYFISH SHELLS AS A FEED SUPPLEMENT

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The spiny cheek crayfish, *Faxonius limosus* (Rafinesque, 1817), is one of the most invasive crayfish species in Europe, threatening river ecosystems by potentially causing the extinction of native species and altering ecosystem functions. Recent research has highlighted the potential of crayfish shells not only for chitin extraction, but also as a valuable source of amino acids and minerals that can serve as a feed supplement. These shells are particularly rich in calcium carbonate, a mineral crucial for the development and maintenance of bone and shell structures in animals. This research evaluated the chemical composition of the crayfish shell, focusing on its amino acids and mineral content. The spiny cheek crayfish were collected from the Begeč region of the Danube River. The shells, separated from the meat, were dried, ground, and analyzed. Amino acid profiling revealed the presence of 17 amino acids, with essential amino acids comprising 8.57 g / 100 g and non-essential amino acids constituting 14.19 g/100 g of the total content. The chemical analysis of the shell revealed 7 elements, with calcium being the most abundant in the dry residue. The calcium content in the crayfish shell was 17.90 g/100g dry weight. The shells also contained other microelements in the following concentrations: Ca > Na > Mg > Mn > Fe > Zn > Cu. The concentrations of heavy metals such as Pb, Cd, Hg, and As were below detection limits. These findings suggest that crayfish shells from this Danube location, rich in amino acids, calcium, and other essential minerals like magnesium and zinc, have significant potential as a cost-effective feed supplement. Using crayfish shells as a feed supplement enhances animal nutrition while being cost-effective and eco-friendly. This approach meets the demand for quality feed and supports environmental sustainability by utilizing waste materials.

**Keywords:** Invasive species, spiny-cheek crayfish, crayfish shells, zero waste, feed supplement

**Acknowledgements:** This research was supported by the Science Fund of Republic Serbia, #GRANT No. 7417. "Reducing the negative impact of invasive crayfish *Faxonius limosus* in the Danube by smart exploitation of their meat and shells" DANUBECare.

## THE POTENTIAL OF ENCAPSULATION OF MARIGOLD EXTRACT (*TAGETES ERECTA*) WITH CALCIUM ALGINATE FOR THE LAYING HEN NUTRITION

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Yolk colour is influenced by dietary carotenoids, and it is common practice in egg production to supplement hens' diets with these pigments. Carotenoids are susceptible to degradation, and their content in the diet decreases during storage. One of the approaches to protect carotenoids is encapsulation, a coating technology widely used in the food and feed industry. The present study aimed to encapsulate marigold extract with calcium alginate and investigate the use of the resulting microcapsules as a pigment source in egg production. Carotenoids from marigold flowers were extracted with acetone using a Soxhlet apparatus, and the dry extract was dissolved in sunflower oil. The carotenoids in oil were encapsulated using the ionic gelation method with sodium alginate and calcium chloride. After drying, the microcapsules were added to a maize-soybean laying hen diet in proportions of 0.5 and 1%. The laying hen trial was conducted in a completely randomized design with 3 treatments (T0 – control, standard laying hen diet, T0.5 – 0.5% microcapsules and T1 – 1% microcapsules (3 treatments×5 cages, 3 hens in each). Eggs were collected in the 4th week, and yolks were analysed for carotenoid profile, oxidative stability in iron-induced oxidation and colour (CIE Lab). The microcapsules contained lutein, zeaxanthin,  $\beta$ -cryptoxanthin and  $\beta$ -carotene (569, 108, 6.27 and 2.98  $\mu\text{g/g}$ , respectively). The addition of microcapsules increased the contents of all carotenoids in yolks compared to the control ( $P < 0.01$ ; total carotenoid content: T0 – 17.17  $\mu\text{g/g}$ , T0.5 – 23.16  $\mu\text{g/g}$ , T1 – 28.13  $\mu\text{g/g}$ ). The addition of microcapsules also increased yolk oxidative stability compared to the control, but there was no difference between the two levels of supplementation (slope: 1.31 vs. 0.81). Contrarily, the yolk colour intensity was considerably lower than the control (a value: 19.52 vs. 7.18). The addition of calcium alginate microcapsules containing marigold carotenoids in the hens' diet increased carotenoid content in yolks, but not to the extent expected, resulting in low colour intensity.

**Keywords:** carotenoids, encapsulation, egg yolk, color, oxidative stability

**Acknowledgements:** This work was supported by the Croatian Science Foundation under the project ColourMaize ("Bioavailability of maize carotenoids in laying hens: effect of grain microstructure and diet composition"; IP-2019-04-9063). The work of doctoral student Dora Zurak has been fully supported by the "Young researchers' career development project – training of doctoral students" of the Croatian Science Foundation.

## **AFLATOXICOSIS IN DOGS - EXAMPLES AND INTERPRETATIONS**

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The presence of aflatoxins in pet food is often neglected in practice, until an incident occurs, which is unfortunately becoming more common lately. On the other hand, perception of the danger of these secondary metabolites of molds in the human food chain is generally at a satisfactory level. Maximum permissible limits are regulated in food for both humans and farm animals, so monitoring programs are part of the strategy of state control of food and feed safety in European countries, including Serbia. However, the mycotoxicology system in pet food differs, with dogs being particularly sensitive to aflatoxins and having fatal consequences as a common outcome. Adding the fact that pets are called "companion animals", it is clear that due to the emotional attachment of the owners, each case presents very sensitive and more complicated challenge and exceeds the financial losses typical of farms. The lack of knowledge about the impact of aflatoxins on the health of dogs and the absence of regulations and control of this type of feed are the most common causes of recent outbreaks. The redirection of contaminated grains from the human food chain into pet food is unacceptable, and the frightening consequences can only be avoided by raising the level of information and educating all stakeholders. Therefore, the aim of this paper is to bring up awareness about the importance of control and conscientious behavior in the pet food industry, as well as to warn feed producers and dog owners about the negative effects that aflatoxins have on these animals.

**Keywords:** aflatoxins, dogs, pet food

**Acknowledgements:** The work is done as part of the Contract No 451-03-66/2024-03/ 200030 with the Serbian Ministry of Science, Technological Development and Innovation.

## IMPACT OF MICROPLASTICS FROM THE FOOD CHAIN ON THE REPRODUCTIVE HEALTH

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Microplastics (MPs) enter animals' bodies (digestive, respiratory system and skin) through contaminated water or feed/food. This can lead to the accumulation of plastics in their bodies, which may then be transferred to humans through the consumption of animal products. MPs have been shown to cause various health issues on all living organisms, such as inflammation, oxidative stress, and cellular damage. Their different distribution routes depend on their physical and chemical properties, and they can also act as a carrier of other harmful substances and microorganisms. MPs have even been discovered in animal products such as milk and eggs. We do not know how harmful microplastics are for the living organism, but there is evidence that small absorbed particles reach the gonads. As a consequence, more expressed ovarian cysts, more frequent corpora lutea presence, dilatation of the oviducts, lower number of growing follicles, a thinner layer of the granulosa cells in the female mouse and rats were noticed. In the testes of male animals an increase in SOD and MDA activities were described as a result of antioxidative response after MPs exposure. Toxicological research has demonstrated that MPs elicit toxic reactions in pig testicular cells, leading to apoptosis and necrosis of testicular tissue. Presence of MPs is connected with downregulation of testosterone, LH (luteinizing hormone), FSH (follicle-stimulating hormone) and AMH (anti-Mullerian hormone) concentrations in mice. Long-term exposure to MPs has been shown to cause a weakening of thyroid endocrine function, leading to a decreased ability to regulate growth, development, metabolism, and reproduction in marine vertebrates. Therefore, more studies should investigate alternative ways in which microplastics are excreted in livestock. In conclusion, microplastics pose a serious threat to animal health, affecting a wide range of species across different environments. Addressing this issue requires a concerted effort from scientists, policymakers, industries, and the public to reduce plastic pollution and protect animal health.

**Key words:** microplastic, food chain, livestock, reproductive health, endocrine system

## QUALITY OF INTERCROPS FORAGE AND MAIN CROP NUTRITIONAL PROPERTIES FROM THE SUBSEQUENT SOWING PERIOD

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Cover crops are mostly cultivated to enhance or build soil quality. Additionally, they expand the choices available to livestock farmers by generating fresh feed sources such as forage and grain, and allowing perennial pastures to rest. The definition of forage and grain quality can be quite diverse and is sometimes not fully grasped. It fluctuates significantly among different types of forage crops and even within them. Likewise, the nutritional requirements differ among animal species and groups. Forage and grain quality significantly impacts animal performance, the value of forage, and ultimately, the profitability of operations. The nutritional quality of forage for ruminants is demonstrated by a harmonious nutrient balance and an economically efficient utilization approach in production. Key indicators of quality include protein content, lignocellulosic fiber levels, and the digestibility of the forage. The goal of the research is to determine the quality and utility value of intercrops and crops as feed, specifically in terms of ruminant nutrition. The tested quality parameters are as follows: the content of protein, cellulose, hemicellulose, neutral detergent fibers (NDF), acid detergent fibers (ADF) and acid detergent lignin (ADL), as well as *in vitro* dry matter digestibility. According to the findings, it can be inferred that planting winter peas alone led to increased starch content in maize grains. Intercropping (triticale + peas) influenced the higher protein content in soybeans. On the other hand, simultaneously planting triticale and peas positively impacted the digestibility of Sudan grass (*Sorghum × drummondii*) and reduced its NDF, ADF and ADL content. In the agro-ecological context of Vojvodina, cultivating winter intercrops with carefully chosen crops for subsequent sowing offers a sustainable alternative to traditional arable farming methods. This proposed cultivation system not only brings positive agronomic benefits but also supports the production of sufficient high-quality feed for ruminants while closing the organic matter cycle within the farm.

**Keywords:** cover crops, grain quality, feed, crop diversification

**Acknowledgements:** This research was financially supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (No. 451-03-66/2024-03/200222).

## CAPILLARY TEST FOR OIL LEAKAGE EVALUATION

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High energy-dense fish feed, e.g. salmon feed with crude fat levels up to 40%, is known for its increased risk of leaking oil post-production, especially during storage at elevated temperatures and physical handling, including pneumatic transport from feed silos to the cages at sea. Oil leakage is a problem as it lowers the nutritional value of the feed, pollutes the environment, reduces water quality, and increases the risk of other quality issues, especially unintended float of the feed pellets. Therefore, salmon feed producers are following well-proven procedures in their manufacturing process to reduce the risk of oil leakage. One formulation-related approach is the addition of saturated fat to the oil mix added during vacuum-coating, thereby increasing the overall oil viscosity and consequently reducing the likelihood of oil leaking out of the feed pellets. As the oil viscosity, or more accurately, its physical stability and strength, is known to correlate with the risk of oil leakage, rheometers have been the obvious choice to describe the rheological behavior of the oil mix. Alternatively, actual feed pellets have been vacuum-coated in pilot coaters and stress-tested by means of centrifuges to quantify the amount of oil leaking out of the pellets. However, both approaches face challenges that lower the probability of obtaining significant and representative results when comparing fluids with different rheological properties. To evaluate the risk of oil leakage under more realistic and standardized conditions, a novel test method has been developed. This method utilizes capillary tubes, each representing a cylindrical pore. Filling the tubes with the oil mix and exposing them to realistic cooling conditions, then centrifuging the tubes and measuring the amount of retained oil, allows for strong statistical comparison of different fluids and selection of the physically most promising oil mix for full-scale aqua feed production.

**Keywords:** Oil leakage, Salmon feed, Physical Pellet Quality (PPQ), Rheology, Fat sealers



## **SUSTAINABILITY AND CIRCULARITY: WHAT IT BRINGS TO THE FEED INDUSTRY**

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Circularity in animal feed production is crucial for several reasons, encompassing environmental sustainability, economic efficiency, and social responsibility. Circular thinking and handling has become a necessity to establish sustainable animal production. Circularity is the driving factor for change, for innovation, for research, for reorganization within the feed industry and its supply chain. Within this key note the importance of circularity in the animal feed sector is approached from these three aspects, addressing several topics as waste reduction, added value creation, improving animal health and food security. Within the European feed industry, companies collaborate with research institutes and universities to implement circularity, resulting in research projects, partially or completely financed by government. Interesting case studies of company research will illustrate the willingness of the animal feed sector to grow towards a higher level of sustainable feed production.

**Keywords:** circularity, sustainability, waste reduction, value creation

## CHARACTERISTICS OF RAW MATERIAL FOR PET FOOD AND AQUA FEED EXTRUSION

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Extrusion is a pivotal process in the production of pet food and aqua feed, where the formulation heavily relies on a careful balance of proteins, carbohydrates, and fats. Each macronutrient plays a crucial role in determining the nutritional value, texture, digestibility, and processability of the final product. When selecting raw materials, manufacturers consider factors such as nutritional content, processability, economic viability, uniformity, fat inclusion, texture, palatability, and appearance. Proteins serve dual functions in aqua feed manufacturing: providing essential nutrition and imparting functional properties during extrusion. They are vital for growth, maintenance, and overall health in pets and aquatic organisms, supplying necessary amino acids for various physiological functions. During the extrusion process, proteins undergo denaturation, enhancing their digestibility. The heat and shear forces applied in the extruder also help reduce anti-nutritional factors present in certain raw materials. Additionally, proteins contribute to the feed's binding properties, significantly impacting texture and palatability. High-quality protein content can enhance the firmness and structural integrity of extrudates, leading to better performance. Carbohydrates serve as the primary energy source for pets and fish, providing the caloric intake necessary to support metabolic activities. During extrusion, starches gelatinize, which not only improves the digestibility of carbohydrates but also enhances the feed's binding properties. Properly extruded starches contribute to the desirable texture and shape of the finished product, making it more appealing to consumers. Fats are a concentrated source of energy, offering more calories per gram than proteins or carbohydrates, making them crucial for maintaining body condition and supporting metabolic functions. The extrusion process can improve the palatability of feeds, encouraging consumption among pets and aquatic species. However, careful management of fat inclusion is essential during extrusion to prevent excessive degradation, which can lead to off flavours, reduced nutritional quality, and compromised pellet durability. The balance and quality of proteins, carbohydrates, and fats are fundamental to optimizing the extrusion process for pet food and aqua feed. By understanding the specific roles of these macronutrients, manufacturers can formulate feeds that not only meet the nutritional needs of pets and aquatic species but also enhance the physical properties of the extrudates. This leads to improved performance, greater consumer satisfaction, and a more sustainable production process.

**Keywords:** Extrusion, Pet food, Aqua Feed, Protein, Carbohydrates, Fats

## HIGH PRESSURE PROCESSING OF FERMENTED SAUSAGES APPLIED AT THE EARLY STAGE OF RIPENING REDUCES LACTIC ACID BACTERIA HETEROFERMENTATIVE ACTIVITY

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Lactic acid bacteria in fermented sausages contribute to fermentation by several positive effects, like acidification, desirable degradation of initial sausage components, competitiveness and production of inhibitory compounds such as bacteriocins. However, heterofermentative microbiota, in addition to lactic acid, produce to a lesser extent also acetic and formic acids, hydrogen peroxide, carbon dioxide and ethanol. Higher quantities of these substances can lead to flavor and textural problems of the final products, like sour taste, hole formation and/or grey discoloration of sausages. Among additional processing measures for improving quality, high pressure technology is used mainly to increase shelf life and the safety of meat products. The aim of the study was to examine heterofermentative activity in dry fermented sausages produced without or with starter culture and processed by high pressure at the early stage of ripening. Application of high pressure at the optimal moment reduced significantly counts of colonies with heterofermentative activity in products without starter culture. Percentage of heterofermentative microbiota decreased from 50.6% to 6.5% in the total population of lactic acid bacteria in pressurized sausages, compared to untreated counterparts. Also, ratio of microbial community members assessed by amplicon 16S rRNA metataxonomics show that in total population percentage of obligate heterofermentative *Leuconostoc* genus clearly decreased (23.3% to 11.6%), while the percentage of *Latilactobacillus* increased (14.9% to 31.3%) in final products after high pressure treatment. Expectedly, in sausages with starter culture (pressurized or not) no colonies with heterofermentative activity were detected. The patterns of changes of main physicochemical parameters (pH and  $a_w$ ) in all sausages during ripening were usual for this type of products. Results showed that the application of high pressure at the optimal moment of the sausage ripening induces reduction of indigenous lactic acid bacteria heterofermentative activity.

**Keywords:** lactic acid bacteria, high pressure processing, sausages, heterofermentation, preservation

**Acknowledgements:** The financial support of the Ministry of Education, Science, and Technological Development of the Republic of Serbia (no. 451-03-68/2023-14/200117) and MASTER (project from the EU Horizon 2020 research and innovation programme (no 818368) is gratefully acknowledged.

## MICROPLASTICS CONTAMINATION IN CROSS-BORDER REGION OF CROATIA AND SERBIA

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Microplastics have become a major environmental concern, contaminating water, soil, and air, and impacting crop quality, food safety, and human health. They enter agricultural systems through irrigation water, soil amendments, flooding, and atmospheric deposition. Microplastics can alter soil structure and fertility, and plants can absorb them, leading to food chain contamination. Domestic animals ingest microplastics through contaminated feed, further exacerbating the issue. Addressing microplastic contamination in agriculture requires policy measures, sustainable practices, waste management, and public education. The EU's strategies, such as the European Green Deal and the EU Plastics Strategy, aim to reduce environmental impacts through regulations like REACH and the Single-Use Plastics Directive, promoting biodegradable alternatives and enhancing recycling. Monitoring microplastics in irrigation water and soil is essential for understanding contamination levels. Standardized detection methods ensure consistent data and reliable assessments. Microplastics pose food safety risks as they accumulate in animal tissues and carry harmful chemicals like persistent organic pollutants and heavy metals. Ingested by humans, these chemicals can cause inflammation, oxidative stress, and endocrine disruption, increasing health risks. The ECO(RE)ACT project, funded by Interreg IPA Croatia-Serbia, aims to reduce microplastics contamination and enhance climate change resilience in Croatia and Serbia through agricultural plastic waste management, ecosystem monitoring, and innovative mitigation solutions. By leveraging cross-border expertise, the project seeks to reduce microplastics contamination and promote sustainable agricultural practices.

**Keywords:** Microplastics contamination, Agricultural systems, Food safety, Environmental health

## PET FOOD INDUSTRY: GETTING CLOSER TO HUMAN FOOD

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Recently, there has been a growing interest in innovation in pet food production. According to data from the European Pet Food Industry Trade Association (FEDIAF) for 2022, there are 352 million pets in Europe with an annual pet food production of 9.9 million metric tons. Pet food is no longer just a simple meal with the necessary ingredients for the growth and development of animals. Still, it approaches food for humans, respecting the principles of a balanced diet with all the required additives and supplements, being visually representative, and focusing on the owner who wants to give his pet the best. A pet is no longer just an animal but a family member who has equal status with the other family members. The latest research shows that profits from the production of pet food have reached extremely high values, and it is predicted that by 2030, this market will generate earnings of 300 billion dollars. In the past five years, about a third of innovative pet foods have come from new companies or brands, far more than what we see in human foods. There are three key global trends in pet food production: personalization of products and recipes, use of natural ingredients, and sustainability. As consumers continue to look for ways to personalize and optimize their health, they can be expected to consider the same for their pets, researching what is the healthiest diet for a pet. On the other hand, as in human food, the trend of using natural products without artificial ingredients is still present in dog and cat food. One way to achieve "naturalness" is frozen or chilled pet food, with the biggest challenge being the transportation, distribution, and storage of this type of food. In the pet food industry, there is an increasing focus on sustainability, especially as it relates to packaging. There are also pet food brands dedicated to regenerative agriculture, in line with human food trends.

**Keywords:** pet food, trends, sustainability, human food

**Acknowledgements:** This research was financially supported by the Provincial Secretariat for Higher Education and Scientific Research AP Vojvodina (Project No. 142-451-3504/2023-01), as well as by the Ministry of Science, Technological Development and Innovation, Republic of Serbia, Institute of Food Technology in Novi Sad (451-03-66/2024-03/200222).

## ASSESSMENT OF RHEOLOGICAL PROPERTIES OF CARP FEED MIXTURES WITH DIFFERENT PROTEIN SOURCES USING MIXOLAB

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Three carp diets with same starch content (approximately 230 g/kg) with the inclusion of different protein sources were formulated. Control feed contained fish meal and soybean meal as a main source of protein which were completely substituted by dried duckweed. All ingredients of the diets were finely ground at the hammer mill and then mixed in twin-shaft paddle mixer. The aim of this paper was to assess the rheological and thermomechanical properties of these mixtures in order to forecast their behavior during subsequent processing. The determination of rheological properties was performed by Mixolab where water absorption capacity was set at a constant value of 100%. The Mixolab curve profiles varied where diet in which the fishmeal was completely replaced by the novel plant protein ingredient, duckweed flour, displayed greatest resistance to deformation, therefore the highest water holding capacity which was in accordance with results obtained by the method used for assessment of water absorption properties. Obtained results demonstrated that Mixolab has a good potential to be used as a tool for screening dry feed mixes when taking into account the amount of water and steam necessary for the successful conditioning management. Further investigation is required to enhance the applicability of these methods in establishing more effective parameters during the conditioning process.

**Keywords:** fish feed, mixolab, starch rheology

**Acknowledgements:** These results are part of the research within the project funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant number 451–03-66/2024–03/200222) and Horizon Europe project IMPRESS – Innovative approaches for marine and freshwater based ingredients to develop sustainable foods and value chains (project number 101084437).

## NUTRITIONAL STRATEGIES FOR LAYING HENS TO ADDRESS ENVIRONMENTAL CHALLENGES BY REDUCING THE NITROGEN EXCRETION

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This research aimed to assess the impact of incorporating *Castanea sativa* powder into laying hen diets, examining reduced crude protein levels and their effects on production performance, health status, nutrients and mineral digestibility, and environmental pollution by nitrogen (N) excretion and absorption. For that, a 6-week trial was developed, with 90 Lohmann Brown laying hens aged 51 weeks, raised in digestibility cages, divided into three groups with 30 hens each. The diets were composed as follows: a control group fed with 17.50% crude protein (CON), an experimental group with a reduced protein level of 15.50% (RPL), and a similar reduced protein group supplemented with 0.5% *Castanea sativa* powder (RPCs) as tannin supplement. The limiting amino acids (lysine, methionine, and threonine) were supplemented to maintain constant equal amino acid concentrations in all experimental diets. Throughout the trial, the laying intensity was higher in the RPCs group (94.12%), followed by RLP (93.65%) and CON (91.11%). However, the CON hens yielded heavier eggs compared to the RPL and RPC groups. Average daily feed intake and feed conversion ratio showed no significant differences among the groups. Health assessments from blood samples taken at the end of the trial showed a significant effect on monocytes, and uric acid among the groups with tendencies for leucocytes, lymphocytes, and heterophiles. Notably, excreted nitrogen levels were significantly reduced (up to 30%) in experimental groups compared with the CON group, showing a promising way of reducing N pollution. On the other hand, the apparent absorption of protein was higher in the groups with lower levels of protein in the diet (RPL and RPCs) compared with the CON group.

**Keywords:** *Castanea sativa*, health status, performances, poultry, protein levels, nitrogen balance

**Acknowledgements:** This research was funded by Romanian Ministry of Research Innovation and Digitalization, Grant number PN 23.20-0301 (Nucleu).

## POSTHARVEST QUALITY CONTROL AND MECHANISM RESEARCH OF SOLANACEOUS CROPS

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The tomato and potato are two representative solanaceous crops widely cultivated in Shandong Province, China. During the post-harvest storage process, tomato fruits are prone to infection by *Botrytis cinerea* or cold stress, leading to significant post-harvest losses. Jasmonic acid is an important small molecule signal in plants, involved in the plant's response to stress. This report introduces how ABC transporters regulate the transport of jasmonic acid within the plant, affecting jasmonic acid signal transduction, and further explores the mechanism by which ABC transporters in tomatoes influence resistance to *B. cinerea* and cold damage. Additionally, potatoes are susceptible to mechanical damage during harvesting, processing, and transportation, which easily leads to enzymatic browning and quality deterioration. The report also discusses the discovery of safe and efficient browning inhibitors and the research on enzymatic browning regulation mechanisms. This provides important theoretical references for the post-harvest storage, processing, and preservation of solanaceous crops.

**Keywords:** *B. cinerea*, ABC transporters, enzymatic browning



## INDUSTRIAL HEMP FOR A SUSTAINABLE FEEDING OF RUMINANTS

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Industrial hemp (*Cannabis sativa* L.) is a versatile crop that offers applications beyond human consumption. Its botanical parts and by-products show potential in the animal feed industry due to their nutritional properties. This study aims to provide a comprehensive overview of industrial hemp as a sustainable feed for ruminants. The research involved the chemical characterization of different hemp varieties by analyzing the whole plant, its botanical fractions (stems, leaves, seeds) and hemp-derived products (cake and oil). Additionally, the use of hemp cake as feed for veal calves was tested to evaluate their performance and behavior. Lastly, the use of the stems as animal bedding material was analyzed. The findings revealed that the proportion of botanical fractions and the chemical composition are significantly influenced by agro-climatic conditions. Hempseed and its derived products contain high amounts of polyunsaturated fatty acids, primarily linoleic and  $\alpha$ -linolenic acids. The amino acid profile showed that arginine and glutamic acid were the most abundant. The mineral content showed high levels of manganese. Tetrahydrocannabinol (THC) levels were very low, indicating minimal to no psychoactive effects. Hempseed had high levels of fat (31.72 %DM) and protein (25.17 %DM). These findings highlight CS, Eletta Campana, and Tisza as the most promising varieties. Including hemp cake in the concentrate feed of Holstein veal calves showed no adverse effects on their health or *in vivo* performance parameters. Furthermore, diluting hemp cake in the milk replacer of Belgian Blue veal calves had no effect on their behavior. Hemp shives, derived from the stems, were found to be suitable as animal bedding material due to their excellent physical characteristics, such as high water and ammonia absorption capacity. In conclusion, hemp cake is a versatile and nutritionally valuable resource for feeding ruminants, offering a sustainable option for the animal feed industry.

**Keywords:** hemp, animal nutrition, hemp cake, hemp seeds, ruminants

**Acknowledgements:** Fondazione Cassa di Risparmio di Padova e Rovigo (CARIPARO).

## IMPACT OF SUPPLEMENTATION OF *CHLORELA VULGARIS* ON THE NUTRITIVE VALUE AND PHYSICAL PROPERTIES OF PELLETTED FEED FOR LAYING HENS

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Microalgae are a promising alternative to conventional feed ingredients like corn and soybean meal, due to their high nutritional value, functional benefits, and potential to improve the sustainability of animal production systems. Therefore, the aim of this study was to evaluate the effect of inclusion of *Chlorella vulgaris* (CH) to the diet of laying hens on the nutritive profile and physical characteristics of pellets. Dried CH was included in a diet formulation based on a corn-soybean-sunflower meal (control group), replacing part of the protein source at levels of 5% (CH5) and 10% (CH10). Feed ingredients were mixed in a two-shaft paddle mixer, and then pelleted in a pellet press with a flat die featuring an opening diameter of 3 mm. It was observed that the inclusion of CH did not affect the level of saturated fatty acids, while it increased the proportion of monounsaturated and polyunsaturated fatty acids at the higher inclusion level (CH10) compared to the control group. Dietary supplementation with CH resulted in an increase in alpha-linolenic acid and up to a 2.5-fold decrease in the n-6/n-3 ratio (from 13.15 to 5.16). The inclusion of CH at 10% elevated the content of essential amino acids, particularly phenylalanine, methionine, and isoleucine. Regarding the physical properties of the feed, the pellet durability index was not affected by the addition of CH, while the hardness of the pellets decreased irrespective of the amount of algae added to the feed. In contrast, bulk density of the pellets improved with the addition of 10% CH. Furthermore, the specific energy consumption of the pellet press was reduced by half with the addition of 5% CH compared to the control group.

**Keywords:** microalgae, fatty acids, pellets, bulk density, specific energy consumption

**Acknowledgements:** This study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (451-03-66/2024-03/200222).

## OPTIMIZED DRY FRACTIONATION PROCESS: A METHOD TO ELEVATE THE NUTRITIONAL VALUE OF SUNFLOWER MEAL FOR FEED FORMULATIONS

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The relatively high protein content and low cost of sunflower meal, a byproduct of hexane oil extraction from sunflower seeds, make it a potentially valuable plant protein source in feed formulations. In this study, the optimal set of parameters for the dry fractionation process, determined in a previous study, was applied to a new batch of sunflower meal with similar characteristics, aiming to enhance its nutritional quality, particularly its protein content. The optimal set of parameters for two-stage grinding was identified as sieve openings of 2 mm, a roll gap of 0.25 mm, a feed rate of 0.2 kg/cm<sup>2</sup>min, and a roll speed of 400 rpm. The predicted values for the desired responses were: protein content of 45.5%(dm), fraction yield of 77.89%, and grinding energy consumption of 8.31 Wh/kg. Additionally, the amino acid content of the improved sunflower meal was analyzed and compared to that of commercially available LowPro soybean meal (protein content of 44%). The optimized dry fractionation process increased the protein content of sunflower meal from 35.64%(dm) to 44.20%(dm), representing a relative protein enrichment of 24%. Grinding energy consumption was 8.44 Wh/kg and fraction yield of the improved sunflower meal was relatively high 78,9%. The dry fractionation process also improved total amino acid content of sunflower meal from 24.62%(dm) to 30.00%(dm), representing a relative amino acid enrichment of 21,85%. While the total amino acid content of the improved sunflower meal was slightly lower than that of the LowPro soybean meal (32.80%(dm)), it exhibited a relatively similar amino acid composition. The study demonstrates that the optimized dry fractionation process can significantly enhance the protein and amino acid content of sunflower meal, supporting its use in high-quality feed formulations.

**Keywords:** Dry fractionation process, sunflower meal, hammer mill, roll mill, protein content

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development, and Innovations, the Republic of Serbia (contracts no. 451-03-66/2024-03/200222 and 451-03-65/2024-03/200134).

## IMPACT OF SOYBEAN MOLASSES ON THE PELLETING PROCESS AND PHYSICAL PELLET QUALITY OF BROILER DIETS

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Soybean molasses is a by-product of the soybean processing industry, is partially used as an energy source, and partially disposed of as waste in landfills. The organic matter in soybean molasses can pollute the air, and its nitrogen content has the potential to contaminate groundwater. This study examines soybean molasses, as a product with potential utility in the animal feed industry. Its relatively low cost offers an opportunity to reduce the overall price of animal feed products. The primary objective of this research was to optimize the production process of broiler feed pellets by varying soybean molasses content (1, 3, 5%), moisture content (13, 15, 17%), and sieve opening diameter of the hammer mill (1, 2, 3 mm). The experiment was conducted using the Box-Behnken experimental design. The measured responses were specific energy consumption of the pellet press (SEC), quantity of fines (QoF), pellet hardness (PH), pellet durability index (PDI), and bulk density (BD). The findings revealed that increasing the soybean molasses content led to a decrease in SEC and an increase in PDI, while having no significant impact on QoF, PH, and BD. Models obtained in this research were used to optimize the input parameters to achieve minimum SEC and QoF, and maximum PH, PDI, and BD. The results of the optimization for the input parameters were: soybean molasses content = 5.00 %, moisture content = 13.00 %, and sieve opening diameter of the hammer mill = 1.00 mm. The corresponding responses were: SEC = 22.0 kWh/t, QoF = 0.83 %, PH = 7.51 Kahl units, PDI = 98.81 %, and BD = 687.14 g/L. The high desirability value of 95.4 % indicates a strong alignment of the responses, demonstrating the efficacy of the optimization process.

**Keywords:** broilers, soybean molasses, by product, optimization, specific energy consumption

**Acknowledgements:** This work was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (451-03-66/2024-03/200222).

## TWIN SCREW EXTRUSION OF AFRICAN CATFISH DIETS CONTAINING BARLEY PROTEIN CONCENTRATE

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It is of utmost importance to secure that aquaculture systems be not just profitable but also sustainable. Dietary manipulation can be one beneficial method toward achieving the sustainability of aquaculture. Barley protein concentrate (BPC) is a relatively new protein source, locally available in Hungary and Serbia, that could at lower costs provide similar protein content as soybean protein concentrate, commonly used protein source for aquatic feeds. Investigation of techno functional property of each novel plant or animal based material should not be omitted when assessing its potential to be used in aquafeeds. The aim of this study was to investigate effect of barley protein concentrate (BPC) inclusion in African catfish (*Clarias gariepinus*) diets on extrusion process parameters and physical quality of obtained feeds. One control and two experimental African catfish diets were formulated to have approximately the same protein (450 g/kg) and fat content (110 g/kg) and BPC was used as a protein source in experimental feed, substituting half and complete of soybean proteins content. This meant that BPC was included in diets at levels of 100 and 200 g/kg and extrusion of each of three diets was done under same conditions using a twin screw extruder. Inclusion of BPC did not have any significant ( $p > 0.05$ ) influence on specific mechanical energy during extrusion, meaning no production energy consumption changed for tested diets. BPC did however significantly ( $p < 0.05$ ) increase expansion ratio when present at 200 g/kg level, which subsequently reduced bulk density. However this was beneficial since it increased floating ability of the feed, important requirement of African catfish pellets. Substituting soybean ingredients with BPC resulted in significant drop ( $p < 0.05$ ) in pellet durability, although it should not be considered as negative aspect of BPC, since all feeds possessed high durability values above 97%.

**Keywords:** African catfish, barley protein concentrate, extrusion, physical quality

**Acknowledgements:** This research is a part of ongoing project funded by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222) and current AQUAEXCEL3.0 TNA Proposal PID27144: Local protein sources for feeding hybrid African catfish. The barley protein concentrate was kindly provided by the company Pannonia Bio Zrt., Dunaföldvár, Hungary.

## ANTIMICROBIAL CONTAMINATION IN FEED ADDITIVES: REGULATORY STRATEGIES AND RISK ASSESSMENT FOR RESISTANCE DEVELOPMENT

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The rise of antimicrobial resistance (AMR) poses global threat to both animal and human health. Although the European Commission (EC) (Regulation EC No 1831/2003) prohibited the use of antibiotic feed additives as growth promoters since 2006, there is still a risk of feed additives contamination with antimicrobials. In the European Union (EU), 24 different antimicrobials may be administered with medicated feed, which poses risk for feed cross-contamination. This contamination could occur at various stages of production, including during the sourcing of raw materials, manufacturing and storage, as well as handling and transportation of feed products. These residues could include antimicrobials used in both human and veterinary medicine, raising concerns about their potential contribution to AMR development. To control the use of antimicrobials in animal feed and prevent contamination, the EU has implemented comprehensive regulations, such as Regulation (EC) No 1831/2003 and Regulation (EU) 2019/4. These regulations mandate that feed additives must be free from harmful levels of antimicrobial residues and emphasize the importance of good manufacturing practices (GMP) and hazard analysis and critical control point (HACCP) systems. Risk assessment is a key element of the regulatory framework, focusing on identifying potential hazards, assessing exposure and evaluating the impact on AMR development. The European Food Safety Authority plays key role in conducting risk assessments and providing scientific advice on the safety of feed additives using the Feed Antimicrobial Resistance Selection Concentration (FARSC) model. Monitoring programs at the national and EU levels are essential for detecting and controlling antimicrobial contamination in feed additives. Effective risk management strategies are essential for mitigating the risk of AMR development due to contaminated feed additives. Key measures include strengthening regulatory oversight, improving manufacturing practices, enhancing surveillance and promoting awareness among stakeholders. By adhering to these strategies, it is possible to contribute to the global effort to combat AMR and safeguard public health.

**Keywords:** antimicrobials, antimicrobial resistance, cross-contamination, feed additives

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia [Grant Numbers 451-03-65/2024-03/200117].

*Smart Food Development and Innovation*

## **CONSUMER CO-CREATION AS THE SOURCE OF INSPIRATION FOR NEW FOOD PRODUCTS**

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Consumers can be an important source of open innovations – external inspirations for food producers, helping design new or improve existing products. The presentation discusses the experiences of EIT Food RIS Consumer Engagement Labs project, leveraging a methodology developed by University of Warsaw. In 2019-2024, Labs were implemented in 19 European countries, with 106 consumer panels and 61 companies, resulting in 34 co-created products. Co-creation processes differ significantly from focus groups, sensory studies or acceptance tests. The Labs rely on a longitudinal process, driving the engagement of consumers, their sense of agency and teamwork. The methodology is implemented with non-expert consumers, through a sequence of steps, starting with projective techniques that explore consumer needs and requirements, moving towards assignments that turn consumers into researchers. Subsequently, a set of creative techniques helps generate ideas for modification of existing products and introduction of previously unknown products. The consumer-driven part of the process is followed by the company, screening and selecting the technically viable and commercially attractive proposals for new product design and development. The methodology is suitable both for food products and packaging, as evidenced by experiences of the Institute of Food Technology in Novi Sad, which worked with Serbian consumers to design more sustainable packaging for takeaway meals. Consumer ideas include proposals unexpected by companies: new product concepts, but also modified formulations and convenience enhancements. The presentation discusses examples of co-created products from Lithuania, Portugal, Croatia and Latvia, and summarizes general tendencies in proposals co-created by consumers.

**Keywords:** food design, co-creation, consumers, innovation, living labs

**Acknowledgements:** The paper presents results of EIT Food RIS Consumer Engagement Labs project that has received funding from the European Institute of Innovation and Technology under Horizon 2020 and Horizon Europe, the EU Framework Programmes for Research and Innovation, grant number: EIT Food/19153.



## AI IN MEAT SCIENCE AND TECHNOLOGY

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Artificial intelligence (AI) is transforming the field of meat science and technology by optimizing production processes, enhancing quality control, and improving supply chain management. In meat processing plants, AI-powered systems are used for real-time monitoring and automation. For example, machine learning algorithms can analyze data from sensors to ensure that meat is processed at the optimal temperatures, reducing waste and enhancing safety. AI-driven robotics are also being employed to handle repetitive tasks such as cutting, packaging, and sorting, which not only increases efficiency but also helps reduce human error and labor costs. AI is also being applied in meat quality assessment, traditionally a subjective and labor-intensive process. With advanced computer vision systems and deep learning algorithms, AI can assess the color, texture, marbling, and other quality parameters of meat with great precision. These systems can detect anomalies and predict spoilage by analyzing patterns in data, ensuring that only high-quality meat reaches consumers. Furthermore, AI-based predictive models can forecast meat tenderness and juiciness, offering significant advantages in product development and consumer satisfaction. In supply chain management, AI is revolutionizing the way meat products are tracked and distributed. AI-powered analytics can predict demand, optimize inventory levels, and ensure timely delivery, reducing waste and enhancing efficiency. Blockchain technology combined with AI is being used to improve transparency in meat sourcing, ensuring that consumers receive sustainably and ethically sourced products. Additionally, AI can help monitor and respond to real-time market conditions, adapting pricing strategies and distribution networks accordingly. Overall, AI is pushing the boundaries of what is possible in meat science and technology, making the industry more sustainable, efficient, and safe.

**Keywords:** automation, predictive modeling, computer vision, blockchain

## Computer Aided Food Engineering: digital tools as design tools for innovative food products and processes

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What are digital tools and how can they be used in designing quality food? How can the use of these tools contribute to new product development (food product design)? These are (some) questions put along this talk. Under the large umbrella of digital tools for the design of food products are included software and applications, coupled with any online or offline resource that can be used with computers, mobile devices or other digital devices, and in which it is possible to incorporate data, data analysis and data prediction. Some of these tools are known and established as methodology (mechanistic modeling, statistical analysis, machine learning tools) and are often applied in research, both at industrial and academic level. The benefit of using digital tools resides in the possibility to accelerate the exploration of different scenarios, then shortening the time-to-market of a new food product. Being based on computation, they can virtually provide answers to complex problems in a short time, exploring scenarios with a very limited cost and – potentially – without any limit. So, in a world running toward sustainable food transition, the use of these tools can open new roads to established food companies but also to start-ups. Another unpayable benefit coming from digital tools is the contribution to transfer knowledge. Technological data available on food properties, from ingredients to final products, are often scattered or uncomplete. In such a scenario, a pure statistical approach could lead to relations among data which are misleading. Digital tools can be used in such cases to guide the interpretation of data relationships and help the transfer of knowledge from product developers and process/system designers. Finally, a visionary perspective on the use of unconventional digital tools (though for other purposes than food design) for the understanding of consumers' perception and needs is presented and discussed.

**Keywords:** digital tools, food product design, statistical analysis, knowledge transfer, consumers' perception

## DEEP EUTECTIC SOLVENTS AND IONIC LIQUIDS AS SUSTAINABLE SOLVENTS FOR THE EXTRACTION OF VARIOUS BIOACTIVE COMPOUNDS

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Deep eutectic solvents (DES) and ionic liquids (ILs) have attracted significant attention as sustainable alternatives to conventional organic solvents, showing effectiveness in the extraction of both low-molecular-weight bioactive compounds and macromolecules such as proteins. Their unique physicochemical properties, including low volatility, tunable polarity, and strong solvation capabilities, make them ideal for green extraction processes. Additionally, DES and ILs are often biodegradable, non-toxic, and environmentally benign, providing a sustainable approach to extracting phenolic compounds, flavonoids, alkaloids, terpenoids, and proteins from various biomass sources. These solvents offer distinct advantages over traditional solvents, including higher extraction efficiency and improved selectivity for complex biomolecules. In our study, we investigated protein isolation from rapeseed press cake using DES, specifically choline chloride (ChCl) combined with urea and glycerol as hydrogen bond donors. The ChCl-Urea system demonstrated superior extraction efficiency, yielding 31.3 g of protein per 100 g of dry matter with a protein purity of 89.2%, surpassing the results obtained with conventional alkaline extraction. The ChCl-Glycerol system also showed promising results, although it achieved slightly lower protein yields compared to the ChCl-Urea system. These findings underscore the potential of DES for the efficient and sustainable extraction of both small bioactive compounds and larger biomolecules. There is strong evidence supporting the use of DES and ILs in bioactive extraction, showing that they outperform conventional solvents in terms of yield, stability, and the preservation of bioactive properties. Moreover, the tunable nature of DES and ILs allows for the optimization of extraction processes by selecting appropriate cation-anion combinations. As research progresses, there is increasing interest in scaling up these eco-friendly solvent systems for industrial applications in the pharmaceutical, nutraceutical, food, and cosmetic sectors, presenting a promising future for sustainable extraction processes.

**Keywords:** green chemistry, deep eutectic solvents, protein extraction

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222), as well as by the Science Fund of the Republic of Serbia, Grant No 6673, PROTein from Rapeseed Oil Processing Waste: Application in Food and Wastewater Treatment – PROTOPOWER.

## ASSESSMENT OF MINERAL AND FATTY ACID PROFILES IN MILK AND FRESH CHEESE: A COMPARATIVE STUDY OF KOMBUCHA AND COMMERCIAL STARTER CULTURES

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Within this study, milk and fresh cheese samples made using kombucha and commercial starter cultures were analyzed to determine their nutritional quality regarding mineral content and fatty acid composition. A total of 27 different fatty acids were identified, with palmitic, oleic, stearic, and myristic acids being predominant at average contents of 30.43%, 28.91%, 11.00%, and 8.81%, respectively. No significant differences in the fatty acid profile were observed among milk, kombucha cheese, and cheese made with commercial starter cultures. The total mineral content in milk was 0.70 g/100 g, while it was higher in fresh cheese samples, ranging from 1.03 g/100 g to 1.51 g/100 g, with the highest content in kombucha cheese. The average contents of phosphorus, calcium, potassium, sodium, and magnesium in the samples were 495.44 mg/100 g, 97.48 mg/100 g, 69.72 mg/100 g, 30.91 mg/100 g, and 9.28 mg/100 g, respectively. Microelements such as zinc, copper, iron, and manganese were significantly higher in fresh cheese compared to milk. Kombucha cheese had higher calcium, phosphorus, magnesium, zinc, copper, and manganese levels and lower sodium content than cheese made with commercial starter cultures. Thus, kombucha as a starter culture for fresh cheese production yields a more nutritionally valuable product than cheese made with commercial starters and milk.

**Keywords:** mineral composition, fatty acid content, fresh cheese, milk, kombucha

**Acknowledgements:** Ministry of Science, Technological Development and Innovation of the Republic of Serbia project numbers 451-03-66/2024-03/ 200134 and 451-03-65/2024-03/ 200134.

## POTENTIAL OF CAMELINA FOR INNOVATIVE FOOD APPLICATIONS THROUGH PLANT BREEDING

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*Camelina sativa*, an ancient oilseed crop from *Brassicaceae* family, has gained attention for its potential in food technology. Archaeobotanical analyses reveal it was the only oilseed cultivated in Serbian archaeological sites from 3<sup>rd</sup> to 6<sup>th</sup> CE. *Camelina* is recognised for its unique fatty acids profile, being low in erucic acid, which makes it applicable as food and feed. On top of that, camelina is rich in polyunsaturated fats, particularly alpha-linolenic acid (up to 50%). *Camelina*'s main use was in biofuels, although camelina holds promise for food industry. Research conducted at the Serbian Institute of field and vegetable crops (IFVCNS) focuses on enhancing camelina's traits to meet the growing demand for sustainable and nutritious food sources. By advancing camelina's breeding, we seek to optimise key attributes such as increased oil and protein content, improved oil quality with favourable fatty acid profiles, and reduced levels of anti-nutrients. These improvements aim to position camelina as valuable ingredient in the food industry. To assess stability of seed yield and oil content, we evaluated two promising camelina lines (L1, L2) across 12 diverse environments (E1-E12). Our results indicate significant variations in performance based on sowing time, with spring-sown lines generally outperforming winter-sown ones. While environments E1-E6 excelled in oil content (40-46%), E10 showed the lowest oil production (29%). Environment E5 emerged as the optimal location for high and stable seed and oil yield. As partner of the CARINA project consortium, IFVCNS is at the forefront of exploring innovative uses of camelina. Our high-quality camelina seed and biomass will be exploited for active compounds extraction. Polysaccharides from camelina cake are being evaluated as stabilizing agent for formulations in food-supplements, pharmaceutical and cosmetic industries. New methods for polysaccharides extraction that are more energy-efficient and operate without chemical solvents are tested in frame of this project.

**Keywords:** *Camelina sativa*, camelina oil, food supplement, polysaccharides

**Acknowledgements:** This research was funded by European commission, CARINA project (*Carinata* and *Camelina* to boost the sustainable diversification in EU farming systems) and Provincial Secretariat for Higher Education and Scientific Research, project No. EB: 142-451-3504/2023-01/01. Study was carried out as part of the activities of the Centre of Excellence for Innovations in Breeding of Climate Resilient Crops—Climate Crops, Institute of Field and Vegetable Crops, Novi Sad, Serbia.

## IMPACT OF ZINC AND SELENIUM ADDITION AS A FORTIFICATION IN AGRO-FOOD BY-PRODUCT SUBSTRATES ON BLACK SOLDIER FLY GROWTH PERFORMANCE

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The objective of this research was to investigate the impact of an inclusion of Zn and/or Se in the growing substrate of black soldier fly larvae (BSFL). Concisely, five experimental substrates were formulated: *i*) A plant-based (Gainsville diet) control substrate (CTR); *ii*) Okara and potato waste-based substrate (OkPa); *iii*) OkPa-Zn enriched with 150 mg/kg of Zn; *iv*) OkPa enriched with 0.3 mg/kg of Se; and *v*) OkPa-Zn+Se enriched with 150 mg/kg of Zn + 0.3 mg/kg of Se. 500 BSFL were used for each diet and five replicates were set up. The larvae were reared under dark condition, at 26 °C with 60% RH and their growth performances analysed. The average weight of the individual larvae was not affected by the type of substrate ( $P>0.05$ ). Contrarily, at the end of the trial the CTR group reached a significantly higher total larval biomass ( $P<0.05$ ). The survival rate showed no difference between the experimental groups ( $P>0.05$ ). These data indicate that okara and potato waste could be used as an appropriate substrate for BSFL breeding and that the inclusion of Zn and Se does not affect their growth performance. Further research is needed to assess the larval proximate composition, the micromineral adsorption and the reproductive performance of the resulting black soldier fly.

**Key words:** Black soldier fly, Microminerals, Alternative substrates

## EFFECTIVENESS OF HYSSOP, WINTER SAVORY AND OREGANO ESSENTIAL OIL MICROCAPSULES IN LOWERING AFLATOXIN M<sub>1</sub> CONCENTRATION IN MILK

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Aflatoxin M<sub>1</sub> (AFM<sub>1</sub>), a carcinogenic metabolite of aflatoxin B<sub>1</sub> (AFB<sub>1</sub>), remains a health risk in milk despite pasteurization and storage. Grains contamination by AFB<sub>1</sub>, produced by *Aspergillus* species, leads to significant global economic and health issues. As global warming intensifies aflatoxin risks, new decontamination methods are needed. This study explores the use of essential oil (EO) microcapsules from hyssop (*Hyssopus officinalis* L.), winter savory (*Satureja Montana* L.), and oregano (*Origanum vulgare* L.) to reduce AFM<sub>1</sub> levels in milk. Microcapsules, with a 1:4 core-to-wall material ratio, were prepared via spray-drying and tested in milk samples heavily contaminated with AFM<sub>1</sub> at a level of 0.46 µg/kg. A concentration of 20 mg/ml microcapsules achieved notable reductions of AFM<sub>1</sub>—24.7% with hyssop, 19.0% with winter savory, and 11.5% with oregano. Lower concentrations (2 mg/ml) also reduced AFM<sub>1</sub>, though less effectively: 7.3%, 3.4%, and 8.4%, respectively. Antifungal activity against aflatoxin-producing moulds *Aspergillus flavus* was assessed, showing that oregano EO and its mixtures provided the most significant inhibition, with some combinations exhibiting synergistic effects. Microcapsules with oregano and winter savory completely inhibited *A. flavus* growth in broth media (Sabouraud Maltose Broth, SMB), while hyssop EO reduced it by 0.5 log CFU/g. The study underscores the potential of EO microcapsules for mycotoxin decontamination, highlighting their effectiveness as an alternative method for enhancing food safety. Further research and technological advancements are crucial for developing more effective solutions for aflatoxin reduction in the food industry.

**Keywords:** Aflatoxin M<sub>1</sub>, hyssop essential oil, winter savory essential oil, oregano essential oil, microcapsules

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia, Contract No: 451-03-65/2024-03/ 200134 and Contract No: 451-03-66/2024-03/200031 and by the Provincial Secretariat for Higher Education and Scientific Research, Republic of Serbia, Autonomous Province of Vojvodina (No. 142-451-3498/2023-01/01).

## REDUCTION OF AFLATOXIN M<sub>1</sub> CONCENTRATION IN MILK BY THE HYSOP ESSENTIAL OIL EMULSIONS

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Aflatoxin M<sub>1</sub> (AFM<sub>1</sub>), a hydroxylated metabolite of the potent carcinogen aflatoxin B<sub>1</sub> (AFB<sub>1</sub>), poses significant health risks, including hepatocellular carcinoma. AFM<sub>1</sub> can persist through milk pasteurization and storage, representing a threat to human health, especially in offspring. AFB<sub>1</sub> contamination, linked to *Aspergillus* species, affects a quarter of global food crops, with increasing risks due to global warming. Various methods, including chemical, physical, and microbial approaches, are employed to combat aflatoxin contamination. This study investigates the effectiveness of hyssop (*Hyssopus officinalis* L.) essential oil emulsions in reducing AFM<sub>1</sub> levels in milk. Milk samples with high AFM<sub>1</sub> contamination (0.46 µg/kg) were treated with emulsions containing 1, 5, and 10% (w/v) hyssop essential oil in water. Two emulsification techniques were used: a one-step homogenization and a two-stage process including ultrasonic treatment. The two-stage emulsification significantly reduced drop diameters and enhanced AFM<sub>1</sub> removal. The highest reduction, 53.3%, was achieved with 10% hyssop oil, while 5% oil led to a 20% reduction. The size of essential oil droplets did not significantly influence AFM<sub>1</sub> degradation for the 5% oil emulsion. Additionally, antifungal activities of hyssop essential oil and 5% oil emulsion were tested against aflatoxin-producing moulds *Aspergillus flavus*. The minimum inhibitory concentration (MIC) and minimum fungicidal concentration (MFC) for the oil were 454.54 µL/mL and for the emulsion were 227.27 and 454.54 µL/mL. While the antifungal potential of hyssop essential oil is limited, it shows promise for mitigating aflatoxin contamination in milk.

**Keywords:** Aflatoxin M<sub>1</sub>, hyssop essential oil, emulsions, milk, *Aspergillus flavus*

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovation of Republic of Serbia, Contract No: 451-03-65/2024-03/ 200134 and Contract No: 451-03-66/2024-03/200031, and by the Provincial Secretariat for Higher Education and Scientific Research, Republic of Serbia, Autonomous Province of Vojvodina (No. 142-451-3498/2023-01/01).



## NUTRITIONAL AND FUNCTIONAL POTENTIAL OF SESAME AND PUMPKIN SEED CAKES IN SPREAD PRODUCTION

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Oilseed cakes are known for their well-balanced nutritional content, featuring high levels of protein and fiber, as well as significant amounts of minerals, vitamins, and phenols. Although they have these beneficial attributes, they are often viewed as by-products of the oil extraction process. The growing interest in alternative protein sources and sustainably produced health foods highlights their potential as valuable resources. This study explores the use of sesame and pumpkin seed cakes in traditional spread production methods, aligning with the principles of a circular economy. Three types of spreads were created: one from pumpkin seed cakes, one from sesame seed cakes, and a third that combines both in equal amounts. The preparation process involved grinding the seed cakes into fine powders and mixing them with suitable binders. The spreads were analyzed using chemical methods, focusing on their amino acid profiles, textural characteristics, and color. Amino acid concentrations were measured with advanced chromatography techniques, while texture analysis evaluated the spreadability. Colorimetric assessments provided information on the visual appearance of the spreads, which is important for consumer acceptance. Combining these high-protein ingredients allows for the implementation of two contemporary strategies, resulting in a product that is rich in essential amino acids such as methionine, lysine, leucine, and isoleucine. These specially formulated spreads meet daily essential amino acid needs, offering a nutritionally balanced and healthy dietary choice. Given the nutritional advantages and consumer appeal of these raw materials, developing such products is a promising area of research.

**Keywords:** oilseed cake, added value spreads, plant protein, amino acid

**Acknowledgements:** This research was funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia, grant number 451-03-66/2024-03/200222. Authors would like to acknowledge to Serbia Accelerating Innovation and Entrepreneurship Project – SAIGE.

## ADVANCING SUSTAINABILITY AND HEALTH THROUGH INNOVATIONS IN CEREAL AND GRAIN SCIENCE: MEETING GLOBAL CHALLENGES

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Today, climate change is driving significant shifts in agricultural practices, food processing, and consumer beliefs. Ensuring food security for the growing global population is increasingly challenging, and even more so when it comes to providing nutritious foods that meet daily nutrient requirements while also playing a role in preventing health issues. Cereals have long been staple foods at the base of the food pyramid, and grains like pulses are gaining importance due to the global demand for plant-based proteins. In this context, cereal- and grain-based foods, especially breads, offer an excellent means of delivering essential nutrients and serving as healthy options tailored to specific dietary needs. Globalization has broadened the range of breads available, from white and whole grain varieties to proofed and non-proofed types, further diversifying the market. The incorporation of flours from various cereals, pseudocereals, pulses, and even powders derived from agri-food by-products introduces opportunities for innovation, combining their technological functionality with their nutritional benefits. However, these innovations often face technological challenges due to the distinct functional properties of each ingredient. Physical treatments such as milling, fractionation, germination, and fermentation alter the physico-chemical characteristics of raw materials, directly impacting both product quality and health benefits. These strategies are unlocking new possibilities for creating innovative baked goods, including gluten-free breads, to cater to consumer demands and address specific nutritional needs. This presentation will review all those strategies and the impact on quality and health aspects of breads, identifying the existing gaps that require further research.

**Keywords:** gluten free, pseudocereals, physical treatments, glycemic index

**Acknowledgements:** The authors would like to acknowledge University of Manitoba, and the Discovery Grant (RGPIN-2023-04289) of the Natural Sciences and Engineering Council (NSERC) of Canada for the financial support. The Accreditation as Center of Excellence Severo Ochoa CEX2021-001189-S funded by MCIN/AEI /10.13039/501100011033 is also fully acknowledged.

## THE PSYCHOCHEMICAL, OXIDATIVE AND TEXTURAL EFFECTS OF DECREASING SATURATED FAT CONTENT OF CHICKEN LIVER PATE THROUGH SUBSTITUTION WITH SUNFLOWER SEED OIL BASED OLEOGELS AND VEGETABLE PUREES

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Liver pâté, is a spreadable meat product consisting of animal fat, liver, salt, and spices. Food is valuable all over the world, it is known for its high nutritional value and technological significance. However, these products contain high levels of saturated fat and cholesterol. The recent shift in consumer behavior regarding the health effects of consumption has resulted in an increased demand for healthier alternatives to traditional foodstuff. This necessitates the research and development of food products with lower saturated fat and improved fatty acid content without compromising the quality and characteristics of the traditional counterpart. Researches present formulation healthier meat products with decreased animal fat content and improved fatty acid composition using novel approaches, such as partially modified plant-oil-based emulsions as substitutes. In this study, the effects of substituting animal-based fat with carnauba wax (10%) and sunflower-oil-based oleogels, along with a vegetable puree blend (17% celery stalk, 50% carrot, and 33% red bell pepper), on quality properties of chicken liver pâté were investigated. The chicken liver content was replaced with the vegetable puree by 15%, while the animal fat was substituted by 50% and 100% oleogel. As a result, the nutritional value of the pâté samples improved, with all experimental groups receiving similar sensory scores. Although oxidative stability slightly declined with oleogel substitution, it improved compared to the control when the vegetable puree was included. The study successfully characterized the nutritional profile of the products without significantly compromising their oxidative, sensory, and technological qualities.

**Keywords:** liver, pate, oleogel, substitution, vegetable-based

## EDIBLE FILMS AND BIOPRINTING MATERIALS AS CARRIERS FOR BIOACTIVE INGREDIENTS

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The food packaging industry is a major contributor to global plastic waste. Traditional food packaging petroleum-based materials, such as polyvinyl chloride (PVC), polyethylene terephthalate (PET) and polyethylene (PE), often provide excellent properties towards food preservation, their non-biodegradability and reliance on fossil fuels have damaging effects on the environment. As sustainability concerns grow, there is a pressing need to develop eco-friendly alternatives. In this context, chitosan, a linear polysaccharide derived from chitin of the exoskeletons of crustaceans, such as shrimp, crab and lobster, offers a range of beneficial properties that make it ideal for sustainable packaging applications. Chitosan, is a non-toxic and biodegradable natural biopolymer which finds many applications for the preparation of edible films as intelligent packaging materials. These chitosan-based films could be further reinforced with natural extracts that possess antioxidant and antimicrobial properties, offering active food packaging solutions that extend shelf life while maintaining product safety and quality. Apart from food packaging, chitosan's versatility extends to advanced material applications, such as the development of nanoparticles via ionic gelation. These chitosan nanoparticles could be exploited for targeted delivery of bioactive compounds in food and medical fields. Additionally, chitosan-based hydrogels could be used as bioinks for 3D bioprinting, enabling the creation of biomimetic scaffolds for tissue regeneration and other medicinal applications. The ability to blend chitosan with other biopolymers such as polylactic acid (PLA) or alginate enhances its mechanical and functional properties, enabling the development of composite materials for diverse applications in food packaging, 3D printing, and beyond. This work highlights the unique characteristics and great potential of chitosan as a sustainable alternative to traditional petroleum-based plastics.

**Keywords:** chitosan, natural biopolymer, edible films, nanoparticles, bioprinting material

**Acknowledgements:** This research has been funded by the Hellenic Foundation for Research and Innovation (HFRI) in the frame of the "2nd Call for H.F.R.I.'s Research Projects to Support Faculty Members & Researchers" under grant agreement No. 3308.

## FROM INVASIVE CRAYFISH TO VALUABLE RESOURCES

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Invasive Alien Species (IAS) are ranked among the top five direct causes of biodiversity loss, posing a substantial threat to humanity in the coming decade. The spiny-cheek crayfish, *Faxonius limosus*, native to Eastern North America, has now spread to over 20 European countries, including Serbia, and is classified as an IAS of Union concern. As a prominent aquatic invader in European inland waters, this species presents significant challenges for biodiversity conservation efforts, particularly in terms of its prevention, control, and eradication. Effective management of invasive species requires innovative strategies that turn these challenges into a variety of eco-products, aligning with the Zero Waste concept. Our study addresses the current problem of the invasive crayfish *F. limosus* in the Danube and its detrimental impact on native crayfish species and biodiversity, for which a systemic solution is lacking. We are turning this challenge into an opportunity by developing novel food and feed products while simultaneously utilizing shells for developing biosorbents, rubber bio-fillers, and bio-based packaging materials. Crayfish meat will be used to develop innovative food, pet food, and feed products. The leftover shells, which are rich in chitin—the second most abundant polymer after cellulose—offer a solution to two major environmental challenges: heavy metal contamination in aquatic ecosystems and plastic pollution. Due to their exceptional adsorption capabilities, crayfish shells can effectively remove heavy metals from industrial wastewater. However, the disposal of saturated biosorbents presents a challenge. To overcome this, the saturated biosorbents will be incinerated, with the resulting ash being utilized as a biofiller for rubber. Additionally, chitin and proteins extracted from the shells will be used to create bio-based packaging materials, offering an eco-friendly alternative to single-use plastics.

**Keywords:** Invasive species, spiny-cheek crayfish, crayfish meat, crayfish shells, zero waste

**Acknowledgements:** This research was supported by the Science Fund of Republic Serbia, #GRANT No. 7417. "Reducing the negative impact of invasive crayfish *Faxonius limosus* in the Danube by smart exploitation of their meat and shells" DANUBEcare.

## OPTIMIZATION OF 3D BIOPRINTING PARAMETERS FOR PRODUCTION OF GELATIN SCAFFOLDS

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3D bioprinting is an advanced technology that enables the precise creation of complex biological structures using bioink – a specialized substance composed of living cells, hydrogels, and other biomaterials. Among hydrogels, gelatin, serves as a carrier for cells within the bioink, allowing them to grow, differentiate, and facilitates the efficient transport of oxygen and nutrients. Due to its exceptional properties including biocompatibility, biodegradability, and non-toxicity, gelatin scaffolds play a fundamental role in tissue engineering and biotechnology. Therefore, optimization of the bioprinting process parameters is essential to improve the mechanical and biological properties of these scaffolds, significantly enhancing their efficiency. In this study, bioprinting process parameters were optimized using an extrusion-based bioprinter the Regemat REG4LIFE. The methodology included preparing commercial gelatin and determining the optimal gelatin concentration for scaffold fabrication. Additionally, key printing parameters, such as scaffold dimensions, printing temperature (24-37°C), flow speed, perimeter speed and infill speed were optimized to ensure the best results. As a result, it was found that a gelatin concentration of 10% is the most suitable for scaffold fabrication, providing good stability and layer adhesion. The optimal temperature, visually assessed during priming, was found to be 29°C, influenced by room and laminar airflow temperatures. The scaffold had a diameter of 20 mm, with a layer height set at 0.3 mm, and 10 layers were printed. Based on the analysis, the optimal bioprinting parameters were determined to be: flow speed of the bioink (2 mm/s), perimeter speed (8 mm/s), and infill speed (8 mm/s). Gelatin, as a hydrogel, has proven to be a suitable material for bioprinting scaffolds, providing adequate stability and mechanical properties. Future research will focus on further optimizing bioink formulation and bioprinting parameters to enhance scaffold functionality and explore its applications.

**Keywords:** 3D bioprinting, optimization, scaffold, gelatin, bioink

**Acknowledgements:** This work was supported by The Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant No. 451-03-66/2024-03/200358, 451-03-65/2024-03/200134 and 451-03-66/2024-03/200222).

## THE INFLUENCE OF POLYPHENOLIC COMPOUNDS FROM KOMBUCHA AND WILD THYME HERBAL DUST ON ANTIOXIDANT ACTIVITY AND REDUCTION IN NUMBER OF *L. MONOCYTOGENES*, *S. AUREUS* AND *E. COLI* IN FRESH CHEESE

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Due to the large number of lactic acid bacteria in its contents, kombucha can induce the fermentation of milk. The valorisation of wild thyme (*Thymus serpyllum* L.) as a by-product of the filter tea industry could be essential for its use as a natural additive in fresh cheese production. Phenolic compounds and other bioactive compounds from milk and kombucha can significantly impact the antioxidant activity and reduce the number of bacteria in fresh cheese during its shelf life. This study aimed to investigate the influence of wild thyme in the form of ground supercritical fluid extract and dry extract on the antioxidant activity and the reduction in the number of artificially added *Listeria monocytogenes*, *Staphylococcus aureus* and *Escherichia coli* in fresh kombucha cheese during a 30-day storage period. The present phenolic compounds from milk and kombucha showed a greater impact on increasing the antioxidant activity of fresh cheese samples when determined by ABTS and FRAP assays. The sample with the best antioxidant activity after 30 days of storage was the sample with wild thyme dry extract, where DPPH, ABTS and FRAP values reached 5.10  $\mu\text{M TE/g}$ , 11.80  $\mu\text{M TE/g}$  and 15.00  $\mu\text{M Fe}^{2+}/\text{g}$ , respectively. Using kombucha inoculum effectively protected fresh cheese samples from contamination with *L. monocytogenes*, *S. aureus* and *E. coli*. The addition of wild thyme, especially in the form of ground had the greatest effect on reducing the number of *L. monocytogenes* in fresh kombucha cheese (from 4.20 to 1.90 log CFU/g). The use of kombucha as a starter culture, herbal dust and wild thyme extracts have great potential to preserve fresh cheese from *L. monocytogenes*, *S. aureus* and *E. coli*. In this way, several benefits are achieved: valorization of food industry waste, preservation of functional dairy products in a natural way and improvement of their antioxidant activity.

**Keywords:** fresh cheese, kombucha, wild thyme, antioxidant activity, bacteria

**Acknowledgements:** This research was funded by Ministry of Science, Technological Development and Innovations of the Republic of Serbia, grant numbers 451-03-66/2024-03/200134 and 451-03-65/2024-03/200134.

## TEXTURAL PROPERTIES OF PESTO SAUCE WITH MEALWORMS

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Insects are becoming more popular in the food industry because of their nutritional value and sustainability. They also showed a good potential for creating new food products and improving existing ones. This research aimed to assess if it is possible to substitute cashews in pesto sauce with baked *Tenebrio molitor* larvae. Three different samples were made: a control sample which had 100% of cashews, a sample with 50% substitution and a sample with 100% substitution. All samples had the same total fat content. Texture analysis was done on TA.XTplus texture analyzer using the back extrusion rig with a 35mm disc. Results have shown that the index of viscosity was not statistically significantly different; likely because total fat content stayed the same across all 3 samples. Firmness and consistency were statistically significantly lower in pesto where 50% of cashews were substituted with insects compared to pesto formulation with 0 and 100% insects. The biggest mean value for cohesiveness was measured in pesto which has 50% of cashews replaced with mealworms. The lowest firmness and consistency at 50% substitution is likely a result of mixing ingredients that contain fats from four different sources (olive oil, milk fat from grana Padano cheese, cashews and insects). The highest cohesiveness in 50% substitution is likely due to the favorable geometry of particles in this formulation. This has led to a structure formation that holds together well without being too dense or crumbly. It can be concluded that when it comes to the texture of pesto sauce mealworms have the potential to replace cashews at a certain percentage. More research is needed to determine which percentage is optimal from the point of consumers' preferences and sensory analysis.

**Keywords:** Pesto, *Tenebrio molitor*, Insects, Texture, Cashews

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03-200222).



## ADDED VALUE SPREADS FROM PROTEIN AND FIBER RICH WALNUT AND BLACKBERRY OILSEED CAKES

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Currently, global society is searching for new plant-based protein sources due to health consciousness and the pursuit of a sustainable diet. Innovative spreads made from protein-rich oilseed cakes, left after pressing oil from seeds, present a potential solution to this need. These oil cakes, rich in fibers, high-value proteins, vitamins, and antioxidants, enhance the nutritional value of spreads. From seeds, is obtained valuable seed oil through cold-pressing procedure and valuable side product (oilseed cake). Oilseed cakes obtained after oil extraction by cold pressing are free of toxic solvents and represent safe ingredient. Due to the high content of dietary fibers, proteins, phenolic, and other bioactive compounds, the oil cake residue can be used as a valuable ingredient for nutritionally rich spreads. Added value spreads are created in a ball mill during a 2-hour production process until reaching satisfactory particle size distribution acceptable for consumers. Afterward, the spreads are chemically, nutritionally, and safety-characterized. Two different formulations of spreads are created: one from only walnut cake and other one from combination of walnut (protein-rich) and blackberry (fiber-rich) oil cake. Amount of proteins in walnut cake is over 45%, whereas amount of fibers in blackberry cake is over 62%. Spreads are made of oilseed cakes, cold-pressed oil and emulsifier. Using oil cake left after pressing oil in a completely new way achieves the zero-waste and circular economy concept. Through the reduction of food waste, our planet is progressively adopting a more environmentally sustainable stance. Moreover, the use of unconventional raw materials aims to produce spreads that can carry nutritional claims such as "high fiber" and/or "source of plant protein."

**Keywords:** oilseed cake, added value spreads, plant protein, fibers

**Acknowledgements:** This research was funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia, grant number 451-03-66/2024-03/200222. Authors would like to acknowledge to Serbia Accelerating Innovation and Entrepreneurship Project – SAIGE.

## THE SOYNUT PROJECT: INCREASING THE NUTRITION VALUE OF SOYBEAN GRAIN

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Soybean (*Glycine max* (L.) Merr.) is the world's leading oilseed crop, a crucial raw material in producing protein-rich food and feed, and a significant source of bioactive compounds with diverse medical benefits. Yet, it's also one of the most controversial crops due to its status as the star of Genetically Modified Foods. The project titled "The evaluation of soybean germplasm: Nutritional quality of grains under climate change conditions (SOYNUT)" was created by an interdisciplinary research team of scientists from the Agricultural Institute Osijek and the Faculty of Food Technology Osijek in response to market demands towards functional foods, especially those enriched with proteins. Through implementation of conventional soybean cultivation during a three-year field experiment and modern analytical methods we aim to profile protein content, individual amino acids, oil content and individual fatty acids for a maximum of 55 soybean genotypes. The main result of the project will be a data platform containing information necessary for the development of soybean cultivars particularly suitable for specific use as raw materials in the food industry and animal feed production. Based on the collected nutritional quality data, parental components will be chosen for crossings to create nutritionally superior soybean cultivars through plant breeding. Through various educational activities and promotions, we intend to increase the general population's awareness of the nutritional and health benefits of soybean.

**Keywords:** soybean, nutritional value, oilseed crop, protein-rich, plant breeding

## PHYSICAL PROPERTIES OF COOKIES FORMULATED WITH NETTLE (*URTICA DIOICA* L.) SEEDS: COLOR, TEXTURAL AND SENSORY EVALUATION

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Evaluating the color, texture, and sensory attributes of cookies is crucial, as these parameters not only influence consumer acceptance and overall appeal but also provide essential insights into the product's nutritional quality, ingredient interactions, and potential market success. These analyses ensure that the final product is both visually attractive and palatable while meeting health and dietary standards. Therefore, the purpose of this study was to determine the color, textural, and sensory attributes of wheat flour-based cookies with the addition of nettle seeds. Product color was determined using a MINOLTA Chroma Meter CR-00 with D-65 illumination, a 2° standard viewing angle, and an 8-mm aperture in the measuring head. The results are expressed as lightness (L\*), the proportion of red color (a\*), and yellow color (b\*) according to the CIE Lab\* system. The textural characteristics of the product were determined using a TA-XT2 Texture Analyzer, using a 3-Point Bending Rig. This setup is used to measure the strength and brittleness. The sensory evaluation of the product was conducted using the scoring method 24 hours after baking by a seven-member panel of experienced evaluators for this product group. The results were compared to a control product (product without nettle seeds). The incorporation of nettle seeds significantly affected the color, texture, and sensory quality of the cookies. It was observed that the nettle seed cookie had lower brightness and a reduced yellow tone in color, as well as being significantly softer and less brittle than the control product. Sensory analysis indicated the produced cookies were acceptable in terms of appearance (shape, homogeneity, surface), smell, taste, texture (structure, firmness, brittleness), with the quality score being 1.24 times higher compared to the control product. The analysis indicated that cookies with acceptable physical characteristics could be produced with partial replacement of wheat flour by nettle seed powder.

**Keywords:** cookies, *Urtica Dioica* L. seeds, color, texture, sensory evaluation

**Acknowledgements:** This research was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia, Scientific Research Funding Program, ev. no. 451-03-65/2024-03/200133.

## INVESTIGATION OF QUALITY CHARACTERISTICS OF RESTRUCTURED BEEF STEAKS PRODUCED BY USING TRANSGLUTAMINASE AND DIFFERENT PROTEIN SOURCES

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In this study, low-value carcass trimmings were converted into steak-like meat products using microbial transglutaminase (MTG) alone or in combination with pumpkin seed protein (PSP) or whey protein (WP), and the color and oxidative stability were evaluated. For this purpose, four different treatments were designed: 1% salt (KK), 1% salt and MTG (MK), 1% salt and MTG + 2% PSP (MKP1), and 1% salt and MTG + 2% WP (MKP2). The color characteristics, lipid (TBARS), and protein oxidation (carbonyl and sulfhydryl groups) were analyzed during storage at -18°C for 3 months. At the beginning of the storage period, KK had the lowest L\* value, while the highest value was observed in MKP1. In addition, MKP1 had the highest b\* values, and MK had the highest a\* values during storage. PSP significantly reduced the redness index, which can be attributed to MKP1 having the highest b\* value among all groups. Except for MKP1, all treatments showed a similar total color difference value. TBARS values of all treatments increased during storage, with the KK and MKP1 groups having the highest values on the first day of storage. Besides that, the MKP1 and MKP2 groups showed the lowest TBARS values at the end of the storage. Specifically, the fact that MKP2 had the lowest TBARS values during storage can be attributed to the antioxidative effect of whey protein. While MK had the highest carbonyl content at the beginning of storage, there was no significant difference between the treatments after the 1<sup>st</sup> month, and MKP1 had the lowest carbonyl content in its 2<sup>nd</sup> and 3<sup>rd</sup> months of storage. During storage, fluctuations in the sulfhydryl groups were observed, while all groups decreased overall. These findings suggest that PSP and WP have antioxidant properties that reduce TBARS values, while MTG promotes protein oxidation by causing the loss of sulfhydryl groups and increasing carbonyl content.

**Keywords:** restructured meat, microbial transglutaminase, oxidation, pumpkin seed protein, whey protein

**Acknowledgements:** The authors are thankful for the financial support of Ege University Scientific Research Projects Coordination under project number FM-YLT-2023-29559.

## RHEOLOGICAL PROPERTIES OF GLUTEN-FREE NOODLE WITH RED KIDNEY BEAN FLOUR: INSIGHTS FROM SMALL AND LARGE DEFORMATION TESTS

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The trend towards gluten-free and high-protein foods is growing, driven by consumer demand for healthier options. Red kidney beans offer a valuable alternative for making gluten-free noodle. Combining red kidney bean flour with rice flour results in different dough structures due to their distinct protein and starch compositions. This study aimed to explore the viscoelastic properties of gluten-free noodle with varying levels of red kidney bean flour substitution (0%, 10%, 30%, 50%, 70%, and 90%). The research utilized a Burgers model, which includes Kelvin and Maxwell models in series with an additional Kelvin element, to analyze experimental compliance and strain responses. This provided coefficients for pure elastic deformation, viscoelastic deformation, and pure viscous flow. The gluten-free dough was tested through large deformation (compression-recovery test; 3 N for 5 seconds compression and 55 seconds recovery) and small deformation (creep-recovery test; 20 Pa shear stress for 100 seconds and recovery at 0 Pa for 100 seconds) tests. Results indicated that the coefficients changed with the level of red kidney bean flour substitution. A 30% substitution reduced dough stiffness by 57.8% compared to the control, while a 90% substitution resulted in the strongest dough structure, characterized by low pure elastic deformation and high pure viscous flow parameters. No significant differences were noted at other substitution levels. The dough's breaking point showed a linear softening effect with increasing red kidney bean flour. Stretchability decreased by up to 45.8% with the addition of red kidney bean flour. In conclusion, both shear and axial compression strain tests showed that a 30% substitution of red kidney bean flour is the limit for maintaining the desired structure of gluten-free noodle before it becomes too stiff. The reduction in stretchability at this level also confirmed the limit for achieving the expected properties of gluten-free noodle.

**Keywords:** Gluten-free food, Red kidney bean, Rheological Properties

## THE BENEFIT-SHARING FUND'S GRAINEFIT PROJECT - TOWARDS DELIVERING WHEAT GRAINS THAT BRING NUTRITIONAL BENEFITS

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Wheat is a crucial staple crop in Serbia and worldwide, significantly contributing to food security and nutrition. Modern wheat breeding efforts that have concentrated solely on increasing yields have led to a decrease in genetic diversity. The Benefit-sharing Fund of the International Plant Treaty on Plant Genetic Resources for Food and Agriculture's project GRAINEFIT aims to improve the use and conservation of old neglected wheat varieties and landraces leading to increased availability of diverse nutrient-rich food, food security, reduced adverse impacts to the environment, and enhanced resilience to climate stress factors. One of the key activities that address these objectives was a detailed research of nutritional quality and agronomic performances of 33 Serbian wheat genotypes and their comparison to modern ones in two-year field trials (2021/2022, 2022/2023). Our studies showed that the total phenolics in traditional varieties and landraces were generally higher compared to modern varieties. Largely underutilized traditional varieties contained significant levels of ferulic acid, with some having three times higher levels than modern counterparts. Besides, vanillic, syringic and p-coumaric acid were measured in old varieties, but not in modern ones. Antioxidative potential varied among old and new varieties, with many old varieties demonstrating stronger DPPH radical scavenging activity. We also evaluated the levels of two essential minerals, iron and zinc, in old wheat varieties using a GF-AAS and identified Stara Banatka with the highest ash content, and iron and zinc concentrations. Consuming an average daily portion of bread (166 g) made from this flour could provide about 90% of the recommended daily iron intake, making it a variety suitable for breeding for high mineral content. The agronomic performances of some of these high nutritional quality varieties showed moderately high yields and yield components, good standing ability, resistance to prevalent diseases, indicating that they could be successfully exploited to develop climate-resilient varieties with improved quality.

**Keywords:** landraces, minerals, nutritional value, old varieties, phenolics, wheat

**Acknowledgements:** This research was funded by the Benefit-Sharing Fund of the International Treaty on Plant Genetic Resources for Food and Agriculture, PR-166-Serbia project: "Redesigning the exploitation of small grains genetic resources towards increased sustainability of grain-value chain and improved farmers' livelihoods in Serbia and Bulgaria-GRAINEFIT".

## INFLUENCE OF BREWERS SPENT GRAIN ON OXIDATIVE STABILITY OF COOKED MEATBALLS

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Meat substitutes are non-meat ingredients with a high protein content that can have a beneficial effect on some product characteristics such as water-holding capacity (WHC), protein content, sensory characteristics, and shelf life. The objective of this study was to determine the influence of brewers spent grain (BSG), by-product of the beer industry, as a meat substitute on oxidative stability of cooked meatballs during cold storage. Meatballs, made from pork meat and solid fatty tissue, were divided in three groups – control (C), and experimental samples BSG3 and BSG6 which contained 3% and 6% of BSG, as meat replacement, respectively. All samples were cooked in the oven (until reaching 72°C) and stored at 4°C for 5 days. BSG had positive influence on protein content, since there was significant difference ( $P < 0.05$ ) between C (19.23%) and BSG6 (19.65%). Cooking loss decreased with the increasing percentage of BSG meat replacement. Instrumental colour measurements showed that during storage period the highest difference in colour ( $\Delta E$ ) was noted for sample C followed by BSG6 and BSG3 (without significant differences between BSG6 and BSG3). TBARs test showed that sample C had the lowest (2.93 mgMDA/kg) and sample BSG6 had the best (0.57 mgMDA/kg) oxidative stability on the fifth day of analysis. Sample C has the lowest oxidative stability during all days of storage, too. Also, difference in thiol content between sample groups and C was significant, where C has the highest value at the end of storing period (55.49 nmol thiol/mg protein). Obtained results may indicate the antioxidant potential of BSG, what can be attributed to different phenolic compounds. Hence, sensory analysis showed that BSG3 has the best scores for colour, taste, juiciness and overall acceptability at the end of storage period.

**Keywords:** meatballs, brewers spent grain, meat substitute

**Acknowledgements** This study was part of the Project No. 451-03-65/2024-03/200134, which is financially supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia.



## PROCESSED MEAT WASTEWATER VALORIZATION: A SUSTAINABLE BIOREFINERY APPROACH FOR FUNCTIONAL PROTEIN AND PEPTIDES

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Processed meat wastewaters contain high levels of organic matter, such as proteins and peptides. This makes them a potentially valuable resource, as these organic constituents can be extracted to derive high-value bioactive compounds. Innovative methods of extraction and purification of these valuable compounds from wastewater become a possibility to increase the economic and value-added of these by-products while reducing their environmental impact. The goal of this research was to develop an integrated, laboratory-scale membrane filtration process to recover valuable fractions from salted pork ham brine wastewater, thereby contributing to the bio-refinery approach for sustainability. The research strategy involved a multi-stage membrane filtration process using microfiltration (MF) and ultrafiltration (UF) of the pretreated, salted pork ham brine wastewater. The wastewater was treated with citric acid, both with and without heat, before the membrane filtration steps. This integrated membrane filtration approach yielded fractions rich in proteins and peptides. These protein and peptide fractions could be used in food, pharmaceutical, or cosmetic applications, adding significant value compared to simply disposing of the wastewater. Notably, the UF permeates and retentates, coupled with the MF step, resulted in fractions with the best bioactive properties, particularly enhanced antioxidant capacity (ABTS and ORAC). In addition to these beneficial bioactive attributes, these fractions displayed favorable functional properties such as high solubility, improved water and fat binding capacity, and enhanced emulsifying and foaming properties. The bioactive and functional properties of these recovered by-product fractions make them highly valuable for incorporation into a diverse array of commercial products. This study shows that meat processing by-products can be a valuable source of functional and bioactive compounds. Extracting these high-value ingredients from wastewater has the potential to improve sustainability and profitability in the processed meat industry by reducing environmental impact and generating new revenue streams.

**Keywords:** Meat industry by-products, Bioactive compound extraction, Membrane technology, Bioactive properties, Functional properties

**Acknowledgments:** The authors would like to acknowledge the Technological University Dublin Researcher Award, 2023, for the PhD grant. This research was also funded by the Department of Agriculture, Food, and the Marine (DAFM) and CircoVal projects.



## PHYTOCHEMICAL COMPOSITION AND ANTIOXIDANT PROPERTIES OF ENCAPSULATED POWDERS OF RED BEET MICROGREEN JUICE WITHIN CARBOHYDRATE CARRIERS

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Red beet microgreen juice is a rich source of bioactive compounds with health-promoting properties that are important for human diets. It may be considered a healthy beverage due to its high antioxidant activity related to phenolic content. However, these compounds are sensitive and easily degradable, while red beet juice has an unpleasant, astringent, and earthy taste. The encapsulation process has recently been successfully used to protect bioactive compounds from harmful environmental influences and to mask the unpleasant taste of juices. This study aimed to encapsulate red beet microgreen juice within maltodextrin and inulin using spray-drying and to characterize the obtained encapsulates in terms of phenolic compounds and antioxidant properties. Total phenolic content (TPC) and antioxidant activity (AA) (ABTS<sup>•+</sup> and FRAP) were determined by spectrophotometric methods, and phenolic compounds were assessed using an ultra-high performance liquid chromatography (UHPLC) system coupled with quadrupole time-of-flight mass spectrometry (Q-ToFMS). The TPC, AA, and phenolic compounds were expressed in mg equivalents of gallic acid, Trolox, gentisic acid, coumaric acid, and apigenin, respectively per 100 g of the encapsulates. Considering the results obtained, both encapsulates from red beet microgreen juice exhibited a high content of phenolic compounds, including various phenolic acid and apigenin derivatives, as well as good antioxidant activity. In our study, the encapsulates with inulin had a higher TPC than those with maltodextrin. There was a similar trend for antioxidant activity wherein the encapsulates with inulin showed stronger antioxidant activity determined by ABTS<sup>•+</sup> and FRAP assays than those with maltodextrin. Concerning the results of the UHPLC Q-ToF MS analysis, several phenolic acid derivatives such as hydroxybenzoic acid, hydroxybenzoic acid hexoside, and dihydroxybenzoic acid pentoside were detected in the encapsulates, while various apigenin C-glycoside derivatives predominated among the flavonoids. In summary, the obtained encapsulates can be used as potential functional additives.

**Keywords:** encapsulation, spray-drying, inulin, maltodextrin, antioxidant activity, phenolics

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant number 451-03-65/2024-03/200116).

## FATTY ACIDS AND ELEMENTAL PROFILING OF NEW SERBIAN VARIETIES OF OIL RAPE, SAFFLOWER, AND MUSTARD SEEDS

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The growing interest in oil plants for human and animal nutrition and various industries has driven the diversification and development of new oil crop varieties. This diversification not only improves resilience to pests and climatic variations but also enhances nutritional quality. To ensure the commercial viability of these newly developed oilseeds, it is crucial to characterize their nutritional and chemical profiles comprehensively. This study evaluates two varieties of safflower and both white and black mustard as alternative oilseed crops, focusing on their fatty acid and mineral compositions, compared to two genotypes of rapeseeds, a traditional oil crop. The analysis revealed that the oilseed samples had a high content of unsaturated fatty acids, ranging from 86.42% to 95.08%, and a low content of saturated fatty acids, ranging from 4.69% to 13.56%. Among the unsaturated fatty acids, monounsaturated oleic acid (C18:1c) was the most abundant, with contents of 62.63% and 67.55% in certain varieties, followed by essential polyunsaturated linoleic acid (18:2n6) and  $\alpha$ -linolenic acid (18:3n3). Linoleic acid constituted about 20%, while  $\alpha$ -linolenic acid was present in smaller amounts, ranging from 5% to 7% of total fatty acids. All analysed samples contained significant levels of essential minerals for human nutrition. The predominant elements were phosphorus, potassium, calcium, and magnesium, in descending order. Additionally, these oilseed crops were rich in microelements such as iron, copper, manganese, and zinc. The new varieties of oil rape, safflower, and mustard seeds demonstrate promising nutritional profiles with high unsaturated fatty acid content and valuable mineral compositions. These findings suggest that safflower and mustard crops can be viable alternatives to traditional oil crops, offering substantial nutritional benefits and the potential for increased oil crop diversity.

**Keywords:** fatty acids, mineral composition, oilseeds varieties, crop development

**Acknowledgements:** This research was supported by Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant numbers 451-03-66/2024-03/200032 and 451-03-65/2024-03/200134.

## ENHANCING SUNFLOWER SEED QUALITY: INSIGHTS FROM KERNEL MORPHOLOGY AND CHEMICAL COMPOSITION ANALYSIS

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Sunflower seeds are vital agricultural commodities, esteemed for their nutritional and industrial value. Despite the diversity of sunflower seed varieties, they are frequently harvested without segregation based on quality and morphological traits, resulting in inconsistent outcomes and diminished product quality and efficiency. This limitation impedes the advancement of the sunflower seed processing industry. Nutritional composition indicators, such as oil content, protein levels, and oleic acid content are crucial in determining the quality and commercial value of sunflower seeds. Concurrently, morphological traits, including kernel size, filling, and pericarp thickness, are essential for assessing yield quality in sunflower production. An overemphasis on either aspect can lead to suboptimal breeding outcomes, with seeds exhibiting excellent nutritional qualities but lacking in physical attributes, or vice versa. Given the importance of both morphological and nutritional properties for agricultural and industrial applications, it is imperative to integrate these indicators comprehensively into breeding programs. This holistic approach ensures the development of sunflower varieties that meet high standards for both physical performance and nutritional value. Examining the correlation between kernel morphology and seed quality can significantly enhance breeding strategies, leading to higher yields, improved product consistency, and greater overall efficiency in the sunflower seed processing industry. This study aimed to analyze the oil, protein, and oleic fatty acid content, as well as assess the physical properties of 20 sunflower seed genotypes cultivated in Serbia. Oil content was determined using the ISO 659, 2009 standard method, while total seed protein content was measured by the Dumas procedure. The oleic content of cold pressed sunflower oils was analyzed via gas chromatography following ISO methods 12966-4:2015. The physical properties test included 50 randomly selected achenes per genotype, with transverse sections of the pericarp taken from the central part of the seed. Morphological measurements were made using calipers, while pericarp observations were conducted with a light microscope. The oil and protein contents exhibited variations, averaging 42.03% and 20.26%, respectively. The oleic acid content in the analyzed oil samples ranged from 11.01% to 62.36%. The results indicated that oil concentration in seeds is negatively correlated with pericarp thickness, but positively correlated with seed thickness. Additionally, oil concentration showed a negative correlation with both the protein content and oleic acid concentration in the seed. In conclusion, integrating morphological traits and nutritional composition indicators in sunflower breeding is essential for improving seed quality and efficiency in processing, as demonstrated by correlations between oil content, pericarp thickness, and seed morphology in this study.

**Keywords:** sunflower seed, protein and oil content, morphology, chemical composition

**Acknowledgements:** This work was carried out as collaborative activities of the Institute of Field and Vegetable Crops, Novi Sad, Serbia and University of Novi Sad, Faculty of Technology Novi Sad, Novi Sad, Serbia. This research was supported by Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant numbers 451-03-66/2024-03/200032 and 451-03-65/2024-03/200134.

## THE INFLUENCE OF HEAT TREATMENT ON THE COMPOSITION OF AMINO ACIDS AND FATTY ACIDS IN THE MEAT OF THE INVASIVE CRAYFISH *FAXONIUS LIMOSUS*

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The spiny-cheek crayfish (*Faxonius limosus*, *Rafinesque*, 1817) is one of the most important aquatic invaders in European inland waters and has been recorded in more than 20 European countries. From the end of the 19th century until now, it has become the most widespread non-indigenous crayfish species in Europe and has been included in the list of Invasive Alien Species (IAS) of universal concern within EU regulations. The aim of the present study was to analyze the effect of heat treatment on chemical composition, profile of amino acids and fatty acids of the meat of the invasive crayfish *Faxonius limosus*. The content of protein, moisture and crude fat in the fresh and thermally treated meat was ranged from 17.92% to 17.39%, 79.92% to 80.81%, and 0.23% to 0.25%, respectively. Fresh crayfish meat was of high nutritional quality with an amount of essential amino acids of 7.18 g/100g in sample. A slight decrease in the content of amino acids threonine, phenylalanine and histidine was observed in thermally treated meat. On the other hand, the crayfish meat was characterized by high levels of essential polyunsaturated fatty acids (PUFA), particularly n-3 PUFA, at an optimal ratio of n-3/n-6 and with low values of atherogenic and thrombogenic indices. The results clearly demonstrate that the meat from crayfish has a high nutritional value and has a good potential to serve as a new food source.

**Keywords:** spiny-cheek invasive crayfish *Faxonius limosus*, nutritional quality, amino acids, fatty acids

**Acknowledgements:** This research was supported by the Science Fund of Republic Serbia, #GRANT No. 7417. "Reducing the negative impact of invasive crayfish *Faxonius limosus* in the Danube by smart exploitation of their meat and shells" DANUBECare.

## WHEAT BRAN – ALTERNATIVE PROTEIN SOURCE

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Wheat bran, a byproduct of wheat milling, has getting increasing attention as an alternative protein source due to its nutritional value, sustainability, and cost-effectiveness. With a protein content of up to 18%, wheat bran offers a compelling solution to the growing demand for protein in both human and animal nutrition. Utilizing wheat bran as a protein source can also contribute to waste reduction and the efficient use of agricultural by-products. The isolation of protein from wheat bran has been performed via pH precipitation and has demonstrated significant potential as a sustainable and efficient method. This technique yields more than 9% of protein with a protein content exceeding 80%. The isolated wheat bran protein powder (WBPP) is rich in glutamic acid and arginine, but more importantly contains all essential amino acids. The limiting one is lysine, but it is present in higher amount than in wheat endosperm protein (4.0 g/100 g). Furthermore, the digestibility of it exceeds 93%, making it highly suitable for nutritional applications. The utilization of WBPP in food applications underscores its potential to improve the nutritional profile of everyday foods and promoting better health outcomes. Like other plant proteins, the addition of WBPP can affect the techno-functional and sensory properties of foods. Especially bitterness is main sensory defect caused by WBPP addition. However, the development of the recipe and the modification of WBPP properties led to the successful production of high-protein pastries and yogurts of acceptable quality.

**Keywords:** Wheat bran protein, nutrition, high-protein food, sustainability

**Acknowledgements:** This research was supported by project no. FCH–S-23-8330 financed by the Ministry of Education, Youth and Sports of the Czech Republic.

## *Nutrition & Health*

## OPTIMIZING TEFF SOURDOUGH IN RYE BREAD TO ENHANCE QUALITY AND NUTRITIONAL VALUE

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The aim of the work was to select a recipe and conditions for making teff sourdough, selecting a recipe for rye bread made with teff sourdough and assessing the quality of the obtained bread. Rye bread with teff flour in amounts of 5, 10 and 15% was made using a two-phase method using teff sourdough. The control sample was 100% rye bread. In rye flours type 580 and 720, as well as in ground teff, the following parameters were determined: moisture, color, protein, ash and total dietary fiber content. The pH value, titratable acidity and organoleptic assessment were measured in the prepared sourdoughs. The sourdough prepared using LV1 starter cultures with a capacity of 250 was selected for baking bread. The following tests were carried out on the baked breads: volume, crumb moisture, crust and crumb color, organoleptic assessment, Instrumental Texture Profile Analysis Test, protein and ash content, as well as total dietary fiber content. The share of teff in rye bread in the form of sourdough in an amount of up to 15% had a positive effect on its quality characteristics and health-promoting value by increasing the weight of the bread, volume, moisture and improving the taste of the bread, as well as increasing the content of protein, ash and total dietary fiber. There was no negative impact of the addition of teff on the quality of the dough and the attractiveness and general acceptability of the resulting bread.

**Keywords:** teff, sourdough, rye bread, quality, nutritive value

## IMPACT OF ADDING LYOPHILIZED FRUITS ON THE ANTIMICROBIAL AND ANTIPROLIFERATIVE PROPERTIES OF RAPESEED HONEY

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Historically, honey and other bee products have been valued as religious symbols, nutritional sources, and medicinal substances, recognized as some of the earliest functional foods. While honey primarily consists of carbohydrates, water, and proteins, it also contains beneficial components such as amino acids, organic acids, vitamins, and polyphenols. Modern research highlights honey's antioxidant, antibacterial, anti-inflammatory, and antitumor properties. Enriching honey with fruits enhances its health benefits, flavor, and stability, making it suitable for functional food applications. This study aimed to evaluate the impact of adding lyophilized fruits to rapeseed honey on its antimicrobial and antiproliferative activities. Seven formulations of rapeseed honey, enriched with 10% lyophilized fruits (sour cherry, strawberry, blueberry, raspberry, blackberry, orange, and pineapple), were compared to a control sample of rapeseed honey to assess differences in these functional properties. Given that rapeseed honey is generally known for its lower antibacterial potential compared to other honey types, it was anticipated that enriching it with fruit lyophilizates would enhance its antibacterial activity due to increased polyphenol content. The results indicated that all honey-based products, particularly those enriched with lyophilized berry fruits, exhibited antibacterial properties, with significant efficacy observed against gram-positive bacteria, specifically *Staphylococcus epidermidis* and *Staphylococcus aureus*. The antiproliferative activity of the honey and honey-based products was tested using human solid tumor cell lines, including estrogen receptor-positive (ER+) breast adenocarcinoma MCF-7, cervix carcinoma HT-29, human cervix adenocarcinoma HeLa, and normal fetal lung fibroblast MRC-5. Honey-based products enriched with pineapple and blueberry lyophilizates showed the highest antiproliferative activity against breast (MCF-7), cervix (HeLa), and colon (HT-29) cancer cell lines. Rapeseed honey exhibited the lowest activity, consistent with earlier studies. The notable effectiveness of pineapple-enriched honey, despite its lower polyphenol content, suggests that additional bioactive compounds, such as bromelain, play a significant role in its antitumor activity. The study demonstrated that adding lyophilized fruits to rapeseed honey enhances its antimicrobial and antiproliferative activities, underscoring the increased health benefits of fruit-enriched honey.

**Keywords:** rapeseed honey, honey-based products, antibacterial activity, antiproliferative activity

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).



## CHANGES IN BIOACTIVE COMPOUNDS STABILITY AND COLOUR OF FUNCTIONAL PLUM SPREAD DURING DIFFERENT STORAGE CONDITIONS

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There is growing interest in utilizing juice pomace, including plum pomace, in new food products as a sustainable strategy for reducing waste and providing an alternative source of dietary fibers and phenolic bioactives in the functional food. Some phenolics, such as anthocyanins are more sensitive and susceptible to degradation processes in fruit preparation during storage. This study aimed to assess the effect of a storage period and temperature on the retention of phenolics, anthocyanins (total and monomeric) and colour of functional plum spread enriched with plum pomace. The spreads were stored at 4 °C (14 days) and at 20 or 40 °C for 14, 28 and 45 days. Control sample, was also subjected to analysis one day after production and obtained measurements for investigated spreads were compared. Results revealed that total phenolics remained more stable than total anthocyanins and total monomeric anthocyanins at room temperature, while a temperature of 40 °C led to the loss of all bioactives. Colour deterioration was observed in all samples, with the least change at 4 °C. Refrigeration of spread was found to significantly slow down the loss of phenolics and anthocyanins and better preserve colour.

**Keywords:** functional plum spread, plum pomace, phenolic bioactive compounds, anthocyanins, colour, storage conditions

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia under the Agreements on the Implementation and Financing of Research (nos. 451-03-66/2024-03/200222).

## NUTRACEUTICAL POTENTIAL OF EDIBLE MUSHROOMS: ANALYSIS OF PHENOLIC COMPOUNDS AND THEIR ANTIPROLIFERATIVE EFFECTS ON MCF-7 CELLS

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Edible mushrooms serve as a valuable source for the development of nutraceuticals and pharmaceuticals with notable anti-tumor, antioxidant, and antimicrobial properties. In addition to their therapeutic potential, mushrooms are an integral component of the human diet, characterized by low fat, high protein, and low caloric content. This study investigates the impact of total phenols (TP) and flavonoids (TF) on the cytotoxic activity of ethanol (EtOH) and water (Aq) crude extracts from various mushroom species (*Meripilus giganteus* - Mer, *Laetiporus sulphureus* - Ls, *Auricularia auricula-judae* - Au, *Fistulina hepatica* - Fh, *Armillaria mellea* - Am, *Coprinellus disseminates* - Cd, *Lactarius controversus* - Lc, *Lycoperdon perlatum* - Lp, *Macrolepiota procera* - Mp) collected in Eastern Serbia (Sikole, near Negotin) against the MCF-7 breast cancer cell line, which is positive for estrogen and progesterone receptors. The TP content in the examined extracts ranged from 16.68 mg GK/g (Aq Lc) to 106.33 mg GK/g (EtOH Mer), while the TF content varied from 0.27 mg KV/g (Aq Am) to 18.0 mg KV/g (EtOH Au). Cytotoxic activity assessments indicated that all mushroom extracts exhibited antiproliferative effects, with the Aq extract of Mp demonstrating the highest activity (IC<sub>50</sub> 144.53 µg/ml) after 24 hours. Among EtOH extracts, Mer showed the best activity (IC<sub>50</sub> 155.02 µg/ml) after 72 hours. A significant negative correlation between TP content and cytotoxicity was observed in the EtOH extracts of Cd ( $r^2 = 0.99$  after 24 hours;  $r^2 = 0.97$  after 72 hours), as well as in the Aq extracts of Mp ( $r^2 = 0.99$  after 24 hours;  $r^2 = 0.98$  after 72 hours) and Fh ( $r^2 = 0.97$  after 24 hours;  $r^2 = 0.95$  after 72 hours). High correlations ( $r^2 = 0.99$ ,  $p < 0.05$ ) between flavonoid content and cytotoxic activity were noted in the EtOH extracts of Fh and Cd in both incubation periods, and in the 24-hour treatment of Mp and Au extracts. For the 72-hour treatment, significant correlations were also observed in the Lp, Lc, and Mer extracts, along with the Aq extracts of Cd and EtOH of Fh showing negative correlations. Our data highlight the significant influence of TP and TF content on the cytotoxic activity of the examined mushroom extracts against the MCF-7 breast cancer cell line.

**Keywords:** mushrooms, phenols, cytotoxic, MCF-7

**Acknowledgments** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## PREPARATION OF LOW MOLAR MASS OAT $\beta$ -GLUCAN SPECIMEN FOR DIETARY INTERVENTION IN THE PORCINE MODEL OF COLITIS

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The incidence of inflammatory bowel diseases, is steadily increasing worldwide, particularly affecting children and young adults. Safe therapies are being sought, and one of them is the protective effect of oat  $\beta$ -glucans, which has been widely documented in the rat animal model of colitis. This study investigates the preparation and therapeutic potential of low molar mass oat  $\beta$ -glucan (OBG) obtained by alkaline water extraction, with particular focus on its effects in a porcine model of 2,4-dinitrobenzenesulfonic acid (DNBS) induced colitis. The gastrointestinal tract of the domestic pig is physiologically similar to that of humans, making it an ideal model for studying human colitis. The main purpose of wet alkaline extraction was to obtain isolated and purified OBG fraction. The steps in the wet alkaline isolation of OBG include freezing of oat bran, grinding and hydrolysis, extraction and enzymatic treatment to remove residues. This isolated OBG, purified of bioactive substances, reduces the number of possible actions that adversely affect colonic inflammation. The 5% OBG solution was prepared by mixing OBG powder with ultrapure water. The solution was heated and mixed to a homogeneous consistency and then sterilized in an autoclave. The OBG solution at 37°C was administered to pigs via an intragastric cannula at two doses corresponding to 100 or 200 mg OBG/kg BW. Our preclinical study showed that this dietary intervention was safe for the animals and reduced colitis symptoms.

**Keywords:** oat  $\beta$ -glucan, alkaline extraction, porcine colitis model, DNBS

**Acknowledgements:** The study was funded by the National Science Centre in Poland, project No 2022/47/B/NZ9/03123.

## NUTRITIONAL AND PHENOLIC PROFILE ANALYSIS OF YELLOW KIWIFRUIT PEEL AND BAGASSE (*ACTINIDIA CHINENSIS*)

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Kiwifruit (*Actinidia spp.*), and particularly yellow or golden kiwifruit (*Actinidia chinensis*), has gained worldwide popularity due to its excellent taste and high vitamin C content. However, its commercialization generates a considerable amount of waste, mainly in the form of peels and bagasse, which poses a significant environmental challenge. This study focuses on a detailed analysis of the nutritional and chemical composition of yellow kiwifruit peel (YKS) and yellow kiwifruit bagasse (YKB). The nutritional composition was performed following AOAC international standards, while heat-assisted extraction (HAE) was used to extract the bioactive compounds present in both samples (YKS and YKB), followed by their identification and quantification by HPLC-ESI-QqQ-MS/MS. The samples were analyzed for the presence of proteins, lipids and their fatty acid profile and mineral content, as well as bioactive compounds such as polyphenols. The results of the study suggest that YKS and YKB are a rich source of nutrients, with YKB containing 6.12 g/100 dw of protein and 5.72 g/100 dw of lipids, of which 35% corresponds to polyunsaturated fatty acids (PUFAs). While YKS stands out for its high mineral content, including K<sup>+</sup>, P<sup>+</sup>, and Fe<sup>2+</sup>. From a chemical point of view, three main groups of compounds stand out: YKS contains 42.5% phenolic acids, 31% flavonoids, and 26.3% of other polyphenols, while YKB contains 60% phenolic acids, 16% flavonoids, and 23.9% of other polyphenols. These data demonstrate that YKS and YKB can become valuable resources for various industries, such as food, cosmetics, and pharmaceuticals. This work also supports the principles of the circular economy, by promoting the sustainable reuse of waste, reducing environmental impact and improving food safety by incorporating by-products into the production chain.

**Keywords:** *Actinidia chinensis*, waste valorization, nutritional analysis, bioactive compounds, heat-assisted extraction

**Acknowledgements:** The research leading to these results has been funded by Xunta de Galicia for supporting the postdoctoral fellowship of A.G. Pereira (IN606B-2024/011) and the predoctoral fellowship of P. Barciela (ED481A-2024-230). The authors gratefully acknowledge national funding from the FCT, Foundation for Science and Technology, through the individual research fellowships of J. Echave (2023.04987.BD) and A.O.S. Jorge (2023.00981.BD).

## ANTIMICROBIAL EFFECTS OF POLYPHENOLS FROM APPLE AND CARROT POMACE

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The pig farming industry faces significant challenges during the weaning period, often resulting in reduced growth rates and higher mortality among piglets. Traditionally, antibiotics and zinc oxide have been used to manage these issues. However, concerns about antibiotic resistance and environmental pollution have led to strict European regulations limiting or banning their use. This has created an urgent need for alternative solutions, with polyphenols emerging as promising candidates due to their bioactive properties, including anti-inflammatory, antioxidant, and antimicrobial effects. These properties are particularly important for preventing weaning-related problems, which are frequently caused by the *E. coli* F4 (K88) bacterium. This study focused on evaluating the antimicrobial properties of polyphenols extracted from apple pomace and carrot pomace, both in their unfermented form and after fermentation with *S. cerevisiae*. The antimicrobial activity was assessed *in vitro* by monitoring bacterial growth through absorbance measurements at different intervals over 24 hours. The results indicated that polyphenols possess significant antimicrobial effects in both their unfermented and fermented states. The extent of bacterial inhibition was influenced by the concentration of polyphenols and the specific types of polyphenols present in the extract. Notably, despite having a lower overall concentration, the fermented polyphenol extracts achieved similar levels of bacterial growth inhibition as the unfermented extracts at higher concentrations. These findings highlight the potential of agro-industrial by-products such as apple and carrot pomace to provide bioactive compounds that could serve as effective alternatives to antibiotics and zinc oxide in livestock farming. By exploiting these natural resources, the industry might move towards more sustainable and environmentally friendly practices while still effectively managing the challenges associated with weaning in piglets.

**Keywords:** weaning, antimicrobial, agro-industrial by-products, polyphenols, *E. coli*

**Acknowledgements:** This work was supported by a grant of the Romanian Ministry of Research and Innovation-UEFISCDI, project number PN-23.20.02.01.

## INFLUENCE OF ARTIFICIAL SWEETENERS SODIUM SACCHARIN AND ACESULFAME POTASSIUM ON THE TASTE, HYDRATION PROPERTIES AND SOLUBILITY OF CAFFEINE

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Recent years have witnessed a surge in the consumption of caffeinated beverages and products worldwide. Caffeine, a widely used psychoactive compound found in coffee, tea, energy drinks, and more, mimics adenosine in the human body, promoting wakefulness by binding to adenosine receptors. This trend parallels the increasing prevalence of artificial sweeteners, favored for their intense sweetness and low caloric content, catering to diabetic and obese populations. Despite ongoing debates about their safety, artificial sweeteners continue to gain market share, often in conjunction with caffeinated beverages. This study focuses on exploring the interaction between caffeine and two aqueous solutions of artificial sweeteners: sodium saccharin and acesulfame potassium. By measuring solubility and physicochemical parameters such as density, sound velocity, and viscosity, it aims to elucidate how these additives affect taste and predict the overall characteristics of these complex systems. Investigations into such systems not only provide essential data for understanding their behavior but also pave the way for predicting taste preferences based on their physicochemical profiles. For instance, the hydration properties and apparent specific molar volume significantly influence their taste perception. The findings from this research contribute to a deeper understanding of how caffeine interacts with artificial sweeteners in beverages and products, offering insights into their combined effects on consumer health and preference. The received results indicate that both artificial sweeteners reduce the bitterness of caffeine, promote self-aggregation and increase caffeine solubility.

**Keywords:** caffeine, artificial sweeteners, taste, solubility, hydration

**Acknowledgements:** The authors would like to gratefully acknowledge the financial support of the Serbian Ministry of Science, Technological Development and Innovation for the (Scholarship for A.V.) and the Secretariat for Higher Education and Scientific Research of the Autonomous Province of Vojvodina, Serbia (Grant No. 142-451-2545/2021-01/2).

## HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC) METHOD FOR THE INVESTIGATION OF BIOACTIVE COMPOUNDS IN TWO MICROGREEN VARIETIES

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Nowadays, we are facing numerous challenges related to nutrition and human health. Pathological conditions caused by inadequate nutrition are particularly prevalent in urban areas and developed countries. The daily diet of the modern population is characterized by insufficient intake of minerals, nutrients, vitamins, and phytochemicals. It is estimated that by 2050, 80% of the world's food will be consumed in cities, which emphasizes the importance of plant production near urban areas and within urban environments. One of the potential sources of a large number of bioactive compounds are microgreens. They are known for their nutritional value and bioactivity and are used in culinary arts for their flavor, texture, and visual appeal. Microgreens are young plants harvested after the first true leaves develop from seeds of vegetables, herbs, and industrial plants, easily grown indoors. They are known for their nutritional value and are used in culinary arts for their flavor, texture, and visual appeal. This research investigates the bioactive content of two Brassicaceae microgreen plants (Sango radish and kale) using HPLC with a DAD/UV detector, including the content of phenolic acids, carotenoids, and vitamin C. Microgreens in question were lyophilized, homogenized as a powder and stored until further analyses. Specific extractions were performed for different assays. Sango radish and kale showed the highest presence of p-hydroxybenzoic acid (510.70mg/100g), and sinapic acid (600mg/100g), respectively.  $\alpha$ - and  $\beta$ - carotene and lutein were found in both microgreen samples and zeaxanthin was only present in kale. While lutein was the dominant carotenoid in Sango radish (511.23 mg/100g), in kale the highest carotenoid content amounted to  $\alpha$ -carotene (1238.68 mg/100g). Ascorbic acid was three times higher in kale compared to radish microgreens (21.31- 66.05 mg/100g). The obtained results highlight the health-promoting properties of investigated microgreens and the importance of their daily intake and expanded future use.

**Keywords:** HPLC, microgreens, phenolics, carotenoids, ascorbic acid

**Acknowledgements:** This research study was funded by the Provincial Secretariat for Higher Education and Scientific Research of Autonomous Province Vojvodina (Serbia) within the project "New ecological phytopreparation for antifungal and antioxidant treatment of selected fruits and vegetables—EcoPhyt" (grant No. 142-451-2732/2023-01/01) and by the Ministry of Education, Science, and Technological Development of the Republic of Serbia (grant No. 451-03-66/2024-03/200134).



## ASSESSMENT OF OLEOGEL AND OLEOGEL EMULSION AS FUNCTIONAL BEEF FAT REPLACERS IN TURKEY MEAT PÂTÉ FORMULATIONS

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Consumer interest in healthier food options is increasing, with individuals seeking balanced and nutritious alternatives. In response to this trend, replacing beef fat in meat products—an integral component of the diet—with structured, healthier plant oils has gained prominence. This study aimed to develop turkey meat pâté by replacing 25% of beef fat (C) with 50% oleogel (OL50), oleogel emulsion (OLE50), 100% oleogel (OL100), or oleogel emulsion (OLE100). Oleogel (O) was prepared with carnauba wax and extra virgin olive oil, while oleogel emulsion (OE) consisted of carnauba wax and extra virgin olive oil in the oil phase, and xanthan gum and salt in the water phase. The study assessed the chemical composition, emulsion stability, water-holding capacity (WHC), peroxide value, and thiobarbituric acid reactive substances (TBARS). Results indicated no significant differences in moisture content ( $p > 0.05$ ). The fat content in pâté samples decreased with the addition of OE. Emulsion stability was lower in treatments using O as a 50% beef fat replacer ( $p < 0.05$ ). In contrast, stability was higher in samples containing OE compared to the control group, regardless of the amount ( $p < 0.05$ ). The control (C) exhibited the lowest WHC ( $p < 0.05$ ). Increasing oleogel levels reduced WHC; however, the highest WHC was achieved with a higher rate of OE addition. The lowest peroxide and TBARS values were observed in samples where beef fat was completely replaced with OE ( $p < 0.05$ ). Sensory analysis revealed that the pâté with 50% beef fat replacement with oleogel was the most preferred in terms of appearance, color, texture, flavor, rancidity, and overall acceptance ( $p < 0.05$ ). Overall, these findings suggest that oleogel and oleogel emulsion are viable alternatives to beef fat, providing healthier options without compromising the quality of turkey meat pâté.

**Keywords:** turkey meat pâté, beef fat replacement, oleogel, carnauba wax, oxidation

**Acknowledgements:** The authors are thankful for the financial support of The Scientific and Technological Research Council of Turkey under project number: 1919B012327105.



## INFLUENCE OF THE FLOUR BLENDS ON THE TEXTURE OF RYE PASTA RICH IN OMEGA-3 FATTY ACIDS

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Producers in many countries often use cheaper common wheat flour/semolina instead of durum semolina, whose specific cultivation requirements and lower yield than common wheat significantly increase its price. However, pasta made from common wheat flour/semolina or from blends of common wheat flour/semolina with durum semolina is considered to be of lower quality than pasta from durum semolina, which is undoubtedly a product with the best quality parameters. The main purpose of this research was to examine the texture (hardness and adhesiveness) of pasta prepared from whole grain rye flour and durum semolina, one group of samples and whole grain rye flour and wheat semolina without and with the addition of rapeseed oil powder. Rapeseed oil powder is added as a raw material which is a source of alpha-linolenic acid. Durum semolina creates a firmer pasta with less stickiness (hardness 13858 g, adhesiveness 320 g\*sec) while wheat semolina creates a less hardness pasta (11405 g) with more adhesiveness (572 g\*sec). The addition of rapeseed oil powder in both samples does not significantly impair the pasta texture properties. The sample paste from whole grain re flour and durum semolina with canola oil powder has the following values for hardness of 13597, and adhesiveness of 505 g\*sec, and the sample pasta from whole grain rye flour and wheat semolina has a value for hardness of 11585 g and adhesiveness 759 g\*sec. The results of these studies indicate that semolina durum can be fully replaced by common wheat flour, which can significantly reduce production costs in countries where access to durum wheat is limited.

**Keywords:** pasta, whole grain rye flour, wheat semolina, durum semolina, rapeseed oil powder

**Acknowledgements:** This research was supported by the Innovation Fund of the Republic of Serbia, Innovation vouchers No. 1505 and Ministry of Science Technological Development and Innovation of the Republic of Serbia, No. 451-03-66/2024-01/200222.

## PASTA WITH RYE FLOUR AND RICH IN OMEGA-3 FATTY ACIDS

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Traditionally, pasta is manufactured from durum wheat (*Triticum durum* D.) which results in a product considered to be of superior quality to pasta made from common wheat (*Triticum aestivum* L.). Wholemeal rye flour can be considered a good candidate for pasta fortification due to its health benefits. Nowadays, its use is limited mainly as a result of the problems arising from its pentosan and water-soluble proteins and with problems about its flavour; rye flavour, is perceived as bitter and intense. Preliminary research determined that the optimal ratio of wheat semolina to whole grain rye flour is 70% to 30% (w/w). Scientific literature documents the fortification of pasta dough with unconventional ingredients. Rapeseed oil, rich in unsaturated fats such as omega-3 and omega-6 fatty acids, is one such ingredient. It has been demonstrated the use of rapeseed oil as a margarine substitute in biscuits, enhancing their nutritional quality and antioxidant properties. However, the industrial application of rapeseed oil in its liquid form remains impractical. The work aims to formulate pasta rich in omega-3 fatty acids with the addition of wholemeal rye flour in the amount of 30% and rapeseed oil powder. The rapeseed oil powder sample had 69.1 mg/g of alpha-linolenic acid (ALA). With the addition of 10% ALA powder to the flour mixture, the content of 0.615 g ALA in 100 g of dry pasta was achieved that fulfills the requirement for the nutritional statement "rich in omega-3 fatty acids". The content of ALA in cooked pasta is 0.303 g/100 d.m. so it can be considered as a source of omega-3 fatty acids. The addition of rapeseed oil powder did not affect the sensorial quality, as well as other pasta physical characteristics.

**Keywords:** pasta, rye flour, rapeseed oil powder,  $\omega$ -3 fatty acids

**Acknowledgements:** This research was supported by the Innovation Fund of the Republic of Serbia, Innovation vouchers No. 1505 and the Ministry of Science Technological Development and Innovation of the Republic of Serbia, No. 451-03-66/2024-01/200222.

## POLYPHENOLIC PROFILE OF RAPESEED PRESS CAKE: KEY COMPOUNDS AND THEIR FUNCTION IN IMPROVING PRODUCT VALUE

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Polyphenols have gained considerable attention and market potential due to their health-promoting effects. In rapeseed, particularly within the press cake remaining after oil extraction, significantly higher concentrations of polyphenols are found compared to other oilseeds. Although polyphenolic compounds can be highly valuable, they also pose significant challenges in further processing. The production of food-grade proteins from rapeseed press cake is very demanding, mainly due to the intense bitter off-taste and dark color associated with polyphenolic compounds, which limit palatability for human consumption. Therefore, the aim of this study was to determine the polyphenolic profile of rapeseed press cake obtained from cold-press oil production of domestic Serbian rapeseed varieties and to evaluate potential strategies to improve the protein isolation process. The results of the HPLC-DAD analysis of the polyphenolic profile of the analyzed rapeseed press cake showed that sinapine and phenolic acids, in both free and esterified forms, were the most dominant. Although the concentration of these compounds depends on rapeseed processing, growing conditions, extraction parameters, and various other factors, sinapine constituted 73% of the total phenolic compounds, which is in accordance with previously published results. Among the phenolic acids in free form, the most dominant were caffeic, gallic, protocatechuic, 4-hydroxybenzoic, syringic, and sinapic acids. Considering that sinapine and sinapic acid have been reported to exhibit a bitter taste, subsequent stages in the protein isolation process should focus on methods to systematically remove these polyphenolic compounds. Given that the proportion of total phenolic acids in the insoluble-bound form was up to ten times lower compared to free and esterified phenolic acids, it can be concluded that the analyzed press cake, with appropriate pretreatments during the protein isolation process, has the potential to serve as a highly suitable starting material for food-grade protein production.

**Keywords:** phenolic compounds, rapeseed press cake, HPLC-DAD analysis

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovations, Republic of Serbia (Contract No. 451-03-66/2024-03/200222), as well as by the Science Fund of the Republic of Serbia, Grant No 6673, PROTein from Rapeseed Oil Processing Waste: Application in Food and Wastewater Treatment – PROTOPOWER.

## CATEGORIZATION OF FOOD BASED ON VITAMIN K-CONTENT FOR PREVENTING ACENOCOUMAROL-FOOD INTERACTIONS

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Acenocoumarol prevents carboxylation of vitamin K-dependent clotting factors II, VII, IX and X, and thus interferes with coagulation. Since vitamin K is a competitive antagonist of acenocoumarol, foods rich in vitamin K reduces the therapeutic effect of the drug. A single vitamin K-rich meal does not have major negative consequences during a 24h-period nor does 50 mcg vitamin K daily have a statistically significant impact on the international normalized ratio (INR) or the levels of uncarboxylated vitamin K-dependent coagulation factors. However, there are case reports about clinically relevant vitamin K rich food interactions with INR and/or acenocoumarol levels especially among geriatric patients with comorbidities. The vague guidelines on the use of acenocoumarol among elderly with food and the consequent INR fluctuations make the therapy less efficient and lead to more adverse reactions. In this study a dietary plan recommendation for patients who are taking acenocoumarol were made. Data on the average content of vitamin K were obtained from the U.S. Department of Agriculture, Agricultural Research Service (<https://fdc.nal.usda.gov/>). FAO tables were used to convert the amount of vitamin K in foods to micrograms per 100 g and divided into 4 categories:

Category I: < 5mcg vit K/100g: cooked potatoes, canned tuna

Category II: 5-10mcg vit K/100g: fat-free milk

Category III: 10-40mcg vit K/100g: carrots

Category IV: >40mcg vit K/100g: broccoli, green leaf lettuce, green beans, spinach, pickled cucumber, asparagus etc.

Raising awareness on the vitamin K-content category of specific foods among acenocoumarol-prescribed patients might reduce the risks of INR oscillations and other serious adverse effects of the drug simply by providing more clarity on vitamin K content of foods to patients.

**Key words:** acenocoumarol, vitamin K, international normalized ratio

## TAKE YOUR MEDICINE WITH WATER: COMMON FRUIT JUICE-DRUG INTERACTIONS

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Fruit juices may interact with drugs in unpredictable ways due to the presence of unknown combinations of structurally diverse secondary biomolecules. In this study some of the known and reported fruit juice interactions with drugs are summarized. Apple juice is being reported to have interactions with beta blocker atenolol, renin inhibitor aliskiren and anti-asthmatic drug montelukast probably due to inhibition of intestinal drug transporter OATP2B1. Grapefruit juice as CYP3A4 inhibitor has reported interactions with anti-hypercholesterolemia drugs as statins, anticonvulsive as carbamazepine, antiarrhythmics such as amiodarone and immunosuppressives such as cyclosporine. The area under the curve and the maximum plasma concentration levels of anti-hypercholesterolemic drug, simvastatin was reported to be more than 10 times higher when applied with grapefruit juice in comparison to control with water. Pomegranate and sour orange juice as CYP3A4 and P-gp inhibitors increase the bioavailability of sildenafil applied in the treatment of erectile dysfunction. Orange juice decreased the bioavailability of medicines such as: atenolol, aliskiren, montelukast and alendronat. Pomelo juice on one hand decreases the bioavailability of sildenafil and its therapeutic effect while on the other hand it significantly increases the area under the curve and maximal plasma concentration of cyclosporine. The reverse effect on cyclosporine has been observed when applied with grape juice: decreased area under the curve and maximal concentration is followed by risk for subtherapeutic effect. Cranberry juice is reported to have interactions with oral anticoagulant drugs including warfarin. Persistent subtherapeutic levels of the antiepileptic drug phenytoin were reported when taken with noni juice. Given the popularity of fruit juices and smoothies, especially among health-conscious individuals seeking nutritious, on-the-go snacks, and being aware of clinically relevant interactions between fruit juices and medicines, it would be advisable to recommend the application of medicines exclusively with water in order to increase patient's safety.

**Key words:** fruit juice-drug interactions, bioavailability, CYP3A4 inhibition, P-gp inhibitors

## COMPARING THE CHEMICAL AND BIOCHEMICAL CHARACTERISTICS OF BERMET AND ITS BASE WINE

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Bermet is a unique aromatic dessert wine from Sremski Karlovci, a well-known winemaking area on Fruška Gora mountain in northern Serbia, which was originally created for medicinal purposes. This wine is made by adding various herbs and spices to the base wine to enhance the maceration of aromatic compounds. While mugwort is the main ingredient in every Bermet, each winery adds its own secret mix of herbs and spices. The aim of this study was to investigate how added herbs and spices influence the chemical composition and biological activity of the base wines and to identify the features they contribute to Bermet wine. For this purpose, 2 white, 1 rosé and 5 red base wines were compared with their respective Bermet wines. The polyphenolic profile was determined with LC-MS/MS and HPLC-UV/VIS methods, while total soluble solids (TSS) were measured with a refractometer. Spectrophotometric methods were used to assess the antioxidant activity by measuring how well the samples scavenge HO<sup>•</sup>, and to evaluate their neuroprotective properties by testing their ability to inhibit the enzyme acetylcholinesterase. From 75 analysed polyphenols, 50 were quantified in examined samples. Bermet wines contained polyphenols that are not characteristic for base wines, however base wines had greater amounts of main polyphenolics, such as anthocyanins, flavan-3-ols and gallic acid. TSS were higher in all Bermet wines, probably due to a higher sugar content. All base wines expressed significantly higher neuroprotective activity, while the antioxidant activity was similar between base and respective Bermet wines. This study highlighted the chemical changes that occur during the making of Bermet and how those changes affect the evaluated biological activities. Each Bermet gained unique characteristics from the added herbs and spices, although the biological potential of base wines has lowered during Bermet production process.

**Keywords:** Bermet, wine, polyphenols, sugar, antioxidants, acetylcholinesterase

**Acknowledgements:** We wish to thank the wineries from Sremski Karlovci, Serbia, for their generous donation of base and Bermet wines analysed in this study. This research work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grants No. 451-03-66/2024-03/200125 & 451-03-65/2024-03/200125).

## COMPARING THE EFFECTS OF SUPPLEMENTS AND FOOD ON VITAMIN C LEVEL ELEVATION IN THE STUDENT POPULATION

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Vitamin C has many important functions in our organism. Besides its antioxidant properties, vitamin C is crucial for the biosynthesis of collagen and L-carnitine and acts as a cofactor for the family of metalloenzymes, being involved in the synthesis of catecholamines and peptide hormones. Its deficiency causes many serious health problems, such as impaired wound healing, bleeding diathesis, osteoporosis and scurvy. Students' population often leads an unhealthy lifestyle where their diet lacks nutritional beverages such as fruits and vegetables, causing lower vitamin C levels. The first aim of this study was to assess the levels of vitamin C in the student population of the University of Novi Sad, Serbia, and establish their everyday habits through a survey consisting of questions about their diet, average sleeping hours, consumption of vitamin C-rich foods and supplements, alcohol and cigarettes. The next aim was to investigate which form of vitamin C intake would best elevate its levels in a one-week period. Fifty respondents included in this study were divided into 4 groups, and their vitamin C levels were determined using a titrimetric method and urine test strips. Three groups were then given 500 mg of vitamin C supplements in the form of tablets, effervescent tablets and powder for 7 days, while the fourth group consumed vitamin C-rich foods. Urine was sampled on the 1<sup>st</sup>, 3<sup>rd</sup> and 7<sup>th</sup> day of the treatment. Results showed that the student population has a vitamin C deficiency. Supplementation elevated vitamin C levels in all 4 groups. Tablets expressed the best effect in the first 24 hours, while at the end of the study, both tablets and effervescent tablets caused the highest elevation. Students' habits also influenced vitamin C levels; cigarette consumption lowered vitamin C levels by approx. 20–30%, as did less sleep and an unhealthy diet.

**Keywords:** vitamin C, ascorbic acid, supplements, student population

**Acknowledgements:** We wish to thank all the students who took part in this study, and the Faculty of Medicine, University of Novi Sad for providing the reagents, equipment and laboratory needed for the realization of this research.

## CHEMICAL CHARACTERISTICS, ANTIOXIDANT ACTIVITIES AND GLUCOSINOLATES OF CAPER (*CAPPARIS SPINOSA*) VARIED BY SOIL SUBSTRATES

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Dryland ecosystems contain different soil conditions that can affect the chemical properties of plants. Caper (*Capparis spinosa*), as an important economic species, can thrive in various soil conditions, yet there is limited information about the effects of environmental conditions on its chemical properties. In this study, we collected *C. spinosa* from both gypsum and non-gypsum soils in southern Iran, where the largest and most economically important population of this species exists. We considered different parts of the plants, including petals, sepals, flags, pistils, fruits, leaves, and seeds. We then determined the total phenol content, total flavonoid content, antioxidant activities (DPPH, FRAP, ABTS), and Glucocapparin in both soil conditions. Based on the findings, plant parts and site had a significant impact on most of the parameters. The highest amount of Glucocapparin was found in the pistils, while the lowest amount was in the petals. Petals had the highest total phenol content, while the lowest was in the seeds. The highest total flavonoid content was in the leaves and fruit, and the lowest was in the seeds. The highest activity of DPPH was found in the leaves, while the lowest was in the pistils. As gypsum levels increased, the activity of DPPH also increased in various plant parts. There was no significant difference between the two sites for FRAP, with the highest activity was found in the sepals. Overall, the gypsum sites showed higher values for most of the parameters. The PCA results showed that pistils were associated with Glucocapparin, petals with DPPH, and the leaf and sepal with total phenol content, FRAP, ABTS, and total flavonoids. Total phenol content had correlations with most of the factors. Additionally, different glucosinolate compounds were found in various parts of the plants. Ours results indicate that *C. spinosa* have valuable chemical compounds in both soil conditions.

**Keywords:** Gypsum soil, *Caper*, biochemical trait, Glucosinolate

**Acknowledgements:** This study forms part of AGROALNEXT (Agroalnext\_2022\_027) programme and was partially supported by MCIN with funding from European Union NextGenerationEU (PRTR-C17.11) and by Generalitat Valenciana, A.D.C. also would like to thank the Erasmus+ Student Mobility for Traineeships that facilitated the mobility between Agricultural University of Athens and CEBAS-CSIC 2022-2023.



## BIOSTIMULATION OF CRUCIFEROUS FOODS FOR HUMAN HEALTH

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Cruciferous vegetables (*Brassicaceae*) are rich in nutrients (vitamins, minerals, folates, etc.) and bioactive compounds (glucosinolates/isothiocyanates – GSL/ITC and phenolic compounds, vitamins, and minerals), associated with their health-promoting potential in modulation of several types of cancer and chronic non-communicable diseases, in which a chronic inflammatory condition is present at metabolic and neurological level. The current research lines on metabolite farming includes multidisciplinary work integrating the food chain – from seed to food and health – evaluating the influence of pre- and post-harvest factors to enrich foods in bioactive phytochemicals for human health, using sustainable practices. In this activity, we evaluated different varieties of cruciferous germinating seeds (red cabbage, red radish, broccoli, mustards, etc.) under controlled environmental conditions of hydroponics using priming, elicitors a selective LED light illumination to obtain sprouts/microgreens enriched in bioactive compounds, as sources of extracts and bioaccessible fractions of metabolites to investigate their anti-inflammatory and chemopreventive effects. The evaluation of bioactivity allowed to demonstrate significant antiinflammatory effects of sulforaphane (at therapeutic dosages), without any toxic effect, with higher potency than the parental glucosinolates (glucoraphanin, glucoraphenin) or phenolic compounds. The bioaccessible fractions of GSL/ITCs of red varieties of sprouts (radish and cabbage) demonstrated health-promoting effects on cellular models of chronic inflammation. This research line continues evolving with the study of the effects of GSL/ITC from cruciferous foods in adult population with overweight, to investigate on the physiological, metabolic, and microbiome factors involved in this chronic disease.

**Keywords:** *Brassica*, biofactory, bioactives, food, inflammation, obesity

**Acknowledgements:** This work was supported at least in part by the Project “SANO” CPP2022-009528, with funds of the MCIU/AEI/10.13039/501100011033 and the European Union “NextGenerationEU”/PRTR”. Additionally, authors would like to thank the support of the AGROALNEXT Program, granted by MCIN with NextGenerationEU funds and the “Fundación Seneca” – Agency for Science and Technology of the Murcia Region (CARM), project PRTR-C17.11.

## IMPACT OF A WATER-SOLUBLE OREGANO-BASED SUPPLEMENT, IN BROILER CHICKENS IN THE FACE OF INTESTINAL CHALLENGE

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Intestinal challenges compromise broiler performance. Phytochemicals may restore performance by improving antioxidant status while also aligning with the increasing resistance to antimicrobial growth promoters and consumer preferences for natural additives that improve meat quality. In the present study, we assessed the efficacy of Ecodiar®, a water-soluble oregano-based supplement (0.5 ml/lit of drinking water). A total of 252 day-old male broilers were divided into three groups with six replicate pens per group: an unchallenged negative control (NC), a challenged positive control (PC), and a challenged oregano-supplemented group (COR). Challenged groups were raised on reused litter and received a 10-fold dose of an anticoccidial vaccine (EVANT®, Hipra) at d7, while the NC group was raised on fresh litter. Over the 42-day trial, we assessed weekly performance parameters and end-of-trial measures of lipid peroxidation (MDA), total phenolic content (TPC), and total antioxidant capacity (TAC) in meat (breast and thigh), liver, and ileal digesta. In 12 birds per group. The COR group had significantly increased BW from d14 onwards, ADG (weeks d7-d14, d14-d21, and d35-d42), FI (week d7-d14), and European Broiler Production Index (EBPI) in comparison to PC with no statistical differences on weekly FCR. Performance metrics were similar between NC and COR birds ( $P>0.05$ ). COR birds demonstrated lower MDA levels, in breast and thigh tissues ( $P<0.05$ ), than both NC and PC birds. Moreover, COR birds had similar TPC in ileal digesta and liver tissue to NC birds and significantly greater than PC birds ( $P<0.05$ ). These results suggest that oregano supplementation can enhance broiler meat antioxidant status and restore performance of challenged birds. This study highlights the potential of integrating natural feed additives into poultry diets to improve the performance under challenging conditions and meet consumer demands.

**Keywords:** broiler chicken, oregano, antioxidant status, intestinal challenge

**Acknowledgements:** The authors acknowledge Ecopharm Hellas S.A., Kilkis, Greece for providing the supplement Ecodiar®.

## IODINE CONTENT IN COMMERCIALY BRANDED SALTS IN CROATIA

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Table salt is the most important dietary source of iodine for all population groups in Croatia due to its mandatory fortification with iodine. Based on our previous research regarding the consumption frequency of salts available on Croatian market, we conducted analysis of iodine content of commercially branded salts available in Croatia. A total of 21 salts were bought in large supermarket chains and web shops. Spectroscopic method based on the Sandell-Kolthoff reaction was used to determine iodine content. Samples contained between 0.0 mg/kg (ppm) and 22.48 mg/kg (ppm) of iodine. Five samples had no iodine (0.0 mg/kg), including one Himalaya salt and one sample declared as "fortified with iodine". Additional six samples have iodine content below the minimum required 15 ppm, ranging from just 0.17 ppm to 13.54 ppm. Finally, 62% of analysed samples do not comply with national regulations on iodine fortification. Iodine plays important role in a range of health aspects, from the synthesis of thyroid hormones, overall metabolism, growth of the body, to neurodevelopment of foetal brain. The gaps we identified should urge national bodies to strengthen product control and introduce educational activities for general public on appropriate dietary sources of iodine.

**Keywords:** iodine; salt; iodine content; market analysis

## THE EFFECT OF FEEDING PRACTICES, SEASONAL VARIATIONS AND LACTATION STAGES ON DONKEY MILK QUALITY

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The purpose of this study was to examine how feeding practices, seasonal variations, and lactation stages affect the composition of donkey milk. Donkeys grazed continuously on the Valjevac pasture within the Zasavica Special Nature Reserve. During spring, they were supplemented with maize grain and clover, while during the winter; they were given meadow hay and maize as feed. Milk samples of donkeys were collected during morning milking sessions at different stages of lactation (early spring, early summer, late summer, and winter: 40, 100, 160, and 220 days post-partum). The following quality parameters were analysed: the basic chemical and mineral composition of the same pasture in various seasons of the year (spring, summer, autumn and winter), meadow hay, clover, and maize grain, as well as the fatty acid profiles during different feeding conditions. This research investigated how the milk composition changes throughout the productive season of donkeys. The highest protein content in the milk during spring resulted from feeding on high-protein pasture supplemented with fresh clover. Due to the pasture quality and the supplementations during spring the donkey milk had increased proportions of monounsaturated fatty acids (MUFA), polyunsaturated fatty acids (PUFA), unsaturated fatty acids (UFA), oleic acid C18:1 n-9 and alpha-linolenic acid C18:3 n-3. Milk in the winter season exhibited higher levels of saturated fatty acids (SFA), likely due to increased consumption of meadow hay and maize grain. The mineral composition of pasture in the autumn affected the highest mineral concentrations in the milk. In summary, this study offers a thorough analysis of the characteristics of donkey milk, pointing out how feeding practices, seasonal changes and lactation stages interact and influence the milk composition. Unique composition of the donkey milk can classify it as functional food or ingredients in novel food products.

**Keywords:** feed, seasons, quality, donkey milk

**Acknowledgements:** This research was financially supported by Ministry of Science, Technological Development and Innovation, Republic of Serbia, Institute of Food Technology in Novi Sad (No. 451-03-66/2024-03/200222).

## THE ROLE OF THE MEDITERRANEAN DIET (AND LIFESTYLE) IN HEALTH AND AGING

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Aging is a complex process influenced by various factors, including genetics, environment, and lifestyle. Healthy lifestyle choices can significantly impact aging and extend the number of years lived free from major chronic diseases. Recent research has increasingly focused on the roles of nutrition and lifestyle in promoting longevity and preventing age-related diseases. Mediterranean diet, and overall Mediterranean lifestyle, are indicated to promote healthy aging and were associated with lower risk of all-cause mortality. The Mediterranean lifestyle encompasses the consumption of foods typical of the Mediterranean diet, adherence to related eating habits, and the incorporation of adequate physical activity, rest, and healthy social interactions. The Mediterranean diet, which includes omega-3 polyunsaturated fatty acids, polyphenols, fermented dairy products, fibers, and other components, has the capacity to improve overall health status by exhibiting antioxidant activity, reducing inflammatory load, and restoring the intestinal microbiota. Not only nutrient intake but also food consumption habits, such as meal timing and eating environment, could significantly influence these effects. Furthermore, other lifestyle factors that are part of the Mediterranean lifestyle may affect the levels of hormones, neurotransmitters, and anti-inflammatory or pro-inflammatory compounds, thereby promoting healthy aging and preventing certain diseases, including metabolic disorders and neurodegenerative conditions. Everyday dietary and lifestyle choices play a crucial role in the aging process. Understanding these influences provides valuable insights for promoting healthy aging through nutrition and other modifiable lifestyle factors.

**Keywords:** Mediterranean diet, lifestyle, quality of life, aging, health

## CAROTENOIDS PROFILE AND TOCOLS CONTENT OF FLOWERS, LEAVES AND STEMS OF THE WILD EDIBLE PLANT, *OXALIS PES-CAPRAE* L.

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Wild Edible Plants (WEPs) are plants that grow without human help, and with what they obtain directly from nature. One of them is *Oxalis pes-caprae* L., belonging to the Oxalidaceae family. They are commonly known as weed. This study aims to investigate the composition of this species and determine its potential health benefits. The plant material, including flowers, leaves, and stems, was collected in Orihuela, Alicante (Spain) in October 2022. The carotenoid profile analysis was made by High-Performance Liquid Chromatography (HPLC) (Dionex UltiMate 3000, Sunnyvale, CA, USA). The results revealed that the leaves had the highest carotenoid concentration (12.5 mg/100 g fresh weight), while the flowers exhibited the lowest concentration (1.47 mg/100 g fresh weight). The majority compounds in the carotenoid profile of the pes-caprae oxalis leaf were: Lutein,  $\beta$ -carotene and  $\beta$ -Cryptoxanthin (4.76, 4.50 and 1.35 mg/ 100 g, respectively). The results in stems maintained the same order of importance as on the sheet; however, in the flowers, zeaxanthin (0.17 mg/ 100g) obtained greater concentration than lutein (0.10 mg/ 100g). Regarding tocopherols, the highest concentration was detected in the leaves (7.20 mg/100 g), predominantly in the form of  $\alpha$ -tocopherol (6.81 mg/100 g fresh weight). In conclusion, these results show underscore the importance of further investigation into these plants as potential sources of bioactive compounds for applications in the food, cosmetic, and pharmaceutical industries.

**Keywords:** nutrition, vitamins, thiamine, riboflavin, bioactive compounds

**Acknowledgements:** ReTos-AAA program Miguel Hernández University.

## CHARACTERIZATION AND EVALUATION OF NUTRIENT-ENHANCED SMOOTHIES PREPARED WITH FROZEN FRUITS AND VEGETABLES

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The increasing consumer awareness of functional foods has sparked significant interest in products with high nutrient profiles and health-protecting qualities. In response, the food industry is developing new products to meet these demands, with smoothies being an excellent example. Smoothies are defined as non-alcoholic beverages prepared from fresh or frozen fruit and/or vegetables. They have a typical semi-liquid, smooth consistency and are prepared by mixing, in appropriate proportions, different ingredients, promoting the daily consumption of fruits and vegetables. Individually Quick Freezing (IQF) technology has the potential to markedly enhance the quality of fruit and vegetable smoothies by preserving their nutritional and sensory attributes. The aim of this study was to identify optimal combinations of frozen fruits and vegetables for smoothie preparation, focusing on taste and nutritional value. Various fruits and vegetables including oranges, peaches, apples, spinach, peas, and lemons were initially blanched to deactivate enzymes, followed by the application of individually quick freezing (IQF) to preserve their freshness and nutritional integrity. Three different combinations of fruits and vegetables were used to prepare the nutrient-enhanced smoothies. The physicochemical properties and the nutritional profile of the smoothies were defined. Also, the concentration and stability of vitamin C in the smoothies over time were assessed using HPLC analysis. Additionally, sensory evaluation was conducted to determine taste, texture, and overall acceptance of the smoothies by a trained panel. The results demonstrated that the combinations of fruits and vegetables provided a high nutritional value, offering rich content of vitamin C. The combinations were found to be highly satisfactory, with balanced taste and texture, making the smoothies ideal for daily consumption. In conclusion, the study highlights the importance IQF technology in enhancing the nutritional and sensory attributes of smoothies, thus facilitating the consumption of healthy dietary choices.

**Keywords:** smoothies, fruits, vegetables, individually quick freezing

**Acknowledgements:** This project was co-funded by Greece and European Union (Project title: Optimization of fruit combinations properties in terms of energy value and nutritional characteristics, using a quick freeze method, M16SYN2-00080).

## DEVELOPMENT OF A GARLIC PROTEIN-DERIVED ACE INHIBITORY PEPTIDE AND EXPLORATION OF ITS ANTIHYPERTENSIVE MECHANISMS

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Renin-angiotensin system is a key system for blood pressure regulation, inhibiting the activity of ACE plays a crucial role in prevention and treatment of hypertension. This study aimed to screen novel angiotensin I-converting enzyme (ACE) inhibitory peptides from garlic proteins and to explore their antihypertensive mechanisms *in vivo*. After simulated hydrolysis and *in silico* screening (based on peptideRanker, molecular docking, ADMET analysis and structure-activity relationships), two novel peptides (MGR and HDCF) were obtained with the highest ACE inhibitory activity ( $IC_{50}$  of 4.50  $\mu$ M and 26.38  $\mu$ M) and competitive patterns. They could bind to key residues in the ACE active site mainly through hydrogen bonding interactions and exhibited excellent stability against high temperature, extreme pH and gastrointestinal digestion. In spontaneously hypertensive rats, MGR and HDCF could effectively lower blood pressure after single or continuous treatments, with no effects on Wistar Kyoto rats. Further analysis found that the antihypertensive activity of these two peptides was mainly achieved by balancing the renin-angiotensin system, improving renal and cardiac impairment, and regulating endothelial dysfunction. The network pharmacology analysis identified five key genes (INS, REN, ACE, NOS3, and EDN1) in the protein-protein interaction network between HDCF and hypertension-related targets. GO and KEGG pathway enrichment analyses of differential genes indicated that high-dose peptide treatment led to significant changes in hypertension-impaired organs (e.g., pancreas and heart). Some endothelial dysfunction-related pathways were substantially regulated, including arachidonic acid metabolism, cortisol synthesis and secretion, arginine biosynthesis, calcium signaling pathway, and cGMP-PKG signaling pathway. In Ang II-induced human umbilical vein endothelial cells, HDCF treatment significantly increased NO levels and cGMP production via activating PI3K/Akt/eNOS and cGMP/PKG signaling pathways. Taken together, the improvement of endothelial dysfunction by HDCF was an important pathway for its antihypertensive effect.

**Keywords:** Garlic proteins, ACE inhibitory peptide, In silico screening, antihypertensive effects *in vivo*, antihypertensive mechanisms

**Acknowledgements:** This work was supported by the Special Fund for Leading Talent in Mount Tai Industry of Shandong Province (No. TSCY20200121).



## THE IMPACT OF DONKEY MILK IN PNEUMONIA TREATMENT OUTCOMES

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The health benefits of milk and dairy products for the growth and development of children and adults are well-documented. Recently, there has been growing interest in the impact of donkey's milk on respiratory health. The aim of this study was to investigate the impact of donkey milk on the clinical outcomes of hospitalized patients with community-acquired pneumonia (CAP). This prospective study included sixty patients with CAP hospitalized at Institute for Pulmonary Diseases of Vojvodina, Serbia, from June 2022 to January 2024. One group of patient aged  $65.17 \pm 10.98$  received standard antibiotic therapy plus 250 ml of pasteurized donkey milk twice daily for one month and control group aged  $63.13 \pm 13.06$  treated with standard antibiotic protocols. The length of hospitalization was observed as well as inflammatory parameters (C-reactive protein - CRP, fibrinogen, leukocytes) and the rate of radiological regression during hospitalization, upon discharge, on the tenth and twentieth days of follow up. The values of CRP were statistically significant higher in control group on the third ( $118.51 \pm 91.11$ ), seventh hospitalization day ( $40.09 \pm 40.07$ ), upon discharge ( $13.95 \pm 13.19$ ) and on the tenth day of follow up ( $6.83 \pm 5.96$ ) compared to group that used milk, respectively ( $78.17 \pm 63.84$ ;  $20.91 \pm 27.28$ ;  $6.53 \pm 7.04$ ;  $3.75 \pm 4.93$ ). On the first hospitalization day the values of CRP did not differ between groups ( $227.93 \pm 123.49$  vs  $209.09 \pm 130.63$ ). Leukocyte and fibrinogen were not significantly different between groups during hospitalization nor on follow up ( $8.63 \pm 3.63$  vs  $7.64 \pm 2.42$ ;  $8.37 \pm 2.55$  vs  $7.80 \pm 2.02$ ). The results of  $\chi^2$  test showed statistically significant better degree of radiological regression on the seventh day of hospitalization, at discharge, as well as on the first follow-up in the group that used milk compared to control group ( $\chi^2=21.08$ ;  $p<0.001$ ). The use of donkey's milk in the treatment of pneumonia leads to a significantly better and faster patients recovery.

**Key words:** community-acquired pneumonia, donkey milk, inflammatory parameters, x-ray, follow up

**Acknowledgements:** This research was funded by the Provincial Secretariat for Higher Education and Scientific Research, Autonomous Province of Vojvodina, Republic of Serbia [grant number 142-451-2637/2021-01/2] as a part of the project entitled "Examination of the impact of goat, mare and donkey milk on the recovery of patients with pneumonia.

## FOODS AND BEVERAGES CONTAINERS AS POTENTIAL SOURCES OF BISPHENOL A – SURVEY AMONG LUNG CANCER PATIENTS

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Lung cancer remains the leading cause of cancer death worldwide, despite the decreasing rates of tobacco smoking. Therefore, there is a noticeable rise of clinical interest in other modifiable risk factors for lung cancer such as alcohol consumption, lack of physical activity, diet, infections and environmental pollution, especially among non-smokers. Bisphenol A (BPA), an endocrine disruptor and environmental pollutant can be found in epoxy resins of food and beverage containers and can leach under normal conditions into the food. BPA enhances the carcinogenesis susceptibility and stimulates in vitro migration and invasion of lung cancer cells. The aim of this study was to evaluate the potential BPA exposure from instant soups, canned food and plastic containers among lung cancer patients. A total of 205 patients out of which 60 (45% female) with small cell lung cancer (SCLC) and 145 (46.21% female) with non-small cell III/IV stage lung cancer (NSCLC) from the Institute for Pulmonary Diseases of Vojvodina, Sremska Kamenica, Serbia were surveyed about selected dietary and lifestyle habits. There were no statistical differences in smoking habits among SCLC and NSCLC groups nor between sexes. The use of bagged instant soups and canned foods was the same across SCLC and NSCLC patients ( $p=0.314$  and  $p=0.177$ , respectively). Male patients with SCLC were more frequently users of canned foods in comparison to females ( $p=0.010$ ), with no differences among other types of cancer between genders. The majority of the respondents in both, the SCLC and NSCLC group never used plastic containers for heating foods ( $p=0.339$ ). Despite tobacco smoking is recognized as the main cause of SCLC type, other factors such as the presence of environmental pollutant BPA should also be considered a major health concern. There is an urgent need for the assessment of BPA presence in biological samples of lung cancer patients.

**Keywords:** lung cancer, bisphenol A, bagged instant soup, canned foods, plastic containers

**Acknowledgements:** This work was supported by the Provincial Secretariat for Higher Education and Scientific Research, AP Vojvodina, Republic of Serbia (Grant No. 142-451-3509/2023-01).

## ALTERNATIVE THERAPY FOR HYPERLIPIDAEMIA-SPIRULINA CASE STUDY

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Potential therapeutic effects of spirulina supplementation against obesity, hypertension and diabetes are reported based on in vitro and in vivo studies. The recent research performed on adult rats suggests that spirulina induced bile acids excretion and gut microbiota changes may be the mechanism of spirulina hypolipidemic activity. However, the anti-hyperlipidaemic effects of spirulina supplementation on a molecular level are still not sufficiently investigated. The aim of this study was to assess the oxidative stress and antioxidant biomarkers in order to investigate spirulina effects against hyperlipidaemia induced impairment of cellular reactions that produce reactive oxygen species (ROS). Biomass of *Spirulina platensis* (strain S2) was used. A group of 40 adult male Wistar rats (approved by the Bioethics committee of University of Novi Sad) were randomly divided into five groups according to feeding regimes (I-normal diet; II-normal diet with spirulina supplementation; III-lipogenic diet; IV-lipogenic diet with concomitant spirulina supplementation; V-lipogenic diet with spirulina treatment). The activity of oxidative stress biomarkers in liver were noticed in all groups. Although, no differences at significant level were obtained between groups on pure atherogenic diet (III) and atherogenic regime with spirulina (IV and V), a reduction in superoxide dismutase and glutathione S-transferase were observed in group V in comparison to group IV. The significant increase in catalase activity was measured in animals on pure atherogenic regime (III) in comparison with groups with implemented spirulina in diet (II and V). It is important to note that the marked reduction in xanthin oxidase, glutathione peroxidase and total antioxidative activity at significant level was also observed in group V in comparison to group IV. The obtain results suggest that spirulina supplementation may decrease ROS production in diagnosed hyperlipidaemia due to the spirulina antioxidative potential.

**Keywords:** oxidative stress, hepatoprotective effects, spirulina, functional food

**Acknowledgements:** This research was funded by Provincial Secretariat for Higher Education and Scientific Research, AP Vojvodina, Republic of Serbia (Grant No. 142-451-3509/2023-01).

## GLUTEN-FREE BREAD ENRICHED WITH NON-GERMINATED AND GERMINATED ALFALFA SEED FLOUR: ASSESSMENT OF NUTRIENT CONTENT IN RELATION TO COMMERCIAL COUNTERPARTS

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Considering that 1–2% of the human population worldwide lives with diagnosed coeliac disease which requires a lifelong adherence to a gluten-free (GF) diet, the research community effort to develop a new range of quality GF products has increased in the last decade. Although advances regarding the technological and sensory quality of GF bakery products were partially achieved, severe drawbacks in terms of nutrient deficiencies (lack of proteins, vitamins, fibres, antioxidants, and minerals) still represent a challenge to combat. Legumes emerge as a compelling strategy to combat nutrient deficiency in GF bread (GFB) and enable shifting towards a more sustainable diet due to their high protein content and low environmental impact. Considering the presented background, this study aimed to explore the effect of non-germinated and germinated alfalfa seed flour addition (5% on maize flour/starch basis) on GFB nutrient (protein, ash, lipid, carbohydrates content and energetic value) and mineral content (Ca, Mg, Fe, Zn), and compare it with control (without alfalfa) and commercial GFB available on the market. A higher content of protein, ash, and lipid was observed in GFB containing alfalfa compared to control and commercial GFB. Conversely, alfalfa-enriched GFBs were characterised by lower carbohydrate content (30.1–31.7 g/100 g) and energetic value ranging from 874 to 894 KJ. As regards to mineral content, a significant increase in Mg (from 342.2 to 908.3 mg/kg), and K (from 968.8 to 1478.9 mg/kg) was observed in alfalfa-enriched GFB, while lower content of Ca (320.0–462.8 mg/kg) and Fe (10.2–10.3 mg/kg) was recorded compared to commercial GFB (33.8 mg/kg). However, Fe content in alfalfa-enriched GFB was still higher than in control GFB (6.6 mg/kg). Significant differences in nutrient and mineral content were not observed between GFBs containing non-germinated and germinated alfalfa seed flour suggesting that both flours of this legume can be used as ingredients for improving nutrient content in GFB.

**Keywords:** gluten-free, bread, alfalfa, germination, nutrients

**Acknowledgements:** The presented study is financially supported by the Provincial Secretariat for Higher Education and Scientific Research of Vojvodina, Republic of Serbia through Project number 000858196 2024 09418 003 000 000 001 04 003.

## SOURDOUGH TECHNOLOGY AS A STRATEGY TO ENHANCE THE APPEARANCE, TEXTURE, AND SENSORY ATTRIBUTES OF WHEAT BREAD CONTAINING NON-GERMINATED AND GERMINATED ALFALFA SEED

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Bringing diversity in protein consumption represents a global challenge to combat public health, climate, and nature-related risks and achieve long-term food security. Legumes are a valuable source of proteins, however relatively long preparation time, digestive discomfort occurrence, presence of antinutrients, and their beany or bitter flavour limit their incorporation into widely consumed staple food such as bread. To counteract the mentioned drawbacks for legume flour inclusion into bread formulations, sourdough fermentation and germination arise as re-emerging trends in healthy food production. These natural, traditional, low-cost and green processes could be a valuable ally in enhancing and tailoring the texture, flavour, nutritional value, and shelf life of legume-containing bread. Among legumes, alfalfa (*Medicago sativa* L.) seed potential for inclusion in bread formulations is scarcely investigated. Therefore, this study aimed to evaluate the effects of sourdough fermentation on specific volume, crust and crumb colour, crumb texture and sensory acceptability of wheat bread containing 5 and 10% convective dried non-germinated (ASF) and germinated alfalfa seed flour (GASF). Sourdough fermentation increased bread specific volume (from 2.63 to 3.62 cm<sup>3</sup>/g) in samples with 5% ASF and GASF. However, a slight volume-depressing effect compared to control sourdough bread was observed in samples containing 10% ASF and GASF. Greater specific volumes of sourdough bread samples containing alfalfa flour were followed by lower crumb hardness (reduced from 3.82 to 1.68 N), especially pronounced in samples with 5% ASF and GASF. Darker crust and crumb colour with a prominent visual difference ( $\Delta E > 2.3$ ) was obtained in bread samples containing ASF and GASF compared to control sourdough bread. Sourdough bread samples containing ASF and GASF were rated as more acceptable compared to control sourdough bread and standard wheat bread (without sourdough) according to a 5-point hedonic scale. Sensory acceptability was the highest for sourdough bread containing 5% ASF (4.46 points), followed by samples with 5 and 10% GASF (4.37 points), and 10% ASF (4.17 points). The reported results showed that sourdough fermentation combined with alfalfa seed flour addition is a winning strategy to enhance specific volume, colour, crumb softness and sensory acceptability of legume-containing bread.

**Keywords:** legumes, germination, colour, crumb texture, sensory evaluation

**Acknowledgements:** This work was done under the framework of CEEPUS network HR-0306-15-2223 "For Safe and Healthy Food in Middle-Europe" (during research mobility of Marijana Djordjević).

## CUSTOMIZED PRODUCTION OF XYLOOLIGOSACCHARIDES USING *ASPERGILLUS TUBINGENSIS* FAT 35 XYLANASES

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Xylooligosaccharides (XOS), sugar oligomers consisting of 2-10 xylose units, are considered as emerging prebiotic compounds that have shown beneficial effect on health. In addition to significant prebiotic properties, XOS also have considerable antioxidant potential. Due to the wide pH range (2.5-8) and temperature stability (up to 100°C) XOS are a desirable ingredient in functional food. The therapeutic daily intake of XOS, which is the lowest of all prebiotics, further justifies intensive research into their production today. Enzymatic production of XOS by xylanase enzymes is the most preferable method. *Aspergillus tubingensis* FAT 35 has proven to be excellent xylanase producer. Cultivation of the fungus *Aspergillus tubingensis* FAT 35 on corn cob, which is rich in xylan, resulted in a highly efficient xylanase complex with an activity of 550 U/mL. Four distinct protein fractions with xylanase activity were isolated after purifying the fermentation extract using precipitation and chromatographic techniques. Of these, two fractions demonstrate endo-xylanase activity, producing XOS with varying degrees of polymerization. Through a controlled xylan hydrolysis reaction, it was possible to obtain custom XOS mixtures. All XOS mixtures were characterized by HPLC and TLC. Due to their high antioxidant properties (evidenced by DPPH and FRAP assays) the produced XOS has shown significant promise as functional food additives. This research justified the need to find new enzymes since it showed that the newly isolated fungal xylanase can be used to obtain high yield XOS with variable composition depending on the reaction and with very good properties. Future research will explore the possibilities of using the obtained XOS in functional food.

**Keywords:** xylanase, xylooligosaccharides, functional food, prebiotic

**Acknowledgements:** This research was supported by the Science Fund of the Republic of Serbia, #7347, Unique enzyme solutions for obtaining added value products from corn – UniCorn and the Ministry of Science, Technological Development and Innovation of Republic of Serbia (Contract numbers: 451-03-66/2024-03/200026 and 451-03-65/2024-03/200168).

## DEVELOPMENT OF FUNCTIONAL NON-ALCOHOLIC BEER ENRICHED WITH CAROTENOIDS USING PUMPKIN ADDITIVES

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In this study, the aim was to develop a functional beer enriched with bioactive compounds, specifically carotenoids, by incorporating pumpkin powder and pumpkin extract into the brewing process. Given that European regulations define functional beverages as those containing no ethanol or an ethanol content not exceeding 0.5% vol., we opted to create a non-alcoholic beer. The fermentation was conducted using *Saccharomyces chevalieri* yeast for two days, after which it was stopped once the ethanol content reached the permissible limit (0,48% vol. for both beers). Throughout the fermentation process, there was an increase in the content of carotenoids and polyphenols, both known for their health benefits, particularly in terms of antioxidant activity. Similar results were obtained for the beer with the addition of pumpkin powder (1.9 mg/L of carotenoids) and for the beer with the addition of pumpkin extract (1.8 mg/L). Importantly, the formation of unwanted vicinal diketones, which can impart off-flavor at higher content, was successfully avoided. All other quality parameters (real extract, apparent extract, real fermentability, apparent fermentability, color, pH, bitterness, soluble nitrogen content) remained within the acceptable ranges for this type of beer, ensuring that the final product not only met the criteria for a functional beverage but also maintained the sensory qualities expected of a non-alcoholic beer. The successful enhancement of the beer with bioactive compounds from pumpkin, regarding carotenoids, demonstrates the potential for developing functional beers that align with regulatory standards while offering additional health benefits. This study contributes to the growing field of functional beverages, providing insights into the formulation and production of non-alcoholic beers with added nutritional value.

**Keywords:** Functional Beer, Non-Alcoholic Beverage, Pumpkin Powder, Pumpkin Extract, Carotenoids

**Acknowledgements:** This research was funded by the Science Fund of the Republic of Serbia for the funding of the PRISMA project 'Nutrition-sensitive Breeding Of Cucurbita Plants', number '#6680' and Ministry of Science, Technological Development and Innovation of the Republic of Serbia under the Agreements on the Implementation and Financing of Research (nos. 451–03-47/2023–01/200222 and 451–03-47/2023–01/200051).



## IMPACT OF WHEY BIOACTIVE HYDROLYSATES ON THE QUALITY OF FAT FILLINGS FOR CONFECTIONERY PRODUCTS

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Whey protein hydrolysates can be used in a wide range of applications because they offer numerous advantages compared to non-hydrolyzed whey proteins. They are more heat stable, with improved foaming and emulsifying properties due to the presence of bioactive peptides of lower viscosity. Whey hydrolysates have improved absorption, digestibility, excellent nutritional and functional properties, and the ability to extend the shelf life of food products. Due to the large differences in the technological and other physicochemical properties of hydrolysates, the addition of whey protein hydrolysates into confectionery products is much more complicated. This research aims to determine the possibilities of enriching filled confectionery products with whey peptides obtained in two ways: through enzymatic hydrolysis of whey protein concentrate and fermentation (using microorganism *Lb. rhamnosus* ATCC 7469). Peptides were added to a fatty milk cream at a 5% concentration. The study was focused on assessing antioxidant activity, physical, rheological, textural, and sensory properties of three fat fillings: C (control without whey peptides), EWP (5% peptides from enzymatic hydrolysis), and MWP (5% peptides from whey fermentation). Enzymatic hydrolysates increased DPPH radical inhibition by 32%, and fermented hydrolysates by 19%. Enzymatic hydrolysates also demonstrated superior inhibition of lipid peroxidation (IC<sub>50</sub> value of 811.54 mg mL<sup>-1</sup>) compared to fermented hydrolysates (IC<sub>50</sub> value of 178.36 mg mL<sup>-1</sup>). EWP showed the highest antioxidant activity. Addition of enzymatic hydrolysates increased filling firmness by 2.5 times, while fermented hydrolysates had reduced firmness compared to the control. Both types of hydrolysates did not adversely affect the size or distribution of the particles in the fat cream. Thixotropic properties of the fat filling remained unchanged post-incorporation. MWP exhibited the most optimal rheological characteristics with the lowest yield stress. The best sensory characteristics (better than control sample) were found in the EWP.

**Keywords:** enzymatic hydrolysis, microbial hydrolysis, rheology, whey protein, antioxidant activity

**Acknowledgements:** This research was supported by the Ministry of Science, Technological Development and Innovations, Serbia, program (451-03-47/2023- 01/200134).



## GERMINATED ALFALFA SEED IN WHEAT BREAD FORMULATIONS: TRACKING THE PROTEIN STRUCTURAL CHANGES BY MEANS OF ATR-FTIR SPECTROSCOPY

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An insight into the chemical composition and molecular structure of heterogeneous food components undergoing structural and conformational changes during product development can provide useful guidelines toward components assembly in creating acceptable new product. This becomes particularly important in products such as bread in which gluten network plays a key role in defining the product structure and where other ingredient involvement tends to impair the gluten network and reflect on the bread's technological quality and acceptance by consumers. The objective of this study was to identify the major components and detect the conformational changes in protein secondary structures in wheat bread induced by the addition of germinated (GASF) and non-germinated (ASF) alfalfa seeds flour (5 and 10%) using Fourier transform infrared spectroscopy with attenuated total reflectance as sampling technique (ATR-FTIR). The ATR-FTIR spectra confirmed the presence of major components in alfalfa flours and resulting breads including protein, starch, and lipids. Further deconvolution of the Amide I protein region revealed the presence of  $\beta$ -sheets,  $\alpha$ -helix and  $\beta$ -turn protein secondary structures in flour samples while in addition to mentioned ones, random coil and aggregates were present in bread samples. Germination of the alfalfa seed induced changes in the protein secondary structure by increasing  $\beta$ -sheets contribution at the expense of  $\alpha$ -helix while  $\beta$ -turn share remained unchanged. The  $\beta$ -sheet conformation prevailed in all bread samples except control and bread with 10% ASF where  $\alpha$ -helix conformation was dominant. Bread samples containing ASF exhibited increase in  $\beta$ -sheet conformation compared to control, however the inclusion level showed great impact on the main protein structural domain which was  $\beta$ -sheets in 5% bread while that place was occupied by  $\alpha$ -helix in the 10% bread formulation. The inclusion of GASF further raised the  $\beta$ -sheet (~50%) as well as random coil share in bread's gluten network regardless of substitution level with simultaneous decrease in  $\alpha$ -helix when compared to control bread. The addition of ASF and GASF promoted substantial changes in structural conformation of proteins thus modifying the gluten network. The observed increase in  $\beta$ -sheet conformation regardless of added type of alfalfa seed flour could indicate altered bread quality in terms of volume, and hardness.

**Keywords:** ATR-FTIR, alfalfa, germination, bread, protein secondary structure

**Acknowledgements:** The authors acknowledge the contribution of the staff from Laboratory for materials in cultural heritage (Faculty of Technology Novi Sad, University of Novi Sad, Serbia) in ATR-FTIR analysis.

## BIOACTIVITY OF METHANOLIC PULP EXTRACT FROM PUMPKIN (*Cucurbita moschata*) IN HUMAN KERATINOCYTES

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The study investigated the potential of pumpkin pulp extracts from two *Cucurbita moschata* accessions grown in Serbia as skin-friendly agents with antioxidant and sun protecting properties. The research aimed to address the underrepresentation of pumpkin pulp in bioactivity studies despite its potential as a rich source of beneficial compounds for skin health. Methanolic pumpkin pulp extracts (MPE) were prepared from two accessions, MO 31 and MO 32, sourced from the breeding collection of the Institute of Field and Vegetable Crops in Novi Sad, Serbia. The MPE were characterized for their carotenoid content using HPLC-DAD, revealing different levels of carotenoids in each accession. Cumulative carotenoid content in MO 31 and MO 32 was 0.29 and 0.09 mg/g, respectively. The cytotoxicity of the extracts was assessed using viability assay MTT in human keratinocyte (HaCaT) cells, and showed no significant decrease in cell viability at concentrations of 10, 100, and 1000 µg/mL after 24 hours of exposure. The antioxidant potential of the extracts was evaluated by H<sub>2</sub>DCEFA assay, measuring their ability to reduce reactive oxygen species (ROS) levels in HaCaT cells co-incubated with 1mM hydrogen peroxide. Both MO 31 and MO 32 extracts significantly reduced oxidative stress at a concentration of 10 µg/mL, with similar effects observed at higher concentrations. Additionally, the in vitro sun protecting factor (SPF) of the extracts was determined using absorbance measurements and the Mansur equation, revealing notable SPF values of 4.15 and 5.16 for MO 31 and MO 32, respectively. Overall, the study demonstrates that the *Cucurbita moschata* pulp extracts possess potential as photoprotective and antioxidant properties, with no observed toxicity to skin cells. These findings suggest the extracts could be promising components in natural cosmetic formulations aimed at combating dermal photoaging and oxidative stress.

**Keywords:** pumpkin pulp, reactive oxygen species, cytotoxicity, photoprotective

**Acknowledgements:** This study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Agreement no. 451-03-66/2024-03/200019).

## TOCOPHEROL CONTENT IN COLD-PRESSED OILS: IMPLICATIONS FOR SHELF LIFE AND NUTRITIONAL VALUE

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Tocopherols play a crucial role in protecting oils from oxidation, thereby extending their shelf life and enhancing their nutritional value. These antioxidants neutralize free radicals, preventing oxidative rancidity and maintaining the flavor and quality of oils. Additionally, tocopherols offer health benefits, supporting immune function and reducing inflammation. Their role in preserving oil quality and providing nutritional advantages highlights their importance in the food industry and dietary applications. This study analyzed eight cold-pressed oils (camelina, saffron, flax, hemp, pumpkin, rapeseed, coconut and sesame) to assess the levels of four tocopherol isomers ( $\alpha$ -,  $\beta$ -,  $\gamma$ -, and  $\delta$ -tocopherols) as well as the total tocopherol content. Oil samples were obtained using a hydraulic press, and tocopherol quantification was conducted via high-performance liquid chromatography with fluorescence detection ( $\lambda_{ex}=280$  nm,  $\lambda_{em}=340$  nm). The results revealed significant variation in tocopherol content across the different oils. Sesame oil had the highest total tocopherol content, primarily  $\gamma$ -tocopherol, exceeding 1258 mg/kg, followed by pumpkin oil with over 537 mg/kg of  $\gamma$ -tocopherol. Rapeseed oil exhibited a balanced tocopherol profile with significant amounts of both  $\alpha$ - and  $\gamma$ -tocopherols, totaling around 430-454 mg/kg. Camelina oil, rich in  $\gamma$ -tocopherol, had 479.97 mg/kg, offering strong antioxidant properties but was lower than sesame and pumpkin oils. Hemp oil had a diverse tocopherol profile, including  $\delta$ -tocopherol, with a total tocopherol content ranging from 505 to 507 mg/kg. Flax oil, though low in  $\alpha$ -tocopherol, maintained moderate total tocopherol due to  $\gamma$ -tocopherol, while saffron oil was predominantly  $\alpha$ -tocopherol and coconut oil contained tocotrienols instead of tocopherols. Sesame and pumpkin oils stood out for their high tocopherol levels, whereas rapeseed and hemp oils were noted for their diverse tocopherol profiles. Flax and saffron oils had lower total tocopherol content, dominated by specific tocopherol isomers. These findings emphasize the importance of selecting oils with high tocopherol content for their antioxidant properties, which play a critical role in extending shelf life, maintaining quality, and offering health benefits in dietary application.

**Keywords:** Tocopherols, cold-pressed oils, nutritional value, oil quality

**Acknowledgements:** This work is supported by the Ministry of Education, Science and Technological Development of Republic of Serbia, grant number 451-03-66/2024-03/200222, and Provincial Secretariat for Higher Education and Scientific Research, project No. EB: 142-451-3504/2023-01/01 by the Science Fund of the Republic of Serbia, the European Commission through Twinning Western Balkans project CROPINNO, grant number 101059784 and Project HelEx, grand number 101081974, Project MIDAS, grand number 101082070 funded by EC.

## ANTIOXIDATIVE PROPERTIES OF ORALLY EFFECTIVE SUPEROXIDE DISMUTASE COMBINED WITH WHEAT GLIADIN

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Superoxide dismutase (SOD) is one of the most important antioxidant enzymes in our body. An orally effective form of superoxide dismutase is a new kind of antioxidant supplement used to fight oxidative stress. The examined antioxidant supplement is made of cantaloupe melon extract rich in SOD combined with wheat gliadin (1 mg=1 IU SOD). To test the antioxidant activity of this supplement (250 IU) we used two supernatants after water sonication and centrifugation. This pilot study explored: prooxidative-antioxidative balance (PAB), total concentration of sulfhydryl group (SHG), total antioxidative status (TAS), total oxidative status (TOS), and ad SOD activity. Oxidative stress parameters were determined in 500µL of serum samples of healthy individuals, two supernatants (S1, S2), a solution of Trolox, a water-soluble analog of vitamin E (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) (2mmol/L) and prooxidant terc-buthyl-hydroperoxide TBH (0.25 mmol/L); serum combined with water, serum with TBH, serum with Trolox, serum with supernatants 1 and supernatant 2 (450µL + 50 µL); and serum mixed with equal quantity of supernatant and TBH( 450 µL serum+ 25 µL (S1 or S2)+ 25 µL TBH). For PAB mean value was 137.5±48.1 HKU, maximal value was measured in TBH solution (192 HKU), minimal in supernatant S2 (40.1 HKU), SHG mean value was 0.211±0.072 mmol/L, with maximal value in serum sample with Trolox (0.332 mmol/L), minimal in supernatant S1 (0.097 mmol/L), TAS mean value was 765.7±484.9 µmol/L, maximal value in Trolox solution (2100 µmol/L) and minimal TBH solution (145.5 µmol/L), TOS mean value was 44.3±33.7 µmol/L with maximal value in TBH solution (130.0 µmol/L) and minimal value in supernatant S2 (12.0 µmol/L). SOD mean value was 121.5±26.5 U/L with maximal value in supernatant S2 (171 U/L) and minimal in serum with TBH 98.5 U/L. These results indicate that the supernatant of the examined supplement has good antioxidant potential with minimal value of PAB, TOS, and maximal SOD activity within tested samples.

**Keywords:** superoxide dismutase, oxidative stress, antioxidant activity

## TAGLIATELLE MADE OF WHOLE GRAIN SPELT RICH IN OMEGA-3 FATTY ACIDS

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Studies have shown that *Triticum aestivum* var. spelt (spelt flour) has a delicate nutty flavor and contains nutrients with health benefits (reduces cholesterol levels, and helps eliminate toxins). Products based on spelt are more digestible than those obtained from common wheat. Spelt flour has been used in the production of pasta for a long time; hence spelt pasta is represented on the market and very well accepted by consumers. Pasta is an excellent matrix for incorporating "nutraceuticals" because it is popular with consumers due to its easy handling, storage, and preparation. Nontraditional ingredients could, however, modify the dough's rheological properties or the final product's sensory acceptability. Therefore, balanced formulations and adequate technological processes must be adopted. This investigation aimed to develop a suitable formulation and appropriate process for producing spelt Tagliatelle rich in omega-3 fatty acids characterized by adequate levels of alpha-linolenic acid (ALA) and good sensory acceptability. Vegetable sources such as flaxseed, canola, and soybean oils could increase the omega-3 fatty acids content in the form of ALA. The research team formulated Tagliatelle made of a mixture of whole grain spelt flour and wheat semolina ratio of 80%:20, which is rich in omega-3 fatty acids adding rapeseed oil powder (10% on the amount of mixture flour). Tagliatelle contains 0.605 g ALA in 100 g of dry pasta, hence is achieved and fulfills the requirement for the nutritional statement "rich in omega-3 fatty acids". The content of ALA in cooked Tagliatelle is 0.343 g/100 s.m. Therefore it becomes a source of omega-3 fatty acids. The addition of rapeseed oil powder does not affect the sensorial quality, as well as other Tagliatelle physical characteristics.

**Keywords:** Tagliatelle, whole grain spelt flour, rapeseed oil powder,  $\omega$ -3 fatty acids

**Acknowledgements:** This research was supported by the Innovation Fund of the Republic of Serbia, Innovation vouchers No. 1505 and Ministry of Science Technological Development and Innovation of the Republic of Serbia, No. 451-03-66/2024-01/200222.

## RESISTOME AND MICROBIOTA ANALYSIS OF A THERMAL SPRING USED AS DRINKING WATER IN ANKARA BY SHOTGUN METAGENOMICS

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Thermal springs are natural outlets where hot water and steam from beneath the earth's surface emerge. Thermal waters can be used for irrigation or animal feeding in specific cases, but they are generally preferred for drinking water or industrial uses. Specifically, the microbial composition of thermal waters can significantly influence their intended uses and environmental effects. These environments support diverse microorganisms such as bacteria, fungi, algae, and others, enabling their growth and proliferation. Modern omics technologies, particularly shotgun metagenomics, have been employed to study and reveal microbial communities in extreme environments, as well as resistomes. Recently, resistome in environmental samples has been another trending topic since antibiotic resistance (AR) has been considered a complex problem affecting human and animal health since AR can be transmitted between humans and animals. The fact that thermal waters can be used as irrigation or feeding water makes it important to identify potential resistance genes that may be present in these waters. The thermal spring water investigated in this research is located in the Kahramankazan district of Ankara. The temperature of the water was  $50.4^{\circ}\text{C} \pm 0.07$  and the pH was  $6.04 \pm 0.01$ . For shotgun metagenome analysis, megahit was used for assembly. According to shotgun metagenomics statistics and results, Q20 and Q30 scores are 97.9% and 94.1%, respectively. 100 % fitted five antibiotic resistance genes belonging to Beta-lactam and Aminoglycoside were detected. 29% of unclassified bacteria 27% of *Pseudomonas* spp., and 4% *Methyloburum* spp., have been detected. Additionally, % 31 of *Nitrososphaera gargensis* has been detected in Archaea.

**Keywords:** Shotgun metagenomics, microbiota, resistome, thermal spring, thermal water

**Acknowledgments:** This project was funded by the Başkent University Research Foundation with PRJ\_2023\_04\_0001 project number.

## NUTRITIONAL AND CHEMICAL PROFILE ANALYSIS OF *FUCUS VESICULOSUS* AND *PELVETIA CANALICULATA* BROWN MACROALGAE

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The changes in the production and consumption system require sustainable approaches. The use of macroalgae is proposed as a strategy for more sustainable food systems. The exploration of brown algae has shown significant growth due to their natural substances with a wide range of bioactive properties, which makes them excellent options for the extraction and development of innovative nutraceuticals. In this study, two species from Fucaceae family (*Fucus vesiculosus* and *Pelvetia canaliculata*) were screened for their nutritional, chemical and biological characteristics. The species were nutritionally characterized following AOAC international standards; heat-assisted extraction (HAE) was applied to obtain extracts with bioactive compounds that were identified and quantified by HPLC-ESI-QqQ-MS/MS. *F. vesiculosus* showed more than twice proteins than *P. canaliculata*, being 10.35 g/100 dw and 4.7 g/100 dw, respectively. Regarding the lipid content, *P. canaliculata* had higher content (6.13 g/100 dw) than *F. vesiculosus* (3.92 g/100 dw). Moreover, the chemical characterization of the ethanolic extracts revealed a total of 42 compounds including phenolic acids, flavonoids, phlorotannins and bromophenols. Following extracts profiling, their properties were assessed by means of antioxidant activity, anti-tyrosinase activity and antimicrobial activity against foodborne pathogens. The results showed promising results for scores for *F. vesiculosus*, with 91.75% of  $\alpha$ -tyrosinase inhibition at 0.0625 mg/mL. Overall, these findings provide insight into the potential use of brown algae from Fucaceae family as a source of active compounds, thus diversifying their industrial outcome.

**Keywords:** Fucaceae, macroalgae, nutritional analysis, bioactive compounds, bioactive properties

**Acknowledgements:** The research leading to these results has been funded by Xunta de Galicia for supporting the postdoctoral fellowship of A.G. Pereira (IN606B-2024/011) and the predoctoral fellowship of P. Barciela (ED481A-2024-230). The authors gratefully acknowledge national funding from the FCT, Foundation for Science and Technology, through the individual research fellowships of A.O.S. Jorge (2023.00981.BD).



## UTILIZATION OF RAPESEED PROTEINS IN SPORTS NUTRITION FORMULATIONS

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Rapeseed press cake, a byproduct of oil production, contains a significant amount of high-quality proteins, making it a promising ingredient for sports nutrition formulations. This study explores the potential of rapeseed proteins as a sustainable alternative to conventional sources like whey and soy in sports nutrition products. Unlike many other plant-based proteins, rapeseed proteins contain all essential amino acids, eliminating the need to mix them with other sources to achieve a complete amino acid profile. Through extraction and purification processes, the proteins were analyzed for their amino acid composition, digestibility, and functional properties, such as solubility and emulsification. Thanks to their techno-functional properties, these proteins have great potential to be incorporated into various food products. Their excellent emulsifying properties make them ideal for formulations like shakes and recovery formulations designed for athletes, while their complementarity with cereal proteins makes them suitable candidates for various bakery and confectionary products, such as protein bars, breads, and flour mixtures. Rapeseed proteins could also support muscle repair and growth, offering substantial benefits to athletes engaged in high-intensity training. Obtaining high-purity rapeseed proteins remains a significant technological challenge, which is likely the reason for their limited commercialization in sports nutrition formulations. On the other hand, their sustainable production and environmental benefits position them as an attractive option for eco-conscious consumers. This research underscores the feasibility of using rapeseed proteins in sports formulations, contributing to both the nutritional needs of athletes and the sustainability goals of the food industry.

**Keywords:** rapeseed proteins, plant proteins, sport nutrition

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovations, Republic of Serbia (Contract No. 451-03-66/2024-03/200222), as well as by the Science Fund of the Republic of Serbia, Grant No 6673, PROTein from Rapeseed Oil Processing Waste: Application in Food and Wastewater Treatment – PROTOPOWER.



## POMEGRANATE PEEL EXTRACT AS A NATURAL ANTIMICROBIAL AGENT TO REDUCE FOOD WASTE AND COMBAT ANTIBIOTIC RESISTANCE

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Pomegranate (*Punica granatum* L.) is renowned for its numerous health benefits, including antibacterial, antioxidant, anti-inflammatory, and anti-diabetic properties. The edible part of the pomegranate, rich in nutrients and bioactive compounds, is widely popular in various food industries, particularly for the production of juices, confections, and as a food additive. However, the peel of the pomegranate, often considered agricultural waste, has also demonstrated significant therapeutic potential. This study explores the antimicrobial properties of pomegranate peel extract (PoPEX) against various bacterial strains, including both Gram-positive and Gram-negative bacteria. The results indicate that PoPEX exhibits strong antimicrobial activity, especially against Gram-positive bacteria such as *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Enterococcus faecalis*. The minimum inhibitory concentrations (MICs) for these bacteria were significantly lower compared to those for Gram-negative bacteria, highlighting the extract's higher efficacy against Gram-positive strains. The antimicrobial effect of PoPEX can be attributed to its high content of phenolic compounds, such as punicalagin, punicalin, gallic acid, and ellagic acid. By utilizing pomegranate peel, this study not only addresses the problem of food waste but also presents a natural alternative to combat antibiotic resistance, potentially reducing reliance on synthetic antibiotics. The findings support the potential use of pomegranate peel extract in developing new antimicrobial agents to tackle the growing challenge of antibiotic resistance.

**Keywords:** Pomegranate peel extract, antimicrobial activity, food waste reduction

## INSECT OIL AS A POTENTIAL SUBSTITUTE FOR FISH OR VEGETABLE OIL IN THE DIETS OF AFRICAN CATFISH HYBRID

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Alternative sources of dietary fish oil are necessary for the growth of the aquaculture industry. In this study, we aimed at examining the potential of black soldier fly (*Hermetia illucens*) larvae oil as a substitute for fish and/or vegetable oil in the diets of African catfish hybrids (*Clarias gariepinus* × *Heterobranchus longifilis*). Three isonitrogenous and isolipidic diets were formulated and included a reference diet (fish oil and rapeseed oil, 1:1 ratio) and two experimental diets: IO50, in which insect oil (IO) replaced 50% of the fish oil and rapeseed oil, and IO100, containing 100% IO. The feeding trial, which lasted seven weeks, involved a total of 900 fish juveniles (average initial weight: 29.1 ± 1.69 g) randomly distributed into the three dietary groups in a recirculation aquaculture system. At the end of the trial, major differences were not observed in fish growth, nutrient retention, body indices, or survival between the dietary fish groups. The whole-body moisture, crude protein, crude lipid, and ash were also similar between the groups. However, the content of whole-body monounsaturated fatty acids (MUFA), eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA), and n-3 polyunsaturated fatty acids (n-3 PUFA) significantly decreased while Arachidonic acid (ARA) content increased with an increase in IO inclusion in the diets. Similar trends in the contents of ARA and EPA were also observed in fish liver, while DHA indicated no major difference between the dietary groups. The hydrolytic activities of intestinal enzymes and the antioxidation capacity of the fish liver were not significantly different between fish fed insect oil-based diets and those fed the control diet. These results indicated that potential benefits similar to those provided by dietary fish and/or vegetable oil can be achieved when black soldier fly larvae oil is used as an ingredient in the practical diets of African catfish hybrid.

**Keywords:** Catfish, African catfish, Insect oil, Black soldier fly, Fatty acid

**Acknowledgements:** This work was supported by the Stipendium Hungaricum Scholarship Program of the Hungarian Government. Authors are thankful to the technical staff of MATE for their support in fish husbandry (Csaba Weber) and laboratory sample analyses (Judit Molnar).

## HEALTH PROMOTING EFFECTS OF *HELICHRYSUM ITALICUM* – FINDINGS FROM *IN VITRO* AND *IN VIVO* STUDIES

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*Helichrysum italicum* (HI) is a Mediterranean plant with well-reported use in traditional medicine for a variety of applications, including digestive and liver disorders, parasitic intestinal infections, wound healing and stomach pain. Despite its traditional use, there is a lack of scientific data to evaluate its effects. We have analysed the biological activities of infusions prepared from HI in cell lines and performed a transcriptomic analysis of human colon cells exposed to the infusion. *In vivo*, the effects of acute and regular consumption of HI infusions were also studied. The infusion had substantial antioxidant potential *in vitro* and showed effective protection against induced oxidative stress in different cell lines. Transcriptomic analysis, where enriched pathways and differentially expressed individual genes were analysed, suggests that the main mode of HI action is in wound healing. In addition to its indirect prevention of diseases resulting from the impaired intestinal barrier integrity, HI also has a direct effect on inflammatory and metabolic processes. In a pilot study with 10 individuals, a single ingestion of HI infusion increased fat oxidation and resting energy expenditure. This was confirmed in cell lines, where genes involved in beta-oxidation were upregulated in HI-treated hepatocytes. A 28-day randomised comparative intervention showed that consumption of HI had a beneficial effect on anthropometric characteristics; significant reductions in body weight, body mass index, visceral and total body fat were observed. Also, LDL levels were lowered and serum antioxidant properties were improved. The observed reduced gastrointestinal symptoms and inflammation can at least partially be explained with the improvement of intestinal wall integrity, since we found up-regulation of tight junctions. Overall, our data show health-promoting effects of HI infusions, suggesting that its consumption could be a simple and readily available profitable habit for people with increased body mass or a deregulated lipid profile.

**Keywords:** metabolic syndrome, antioxidative potential, inflammation

## THE EFFECTS OF USING DIFFERENT NON-CONVENTIONAL PROTEIN SOURCES ON THE ACTIVITY OF ANTIOXIDANT ENZYMES IN THE COMMON CARP

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In response to the shortage of fishmeal and its high price, non-conventional protein sources are being developed. Mealworms, earthworms and zooplankton are good substitutes for fishmeal due to their high protein and fatty acid content. Some previous studies have investigated the effects of using alternative protein sources on the growth parameters of fish. However, there is a lack of information on the effects of non-conventional feed on the activity of antioxidant enzymes. The aim of this study was to investigate the effects of replacing fishmeal with different non-conventional protein sources on the activity of antioxidant enzymes in carp. The fish were obtained from the Centre for Fisheries and Applied Hydrobiology "Mali Dunav" of the Faculty of Agriculture. Three different feeds (mealworms, earthworms and zooplankton) were fed instead of fishmeal for 90 days under controlled conditions in a flow-through tank. The activity of antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT) and glutathione S-transferase (GST) was determined using standard spectrophotometric assays. The results for SOD showed no differences between the fish fed different diets. The fish fed with the control diet had the highest CAT value, while the fish fed with the mealworm diet had a higher CAT value than the other samples. Both enzymes play an important role in neutralising reactive oxygen species (ROS) and contribute to improving the stress response. In contrast, there were no differences between the control fish and the fish fed non-conventional feed, with the exception of the zooplankton-fed fish, which had the lowest GST levels. Our results suggest that the use of non-conventional protein sources in the fish diet can stimulate the activity of the enzymes SOD and CAT, which has an impact on health performance.

**Keywords:** *Cyprinus carpio*, non-conventional diets, SOD, CAT, GST

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant number 451-03-65/2024-03/200116 and 451-03-66/2024-03/200007).

## INVESTIGATION OF THE INFLUENCE OF A COMPLETE REPLACEMENT OF FISHMEAL BY ANIMAL PROTEIN SOURCES ON PRODUCTION PERFORMANCE AND THE CONDITION FACTOR

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The feed industry has recently been faced with the problem of producing commercial fish feeds such as the most commonly used fishmeal and fish oil. This problem affects the aquaculture sector, which uses fishmeal as the main source of protein for fish farming. To overcome these problems, many plant and animal protein sources are used to replace fishmeal. In addition, insect meal is rich protein source, whose protein can serve as a future protein source for fish feed. On the other hand, insects are approved by the European Commission for use in animal feed. The aim of this study was to investigate the possibility of completely replacing fishmeal with alternative protein sources, such as zooplankton (ZO), mealworm (TM) and earthworm (EF), and their effects on the production parameters of common carp. The experiment was conducted in a cage system at the Radmilovac experimental farm of the Faculty of Agriculture, University of Belgrade. Body weight gain (BWG), specific growth rate (SGR), feed conversion ratio (FCR), and condition factor (CF) were calculated using known equations. The highest BWG value was achieved by the fish fed with FM, followed by TM and EF, while the lowest BWG value occurred in the fish fed with ZO. The same trend was observed for SGR. For the other parameters of production efficiency such as FCR, the lowest FCR value was obtained from fish fed with TM, while the highest value was achieved by fish fed with ZO. The results for CF were typical for fish species and ranged from 1.8 to 2. Considering the results for production efficiency, the alternative animal protein sources including mealworms and earthworms can successfully replace fishmeal in aquafeeds.

**Keywords:** common carp, growth parameters, condition factor, insects

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant number 451-03-65/2024-03/200116, 451-03-66/2024-03/200007 and 451-03-66/2024-03/200222).

## AGRONOMIC AND BIOCHEMICAL PROPERTIES OF AUTUMN GARLIC FROM IFVCNS COLLECTION

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Garlic (*Allium sativum* L.) is one of the most important *Allium* species considering global production, consumption, and numerous human health benefits, mainly due to its diverse bioactive compounds and strong antioxidant properties. From the garlic collection of the Institute of Field and Vegetable Crops (IFVCNS), 20 accessions were selected for analysis with the aim to assess the variability of agronomic and biochemical traits. The experiment was set up during two growing seasons (2017/18 and 2018/19), on the Rimski šančevi site. A sample of 30 bulbs was analyzed for the following agronomic traits: bulb mass (BM), number of cloves per bulb (NC) and clove mass (CM). Lyophilized garlic cloves were further used for biochemical analyses of total phenolics (TP), total flavonoids (TF) and antioxidant activity (DPPH). The average value of BM within the collection was 46,1 g while the values varied from 26,6 g to 64,0 g. The smallest NC in collection was 6,3 and the highest was 19, with the average value of 10,4. Variability in collection was also observed for CM ranging from 2,6 to 7,7. The average value for this trait was 4,6. Greater variation among samples in the collection was found for biochemical properties. Values of total phenolics varied from 0,49% to 3,09% with average value of 1,53% while total flavonoids ranged from 0,16% to 0,77% with average value of 0,45%. Antioxidant activity also varied significantly among the samples, with the lowest value of 0,93 and the highest of 3,99 µg AAE/g, while average value was 1,91 µg AAE/g. The results of the study indicated the diversity within garlic collection based on agronomic and biochemical traits. Variability of garlic collection in analyzed traits provides an opportunity for selection of genotypes with enhanced bioactivity and higher yield potential, as one of the most important breeding goals.

**Keywords:** yield components, biochemical traits, variability

**Acknowledgements:** This research was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant number: 451-03-66/2024-03/200032. The authors are grateful to Boris Popović, PhD, Full Professor and Bojana Blagojević, PhD, Teaching Assistant (University of Novi Sad, Faculty of Agriculture, Novi Sad, Serbia) for the help with lyophilization of plant material used in performed biochemical analyses.

## **SUBCRITICAL WATER EXTRACTION OF DANDELION (*Taraxacum officinale*) FLOWERS: INFLUENCE OF TEMPERATURE ON POLYPHENOLS CONTENT AND ANTIOXIDANT ACTIVITY**

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Dandelion (*Taraxacum officinale*) has a long history in traditional herbalism worldwide. Described as a non-toxic, the herb has been consumed in various forms as a valuable source of nutrients, minerals and vitamins, the consumption of which may help to prevent or reduce the risk of complex diseases such as cancer, obesity, arthritis, hepatitis, cardiovascular and gastrointestinal disorders. All parts of the dandelion herb are edible and contain flavonoids, phenolic acids, alkaloids and terpenes, with the best studied extracts being from leaves and roots. The most abundant phenolic acids are hydroxycinnamic acid derivatives, especially chicoric acid, chlorogenic acid and caffeic acid. Luteolin and its glucosides are mainly enriched in extracts from dandelion leaves and flowers. The aim of this study was to investigate the influence of temperature of subcritical water extraction of dandelion flowers in the interval of 110-160° C. The parameters analysed were total phenolic content (TPC), total flavonoid content (TFC), total antioxidant capacity (TAC) and the DPPH radical scavenging activity, determined by UV-spectrophotometry. The highest TPC (36.37 mg GAE/g DW) and TAC (76.80 mg AAE/g DW) was noted in extracts obtained at 140° C, while the highest TFC of 10.95 mg RE/g DW was observed when extraction was performed at 130° C. As for DPPH scavenging activity, the highest value was for the extraction temperature of 110° C (0.906 mg AAE/g DW), and the lowest for the extract obtained at 160° C (0.718 mg AAE/g DW). The results of this study suggest that dandelion flower extracts obtained at moderate temperatures (130-140° C) with subcritical water have the highest polyphenol content and antioxidant activity.

**Keywords:** Dandelion flowers, subcritical water extraction, polyphenolic content, antioxidative activity

**Acknowledgements:** The Science Fund of the Republic of Serbia (project *In situ* pollutants removal from waters by sustainable green nanotechnologies – CleanNanoCatalyze, Grant No 7747845) and the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (project 451-03-66/2024-03/200134) are acknowledged.

## TOTAL PHENOLIC AND FLAVONOID CONTENT IN *BOSWELLIA SERRATA* RESIN EXTRACTS OBTAINED WITH SUBCRITICAL WATER

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*Boswellia serrata* is a tree found mainly in arid regions of India and its oleoresin, internationally known as frankincense, is used in Ayurvedic, traditional Arabic and Chinese medicine. This gum resin contains 15-20% of boswellic, lupeolic and pentacyclic triterpene acids, of which the boswellic acids (beta-boswellic acid, keto-beta-boswellic acid and acetyl-11-keto-beta-boswellic acid) have been shown to have anti-inflammatory, anti-cancer and anti-diabetic properties and are used in modern pharmaceutical industry. In addition to its ability to prevent and treat various diseases (rheumatoid arthritis, osteoarthritis, Chron's disease, ulcerative colitis and asthma), other biological functions of *B. serrata* resin should not be neglected. The aim of this study was to analyse extracts of *B. serrata* resin obtained by subcritical water at different temperatures (110° C - 190° C) for phenolics and flavonoids contents. The total phenolic content (TPC) was determined by UV-spectrophotometry using the Folin-Ciocalteu method. The total flavonoid content (TFC) was also determined by UV-spectrophotometry using a colourimetric method with AlCl<sub>3</sub>. With increasing extraction temperature, the TPC increased from 3.76 mg GAE/g resin at 110° C to 13.78 mg GAE/g resin at 190° C. The highest TFC was observed in the extract obtained at 170° C (8.56 mg RE/g resin). The results of this study suggest that extracts of *B. serrata* resin obtained with subcritical water are rich sources of bioactive compounds that can be used in dietary supplements and functional foods.

**Keywords:** *Boswellia serrata*, subcritical water extraction, total phenolic content, total flavonoid content

**Acknowledgements:** The Science Fund of the Republic of Serbia (project *In situ* pollutants removal from waters by sustainable green nanotechnologies – CleanNanoCatalyze, Grant No 7747845) and the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (project 451-03-66/2024-03/200134) are acknowledged.



## **SURVIVABILITY OF MICROENCAPSULATED *LACTOBACILLUS PLANTARUM* DURING KOMBUCHA FERMENTATION**

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Kombucha is a traditional beverage, which is usually prepared by fermenting sweetened black tea using a symbiotic consortium of acetic acid bacteria and yeast. Kombucha exhibits several functional properties such as antioxidant, antimicrobial, anticancer, etc. Most often kombucha can not be labeled as a probiotic product, since the number of probiotic microorganisms usually does not reach the specified criteria. Lactic acid bacteria (LAB) are major microorganisms used for probiotic purposes. For LAB to exhibit health benefits, they should be able to survive sometimes harsh conditions during food processing, fermentation, storage, and passing through the human gastrointestinal tract. Several methods of microencapsulation of probiotic microorganisms are widely used to improve their viability in specified conditions. In this study, the probiotic strain *Lactobacillus plantarum* ATCC 14917 was encapsulated in three materials: pectin, inulin, and a mixture of maltodextrin and glucose by emulsion method and separately added at the beginning of kombucha fermentation. The survivability of *L. plantarum*, as well as chemical (pH and titrable acidity) and microbiological (number of yeasts and acetic acid bacteria) parameters, were monitored during fermentation. The number of LABs was also determined during the storage of the finalized product. Encapsulation efficiency (EE) was measured as follows, 93.17 % for pectin, 86.34 % for inulin, and 88.68 % for maltodextrin and glucose mixture. Pectin demonstrated the best potential for microencapsulation of *L. plantarum* since the bacteria were present in the beverage until the end of fermentation and during 4 days of storage in an amount of approximately  $10^4$  CFU/ml. As for the remaining two used materials, LAB was present until the third day of fermentation, after which they were no longer detected. The obtained results show that with further research, pectin could be a promising agent for the microencapsulation of probiotic bacteria to obtain kombucha beverages with probiotic characteristics.

**Keywords:** kombucha, microencapsulation, probiotics, fermentation

**Acknowledgments:** This research was funded by the Republic of Serbia program of the Ministry of Science, Technological Development and Innovation, grant numbers: 451-03-66/2024-03/200134 and 451-03-65/2024-03/200134.

## HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC) METHOD FOR THE INVESTIGATION OF BIOACTIVE COMPOUNDS IN TWO MICROGREEN VARIETIES

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Nowadays, we are facing numerous challenges related to nutrition and human health. Pathological conditions caused by inadequate nutrition are particularly prevalent in urban areas and developed countries. The daily diet of the modern population is characterized by insufficient intake of minerals, nutrients, vitamins, and phytochemicals. It is estimated that by 2050, 80% of the world's food will be consumed in cities, which emphasizes the importance of plant production near urban areas and within urban environments. One of the potential sources of a large number of bioactive compounds are microgreens. They are known for their nutritional value and bioactivity and are used in culinary arts for their flavor, texture, and visual appeal. Microgreens are young plants harvested after the first true leaves develop from seeds of vegetables, herbs, and industrial plants, easily grown indoors. They are known for their nutritional value and are used in culinary arts for their flavor, texture, and visual appeal. This research investigates the bioactive content of two Brassicaceae microgreen plants (Sango radish and kale) using HPLC with a DAD/UV detector, including the content of phenolic acids, carotenoids, and vitamin C. Microgreens in question were lyophilized, homogenized as a powder and stored until further analyses. Specific extractions were performed for different assays. Sango radish and kale showed the highest presence of p-hydroxybenzoic acid (510.70mg/100g), and sinapic acid (600mg/100g), respectively.  $\alpha$ - and  $\beta$ - carotene and lutein were found in both microgreen samples and zeaxanthin was only present in kale. While lutein was the dominant carotenoid in Sango radish (511.23 mg/100g), in kale the highest carotenoid content amounted to  $\alpha$ -carotene (1238.68 mg/100g). Ascorbic acid was three times higher in kale compared to radish microgreens (21.31- 66.05 mg/100g). The obtained results highlight the health-promoting properties of investigated microgreens and the importance of their daily intake and expanded future use.

**Keywords:** HPLC, microgreens, phenolics, carotenoids, ascorbic acid

**Acknowledgements:** This research study was funded by the Provincial Secretariat for Higher Education and Scientific Research of Autonomous Province Vojvodina (Serbia) within the project "New ecological phytopreparation for antifungal and antioxidant treatment of selected fruits and vegetables—EcoPhyt" (grant No. 142-451-2732/2023-01/01) and by the Ministry of Education, Science, and Technological Development of the Republic of Serbia (grant No. 451-03-66/2024-03/200134).

## FRUIT PRODUCTS AND ITS POTENTIAL PREVENTIVE PROPERTIES AGAINST CHRONIC NON-COMMUNICABLE DISEASES

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Oxidative stress is a main cause for development of chronic non-communicable diseases. It can be prevented by antioxidants, especially those from berry fruit and its products. Blueberry is rich source of nutrients and biologically active compounds, but unfortunately it is not available in the fresh form during the whole year. Blueberry wine is a product obtained after alcoholic fermentation of blueberries, and during this process in wine are preserved all thermo labile biologically active compounds. This study is aimed to show phenolic profile, antioxidant properties and activity of fruit wine on enzymatic systems in vitro and lipid peroxidation. Blueberry wines were produced in different controlled microvinification procedures. Phenolic profile was analysed by UPLC/MS-MS system, while total phenolic content (TPC) determined by Folin-Ciocalteu method (mg GAE/L). Antioxidant activity was detected by FRAP (mmol/L Fe<sup>2+</sup>) while antiradical by usage of stabile DPPH radical (expressed as IC<sub>50</sub>). Level of lipid peroxidation (malondialdehyde (MDA) level) and activity of enzymes of antioxidant protection superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx) were evaluated on synaptosomes. Synaptosomes were isolated from the brain of Wistar albino rats. Microvinification procedure significantly affected on phenolic profile, antioxidant and antiradical activity. Blueberry wines showed ability to activate enzymes of antioxidant protection and decrease MDA level in synaptosomes in which was experimentally induced oxidative stress. Among phenolic compounds were the most predominant phenolic acids and flavonoids. The TPC as well as FRAP values were significantly different between different microvinification. In oxidative stress induced synaptosomes which were treated with blueberry wine values for MDA were decreased while SOD, GPx and CAT activities were increased. As a rich source of antioxidants blueberry wine could prevent oxidative stress which is responsible for development of chronic non-communicable disease.

**Keywords:** blueberry wine, oxidative stress, phenolic compounds, antioxidant properties, enzymes of antioxidant protection

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia through Grant Agreement with University of Belgrade-Faculty of Pharmacy No: 451-03-66/2024-03/200161.

## OPTIMIZATION OF THE MICROWAVE-ASSISTED EXTRACTION OF BIOACTIVE COMPOUNDS FROM *Satureja montana* L. BY VARYING EXTRACTION SOLVENT

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Numerous reports have been published on the antioxidant activity of bioactive plant extracts, strongly inspired by an increasing consumer interest in 'natural' food additives (e.g. preservatives, antioxidants, colours and flavours), as an alternative to the use of synthetic products. The genus *Satureja montana* L. belongs to the Lamiaceae family, and comprises 30 species, whose centre of distribution is located in the eastern part of the Mediterranean area. The presented study aimed to optimize polyphenol extraction from *S. montana* (winter savory) grown at the Institute for medicinal plant research „Dr Josif Pančić“ in Pančevo, Republic of Serbia experimental field, as the antioxidant activity of non-volatile plant extracts has been attributed to the presence of phenolic compounds. All extracts were produced using the microwave-assisted extraction method in the microwave extractor (SR-15 rotor segment of the Milestone Ethos X microwave extractor) by varying the solvent (water, ethanol (30%, v/v; 70%, v/v)), while the process temperature and extraction time were adjusted at 100 °C/1 min and controlled automatically by the internal software. The solid-to-solvent ratio was defined before analysis and fixed at 1:40. The obtained extracts were characterized regarding the total phenol content (TPC), total flavonoid content (TFC), and *in vitro* antioxidant activity measured by DPPH (2,2-Diphenyl-1-picrylhydrazyl) radical scavenging activity. The TPC and TFC, in respect to the implemented solvent, were between 0.897 and 1.004 mg GAE (gallic acid equivalent)/g and 0.226 to 0.303 mg CE (catechin equivalent)/g, respectively. The highest antioxidant activity (determined by DPPH-IC<sub>50</sub>) were obtained in the extracts where the ethanol (70%, w/v) was used as a solvent, approximately 8.95 µg/mL. The performed investigation revealed that winter savory extracts obtained by an advanced extraction technique represent a source of natural antioxidant substances with potential application in pharmaceutical and food industries.

**Keywords:** Winter savory, polyphenols, flavonoids, free radicals

**Acknowledgements:** The authors acknowledge their gratitude to the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, contract number 451-03-66/2024-03/200003 and 451-03-47/2024-01/200019.

## DESIGN AND PREPARATION OF ASTAXANTHIN-TARGETED DELIVERY SYSTEMS FOR COLITIS ALLEVIATION

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Orally targeted strategy of anti-inflammatory agents has attracted tremendous attention for reducing highly health-care costs and enhancing the intervention efficiency of ulcerative colitis (UC). Herein, we developed a new kind of sequence-targeted astaxanthin nanoparticles for UC treatment. Astaxanthin nanoparticles were firstly designed by self-assembly method using (3-carboxypentyl) (triphenyl) phosphonium bromide (TPP)-modified whey protein isolate (WPI)-dextran (DX) conjugates. Subsequently, lipoic acid (LA) modified hyaluronic acid (HA) was coated on the surface of the nanoparticles by double emulsion evaporation method. Exhilaratingly, the constructed sequence-targeted astaxanthin nanoparticle exhibited excellent macrophages and mitochondria targeting ability, with a Pearson's correlation coefficient of 0.84 and 0.92, respectively. In vivo imaging elucidated an obvious accumulation of the sequence-targeted nanoparticles in colon tissues in UC mice. Meanwhile, the reduction stimulus release features of astaxanthin were observed in the presence of 10 mM of glutathione (GSH) at pH 7.4. Most importantly, in vivo experiments indicated that sequence-targeted astaxanthin nanoparticles could markedly alleviate inflammation by moderating the TLR4/MyD88/NF- $\kappa$ B signaling pathway. What's more, the composition of gut microbiota and the production of short chain fatty acid were also improved upon the uptake of sequence-targeted astaxanthin nanoparticles. Our results suggested this novel astaxanthin nanoparticles, which showed sequence-targeted ability and reduction response feature, could be exploited as a promising strategy for effective UC treatment.

**Keywords:** Oral administration, Ulcerative colitis, Astaxanthin, delivery system

**Acknowledgements:** This work was supported by the National Science Fund for Distinguished Young Scholars of China (31925031) and Liaoning Revitalization Talents Program (XLYC1902120).

## SESAMIN ALLEVIATES IGE-MEDIATED FOOD ALLERGIES BY SUPPRESSING SPLEEN TYROSINE KINASE ACTIVATION

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IgE-mediated food allergy is a type I immune response triggered by allergens in food. The increasing prevalence of food allergies worldwide presents a significant health concern. Natural phenolic compounds have attracted considerable attention for their potential to alleviate allergies and modulate immune imbalance. However, the diverse structural nature of phenolic compounds results in variations in their specific effects. To address these challenges, we used computational chemistry to conduct preliminary screening of anti-allergenic phenolic compounds and to elucidate their mechanisms. Spleen tyrosine kinase (Syk) plays a critical role in amplifying and propagating signals within the FcεRI and BCR signaling pathways, making it a promising target for suppressing allergic responses. In this study, a total of 300 phenolic compounds were examined for their ability to interact with the ATP-binding pocket of Syk using computational chemistry approaches (i.e., virtual screening, molecular docking, and molecular dynamics simulations). Sesamin and its metabolites, SC-1 and SC-2, were identified as potential Syk inhibitors. Sesamin significantly inhibited the degranulation of RBL-2H3 cells, resulting in 54% inhibition of β-hexosaminidase release and a 58.45% reduction in histamine levels compared to the control. In vivo, sesamin reduced Evans blue dye leakage and tissue edema in the ears of mice with passive cutaneous anaphylaxis. Additionally, it alleviated allergic symptoms in mice with active systemic anaphylaxis, as demonstrated by improvements in body weight, rectal temperature, spleen weight ratio, and clinical scores. Sesamin also suppressed the synthesis and release of immunoglobulins and pro-inflammatory factors, restored the balance of T helper cells differentiation in the spleen, and inhibited Syk phosphorylation, lowering the phosphorylation levels of downstream ERK, P38, and P65. Overall, these findings suggest the potential of sesamin as a natural compound for alleviating IgE-mediated food allergies.

**Keywords:** food allergy, phenolic compounds, mast cell degranulation, spleen tyrosine kinase

**Acknowledgments:** This research received funding from Incubation Program of Youth Innovation in Shandong Province.

**A COMPARATIVE STUDY OF THE ANTIOXIDATIVE AND LIPID-LOWERING EFFECTS OF ARTICHOKE LEAVES EXTRACTS OF STUNJAN ARTICHOKE AND THE TWO MOST COMMON ITALIAN ARTICHOKE VARIETIES ROMANESCO AND VIOLETO DI ROMAGNA**

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In recent years, the artichoke has seen renewed interest in functional foods. While the effects of biotic factors, genotype, and plant parts on the polyphenolic concentration and bioactivity have been extensively studied in many Italian varieties, the Slovenian Strunjan artichoke has not yet been studied at all, neither of capitula nor leaves. Since artichoke leaves extracts (ALE) are traditionally used and since phenolic compounds in the leaves has been documented as an active principle of this plant, we aimed to compare the Strunjan ALE with commercially more common available Romanesco and Violeto di Romagna artichokes in terms of cytotoxicity and protective effect from oxidative stress induction and also in terms of lipid-lowering properties. Romanesco ALE exhibited the highest radical scavenging activity (EC<sub>50</sub> for Romanesco ALE was 13 µg/mL, for Strunjan ALE 18.5 µg/mL and for Violeto di Romagna 20 µg/mL); however, Strunjan ALE was more efficient in protecting different cell lines, especially primary colon cells CCD112CoN, from induced oxidative stress. Additionally, Strunjan ALE and Violeto di Romagna exerted lipid-lowering activity in HepG2 cells, while Romanesco ALE did not.

**Keywords:** artichoke leaves extracts, antioxidative potential, phenolic compounds, lipid-lowering activity



## JUNEBERRY (*AMELANCHIER LAMARCKII*) POMACE AS SOURCE OF BIOACTIVE COMPOUNDS; EFFECT ON GASTROINTESTINAL DIGESTION AND ANTIMICROBIAL PROPERTIES

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Juneberry is a deciduous shrub native to the northern prairies and plains of North America, the fruit belongs to the *Rosacea* family. The berries are sweet, edible, quite seedy, and are an excellent source of bioactive compounds such as anthocyanins, flavonols, procyanidins and phenolic acid. In this work, the bioaccessibility of compounds in Juneberry (*Amelanchier Lamarckii*) pomace was evaluated by measuring the relevant bioactives, before and after gastrointestinal digestion *in vitro*, by the INFOGEST 2.0 protocol. The extraction was carried out by maceration with ethanol/water (40:60, v/v) for two hours at room temperature. The chemical composition, antioxidative and antimicrobial effect were evaluated; in addition, the profile of phenolic compounds was also determined by HPLC-MS. Moreover, the extracts were also incorporated into tagliatelle pasta and the goal was to determine if these compounds could improve colour parameters and their effect on nutritional composition. Mean and standard deviation of triplicate of total phenolic compounds before and after gastric and intestinal phases of digestion of the ethanolic juneberry pomace extracts were  $210.1 \pm 0.8$ ,  $170.1 \pm 3.8$ ,  $72.9 \pm 0.3$  mg/g, respectively. It was observed that after simulated digestion, the total phenolic and flavonoid content of juneberry pomace extracts decreased significantly during the gastric and intestinal phases compared to the crude extracts. With regard to antioxidant activity, there was a reduction of all studied parameters after gastric digestion. Due to the high content of natural colorants and bioactive compounds in the pomace of *Amelanchier Lamarckii*, this species could be useful for the food industry in the future, as potential ingredients for functional food formulations, flavours or colourants.

**Keywords:** juneberry pomace, antioxidative, bioaccessibility, phenolic compounds

**Acknowledgements:** This study was financially supported by the Slovenian Research and Innovation Agency (research program P1-0005 "Functional foods and food supplements").



## INFLUENCE OF THE COMPOSITION OF FEED MIXTURES IN CHICKEN FATTENING ON THE FATTY ACIDS PROFILE AND NUTRITIONAL INDICES IN BREAST MEAT

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Omega-3 eggs and omega-3 chicken meat can be placed on the food market as functional products from poultry production. The aim of this research was to determine the influence of designed feed mixtures used in chicken fattening on the fatty acid profile and lipid quality in breast meat. For research purposes, 700 Ross 308 chickens were divided into two groups, control and experimental (K and P). The fattening lasted 42 days. In the first three weeks, all chickens consumed the same starter mixture (23.45% crude protein), and in the last three weeks the finisher mixture (19.34% crude protein), which differed by the type of oil used. The control group received a mixture with 5% sunflower oil, and the experimental group received a mixture with 1.0% fish + 3.0% linseed + 1.0% rapeseed oil. The composition of the mixtures influenced the profile of fatty acids in breast meat. A significantly higher ( $P < 0.05$ ) content of  $\sum$ MUFA (35.95% : 30.35%) and  $\sum$ n-3 PUFA (12.52% : 0.80%) was determined in the P group, while in the K group significantly higher ( $P < 0.05$ ) content of  $\sum$ n-6 PUFA (38.62% : 22.38%) was found. The more favourable content of n-3 PUFA in P group compared to the K group influenced the  $\sum$ n-6/n-3 PUFA ratio, which was better in the P group (1.79 : 48.27;  $P < 0.001$ ). Atherogenic ( $P > 0.05$ ) and thrombogenic ( $P < 0.001$ ) indices were lower in the P group, while the hyper/hypocholesterolemic ratio was higher in the P group, but the difference was not significant ( $K=3.397:P=3.444$ ;  $P > 0.05$ ). The nutritional value index was uniform in both groups ( $P > 0.05$ ). The results indicate that the use of designed feed mixtures in chicken fattening can increase the content of omega-3 fatty acids in breast meat, with the recommendation that such meat is desirable in human nutrition because can reduce the possibility of blood clots occurrence in humans.

**Keywords:** chickens, breast meat, fatty acids, nutritional indices

**Acknowledgments:** The data for the work are part of the research project "Profile of fatty acids in oils and animal samples", which is carried out as part of the IT ProFAOAS activity.

## ENHANCING YOGURT WITH FREEZE-DRIED BOVINE COLOSTRUM POWDER

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Colostrum is the initial milk secreted by mammals following parturition, characterized by its dynamic and variable composition and physicochemical properties. Due to its high concentration of immunoglobulins, antimicrobial peptides, and growth factors, colostrum plays a crucial role in the nutrition, growth, and development of newborn infants, significantly contributing to their immunologic defense. The process of freeze-drying is an effective method of preserving the bioactive compounds present in colostrum, making it a convenient and stable ingredient for use in food fortification. This study investigated the feasibility of fortifying yogurt with 1-5% freeze-dried bovine colostrum powder from the first day of lactation, to enhance its nutritional value and properties. Fortified yogurts were evaluated for their microstructure, rheological properties, water holding capacity and acidification kinetics. The objective was to determine the impact of freeze dried colostrum powder on these attributes, ensuring that the fortification process maintains or improves yogurt quality. Results indicated that the presence of colostrum did affect the acidification process by the starter culture. Rheological analysis of yogurts revealed a slight increase in apparent viscosity, contributing to a creamier texture. Additionally, the water retention capacity of fortified yogurts was significantly enhanced, ensuring better consistency. In conclusion, fortifying yogurt with freeze-dried bovine colostrum powder, in all selected concentrations, enhances its physicochemical and rheological properties, making it a superior functional dairy product. This research contributes to the advancement of functional foods, presenting innovative opportunities for health-enhancing dairy products.

**Keywords:** colostrum, freeze drying, fortified yoghurt, rheology

**Acknowledgements:** This project was co-funded by Greece and European Union (Project title: Yogurt product from lyophilized colostrum, M16SYN2- 00077).

*Food Quality and Safety*

## POTENTIAL BIOAGENTS FOR MAIZE SEED BIOPRIMING

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Chemical seed treatments are still the most common tool against seed and soil-borne plant diseases. Due to the negative impact on the environment, human and animal health, it is necessary to find other solutions. Seed biopriming is an advanced seed treatment technique that can be a sustainable alternative. This research aimed to examine the effect of different microbial biocontrol agents on the reduction of seed-borne pathogens in maize. Microorganisms that were used as biopriming agents were: *Bacillus amyloliquefaciens* (BA1), *Bacillus amyloliquefaciens* (ABO2), *Pichia* sp. (P1), *Trichoderma asperellum* (T251/21), and a mixture of five *Trichoderma harzianum* isolates. Two samples of maize seeds previously confirmed to be naturally contaminated with *Fusarium* spp. (over 5%) were used as a plant material. Biopriming was performed by submerging the seeds in the cell suspension of the agent. Subsequently, the seeds were placed on PDA and incubated at room temperature for 7 days. Infection rate and germination parameters were rated to determine the effects of the biopriming. The highest reduction of infection rate in both seed samples was detected on seeds primed by *T. harzianum* mixture and *T. asperellum* (0% infected seeds), while the lowest reduction was recorded in treatments with *B. amyloliquefaciens* (BA1) (6,5% infected seeds). Analysis of variance revealed the existence of significant differences ( $p \leq 0.05$ ) in the length of the primary root and mesocotyl between the treatments. The shortest root and mesocotyl in both samples were observed in treatment with *B. amyloliquefaciens* (ABO2) (16,38 mm; 26,4 mm, respectively), while the longest root and mesocotyl were measured after biopriming with *Pichia* sp. (P1) (58,25 mm, 40,03 mm respectively). The results show that the application of *Bacillus*, *Trichoderma*, and *Pichia* bioagents can benefit the control of maize seed pathogens when applied as a seed treatment, while *Pichia* can also significantly improve root and mesocotyl development.

**Keywords:** biocontrol, seed biopriming, maize

**Acknowledgements:** This research was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grant Nos: 451-03-65/2024-03/ 200117, and 451-03-66/2024-03/ 200117).

## LIPID QUALITY OF PLANT-BASED BUTTERS

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Plant-based butters made from nuts and seeds have steadily increased in consumer popularity due to their unique flavors and healthy nutritional properties. The health benefits of consuming plant-based butters are predominantly associated with their lipid composition. They are rich sources of polyunsaturated fatty acids (PUFA) and monounsaturated fatty acids (MUFA), compounds widely acclaimed for their proven positive impact on human health. Additionally, plant-based butters are excellent sources of protein, dietary fiber, vitamins, minerals, and trace elements. This study assessed the lipid quality of plant-based butters available on the Serbian market through fatty acid composition and atherogenic index (AI). A set of 16 nut and seed butters were analyzed, including 6 peanuts, 3 hazelnuts, 2 pumpkins, 2 sunflowers, 1 almond, 1 cashew, and 1 sesame sample. The fatty acid profile was determined by gas chromatography-mass spectrometry, and AI was calculated. The investigated samples were characterized by a low content of saturated fatty acid (3.63-19.78% on average) and a high content of MUFA and PUFA, where oleic and linoleic acids were dominant. The highest content of PUFA was found in sunflower (64.16%), sesame (49.21%), and pumpkin butter (47.22%), while the highest content of MUFA was found in hazelnut butter (83.76%). Among the peanut butter samples, three were characterized by a very high content of MUFA (82.9-84.6%), while others had approximately 40% MUFA, indicating the use of both standard and high-oleic peanut varieties in production. The very low AI values (0.03-0.12) in the analyzed samples suggest that consuming these plant-based butters could have a favourable effect on human health.

**Keywords:** plant-based butter, nuts, seeds, fatty acid content, atherogenic index

**Acknowledgements:** Ministry of Science, Technological Development and Innovation of the Republic of Serbia project numbers 451-03-66/2024-03/ 200134 and 451-03-65/2024-03/ 200134.

## NUTRITIONAL AND CHEMICAL CHARACTERIZATION OF SAFFLOWER SEEDS (*Carthamus tinctorius* L.): IMPLICATIONS FOR FOOD, FEED, AND OIL INDUSTRIES

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Safflower seeds (*Carthamus tinctorius* L.) are increasingly recognized for their nutritional value and diverse applications in food, feed, and oil industries. A comprehensive analysis of the chemical composition, mineral content, and fatty acid profile of safflower seeds is crucial for understanding their health benefits, optimizing agricultural practices, and supporting various applications in the food and feed industries. Additionally, such analysis provides valuable insights for improving crop quality and enhancing the economic value of safflower in the global market. In this study, the safflower seed variety developed through the breeding program of the Institute of Field and Vegetable Crops was thoroughly analysed. The chemical composition was assessed using AOAC methods, mineral content was measured via atomic absorption spectrometry (SRPS EN ISO 6869:2008), and fatty acid methyl esters (FAME) were prepared and analyzed through gas chromatography according to SRPS EN ISO 12966-2:2017. The analysis revealed a moisture content of 7.03%, crude protein of 12.85%, crude fat of 18.41%, crude ash of 2.10%, and crude fiber of 8.37%. Mineral analysis indicated substantial levels of potassium (3523.54 mg/kg) and calcium (1463.31 mg/kg), alongside trace elements such as phosphorus (0.46 mg/kg), magnesium (1328.81 mg/kg), iron (26.82 mg/kg), manganese (9.18 mg/kg), zinc (23.53 mg/kg), and copper (6.31 mg/kg). The lipid profile highlighted a predominance of polyunsaturated fatty acids (75.6%), mainly linoleic acid (75.4%), with saturated fatty acids at 8.1% and monounsaturated fatty acids at 16.3%. The obtained results will enhance understanding of safflower seeds' nutritional profile, guiding their effective use in food and feed applications. This knowledge will inform health professionals and agricultural stakeholders, promoting healthier diets and sustainable farming practices.

**Keywords:** safflower seeds, nutritional composition, chemical analysis, mineral content, fatty acid profile

**Acknowledgements:** This work was carried out as collaborative activities of the Institute of Field and Vegetable Crops, Novi Sad, Serbia and University of Novi Sad, Institute of Food Technology, Novi Sad, Serbia. This research was supported by Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant numbers 451-03-66/2024-03/200032 and MIDAS project (Marginal lands and industrial crops for the European bioeconomy), No. 101082070. and Provincial Secretariat for Higher Education and Scientific Research, project No. EB: 142-451-2609/2021-01. Study was carried out as part of the activities of the Centre of Excellence for Innovations in Breeding of Climate Resilient Crops—Climate Crops, Institute of Field and Vegetable Crops, Novi Sad, Serbia.

## **SURVIVAL ABILITY OF BIOFILM-PRODUCING *ESCHERICHIA COLI* AND *SALMONELLA* TYPHIMURIUM IN THE SOIL**

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The incidence of food-borne diseases caused by the consumption of fresh or minimally processed fruits and vegetables is constantly increasing. Bacteria *Salmonella* Typhimurium (*S. Typhimurium*) and *Escherichia coli* (*E. coli*) are considered as the most important pathogenic contaminants of fresh vegetables due to the intensity of symptoms and low infection doses. These pathogens are excreted into the external environment from the digestive tract of humans and animals, which opens the way for their unimpeded entry into the soil and watercourses. Bacterial cells can survive for more than a year by adhering to soil and sediment particles and form a biofilm. After infecting the soil, bacterial cells can consequently adhere firmly to the plant surface with the potential biofilm formation. The aim of the present study was to investigate the survival ability of *E. coli* and *S. Typhimurium* in the soil during 120 days under constant (T=23.0 °C; H=70.0%) and variable (T=22.0÷25.0 °C; H=40.0÷50.0%) condition of temperature (T) and relative humidity (H). The biofilm formation of both microorganisms was examined using the crystal violet test at 25 °C by measuring the optical density at 630 nm. Results showed that both bacteria are strong biofilm producers, based on the mean value of absorbance which was 0.510±0.068 for *E. coli* and 0.651±0.042 for *S. Typhimurium*. The presence of viable cells of both microorganisms was detected after 120 days of incubation. Moreover, the survival rate of *S. Typhimurium* was higher compared to *E. coli* under constant and variable conditions and was 0.60% and 0.27%, respectively, and for *E. coli* was 0.40% and 0.03%, respectively. The obtained data provide evidence that bacteria *E. coli* and *S. Typhimurium* could survive for several months in the soil, which increases the possibility of crop contamination.

**Key words:** *Escherichia coli*, *Salmonella* Typhimurium, survival, soil, biofilm

**Acknowledgements:** This research was financially supported by Ministry of Science, Technological Development and Innovation Republic of Serbia, Institute of Food Technology in Novi Sad (Grant Number: 451-03-66/2024-03/200222).

## DEVELOPMENT OF GREEN ANALYTICAL METHOD COMBINED SELECTIVE EXTRACTION AND LIQUID CHROMATOGRAPHY FOR ANALYSIS OF FOOD SAMPLES

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Analysis of food involves sample preparation, analysis, and detection of major food components (e.g. carbohydrates, phenolic compounds, aroma compounds, etc.) and miscellaneous components (preservatives, colorants, and other). A large number of different analytical methods have been developed to analyse the properties of food because of the complexity and diversity of food components. Increased method selectivity can be achieved by applying appropriate extraction technique. Molecularly imprinted polymers (MIPs) are synthetic materials that specifically recognize and selectively adsorb certain target molecules, template or their structural analogues. MIPs are a promising tool for the extraction of substances from complex samples. The aim of this work was to develop green analytical method combined with selective extraction and liquid chromatography for analysis of citrus fruits (lime, grapefruit, tangerine, and other). Deionized water, ethanol, and deep eutectic solvent-like mixtures were tested for extraction, a solid phase extraction with MIP-based sorbents was used for analytes cleaning and reversed phased high performance liquid chromatography method for extract analysis. The proposed method demonstrated a linear range of 50-1000 ng.mL<sup>-1</sup>, the limits of detection in the range of 8.2-44.0 ng.mL<sup>-1</sup> for selected coumarins, and the recovery more than 85% (RSD ≤ 4.8%). Umbelliferone was detected in lime and in grapefruit, herniarin was detected in lime. The environmental character of the analytical method was evaluated using the Analytical Eco-Scale.

**Keywords:** citrus fruits, coumarins, MIP-based extraction, HPLC

**Acknowledgements:** This work was supported by the Slovak Research and Development Agency under the Contract no. SK-SRB-23-0015 and project VEGA (1/0332/25). A. Špačková thanks for financial support from the call for doctoral students and young researchers of Slovak University of Technology in Bratislava to start a research career (Grant 23-04-01-A).



## DETECTION OF BIOFILMS IN BREWERY: TRADITIONAL VS NOVEL METHODS

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Biofilms, complex populations of microbes attached to surfaces, cause substantial issues across numerous industrial settings, including breweries. In the brewing sector, biofilms develop on many surfaces, including tanks, pipelines, heat exchangers, and packaging equipment. The microbial communities consist of bacteria, yeast, and fungus. Their creation can cause various operational and product quality problems such as contamination, spoiling, and equipment fouling. Due to the persistent character of these biofilms, the identification and removal of biofilms have become crucial considerations to ensure product safety, maintain quality, and comply with strict regulatory criteria. Brewers have relied on traditional procedures like microbiological culture and staining techniques to detect biofilms for many years. Although these methods offer vital insights into the existence and makeup of biofilms, they can require a significant amount of time and effort, and their sensitivity and accuracy are generally restricted. Moreover, these techniques generally only identify species that can be grown in a laboratory, so failing to discover a significant fraction of the microbial population present in the biofilm. The constraints of conventional approaches have prompted the quest for more efficient, expeditious, and accurate methodologies to detect and track biofilms in brewery settings. Novel methodologies, such as polymerase chain reaction (PCR)-based methods, provide improved sensitivity, faster detection, and a more thorough comprehension of biofilm composition. This paper aims to compare traditional biofilm detection methods with novel, cutting-edge techniques in the context of brewery operations. The results were obtained by combining standard methods (microscopy of live preparations, Pour Plate, NBB-c specific substrates) and modern methods (PCR). During microbiological control of the equipment, the presence of *Lactobacillus sp.* was confirmed in a significant number of samples, while wild yeasts of different morphological characteristics were detected in the other. When the PCR method was used, it identified 40% more beer-spoilage contaminants than standard methods.

**Keywords:** beer, biofilm, isolation, identification

**Acknowledgements:** This research was funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia, University of Belgrade—Faculty of Agriculture (Agreements No. 451-03-47/2023-01/200134, 451-03-47/2023-01/200116, and 451-03-1202/2021-09).

## ANALYSIS OF HMF CONTENT IN CHERRY CONCENTRATE: EVALUATION OF PRODUCT QUALITY AND SAFETY

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The content of hydroxymethylfurfural (HMF) in fruit concentrates, including cherry concentrate, is a key indicator of product quality. HMF forms during thermal processing and storage of fruit products as a result of the Maillard reaction between sugars and amino acids. Monitoring HMF levels is essential in the fruit product industry as it can provide insights into product quality and safety, highlighting potential issues in the production or storage process. High levels of HMF may indicate excessive thermal processing, improper storage, or degradation of the nutritional and organoleptic properties of the fruit product. Excessive exposure to high temperatures or prolonged storage can lead to increased HMF concentration, potentially affecting the quality and safety of the product. Therefore, HMF analysis becomes crucial for ensuring that fruit concentrates meet quality and safety standards. In this research, the HMF content in cherry concentrate was analysed using test strips, known for their speed and efficiency in detecting the compound in question. The cherry concentrate sample was prepared and analysed under controlled laboratory conditions, with the results compared to applicable standards for fruit concentrates. The test strips allowed for precise and straightforward assessment of HMF levels, facilitating the evaluation of product quality. In accordance with European Union regulations, which are also adopted by Serbia, the maximum allowed concentration of HMF in fruit concentrates is 60 mg/L. Based on this research, the analysis results indicated that the HMF content in the investigated cherry concentrate sample was below the regulatory threshold value. This finding demonstrates that the cherry concentrate underwent appropriate thermal processing and was stored under suitable conditions, contributing to the preservation of high product quality. The low HMF level also suggests that the nutritional values and sensory characteristics of the concentrate were maintained, confirming that the product is safe for consumers or further processing and meets quality standards.

**Keywords:** HMF, cherry concentrate, technological process

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovations of the Republic of Serbia under the programs 451-03-66/2024-03/200134, 451-03-65/2024-03/200134.

## PESTICIDES RESIDUES IN FOODS IN THE REPUBLIC OF NORTH MACEDONIA

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North Macedonia is an agricultural country with a large production of different types of vegetables and fruits in open fields and in greenhouses. Beside domestic products there are also imported food, particularly citrus and other fruit like bananas. The present study investigates pesticide residues in foods from domestic and import origin from January 2020 to December 2023. The easy and quick QuEChERS procedure was employed for sample preparation and gas chromatography with mass spectrometry (GC-MS) techniques were applied for identification and quantification of pesticides. The results of the study showed that some products have constantly presence of the same pesticides residues. Based on the results from the investigation, it was concluded that the most common pesticides detectable with GC-MS are bifenthrin, pirimiphos methyl, chlorpyrifos and boscalid. From the total number of samples, it was noted that the most abundant pesticide in bananas is bifenthrin and in cereals and their products is pirimiphos methyl. The results of the study showed that all of the tested samples contained residues below the MRLs.

**Keywords:** pesticides, foods, GC-MS

## MONILIFORMIN: ADVANCES AND CHALLENGES IN RESEARCH

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Moniliformin is a chemical contaminant that belongs to the group of emerging mycotoxins. It is produced by several species of the genus *Fusarium*, as well as one species from the genus *Penicillium*. Moniliformin has attracted increasing attention in the scientific community in recent years due to its potential toxicity and high presence, especially in maize and other cereals. Certain changes in weather patterns, especially in terms of temperature and precipitation, have the potential to stimulate the growth of moniliformin-producing fungi, thereby increasing the risk of contamination. The main symptoms of moniliformin toxicity in experimental animals include haematotoxicity and cardiotoxicity, accompanied by muscular weakness and respiratory changes. Moniliformin induces chromosomal aberrations *in vitro*, but there are no data on genotoxicity *in vivo*, and no data on carcinogenicity. Given that moniliformin is frequently detected in maize, and due to its potential negative health effects, the development of moniliformin analysis methods is important for monitoring purposes. Techniques such as liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS) allow accurate and efficient detection of low concentrations of moniliformin. The natural co-occurrence of moniliformin and other regulated and unregulated mycotoxins indicates that it is necessary not only to use the developed single-method approach for its detection, but also to integrate it into multi-mycotoxin screening methods to ensure comprehensive monitoring and accurate risk assessment. In addition to analytical methods for mycotoxin determination, it is important to study the influence of weather conditions on moniliformin production to understand the risk factors and develop effective strategies for the prevention of food contamination. Furthermore, physical reduction methods, including milling and extrusion, can find their application in reducing moniliformin content. Based on the above, it can be concluded that the use of integrated approaches for mycotoxin control is essential to maintain food safety and protect human and animal health.

**Keywords:** moniliformin, method development, monitoring, weather patterns, reduction methods

**Acknowledgements:** This research was funded by the Institute of Food Technology in Novi Sad, under the Seed Research Grant Program for young scientists ("Development of a high-efficiency LC-MS/MS method for fast and accurate detection of mycotoxins in maize - MycoScan"), through the financial support of the Serbia Accelerating Innovation and Entrepreneurship (SAIGE) Project, a joint investment by the Republic of Serbia, Ministry of Science, Technological Development and Innovation, the World Bank and the European Union; and by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## SERS-TAG TECHNOLOGY IN FOOD SAFETY AND DETECTION: SENSING FROM THE "FINGERPRINT" REGION TO THE "BIOLOGICAL-SILENT" REGION

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Surface enhanced Raman spectroscopy (SERS) offers many advantages for the fast and sensitive detection of specific hazard factors in foods, motivating to its increasing utilization in food safety. SERS detection platforms with high signal-to-noise ratio in the "biological-silent" region (1800-2800 $\text{cm}^{-1}$ ) are presently being developed for sensing and imaging applications, overcoming the limitations of traditional SERS studies in the "fingerprint" region. However, the bottleneck to SERS becoming a mainstream optical readout technology for single-sampling, "one-pot" readout, interference-free analysis is the need to produce novel Raman reporters (RRs) with suitable specificity for multiple target analytes, mutual non-interference, and Raman peaks in the "biological-silent" region. To date, the number of such reporters is still relatively small. To address this issue, customized RRs are urgently needed. Herein, a series of cyano-programmable RRs operating in the "biological-silent" region were designed based on 4-mercaptobenzoni derivatives and then embedded in core-shell Au@Ag nanostars using a "bottom-up" strategy to provide SERS enhancement and encapsulation protection. The approach enabled the "one-pot" readout interference-free detection of multiple bioamines (histamine, tyramine, and  $\beta$ -phenethylamine) based on aptamer-driven magnetic-induced technology. Three cyano encoded SERS tags resulted in separate SERS signals for histamine, tyramine, and  $\beta$ -phenethylamine at 2220, 2251, and 2150  $\text{cm}^{-1}$ , respectively. A target-specific aptamer complementary DNA competitive binding strategy allowed the formation of microscale core satellite assemblies between  $\text{Fe}_3\text{O}_4$ -based magnetic beads and the SERS tags, enabling multiple SERS signals to be observed simultaneously under a 785 nm excitation laser. The LODs for detection of the three bioamines were  $0.61 \times 10^{-5}$ ,  $2.67 \times 10^{-5}$ , and  $1.78 \times 10^{-5}$   $\text{mg L}^{-1}$ , respectively. The SERS-encoded platform utilizing programmable reporters provides a fast and sensitive approach for the simultaneous detection of multiple biomarkers, paving the way for routine SERS analyses of multiple analytes in complex matrices.

**Keywords:** SERS, Bioamines, "Biological-silent" region, Simultaneous detection

**Acknowledgements:** This work was financially supported by the National Natural Science Foundation of China (Grant 32372431) and Innovation Ability Improvement Project of Small and Medium-Sized Technology-Based Enterprises of Shandong Province (Grant 2022TSGC2409). G.I.N.W. acknowledges funding from the MacDiarmid Institute for Advanced Materials and Nanotechnology.

## **AN INNOVATIVE RAPID TWO-INCUBATION IMMUNOASSAY TO ELIMINATE PRE-CLEAN UP COLUMNS AND OPTIMIZE THE RECOVERY LEVELS OF OCHRATOXIN A IN OVERPARTICULAR MATRICES**

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Ochratoxin A produced by several species of *Aspergillus* is a potent nephrotoxin which causes acute and chronic effects in the kidneys of all mammalian species. OTA has genotoxic and teratogenic characteristics and is considered carcinogenic. Ochratoxin A is legislated therefore accurate and rapid ways of detection in food and feed commodities are of a paramount importance. Many ELISA tests on the market use organic solvents (70% methanol) for the extraction and filtration to achieve the clean-up of some commodities and achieve the quantification of the toxin. In principle these tests are based on the horseradish peroxidase (HRP) enzyme. The wells are coated with OTA specific antibodies and with the use of known OTA concentrations standards they achieve the quantification of the sample through an ELISA protocol. The quantification is achieved with a photometer after measuring the absorbance that is generated in the method with a use of a chromogen substrate. Some of the issues ELISA methods are facing are the low recovery levels of OTA especially in the presence of overparticular matrices like pepper, dates, dry fig, almond and peanut butter. That leads to a pretreatment of the sample with pre cleanup columns which lead to HRP tolerance issues, insufficient OTA recovery levels and protocol limitations from reduced antibody antigen competition. The aim of this project was to examine and introduce an alternative way to improve the recovery levels of OTA in overparticular complex matrices by avoiding the use of the pre-column treatment Bio-shield Ochratoxin ELISA test kit that was developed by Prognosis Biotech using high affinity monoclonal antibodies following an initial protocol dilution and having two different incubation(competition) steps managed to maintain very high sensitivity levels without the use of a pre-clean column resulting in a valuable tool for Ochratoxin analysis with more economic, effortless and rapid characteristics.

**Keywords:** Ochratoxin, Mycotoxins, ELISA, Immunoassays

## BRIDGING THE GAP: MYCOTOXIN REFERENCE MATERIALS AND THE PERCEPTION OF MEASUREMENT

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This study presents a thorough evaluation of liquid mycotoxin reference standards, focusing on 30 distinct standards from 10 leading global manufacturers. Each standard consisted of 10 samples containing three mycotoxins: Aflatoxin B1 (AFB1), Deoxynivalenol (DON), and Zearalenone (ZON). To ensure comparability, all standards were adjusted to a uniform concentration level. The quality attributes of these standards were assessed using LC-MS/MS, HPLC-DAD, and LC-HRMS techniques. The findings revealed that 30% of the reference standards fell outside the acceptable limits, as determined by both LC-MS/MS and HPLC-DAD methods. Additionally, the study identified a total of 30 impurities across the standards: 12 in DON, 10 in AFB1, and 8 in ZON, with variations observed among suppliers. Based on these results, the study advocates for revisions to the ISO 17034 standard, suggesting that the purity of raw materials be determined by q-NMR analysis instead of the commonly used HPLC-UV or LC-MS/MS methods. Furthermore, it is recommended that liquid standards with a shelf life of  $\leq 1$  year should have a total uncertainty not exceeding 3%, while those with longer shelf lives should not exceed 5% uncertainty. The study also underscores the necessity of stability monitoring, highlighting that standards should be subjected to continuous long-term evaluation to prevent deviations in target values, as exemplified by a case where the target value was only 80%. It is also proposed that certificates for each released batch include evidence of HPLC and LC-MS/MS analysis to ensure the reliability of the standards.

**Keywords:** quality control, certificate of analysis, ISO 17034, impurity, stability

**Acknowledgements:** LVA GmbH wants to thank all collaboration partners. This work was supported by the Hungarian National Research, Development and Innovation Office (NKFI grants K 139312).

## **PATHOGENICITY OF *Fusarium* spp. ON POTATO TUBERS**

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Dry rot caused by several species from genus *Fusarium* is a devastating fungal disease affecting potatoes worldwide. Infected tubers and seed pieces rot after harvest or planting. Losses from dry rot during storage are estimated to range from 6 % to 25 %, and occasionally rise up to 60 % in extreme cases. Symptoms of *Fusarium* dry rot first appear on tubers at wound sites as shallow, small brown lesions after about a month of storage. The initial infection may spread in all directions forming necrotic concentric rings within which tissue dries out. Cavities below the brown, dry decay areas are usually lined with mycelium. Susceptibility to dry rot caused by *Fusarium* species can vary in different production areas. Infections of potato tubers by certain *Fusarium* species are usually accompanied by the production of mycotoxin contamination. The task of this work is to check the pathogenicity and determine the difference in virulence of twenty isolates originating from potato tubers with symptoms of tuber dry rot, which were identified as *Fusarium* spp. based on morphological and cultural characteristics. The pathogenicity test for each isolate was performed by wounding and artificially inoculating potato tubers of the Orchestra variety. The inoculated tubers were placed in plastic boxes and incubated in humid conditions at room temperature for 21 days. After the incubation period, the horizontal and vertical diameters of the necrosis were measured. Subsequently, the pathogen was re-isolated from the infected tuber. Data analysis was done using ANOVA (Analysis of variance) followed by Tukey HSD test. Significant differences ( $p \leq 0.05$ ) were observed in the diameter of necrosis caused by different *Fusarium* spp. isolates. The highest virulence was detected in isolates FK47 and FK39, both from Zobnatica, whereas isolate FK48 (Pivnice) had the lowest degree of virulence. The remaining isolates were within the group with medium virulence. This research is part of a study on potato dry rot in Serbia, a significant issue due to the substantial quantitative losses and qualitative damages it causes.

**Keywords:** *Fusarium* spp, potato, dry rot

**Acknowledgements:** This research was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grant Nos: 451-03-65/2024-03/ 200117, and 451-03-66/2024-03/ 200117).



## CONTAMINATION OF FOOD OF ANIMAL ORIGIN WITH AFLATOXIN - PERSPECTIVES IN THE LIGHT OF CLIMATE CHANGE

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From the beginning of the 21st century, climate change is believed to have significantly contributed to the increase of growth of aflatoxin-producing molds on crops in Europe. *Aspergillus flavus* and *A. parasiticus* produce several groups of aflatoxins, where the most common are AFB1, AFB2, AFG1 and AFG2 on plants and grains, which after being ingested convert into their metabolite AFM1 that is excreted through milk. Aflatoxins are proven to have carcinogenic potential, being harmful to the liver, lungs, kidneys, brain, and heart. AFM1 is considered to have a higher carcinogenic potential than AFB1 and other AF groups. Among foods of animal origin, the most common is the findings of AFM1 in the milk of animals fed with contaminated feeding stuff. Concerning meat and meat products, EFSA emphasizes that the real contribution of meat and meat products to the human exposure to aflatoxins is negligible. However recent studies in some countries from subtropical climate range indicate that significant amounts of aflatoxins, both B1 and M1 could be found in beef and chicken meat, as well as in eggs. Bearing in mind that climate change affects the southern parts of Europe with summer temperatures being similar as those in subtropical regions, not only milk, but also meat and egg producing sectors need to be aware of such increasing risk.

**Keywords:** aflatoxin, climate change, milk, meat, eggs

**Acknowledgements:** The study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Contract number 451-03-66/2024-03/200143).

## THE IMPACT OF MINT ESSENTIAL OIL ADDITION ON THE ANTIFUNGAL PROTECTION OF PHYLLO PASTRY

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One of the developmental directions in the field of food protection from microorganism growth is the application of natural antimicrobial agents as substitutes for synthetic preservatives in industrial food production. Therefore, the aim of this study was to investigate the effects of using mint essential oil in the antifungal protection of phyllo pastry made from white wheat flour and phyllo pastry with the addition of 10% whole wheat flour. The phyllo pastry were stored and examined at intervals of 0, 5, 7, 14 and 21 days at a temperature of 8°C. The examinations included mycological analyses to determine the total mold count and the identification of isolated species. The mycopopulation of the samples of phyllo pastry made from type 500 wheat flour and those with the addition of 10% whole wheat flour, without the addition of essential oil, was classified into 3 genera and 4 species. The genus *Penicillium* was represented by two species: *P. aurantiogriseum* and *P. expansum*, while the genera *Aspergillus* and *Cladosporium* were each represented by one species, *A. candidus* and *C. cladosporioides*, respectively. When mint essential oil was applied at concentrations of 0.17% and 0.255%, it exhibited the best antifungal effects on the examined phyllo pastry during 7 days of storage. The greatest differences in the total mold count of the phyllo pastry compared to the control samples were observed, being 1.2 log cfu/g and 1.4 log cfu/g, respectively, for the phyllo pastry made from white wheat flour, and 1.0 log cfu/g and 1.1 log cfu/g, respectively, for the phyllo pastry with the addition of 10% whole wheat flour. The shelf life extension of phyllo pastry made from type 500 white wheat flour and those with the addition of 10% whole wheat flour up to 7 days was achieved by applying mint essential oil at concentrations of 0.17% and 0.255%. These studies represent the base for further research on the effects of essential oils on other bakery products using different concentrations, combinations of essential oils, as well as combinations of essential oils with various packaging conditions.

**Keywords:** molds, essential oils, phyllo pastry, shelf life

**Acknowledgements:** This research was financially supported by Ministry of Science, Technological Development and Innovation Republic of Serbia, Institute of Food Technology in Novi Sad (Grant Number: 451-03-66/2024-03/200222).

## PHARAMACO-KINETICS PROPERTIES OF LINEAR POLYETHYLENE TEREPHTHALATE OLIGOMERS PREDICTED BY USING ADMET PREDICTION TOOLS

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About 95 % of polyethylene terephthalate (PET) is used in food industry, mainly (about 70%) for the production of bottles containers for beverages, such as water, carbonated soft drinks and juices. Side-products of PET polymerization are cyclic and linear oligomers also detected in human blood, and they are recently recognized as the novel non-intentionally added substances in food (NIAS), due to their proven and frequent migration to food from the food packaging materials. In order to get insight does size and end-group chemistry of linear PET oligomers matter in chemical risk assessment, pharamaco-kinetics properties of eight PET oligomers (non-methylated and methylated linear monomers, dimers and trimers) are predicted by using several ADMET (Absorption, Distribution, Metabolism, Excretion, and Toxicology) prediction tools (SwissADME, ADMETlab, admetSAR, pkCSM and Lazar). Methylated oligomers and oligomers with more than one aromatic ring have higher potential for adsorption. Methylated oligomers have higher potential to bound plasma proteins, while monomers, as the most soluble, have highest potential to be in unbound fraction. Probability to be inhibitors and substrates of metabolic enzymes is higher for methylated oligomers, and it is higher for oligomers with higher number of aromatic rings. Clearance was predicted to be higher for methylated oligomers, and increase with increase of aromatic rings number and decrease in solubility. All tested oligomers have potential to be hepatotoxic, particularly methylated and oligomers having higher size, but all have very low probability for carcinogenicity and mutagenicity. Methylated oligomers with more than one aromatic ring are predicted to be oestrogen and androgen receptor binders, particularly dimethylated one. According to *in silico* predictions using several ADMET tools chemical risk of linear PET oligomers, upon their potential migration to food/beverages, increase with methylation of end-groups and increase in number of aromatic rings.

**Keywords:** polyethylene terephthalate, linear oligomers, ADMET, pharamaco-kinetics properties

**Acknowledgements:** This research was supported by the Science Fund of the Republic of Serbia, Grant No. 7275, Exploration of PETase side activity of digestive enzymes of human gastrointestinal tract acting on micro- and nanoplastics: mode of action and products characterization – XPACT and by Ministry of Science, Technological Development and Innovation of the Republic of Serbia, Contract number: 451-03-47/2023-01/200168.

## SAFETY ASSESSMENT OF MILK OBTAINED FROM COWS TREATED INTRAMAMMARY WITH ESSENTIAL OIL-BASED PREPARATION FOR MASTITIS

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Determining the withdrawal period (WP) for veterinary products is performed by evaluation of time-related accumulation patterns of residues of interest in selected animal products used in human diet, or in chosen animal tissues. While essential oils (EOs) are mostly considered as safe, data on their WP are lacking, especially in case of application in dairy cows. This study aimed to evaluate WP of EOs-based pharmaceutical formulation (Phyto-Bomat) containing mixture of four EOs (obtained from the following Lamiaceae species: *Thymus vulgaris*, *Thymus serpyllum*, *Origanum vulgare*, and *Satureja montana*) diluted with marigold and St. John's wort oil macerates. The preparation was applied intramammary (twice daily, for five consecutive days) in cows with mastitis, after which the levels of thymol and carvacrol, as chosen EO residues, were monitored in milk and plasma of treated cows. The selected monoterpenes were quantified by application of analytical method based on gas chromatography–mass spectrometry technique which was validated in term of selectivity, linearity, precision (repeatability, reproducibility), accuracy, limits of detection (LOD) and quantification (LOQ). The obtained LOQs for thymol in plasma and milk samples were 0.1 and 0.03 µg/mL, while for carvacrol were 0.3 and 0.1 µg/mL, respectively. The obtained results indicated that Phyto-Bomat treatment leads to minimal residues of thymol (0.04-0.36 µg/mL) and carvacrol (0.11-0.37 µg/mL) in milk, which decrease to pre-therapy levels 24h after treatment, while both compounds tend to reach their maximum concentrations within the first 12h after Phyto-Bomat intramammary application. Similarly, plasma samples also demonstrated time-dependent changes in accumulation levels of thymol (0.15–0.38 µg/mL) and carvacrol (0.31–0.66 µg/mL), but also suggested the presence of these compounds 24h after treatment. The study indicates that Phyto-Bomat application for treatment of bovine mastitis results in minimal residual levels of thymol and carvacrol in milk, thereby ensuring its safety for human consumption and leading to no undesirable changes in organoleptic characteristics.

**Keywords:** withdrawal period, thymol, carvacrol, GC–MS, essential oil

**Acknowledgements:** This work was supported by the Science Fund of the Republic of Serbia, PROMIS, #GRANT No 6066966, InfoBomat.

## ANTIFUNGAL PROPERTIES OF *SATUREJA MONTANA* L. ESSENTIAL OIL ON TOXIGENIC *ASPERGILLUS PARASITICUS* AND *A. OCHRACEUS*, *IN VITRO* AND IN FOOD SYSTEM

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Fungal contamination is one of the leading causes of food spoilage. Beyond producing a range of enzymes that degrade the sensory and nutritional quality of food, fungi are also producers of mycotoxins, toxigenic secondary metabolites. On the other hand, some preservatives have harmful effects on human health, and a range of physical methods such as drying, heating, cooling, and others can alter the sensory and nutritional quality of products. A potential solution includes the implementation of alternative preservation methods, such as application of essential oils from aromatic plants. These techniques not only contribute to the microbiological safety of food but also benefit human health because many aromatic plants are known for their anti-inflammatory, antioxidant, and anticancer properties. Therefore, the aim of this study was to examine the impact of *Satureja montana* L. essential oil (organic origin) as a potential antifungal agent on selected toxigenic *Aspergillus* species (*Aspergillus parasiticus* and *A. ochraceus*) under *in vitro* conditions and in food system. The *S. montana* L. essential oil was obtained through hydrodistillation and the dominant components were carvacrol, p-cymene, and  $\gamma$ -terpinene. The impact of *S. montana* L. essential oil on the rate and inhibition of *A. ochraceus* and *A. parasiticus* growth was assessed using the agar plates method. Growth of *A. parasiticus* was completely inhibited at essential oil concentration of 1.78  $\mu\text{L/mL}$ , while the growth of *A. ochraceus* was completely inhibited at concentration of 0.89  $\mu\text{L/mL}$ . In the food system, the application of *S. montana* L. essential oil has shown a significant impact in reducing fungal growth compared to the control samples in which essential oil was not added. The obtained results showed that *S. montana* L. essential oil has significant antimicrobial properties and could be a potential antifungal agent for food preservation.

**Keywords:** *S. montana* L. essential oil, antimicrobial, toxigenic fungi, *Aspergillus*, food preservation

**Acknowledgements:** This research was funded by Provincial Secretariat for Higher Education and Scientific Research, Republic of Serbia, Autonomous Province of Vojvodina (No. 142-451-3498/2023-01/01), and by the Program of the Ministry of Education, Science and Technological Development of the Republic of Serbia (No. 451-03-47/2023-01/200134).

## APPLICATION OF QuEChERS IN DETERMINATION OF CONTAMINANTS OF EMERGING CONCERNS IN SAMPLES OF SUGAR BEET AND SOIL

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Unregulated and not regularly monitored synthetic or natural chemicals that have a potentially or proven harmful effect on environmental and human health are classified as contaminants of emerging concern (CECs). CECs includes many different classes of compounds such as pharmaceuticals, personal care products, illicit drugs, hormones, pesticides in current usage, per- and polyfluoroalkyl substances, micro- and nano-plastic, and many others. CECs can enter the environment from point sources with wastewater discharges as main ones and diffuse pollution sources such as terrestrial runoff from agricultural land. Fate of CECs in the environment is depends on their physical and chemical properties as well as of environmental conditions; transformation products of CECs additionally increase the complexity of environmental "CECs cocktails". Surface water represents the main recipient of wastewater discharges, so numerous CECs can be further transferred to agricultural soils if surface water is used for irrigation. The presence of CECs in agricultural lands has led to increased concern about the potential to accumulate in plants because of their tendency to uptake chemicals present in the soil. This leads to the introduction of pollutants into the food chain with potentially harmful effects during acute and chronic human exposure through food consumption. The aim of this work is to determine the efficiency of sample preparation methods based on QuEChERS extraction for the investigation of the CECs levels in sugar beet root. This method previously showed a good efficiency when applied to soil samples. The method was applied for the determination of CECs in spiked and real samples of sugar beet root, as well in real samples of soil. The content of CECs in water used for irrigation was also determined applying another sample preparation method based on solid-phase extraction. All samples were analyzed by high- performance liquid chromatography coupled with triple quadrupole mass spectrometer.

**Keywords:** contaminants of emerging concern, sugar beet, soil, surface water, high performance liquid chromatography

**Acknowledgements:** Funded by the European Union. Views and opinions expressed are however those of the author(s) only and do not necessarily reflect those of the European Union or EU executive agency. Neither the European Union nor the granting authority can be held responsible for them. This study is conducted under the project TwiNSol-CECs that has received funding from Horizon Europe programme under grant agreement no. 101 059867.

## EVALUATION OF pH EFFECT ON THE REMOVAL OF PESTICIDES FROM WATER USING NANOFILTRATION MEMBRANE

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Pesticides are mainly used in order to maintain food production and raw material supply due to a constant population growth. However, persistency and excessive use of pesticides in the agricultural domain has raised an issue of the potential harmful effects of pesticides on the environment. Pesticides are often detected in various water sources and, therefore, their efficient removal from water is required. The aim of this study was to determine efficiency of a nanofiltration membrane in the removal of malathion (insecticide) and propiconazole (fungicide) at three pH values. Experiments were conducted in a METCell dead-end stirred cell unit from Evonik Industries AG (Germany). High pressure nitrogen gas cylinder was used for obtaining 3 bar pressure in the system. Nanofiltration membrane, NF270 (FilmTec™), with molecular weight cut off 400 Da was used for the removal of two pesticides at pH 4, pH 7 and pH 10. Apparent 100% rejection was obtained for propiconazole with the NF270 membrane at all tested pH values. However, malathion rejection using nanofiltration membrane decreased from 100% to 87.54% at pH 4 and pH 7, respectively. Additionally, malathion was not detected in sample solutions, including feed, permeate and retentate, at pH 10. Molecular weight of malathion and propiconazole is 330.36 Da and 342.22 Da, respectively, however dissociation constant (pKa value) of malathion and propiconazole is 6.8 and 1.09, respectively. Neutral charge of malathion at pH 7 could cause lower rejection of malathion at pH 7 compared to pH 4, due to absence of electrostatic interactions. Degradation of malathion increases with an increase in pH value and, therefore, malathion was not detected in sample solutions at pH 10 due to intense hydrolysis. Size exclusion and electrostatic interactions have an evident role in the removal of pesticides from water with nanofiltration membranes.

**Keywords:** nanofiltration, water treatment, pesticides, malathion, propiconazole

**Acknowledgements:** Funded by the European Union. Views and opinions expressed are however those of the author(s) only and do not necessarily reflect those of the European Union or EU executive agency. Neither the European Union nor the granting authority can be held responsible for them. This work is conducted under the project TwiNSol-CECs that has received funding from Horizon Europe programme under grant agreement no.101059867.



## EVALUATION OF TOCOPHEROL VARIABILITY IN WILD HELIANTHUS SPECIES: A GENETIC RESOURCE FOR SUNFLOWER BREEDING

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Tocopherols are one of the most important bioactive compounds which improve the quality of the oil regards to their antioxidant properties. Considering that wild *Helianthus* species are a very important source of genes for the breeding of cultivated sunflower the aim of this research was to evaluate the variability in tocopherol content and composition within a germplasm collection. The evaluated germplasm included seven annual *Helianthus* species (*H. annuus*, *H. argophyllus*, *H. debilis*, *H. neglectus*, *H. niveus*, *H. petiolaris* and *H. praecox*). Oil samples were extracted using a hydraulic press. Tocopherol quantification was performed using high-performance liquid chromatography (HPLC) on a Nucleosil 100-5 NH<sub>2</sub> column with fluorescence detection ( $\lambda_{\text{ex}}=280$  nm,  $\lambda_{\text{em}}=340$  nm). The mobile phase comprised n-hexane/ethyl acetate (70/30, v/v) at a flow rate of 1 ml/min. Tocopherols were identified and confirmed based on their relative retention values and maximum absorption peaks, using 20  $\mu$ l injection volumes. The obtained results of the comparative tocopherol analysis revealed significant variability among wild *Helianthus* species. Tocopherol content averaged 333.5 mg kg<sup>-1</sup> seed in the all analyzed species, with an average profile of 328.56 mg kg<sup>-1</sup> (98.5%) of alpha-tocopherol, which is dominant. Only in species *H. annuus* and *H. niveus*, in small concentrations (6.83 mg kg<sup>-1</sup> and 6.65 mg kg<sup>-1</sup>), beta-tocopherol was identified. By applying of the Principal Components Analysis the species with the lowest tocopherol content (*H. praecox* 133.59 mg kg<sup>-1</sup> and *H. niveus* mg 238.23 kg<sup>-1</sup>) and the species with the highest content (*H. annuus* 458.11 mg kg<sup>-1</sup> and *H. neglectus* 459.35 mg kg<sup>-1</sup>) were clearly distinguished. The obtained results could serve as a tool to select species with high concentration of these bioactive compounds in design management strategies to obtain them in the breeding programs of cultivated sunflower.

**Keywords:** Wild sunflower, germplasm, oil, tocopherol

**Acknowledgements:** This work is supported by the Ministry of Education, Science and Technological Development of Republic of Serbia, grant number 451-03-68/2022-14/ 200032, by the Science Fund of the Republic of Serbia, through IDEAS project "Creating climate smart sunflower for future challenges" (SMARTSUN) grant number 7732457, by Centre of Excellence for Innovations in Breeding of Climate-Resilient Crops - Climate Crops, Institute of Field and Vegetable Crops, Novi Sad, Serbia, and the by the European Commission through Horizon Europe programme project CARINA (grant agreement 101081839) and Twinning Western Balkans project CROPINNO (grant number 101059784). Project HelEx, grand number 101081974, funded by EC.



## DYNAMIC ALTERATIONS IN FATTY ACID PROFILES AND TOCOPHEROL COMPOSITION IN OILSEED RAPE SEEDS UNDER CONTROLLED ENVIRONMENTAL CONDITION

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Oilseed rape, renowned for its high oil content predominantly composed of beneficial monounsaturated and polyunsaturated fatty acids, plays a pivotal role in human and animal nutrition. It is rich in omega-3 and omega-6 fatty acids, enhancing its nutritional value for both dietary and agricultural applications. The versatility of oilseed rape seed lies in its balanced fatty acid profile, crucial for the production of high-quality edible oil, animal fodder, and biofuels. However, the susceptibility of its unsaturated fatty acids, such as oleic and linoleic acids, to oxidation affects its shelf life and industrial utility. This study focused on analyzing the lipid composition of five rapeseed varieties after 6 and 12 months of controlled storage conditions. Gas chromatography (Konik HRGC 4000) facilitated the determination of fatty acid profiles, revealing a notable exponential decline in most acids over time. Specifically, oleic, linoleic and arachidonic acids exhibited a decrease at 6 months followed by a subsequent increase at 12 months, albeit not reaching initial levels. In the case of palmitic and linoleic acids, the content increased over time and then decreased. Furthermore, tocopherol levels were quantified using high-performance liquid chromatography (Nucleosil 100-5 NH<sub>2</sub> column with fluorescence detection ( $\lambda_{ex}=280$  nm,  $\lambda_{em}=340$  nm)), and glucosinolate content estimated based on sulfur levels. These analyses underscored the dynamic nature of rapeseed oil composition during storage, highlighting potential fluctuations in fatty acid content contingent upon storage conditions. While degradation is common in suboptimal storage environments, improvements can mitigate these changes, suggesting that careful management could stabilize or even reverse the decline in fatty acid content over time. In conclusion, understanding the temporal changes in rapeseed oil composition under different storage conditions is crucial for optimizing its nutritional and industrial applications, ensuring its sustained value in various sectors.

**Keywords:** oilseed rape seed, fatty acid, tocopherols, glucosinolate

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant number: 451-03-66/2024-03/200032 and by CROPINNO Grant No. 101059784 funded by the European Commission. This work was done as a part of activities of the Centre of Excellence for Innovations in Breeding of Climate-Resilient Crops - Climate Crops, Institute of Field and Vegetable Crops, Novi Sad, Serbia.

## SNACK PRODUCTS FROM WHOLE-GRAIN RED SORGHUM FLOUR

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Among the cereals, sorghum (*Sorghum bicolor* L. Moench) is a member of the gluten-free cereal family. Its strong resistance to heat and drought and its high photosynthetic efficiency, this crop is widely cultivated in a wide range of geographic locations. Rich in macronutrients (proteins, fats, and carbs) and micronutrients (minerals, vitamins), sorghum also contains phenolic compounds (tannins, phenolic acids, and flavonoids), which have antioxidant properties. Because of its many health benefits, including its ability to suppress the formation of cancer cells and reduce obesity, heart disease, and diabetes, sorghum is used for both human and animal consumption. It can be used for baking, extrusion, and different cereal-based products such as bread, cookies, pasta, expanded snacks, and breakfast cereals. However, in southeastern Europe is mainly used as animal feed. This study aimed to develop four types of snack products based on red sorghum flour (95, 92, 90 and 89%) with the addition of 1. a mixture of sweet and hot ground red pepper (5, 8 and 10%), 2. cocoa powder (5, 8 and 10%), and 3. a mixture of cocoa powder (10%) and cinnamon (1%), while a 100% red sorghum snack product served as a control sample. The following extrusion processing parameters were used: feeding rate of 50 kg/h, screw speed of 800, 850 and 900 rpm, and the material moisture content in the extruder barrel ranged from 13 to 14%. The following quality attributes of snacks obtained were determined: Expansion Ratio, Bulk Density, Water Absorption Index, Water Solubility Index, Color and Texture (Hardness, Number of fractures, Crispiness work, Crispiness index). Based on quality indicators, all types of snack products obtained in this study have the potential for commercialization. However, before commercialization, consumer acceptance tests and preference tests need to be conducted.

**Keywords:** whole-grain red sorghum snacks, pepper, cocoa, twin-screw extruder, quality properties

**Acknowledgements:** This work was funded by the Ministry of Science, Technological Development and Innovations, Serbia, contract no. 451-03-66/2024-01/200222.

## THE ROLE OF LACTIC ACID BACTERIA AND THEIR METABOLITES IN INHIBITING BIOGENIC AMINE FORMATION IN FOOD

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Biogenic amines (BAs) are nitrogenous compounds formed by the decarboxylation of amino acids and are commonly found in fermented and protein-rich foods such as fish, meat and dairy products. High levels of BAs, including histamine, tyramine and putrescine, cause vital health risks, ranging from mild allergic reactions to serious toxicological impacts. Preventing the accumulation of BAs in food is hence crucial to ensure food safety and consumer health. Lactic acid bacteria (LAB), widely used in food fermentation, have been recognised for their potential to inhibit BA formation through various direct and indirect mechanisms. LAB can inhibit the growth of BA-producing microorganisms by producing organic acids such as lactic acid and acetic acid, which lower the pH of the environment and create unfavourable conditions for decarboxylase enzyme activity. In addition, LAB-derived bacteriocins and other antimicrobial peptides specifically target and inhibit BA-producing bacteria, thereby lessening amine formation. Besides, the enzymatic activity of LAB, particularly amino oxidases, can degrade biogenic amines, lowering their accumulation in food. Recent studies have underlined the importance of LAB strains with specialised metabolic pathways that are particularly effective in lessening BAs in fermented foods. These findings have led to increased interest in the selection and use of LAB strains with enhanced anti-amine capabilities in food production. In addition, the potential of LAB as biocontrol agents in new food preservation strategies is being explored as a natural and consumer-friendly alternative to chemical preservatives. This work discusses the role of LAB and their metabolites in controlling biogenic amine formation, focusing on recent research about strain-specific characteristics, metabolite production and their application in different food matrices. By understanding these mechanisms, the food industry can develop safer and higher quality fermented products, thereby lowering the health risks linked with BA accumulation.

**Keywords:** Lactic acid bacteria, biogenic amine, food prevention, fermented food, health risks

## THE EFFECT OF BARBERRY EXTRACT ON OXIDATIVE AND NITROSATIVE STABILITY ON FERMENTED SAUSAGES

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Processes like fermentation and curing extend the shelf life of meat products such as fermented sausages, but they also make processed meats prone to lipid and protein oxidation. Additionally, compounds formed from nitrosative stress can trigger the oxidation of lipids and proteins. It is known that these reactions have many adverse effects on quality and health. Using natural antioxidants in product formulations is an effective solution to minimize these reactions. From this perspective, this study investigated the effect of increasing concentrations of barberry (*Berberis vulgaris*) extract (0 ppm - C, 200 ppm - B2, 300 ppm - B3, and 400 ppm - B4) on the oxidative and nitrosative stability of fermented sausages. Barberry extract (BE) was characterized by 46.33 mg GAE/g total phenolic compounds and 92.93% DPPH radical scavenging activity. The addition of BE did not alter the chemical composition of the fermented sausages but resulted in lower pH values due to the low pH (3.80) of BE ( $p < 0.05$ ). However, during production, the decrease in pH of BE-added samples was slower compared to the control, likely due to BE's potential antimicrobial effects. Higher concentrations of BE led to significantly lower peroxide values ( $p < 0.05$ ), and BE reduced TBARS values regardless of the concentration. Additionally, BE significantly retarded protein oxidation, particularly at concentrations above 300 ppm. The residual nitrite content was similar in C, B2, and B3 samples, but significantly lower in B4 ( $p < 0.05$ ). The presence of 3-nitrotyrosine, a marker of nitrosative stress, was also reduced with BE addition ( $p < 0.05$ ). These findings suggest that barberry extract is a promising natural ingredient for enhancing the oxidative and nitrosative stability of fermented sausages. Furthermore, the study revealed a correlation between 3-nitrotyrosine content and oxidative stress products, indicating that BE could effectively mitigate these reactions, thereby improving the quality and safety of fermented meat products.

**Keywords:** Barberry extract, fermented sausages, oxidative stability, nitrosative stress

**Acknowledgements:** This research was supported by Ege University Scientific Research Projects Coordination Unit under Project No: 29602. The authors extend their deepest gratitude for their invaluable contribution, enabling this comprehensive exploration into the beneficial effects of barberry extract on meat product stability.

## FOOD SUPPLEMENTS - CHALLENGES IN DEVELOPMENT OF NEW PRODUCTS AND QUALITY CONTROL

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Challenges in development of new food supplement products and quality control are related to the endless possible combinations of the bioactive ingredients (bioactive compounds, vitamins, minerals, plant extracts or even several plant extracts in one food supplement product, etc.) and excipients in different formulations such as hard/soft capsules, tablets, and liquids. This diversity of possible ingredients and formulations can cause many problems in development of analytical methods that are used from the from the conception through to the final food supplement product - starting with the selection of the ingredients (plant materials, pure compounds, etc.), selection of plant varieties (giving the best yields), selection of extraction procedures, quality control of the raw materials (identification, chemical and biological fingerprint), and final products (purity, uniformity, content, stability testing). It is hard to imagine quality by design of the food supplements products and also ensuring consumer safety without chromatographic techniques. The lack of available reference standards, SRMs, and marker compounds of particular plant materials, plant extracts, or plant extract fractions, as well as a variety of chemical structures, including isomeric compounds can present further difficulties in the analytics. This presentation will focus on challenges related to development and validation of new analytical methods for determination of active ingredients (e.g. procyanidins, stilbenes, carotenoids) in food supplement products. Issues related to stability of bioactive ingredients during analysis (from the sample preparation step till detection) and the fact - that a food supplements' bioactive ingredient can be present as a minor compound with much higher concentrations of other bioactive compounds will be discussed. The presentation will show that the combined use of different chromatographic techniques (HPTLC-densitometry, HPTLC-image analysis, HPLC-UV/VIS) and their hyphenation to mass spectrometry (HPTLC-MS/(MS) and spectrophotometry, is indispensable in providing support to manufacturers of food supplements as well as to regulatory authorities.

**Keywords:** food supplements, quality control, bioactive ingredients, chromatography, mass spectrometry

**Acknowledgements:** This study was supported by the Slovenian Research and Innovation Agency (ARIS; research core funding No. P1-0005, "Young Researchers" program - Maja Bensa).

## FULLERENOL NANOPARTICLES POTENTIAL TO IMPEDE MYCOTOXIN CONTAMINATION: A NOVEL APPROACH TO FUNGAL METABOLITE SUPPRESSION?

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The nanotechnology era has fully emerged, with carbon-based nanomaterials occupying a prominent position due to their versatility and biocompatibility. Hydroxylated fullerene derivatives ( $C_{60}$ ), called fullerols/fullerenols ( $C_{60}(OH)_x$ ), have shown significant benefits in mycotoxin mitigation strategies over the last years. Mycotoxin contamination remains a global concern, exacerbated by climate change and environmental stressors. The effects of fullereneol  $C_{60}(OH)_{24}$  nanoparticles (FNP) at concentrations of 0, 1, 10, 100, and 1000 ng/mL on secondary metabolite production by selected foodborne mycotoxigenic fungi were examined *in vitro*, using the LC-MS/MS dilute and shoot multimycotoxin method of Sulyok et al. (2020). Results indicate that FNP treatment significantly reduced the concentrations of examined secondary metabolites in a concentration- and genus-dependent manner. These findings provide a basis for further research on fungi-nanoparticle interactions, with implications for food and feed safety. Future studies will focus on elucidating the mechanisms underlying these interactions and their potential applications in contamination control.

**Keywords:** fullereneol nanoparticles, foodborne mycotoxigenic fungi, mycotoxins, secondary metabolism

**Acknowledgements:** The authors gratefully acknowledge the financial support of the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grants No. 451-03-66/2024-03/ 200125 & 451-03-65/2024-03/200125).

## DIETARY INTAKE OF IRON FROM DIFFERENT MEATS AND LIVERS IN SERBIAN ADULTS

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Meat and meat products are an important part of the human diet in Serbia. They are a convenient source of high-value proteins, essential vitamins, and important minerals needed for good health throughout life. Iron (Fe) is found especially in red meat, and it is indicated that vegetarians may be at risk of iron deficiencies. The aim of this study was to analyse beef, lamb, pig, horse, chicken, and turkey meats, as well as livers from the same animal species, regarding the iron content. Samples were gathered from different meat processing facilities in Serbia during 2023. The level of iron was determined by inductively coupled plasma mass spectrometry (ICP-MS). The estimated daily dietary intake (EDI) of Fe was calculated using data of Fe levels obtained in this study as well as data of dietary intake of estimated meats and livers from the European Food Safety Authority (EFSA) database. The following mean levels of Fe were found ( $\mu\text{g g}^{-1}$ ) in meat: beef 20.2, lamb 17.8, pig 7.90, horse 38.8, chicken 4.43, and turkey 6.74. The highest Fe mean level was obtained in horse liver ( $330.00 \mu\text{g g}^{-1}$ ) while the lowest level was obtained in beef liver ( $57.7 \mu\text{g g}^{-1}$ ). The results for EDI are expressed as percent of the Recommended Dietary Allowance (RDA) for adults (male: 8 mg/day; female: 18 mg/day). The obtained results showed that the contribution was dependent on the type of meat and liver. The analysed meat and liver provide in total 16.2% and 7.20% for men and women respectively of the RDA for Fe. The obtained results showed that the estimated meats and livers can be considered as important dietary sources of Fe. Hence, other food types are clearly necessary to provide adequate dietary levels of Fe for the Serbian adult population.

**Keywords:** iron, meats, livers, Serbian adult, intake

**Acknowledgements:** This study was supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia, Grant No. 451-03-66/2024-03/200050 from 05.02.2024.



## INFLUENCE OF *BACILLUS SAFENSIS* STRAIN P114 ON PEPPER FRUITS IN TERMS OF BIOACTIVE COMPOUNDS AND ANTIOXIDANT CAPACITY

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Sweet pepper as a commercial type of vegetable is one of the most important economic crop that includes a large number of different varieties. The nutritional value of pepper fruits is significant due to the content of bioactive compounds and antioxidants such as ascorbic acid, phenols, lycopene, carotenoids, chlorophylls and antioxidant activity. Some bacteria from the genus *Bacillus* are well known to be effective in controlling plant diseases and are associated with increased yields and plant resistance. The aim of this work was to test the influence of *Bacillus safensis* (strain originated from pepper seed, coded as P114) on total anthocyanins, flavonoid synthesis and antioxidant capacity. Inoculum of bacterium was prepared in sterile distilled water and adjusted to  $10^8$  CFU mL<sup>-1</sup> before treatment of pepper plants. Plants were initially treated in growth stage: leaf development and then periodically, every 10 days until the fruits were formed. No treated pepper plants served as control. The content of flavonoid was extracted in 70% acetone and measured spectrophotometrically at 360 nm. Total anthocyanin content was extracted with 2% (v/v) HCl in methanol and analyzed spectrophotometrically. The effect of strain P114 was significant in anthocyanin synthesis compared to control. The content of anthocyanin in the fruits was  $887.98 \pm 32.55$  ug CGE/g dm in the treated peppers, while in control was  $744.77 \pm 24.75$  ug CGE/g dm. The content of flavonoid in fruits was  $11.05 \pm 0.09$  µg/g in the treated pepper, while in control was  $11.53 \pm 0.66$  µg/g and no statistical difference was found. The antioxidant capacity in pepper fruit was similar from both, treated and untreated plants and amounted to  $85.34 \pm 0.78$  mmol Trolox/kg. The obtained results indicate that treatments of pepper plants with *B. safiens* strain P114 increased significantly anthocyanins contents in the pepper fruits.

**Keywords:** bioactive compound, *bacillus safensis*, pepper

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia [451-03-66/2024-03/200216, 451-03-66/2024-03/200040; 451-03-66/2024-03/200088; 451-03-66/2024-03/200010].



## APPLICATION OF ATOMIC ABSORPTION SPECTROMETRY USING THE DIRECT SAMPLING OF SOLID AND LIQUID SAMPLES IN THE DETERMINATION OF SELECTED ELEMENTS IN PROTEIN POWDER SAMPLES

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The work deals with elemental analysis of protein powders, which are among popular food supplements. In total, 40 samples of protein powders of different types were analyzed, in which 15 elements were determined. The preparation of the samples consisted in its mineralization using microwave digestion where nitric acid was used as a decomposition agent. Flame and electrothermal atomic absorption spectrometry methods (FAAS and ETAAS) were used to determine the elements. The conditions were optimized for the determination of the elements Mg, Cr and Pb. FAAS was used to determine Mg, using burner height of 10 mm and an acetylene-air fuel flow rate of 80 L.h<sup>-1</sup>. ETAAS was used to determine Cr where the pyrolysis temperature was 1650 °C and the atomization temperature was 2700 °C. In the case of Cr, the LOD, LOQ and repeatability were 0.61 µg.L<sup>-1</sup>, 2.05 µg.L<sup>-1</sup>, and 4.6%, respectively with the linear range of 1-100 µg.L<sup>-1</sup>. For the determination of Pb, the possibility of direct sampling was used, which provided a satisfactory repeatability of 7.6%. The pyrolysis temperature in this determination was 1100°C and the atomization temperature 1800°C. Based on the principal component analysis, it was found whey proteins regarding the content of elements, show very similar characteristics, forming one group. Another group was formed by rice proteins, with very similar characteristics. The third group was made of mixed proteins which contain soy that also have a specific elemental composition and a high degree of similarity. In contrast, the pumpkin protein was markedly different from all other groups.

**Keywords:** protein powder, ETAAS, FAAS

**Acknowledgements:** This work was supported by the Slovak Research and Development Agency under the Contract no. SK-SRB-23-0015, APVV-23-0066 and VEGA - Scientific Grant Agency of the Ministry of Education under the project number 1/0017/23.

## INHIBITION OF TOXIGENIC *Aspergillus flavus* GROWTH BY GREEN EXTRACTED POLYPHENOLS FROM AROMATIC PLANTS

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*Aspergillus flavus* is a pathogenic fungus associated with food safety issues worldwide. Food contamination with aflatoxins, which are produced by mycotoxigenic strains of *Aspergillus* spp., leads to serious health and economic consequences. Biological metabolites are regarded as a safe, effective, and environmental-friendly method for preventing fungal growth. Aromatic plants such as caper (*Capparis spinosa* L.) and immortelle (*Helichrysum italicum* L.) are considered as valuable sources of natural antioxidants - polyphenols. This study investigates the efficacy of phenolic plant extracts prepared using food-grade solvents (ethanol-water mixture and natural deep eutectic solvents-NADES) to inhibit the growth of aflatoxigenic strain *A. flavus*. The antifungal activity of plant extracts against *A. flavus* mycelia growth was based on agar dilution test using PDA medium. Effect of different plants extracts concentrations on *A. flavus* mycelia growth was also studied. Immortelle ethanol extracts inhibited the mycelial growth of *A. flavus* (43.42%) more effectively than those of caper (11.37%) after 10 days of incubation. The growth of *A. flavus* could be completely inhibited by immortelle NADES extracts at 300-450 mg/mL, depending on the extraction solvent. Different concentrations of the extracts also influenced the macroscopic characteristics of the colony, including changes in color, structure, and particularly the formation of sclerotia. A range of polyphenols, including flavonoids and phenolic acids, were identified and quantified in the extracts. Among the phenolic compounds, rutin was the most dominant in caper extract, while phenolic acids such as chlorogenic acid, caffeic acid, rosmarinic acid, and syringic acid were the most prevalent in the immortelle extract. The findings provide novel information on antifungal activity of green polyphenols from aromatic plants and their potential usage to improve food safety and extend shelf-life. Future research should elucidate mechanism of action, and the effect of these extracts on aflatoxin production.

**Keywords:** *A. flavus*, aromatic plants, green extracted polyphenols, safety application

**Acknowledgements:** The study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Contract Number 451-03-66/2024-03/200222).

## MYCOTOXINS IN CEREALS: PUBLIC HEALTH SIGNIFICANCE, OCCURRENCE AND CONSUMER EXPOSURE

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Mycotoxins represent a structurally heterogeneous group of secondary mould metabolites that can contaminate various cereals not only directly in the field, but also during storage, transport and processing, with most of them proving to be chemically stable and resistant to various food processing methods. They enter the consumer's organism either directly via the consumption of contaminated food or indirectly via feed (carry-over effect) and cause harmful effects ranging from acute poisoning to long-term and chronic effects on the human body. Mycotoxins are one of the main hazards responsible for the increasing number of notifications under the "Rapid Alert System for Food and Feed" (RASFF) over the last ten years. It is well known that agro-ecological factors can significantly influence the presence and content of mycotoxins in agricultural products, with the level of contamination largely dependent on weather conditions, particularly temperature and precipitation. Climate change may not only exacerbate already documented mycotoxin contamination, but also introduce new mycotoxins that are found all over the world. Higher temperatures, long dry periods and heavy rainfall at or just before grain harvest may lead to an increased occurrence of highly toxic aflatoxins. Further research is needed to understand the impact of climate change on their occurrence and to develop cereal varieties and hybrids that are genetically more resistant to toxicogenic mould infections and highly resilient in a variety of environments. Also, of great importance is to investigate the possibility of early detection of mould infestation to predict mycotoxin contamination to ensure sustainable self-sufficiency of grains as a raw material. Given the high incidence of mycotoxins, systematic control with monitoring of key influencing factors, especially weather conditions, is needed to minimise consumer exposure and potential health risks from the consumption of cereals and cereal products.

**Keywords:** mycotoxin contamination, cereals, health risk, climate change, consumer exposure

**Acknowledgements:** The studies were funded by the Croatian Veterinary Institute and the European Union NextGenerationEU with the support of the Ministry of Science and Education of the Republic of Croatia within the project No. NPOO 10 of the Croatian Veterinary Institute entitled „Biological diversity of molds and occurrence of *Fusarium* mycotoxins and their metabolites in the most important cereals grown in the Republic of Croatia“ (MOULD\_FUSion).

## CLIMATE CHANGE AND MYCOTOXINS: A COMPARATIVE PERSPECTIVE BETWEEN CHINA AND SERBIA

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Climate change is a global concern with significant impacts on various aspects of life, including agriculture and food safety. Chemical contamination is identified as a burning issue in food safety, with mycotoxins, toxic secondary metabolites of fungi, posing an ongoing challenge and increasing threat due to recent climate change. Mycotoxins are key indicators of food safety due to their health risks, toxicity, widespread contamination of food and feed, and substantial economic impact. Both China and Serbia have faced climate changes that have led to significant challenges in food safety, particularly with an increased occurrence of mycotoxins. Over the past two decades, Serbia has experienced notable climate changes, including a substantial rise in air temperatures, an increase in the number of tropical days, and more frequent heatwaves and droughts. Conversely, in certain years, high levels of precipitation have led to flooding and record-breaking rainfall. These climatic variations have significantly impacted food safety in Serbia, contributing to a rise in mycotoxin contamination, particularly the more frequent occurrence of aflatoxins in maize, maize-derived products, milk, and dairy products. In China, the diverse geographical regions exhibit distinct climate characteristics, with the southern region experiencing relatively humid weather and the northern region facing more arid conditions. Pronounced climate changes, such as sudden temperature increases, heavy rainfall, and drought, have significantly impacted mycotoxin levels across various areas of China, particularly deoxynivalenol and aflatoxins in cereals. Both countries are experiencing increased risks due to intensified and more frequent climate changes, leading to higher exposure to multiple mycotoxins in food. This situation amplifies the risks to the food supply chain and human health, highlighting the urgent need for ongoing monitoring and the development of adaptive strategies to manage mycotoxin contamination in the face of climate change.

**Keywords:** Climate change, mycotoxins, Serbia, China

**Acknowledgements:** This work was financially supported by Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222), Mobility projects for the 6<sup>th</sup> Session of the Joint Committee Meeting on Scientific and Technological Cooperation between the Republic Serbia and the People's Republic of China "Food Safety in a Changing Climate: Challenges and Solutions in terms of mycotoxins" and Exchange program of the 6<sup>th</sup> Regular session of China-Serbia Science and Technology Cooperation Committee (Grant No.6-17).

## MYCOTOXINS: TRENDS, CHALLENGES, AND PERSPECTIVES IN THE CONTEXT OF CLIMATE CHANGE IN SERBIA

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Observed climate changes in Serbia over the past two decades have been characterized by temperature fluctuations and altered precipitation patterns. These changes are marked by rising air temperatures, more frequent drought conditions during the summer months, and sporadic instances of extremely high precipitation. Such trends toward warmer and drier conditions have already started to affect agricultural productivity, as well as food and feed safety. Among the various factors influencing fungal growth and mycotoxin occurrence, numerous studies highlight climate change as one of the key contributors. In Serbia, over the past two decades, there has been a concerning increase in the occurrence of mycotoxins in certain agricultural products, particularly cereals. In the last 15 years (2009 to 2023), a significant correlation has been observed between climate change and the occurrence of aflatoxins in maize, the country's primary field crop. In the years 2013, 2017, and 2023, between 20% and 50% of maize samples were contaminated with aflatoxins, while in the years 2012, 2015, and 2021, more than 50% of samples were contaminated. During these years, maize contamination significantly impacted the presence of aflatoxin M1 in milk and dairy products. In addition to aflatoxins, other mycotoxins were present in maize samples, depending on the weather conditions during the growing seasons. During hot and dry years, *Aspergillus* mycotoxins were dominant, whereas, in years with increased rainfall, *Fusarium* toxins prevailed. Furthermore, in certain years, maize from Serbia contained several dozen different mycotoxins. Given these trends, there is a clear need for future strategies to address the challenges posed by climate change. Advances in analytical methods, particularly multi-toxin approaches, are also necessary to simultaneously detect the presence of multiple mycotoxins in various samples. Urgent action is required to enhance, invest in, and initiate educational programs across all levels of agricultural and food production in Serbia. This should be done while considering the documented climate change, projected future scenarios, and their impact on mycotoxin occurrence, and exploring future perspectives to effectively manage these challenges.

**Keywords:** mycotoxins, climate change, maize, milk

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## USEFULNESS OF HPLC IN FOOD ANALYSIS BY SPECTROMETRIC METHODS COMBINED WITH CHEMOMETRICS

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HPLC is a significant method of food analysis. In the case of liquid samples, e.g. beverages, it often enables direct analysis. Samples with a complicated matrix, or samples with low analyte content, need to be treated using a wide variety of sample preparation techniques. Due to its good selectivity, sensitivity and ability to provide information on a large group of compounds, HPLC has become a useful tool in proposing solutions to various problems in food analysis using spectrometric methods often combined with chemometrics. Traditional spectrometric methods such as UV-VIS absorption, fluorescence, infrared and Raman spectrometry allow to distinguish fresh from long-term stored food, to detect spoiled products, to classify products according to geographical origin, variety, and others. After developing the chemometric models allowing the products classification itself, it is necessary to determine the properties that cause the differences between the products observed in the spectra. Preliminary estimation of groups of chemical compounds related to the observed differences is possible based on the spectral characteristics of the samples, but the assignment of individual compounds is more reliable based on the comparison of spectra profiles with the results of HPLC separation. Another area of use of HPLC is the clarification of differences in the spectra of samples that are commonly named as outliers, i.e. having atypical properties. The third area is the use of HPLC as a reference method in the development of multivariate calibration models for the determination of individual compounds in food using spectrometric methods. The usual procedure is: (1) to register the spectra of the samples by the spectrometric method, (2) to determine the target analytes in the samples by the HPLC method, (3) to use the results from the quantitative HPLC analysis as dependent variables and the spectral data as independent variables in multivariate chemometric models.

**Keywords:** products classification, food quality, target analysis, untarget analysis

**Acknowledgements:** This work was supported by the Slovak Research and Development Agency under the Contract no. SK-SRB-23-0015 and project VEGA 1/0332/24.

## WINE BRANDY CLASSIFICATION BY GEOGRAPHIC ORIGIN BASED ON SYNCHRONOUS FLUORESCENCE SPECTRA AND CHEMOMETRICS

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Brandy is an alcoholic beverage produced by the distillation of fermented grapes followed by aging wine distillate in wooden barrels. Brandies have traditionally been classified by region of production, although soil conditions, climate and grape cultivars used for wine production have less impact on the final product than the distillation and ageing process. A geographical indication identifies a spirit drink as originating in the territory of a country, where a given quality or other characteristic of that spirit drink is essentially attributable to its geographical origin. The best known is Cognac, the style of brandy produced in France region Cognac. Slovak brandy with geographical indication of origin is a Karpatské brandy špeciál produced since 1975 in the Malé Karpaty wine region. Various analytical methods as gas chromatography-mass spectrometry, gas chromatography-olfactometry and infrared spectrometry have been used to classify brandies by region of origin. The application of fluorescence spectroscopy to the analysis of beverages is particularly attractive due to the high sensitivity. The aim of this work was to develop an eco-friendly method for distinguishing Karpatské brandy špeciál from foreign brandies using synchronous fluorescence spectra (SFS). In scanning of bulk and diluted samples, the excitation wavelength was varied from 250 to 500 nm and the wavelength interval was ranged from 20 to 100 nm. Partial least squares-discriminant analysis (PLS-DA) was done on individual SFS, on unfolded SFS and on variables selected by the variable importance in the projection (VIP) algorithm. The performance of the developed PLS-DA models was evaluated by the estimate of the sensitivity, specificity and efficiency. High sensitivity, specificity and efficiency (all 1.0) were obtained using VIP-PLS-DA modeling of SFS of diluted samples.

**Keywords:** eco-friendly method, sensitivity, beverage classification

**Acknowledgements:** This work was supported by the Slovak Research and Development Agency under the Contract no. SK-SRB-23-0015.



## **CAMPYLOBACTER REDUCTION AND MODE OF ACTION OF THE POTENTIAL PROBIOTIC *BACILLUS SUBTILIS* PS-216 *IN VITRO* AND *IN VIVO* IN THE CHICKEN HOST**

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*Campylobacter* sp. is the cause of the most widespread bacterial gastroenteritis, campylobacteriosis. Antibiotic resistance and the spread of *Campylobacter* sp. in poultry and subsequently in the food chain, are a growing problem worldwide, causing enormous economic losses. Regulations on antibiotic use, health and economic concerns have opened the door for more intensive research into alternative solutions to this problem, such as the use of probiotics. In our research we focus on the use of *Bacillus subtilis* PS-216 as a potential probiotic for *Campylobacter* sp. reduction. In co-cultivation experiments *in vitro*, we found a strong inhibition of *C. jejuni* in different model environments imitating the chicken intestines, chicken litter and chicken breast meat. We confirmed this strong inhibition on 20 *C. jejuni* strains (from 1,5 to 6,5 log reduction). We found the main mode of action of *B. subtilis* PS-216 is the production of antimicrobials bacillisine and bacilaene. However, *C. jejuni* strains with an active type 6 secretion system were significantly ( $p < 0,05$ ) less sensitive to PS-216. Furthermore, we investigated the efficacy of *B. subtilis* PS-216 to control *C. jejuni* *in vivo* in broiler chickens challenged with *C. jejuni* (up to 21-days of age). We confirmed that *B. subtilis* PS-216 significantly reduced the level of *C. jejuni* in the cecum content of broilers when treated for the entire period (21 days) for 1,3 log CFU/g cecum content, although preventative (treatment pre-challenge) and therapeutic measures (treatment only post-challenge) did not result in significant differences. Interestingly, the addition of spores resulted in significantly increased weight of broilers, compared to the untreated group. We conclude that *B. subtilis* PS-216 has the potential to be used as a probiotic in poultry to reduce foodborne pathogens and thus increase food safety.

**Keywords:** *Bacillus subtilis* PS-216, probiotic, *Campylobacter jejuni*, food safety, chicken



## NUTRITIONAL PROPERTIES AND OXIDATIVE STABILITY OF COLD-PRESSED OIL BLENDS

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The blending of refined or cold-pressed vegetable oils with the aim of improving their nutritional value or oxidative stability is increasingly used today. In this study, walnut, hazelnut and almond kernel oils obtained by cold pressing were used. The oils were blended in certain proportions, with walnut kernel oil predominating in the blends. Cold-pressed walnut oil is nutritionally very valuable as it is a good source of the omega-3 fatty acid linolenic acid, but is very susceptible to oxidation due to its high content of PUFAs. By increasing the proportion of hazelnut and almond kernel oil in the blends, the content of polyunsaturated fatty acids (PUFA) was significantly reduced and the proportion of monounsaturated fatty acids (MUFA) increased. This naturally contributed to a better oxidation stability of the blends, which was proven by the Schaal oven test and the induction period (h) on the Oxitest device. The basic quality parameters peroxide (PV) and acid value (AV) were determined by standard methods, the composition of the fatty acids was determined by GC. The PV and AV values of the oils and blends tested were within the legally prescribed limits, indicating that the raw material was of good quality and that no oxidation had taken place during cold pressing. The results show that the addition of hazelnut and almond kernel oil in amounts of 25 % and 10 % and 20 % and 10 %, respectively contributes significantly to the nutritional value of the blends as well as to a better oxidative stability of the cold-pressed walnut oil.

**Keywords:** nut-kernel oils, oil blends, oxidative stability, Oxitest

**Acknowledgements:** The authors are grateful to the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, Contract number 451-03-65/2024-03/200116.

## UNSEEN THREATS ON OUR PLATES: PESTICIDE RESIDUES IN HERBS AND SPICES

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Valued for their distinctive aromatic and medicinal properties, herbs and spices are integral to global cuisine. The cultivation and processing of herbs and spices can expose them to various environmental threats. Pesticides are used to protect crops, however, residues of these chemicals or their by-products can remain on/in crops and thus enter the food chain. The EU Rapid Alert System for Food and Feed (RASFF) served as a database for an overview of pesticide contamination in herbs and spices over 2011-2023. The majority of a total of 557 notifications resulted from border control (57.1%), with the highest number of products originating from India (205), as well as from Egypt, Morocco, Israel, etc. Among 122 pesticide residues detected, the most frequently encountered were organophosphates chlorpyrifos and chlorpyrifos methyl with 180 records, related predominantly to cumin, mint, curry and parsley. Ethylene oxide, a class 1 carcinogen, followed with 135 notifications, related to various spice mixtures, paprika, turmeric, etc. A significant number of notifications recorded the presence of carbamate carbendazim/benomyl (51), organophosphates profenofos (50), triazophos (41) and ethion (40), neonicotinoids acetamiprid (41), imidacloprid (29) and thiamethoxam (29) and bifenthrin (37), a pyrethroid ester. Carbendazim and bifenthrin are considered possible human carcinogens. About 44.5% of notifications were linked to serious risk, resulting in border rejection of approximately half of the shipments (49.2%). A diverse spectrum of pesticides used could cause a plethora of harmful effects on human health, making them not only an analytical challenge but also a risk assessment nightmare, because of their frequent co-occurrence – around one third of the notified products was simultaneously contaminated with more than one pesticide residue, of which one third had five or more residues (mostly curry and cumin), reaching a maximum of 13. Nevertheless, a well-functioning food safety system must ensure safe food environment for all.

**Keywords:** food safety, pesticides, public health, RASFF

## A PUBLIC HEALTH PERSPECTIVE OF A DECADE OF RESEARCH ON MYCOTOXIN CONTAMINATION OF CEREAL FOOD AND ASSOCIATED HEALTH RISKS

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From a public health perspective, mycotoxins in food are chronic risk factors in the human diet, with the potential to exert extremely diverse adverse health effects. Cereals, the staple food, are susceptible to the growth of miscellaneous fungi, which produce various mycotoxins. Food contaminated with mycotoxins is unsafe and therefore not food – is that always the case? Data related to mycotoxin contamination of more than 300 cereal products (flour, bread, breakfast cereals, pasta, crackers, biscuits) obtained from multi-year sampling campaigns carried out during the last decade on the market of Vojvodina (obtained by standard liquid chromatography analyzes) were collated and combined with national food consumption data in order to assess the extent of human exposure and consequent threat to public health. The overview of the occurrence data revealed a fairly high frequency of most of the principal mycotoxins, but in low concentrations, except in years marked by drastic climatic conditions during sensitive periods of crop vegetation: hot and dry as opposed to cold and rainy caused contamination with aflatoxins versus fusarium toxins. Regarding the chronic health risk, mycotoxins other than aflatoxins (ochratoxin A, zearalenone, deoxynivalenol, fumonisins) did not pose concerns in any of the population groups, despite the non-compliance of a number of samples with food safety regulations. On the other hand, comparison of the aflatoxin exposure with respective toxicological threshold caused uneasiness, especially in case of toddlers, children and adolescents, while among adults the highest exposure was noted in vegetarians. The findings were similar for regular cereal foods and those labelled as gluten-free. Further concerns relate to aggregate (one mycotoxin from multiple dietary sources consumed in relevant time-frame) as well as cumulative exposure (multiple mycotoxins simultaneously from one or more dietary sources). To effectively protect public health, evidence-based risk prioritization should be incorporated into the national food safety system.

**Keywords:** aflatoxins, ochratoxin A, zearalenone, deoxynivalenol, fumonisins, risk assessment

## COLORIMETRIC VS. REAL-TIME LAMP FOR DETECTION OF GENETIC MODIFICATIONS IN SOYBEAN (*GLYCINE MAX*)

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Genetically modified food is becoming one of the greatest concerns in agriculture, especially owing to the continuously growing diversity of genetic modifications (GM), which is directly contributing to the difficulty of their monitoring and detection. Long-term impacts on human health and environment are yet to be revealed but recent research efforts indicate possible harmful effects. The aim of this study was to compare two approaches of GM detection: a naked eye detection via colorimetric LAMP method and a fluorescence-based Real-Time LAMP method. In order to imitate GMO samples, we prepared soybean leaf and seed gDNA spiked with gBlocks (P-35S and P-FMV elements which are most commonly included in transgenic constructs of GM plants). Additionally, we analyzed how different approaches for soybean gDNA isolation, i.e. robust Chelex-based method applicable in PON settings and a kit-based isolation suitable for laboratory conditions, influence LAMP amplification dynamics. There were no observable differences related to DNA isolation method in case of colorimetric LAMP, whereas Real-Time LAMP showed evidently faster amplification for DNA samples isolated with a commercial kit. Both LAMP approaches showed success in detection of GM for the threshold of 0.001 ng/μl of gBlocks in 1 ng/μl of total gDNA. We determined the same LOD for both Colorimetric and Real-Time LAMP methodologies, but this limit can be significantly enhanced in case of Real-Time LAMP, since this method has great potential for integration in biosensors with different signal readout systems (e.g. electrochemical or MXene-based). These results indicate the supremacy of Real-Time LAMP over colorimetric LAMP in terms of better insight into result accuracy and better reliability.

**Keywords:** GMO, g-BLOCKS, P-35S, P-FMV, LAMP, soybean

**Acknowledgements:** This research was supported by the Science Fund of the Republic of Serbia, #6710, LAMP-based biosensors for detection of GMOs in agriculture – LABOUR, ANTARES project that has received funding from the European Union's Horizon 2020 research and innovation program under grant agreement SGA-CSA. No. 739570 under FPA No. 664387., H2020 MSCA RISE - IPANEMA GA 872662 and the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grant No. 451-03-66/2024-03/200358).

## ASSESSMENT OF TOXIGENIC PROPERTIES IN BLACK ASPERGILLI FOR SAFE ENZYME PRODUCTION IN FOOD PROCESSING

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Black aspergilli (*Aspergillus* section *Nigri*) represent a diverse group of species utilized for enzyme production. While many are beneficial, some produce mycotoxins - low molecular weight secondary metabolites from filamentous fungi that are acutely or chronically toxic. These mycotoxins pose significant health hazards to humans and vertebrates when ingested, inhaled, or absorbed through the skin, making them a critical concern for food safety in industrial applications of fungi for enzyme or bulk metabolite production. Only non-toxigenic strains can be used as source organisms for enzyme preparations intended for food use. Therefore, discovering new potential enzyme producers from the environment is as crucial as enhancing enzyme production with known strains. Ensuring the safe use of enzymes provided by aspergilli in the food industry takes precedence over their enzyme production capabilities. In this study, we present a comprehensive assessment of the toxigenic potential of black aspergilli, combining analytical and genetic approaches to screen newly isolated fungal enzyme producers. We identified and analyzed 39 isolates collected from various substrates in Serbia. The most common species were *Aspergillus tubingensis* (51.2%), followed by *A. niger* (23.1%), *A. welwitschiae* (23.1%), and *A. uvarum* (2.6%). Given that some *A. niger* and *A. welwitschiae* can produce mycotoxins ochratoxin A (OTA) and fumonisins (FB), we assessed the toxigenic potential of selected enzyme producers both analytically and genetically. Since fungal enzyme producers can be considered food-safe only after confirming the absence of mycotoxins in the enzyme preparation (analytically) and the absence of gene clusters responsible for mycotoxin production (genetically), we selected two promising enzyme producers, isolates of *A. welwitschiae*. The absence of both OTA and FB production capability in these isolates was molecularly confirmed by the lack of complete and critical parts of the biosynthetic gene clusters, respectively, making them suitable candidates for further enzyme production development. Although this study focused on local isolates, the approach is applicable globally.

**Keywords:** ochratoxin, fumonisin, *Aspergillus* spp., enzyme, food

**Acknowledgements:** Ministry of Science, Technological Development and Innovation of Republic of Serbia (Contract numbers: 451-03-66/2024-03/200026, 451-03-65/2024-03/200168, 451-03-66/2024-03/200214 and 451-03-65/2024-03/200116).

## MICROBIOLOGICAL HAZARDS IN CULINARY HERBS AND SPICES

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Microorganisms (bacteria, fungi, viruses and protozoa) naturally inhabit the environment. Therefore, contamination of food by microorganisms is not uncommon, and herbs and spices, a food category with a long tradition of use in cooking, are not an exception. Pathogenic microorganisms can seriously endanger health of consumers. One important tool for safeguarding of public health in the European Union is the Rapid Alert System for Food and Feed (RASFF). The aim of the present study was to obtain an insight into microbiological hazards in herbs and spices, both pathogenic and non-pathogenic, as reported in the RASFF database (2011-2023). Out of a total of 848 notifications, 807 were related to pathogenic microorganisms, with products most frequently contaminated by Salmonella (694), followed by a much smaller number of cases registering the presence of Escherichia coli (83) and Bacillus cereus (44). Non-pathogenic contamination of products included presence of moulds or Enterobacteriaceae. The bases for notification were dominantly border controls (62.0%) and official controls on the market (27.2%), while food poisonings cases (0.6%) and consumer complaints (0.2%), although sporadic, are indicators of non 100% functioning of the system. The product most frequently contaminated with microorganisms was pepper (43.2%), while significant numbers of cases were also reported for paprika (7.8%) and basil (5.8%), with the raw materials mainly originated from Brazil (322 consignments, mostly pepper), but also from other countries – India, Vietnam, China etc. Since 76.6% of notifications were characterized as serious public health risks, more than half of the cases resulted in border rejections (54.5%). Substantially smaller proportions of notifications were classified as information (24.2%) and alerts (21.3%). These findings are a significant resource that enables a better understanding of microbiological contamination of spices, thus enabling the improvement of preventive measures and more efficient risk management in the field of food safety.

**Keywords:** Pathogenic microorganisms, Non-pathogenic microorganisms, Food safety, Public health, RASFF

## ENVIRONMENTAL POLLUTANTS IN CULINARY HERBS AND SPICES - WHAT GOES AROUND COMES AROUND

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Environmental pollutants are substances that pollute the natural environment, causing adverse effects on ecosystems as well as human health. The most common types of such pollutants, originating both from the environment and from anthropogenic activities, are polycyclic aromatic hydrocarbons (PAHs) and toxic metals. As many as 16 of the many compounds that structurally belong to the PAHs group, including the most famous benzo(a)pyrene, have been classified as human carcinogens. Regarding toxic metals, cadmium and arsenic are also proven carcinogens, while lead is classified as probable. This study presents an overview of the contamination of culinary herbs and spices with PAHs and toxic metals, based on thirteen years (2011-2023) of data recorded in the Rapid Alert System for Food and Feed (RASFF). RASFF is an EU-based system established to coordinate responses and action taken in order to protect public health. The company's own check (47.9%) and official control on the market (40.8%) were the mechanisms by which the majority of 71 violations were detected, of which 43 were related to the presence of PAHs and 24 to toxic metals. PAHs were found mostly in ginger, paprika and bay leaf. Among the toxic metals, lead was recorded the most, with 16 notifications (of which one third related to turmeric), followed by cadmium (6), mercury (4) and arsenic (1). As a whole, products such as paprika (11.3%), pepper (8.4%), bay leaf (7.0%) and many others, with raw materials mostly originating from China (18.3%), but also from India, Vietnam and other countries, stood out as being of public health concern, in most cases (85.9%) serious, causing about two-thirds of notifications to be classified as warnings (67.6%). The severity of the toxic effects caused by environmental pollutants, including carcinogenicity, strongly argues for risk management actions aimed at ensuring food safety and thereby protecting public health.

**Keywords:** Polycyclic aromatic hydrocarbons, Toxic metals, Food safety, Public health

## PCR DETECTION OF GLUTEN CONTAMINATION IN ORGANIC FOODS

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Wheat, barley, and rye are among the most commonly consumed cereals in the world and are frequently used in food processing. Gluten is a general term for the water-insoluble storage proteins found in these cereals. Glutenin is specifically found in wheat, while gliadin, hordein, secalin, and avenin are found in wheat, barley, rye, and oats respectively. While most people can tolerate gluten in their diet, there are several health issues associated with gluten consumption, such as celiac disease, gluten sensitivity, and wheat allergy. The primary dietary recommendation for individuals with these conditions is to avoid products containing gluten, particularly those made from wheat. Molecular methods based on PCR are increasingly being used for gluten detection. This study aimed to examine the contamination of products labeled as "gluten-free" on the Serbian market using the PCR method. The survey was conducted on 20 samples of products labeled as "gluten-free" collected from the domestic market. The CTAB method was used for genomic DNA extraction. The quality of the extracted nucleic acids was determined by PCR analysis using eukaryotic universal primers for 18S rDNA. In 6 samples the reaction was not satisfactory due to food processing. The remaining samples were screened using primers WBR11/WBR13 for gluten. Wheat DNA fragments were sized at 201 bp, rye DNA fragments at 201 bp, and barley DNA fragments at 196 bp. TR01/TR02 primers specific to wheat were used to differentiate between wheat and rye products. PCR amplification using primers WBR11/WBR13 and TR01/TR02 showed the presence of gluten in 3 samples, originating from wheat. All samples are from the same manufacturer, and the product declarations do not emphasize the possible presence of wheat allergens. Therefore, careful food handling is crucial because contamination could occur at any step of the food production chain.

**Keywords:** gluten, gluten-free, organic food, celiac disease, PCR

**Acknowledgements:** This research was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant number: 451-03-66/2024-03/200032, and by the Provincial Secretariat for Agriculture, Water Management and Forestry, Vojvodina, Serbia, grant number 104-401-7364/2022-01.



## APPLICATION OF MOLECULAR METHODS FOR INVESTIGATING THE VARIABILITY OF *PENICILLIUM* SPECIES ISOLATED FROM CITRUS FRUITS

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Species of the genus *Penicillium* are among the most significant agents of citrus fruit spoilage, with the ability to produce mycotoxins that pose a serious risk to food safety and public health. The aim of this study was to use molecular methods to identify and characterize *Penicillium* species isolated from citrus fruits stored at room temperature for seven or more days. The isolates were collected during the winter months of 2021 to 2024. Genomic DNA was extracted from the mycelium of all tested isolates, and PCR amplification was performed using specific primers targeting two different genetic regions: ITS1-5.8S rDNA-ITS2 and  $\beta$ -tubulin gene (BenA). Universal fungal primers ITS1 and ITS4 were used for the amplification of the ITS region, producing fragment sizes between 500–600 bp. To amplify part of the  $\beta$ -tubulin gene, the primer pair Bt2a/Bt2b was used, which resulted in fragment sizes of 500–550 bp for all tested *Penicillium* isolates. Preliminary morphological identification and molecular analysis results revealed that most isolates belong to *Penicillium digitatum* and *Penicillium italicum*, the most common causes of blue and green mold on citrus fruits. The primers used in this study proved to be effective for detecting genetic variability among the tested *Penicillium* isolates. Further research will focus on the application of restriction enzymes in RFLP analysis to achieve more precise identification at the species level. These molecular methods represent a valuable tool for identifying and analyzing variability among *Penicillium* species, which is crucial for tracking contamination sources, improving food safety control, and reducing the risk of mycotoxins in the food chain.

**Key words:** *Penicillium*, citrus fruits, ITS region,  $\beta$ -tubulin, genetic variability

**Acknowledgement:** This work was financially supported by the Ministry of Science, Technological Development and Innovations of the Republic of Serbia (Agreement on the implementation and financing of scientific research work: 451-03-66/2024-03/200222).

## EVALUATION OF THE EFFECTIVENESS OF VARIOUS DISINFECTANTS TOWARD *ESCHERICHIA COLI* AND *STAPHYLOCOCCUS AUREUS* IN PLANCTONIC AND BIOFILM FORMS

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The objective of this research was to evaluate the efficacy of selected food disinfectants on planktonic populations of *Staphylococcus aureus* and *Escherichia coli*, as well as on these microorganisms (MOs) incorporated into biofilms. Two disinfectants were tested: one based on peracetic acid (P) and the other on benzalkonium chloride (D). Their effectiveness on the chosen MO populations was assessed using a quantitative suspension test. The standard colony counting method was utilized to determine their efficacy on bacterial suspensions in tryptone soy agar (TSA). The germicidal effect (GE) of the disinfectants was measured by the decimal reduction ratio. For both MOs, a 100% GE was achieved at the lowest concentration (0.1%) within the shortest exposure time (5 minutes). Biofilm production was confirmed with a crystal violet assay on microtitre plates. Both *E. coli* and *S. aureus* demonstrated strong biofilm production at 25°C, with *E. coli* showing a significantly higher adherence capacity. The GE of both disinfectants was significantly lower on 48-hour biofilms compared to the GE on planktonic cells of the same MOs at the same concentrations. Complete destruction of viable biofilm cells was observed after 5 minutes of exposure to the highest tested concentration (2%) for both disinfectants and MOs. The anti-quorum sensing (anti-QS) activity of disinfectants P and D was evaluated using a qualitative disc diffusion method on the biosensor bacterial strain *Chromobacterium violaceum* CV026. The results indicated that the disinfectants had no anti-QS effect, as the inhibition zones around the discs only represented their antimicrobial activity.

**Keywords:** quaternary ammonium compound, peracetic acid, biofilm, *Escherichia coli*, *Staphylococcus aureus*

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia under the Agreements on the Implementation and Financing of Research (nos. 451-03-47/2023-01/200222 and 451-03-47/2023-01/200051) and Provincial Secretariat for Higher Education and Scientific Research, Autonomous Province of Vojvodina, Republic of Serbia [grant number 142-451-3163/2022-02].

## DEVELOPMENT OF AN ANALYTICAL METHOD FOR MYCOTOXIN DETECTION IN SERBIA: ALIGNING WITH GLOBAL TRENDS

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Fungi commonly contaminate food and feed, producing various metabolites, among which mycotoxins, small-molecule secondary metabolites, pose a significant threat to food safety and human and animal health. Scientific data from Serbia over the past decade have shown an increasing mycotoxins prevalence in agricultural raw materials and food products, with climate change exacerbating this issue by promoting fungal growth and mycotoxin production. To ensure the safe food and feed production, reliable and sufficiently sensitive instrumental methods are essential for mycotoxin detection. Initially, the enzyme-linked immunosorbent assay (ELISA) was commonly used for mycotoxin analysis, as noted in most published studies from Serbia. However, it has largely been replaced by more sensitive and selective techniques, such as high-performance liquid chromatography (HPLC) with diode-array (DAD) and fluorescence detection (FLD), which allow for the reliable detection of individual or a few mycotoxins. Nevertheless, ELISA remains widely used in accredited Serbian laboratories. Over the past decade, more advanced methods, such as HPLC coupled with tandem mass spectrometry (MS/MS), have increasingly been employed for the simultaneous detection of multiple structurally diverse mycotoxins. However, literature data suggest a lack of application of HPLC-MS/MS methods for the simultaneous detection of multiple mycotoxins in Serbia, as developed methods have primarily focused on a smaller number of mycotoxins. Therefore, the aim of this research was to develop an HPLC-MS/MS method for the simultaneous determination of multiple mycotoxins, including regulated, non-regulated, and emerging mycotoxins. Given the crucial economic importance of maize production in Serbia, maize was selected as the target matrix for analysis. The research focused on optimizing sample preparation and enhancing analytical methods to ensure precise results. The successful development of these advanced analytical techniques for mycotoxin detection has significant strategic value for the future of agriculture and public health in Serbia and its neighboring regions.

**Keywords:** food safety, mycotoxins, instrumental methods, Serbia

**Acknowledgements:** This research was funded by the Institute of Food Technology in Novi Sad, under the Seed Research Grant Program for young scientists, ("Development of a high-efficiency LC-MS/MS method for fast and accurate detection of mycotoxins in maize - MycoScan"), through the financial support of the Serbia Accelerating Innovation and Entrepreneurship (SAIGE) Project, a joint investment by the Republic of Serbia, Ministry of Science, Technological Development and Innovation, the World Bank and the European Union; and by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## MYCOTOXIN DETERMINATION IN NATURALLY CONTAMINATED MAIZE: CHALLENGES IN ACHIEVING SAMPLE HOMOGENEITY

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Ensuring sample homogeneity for mycotoxin analysis is particularly difficult because mycotoxins, toxic secondary metabolites produced by fungi, are often unevenly distributed within naturally contaminated maize grains. This uneven distribution is influenced by several factors, including environmental conditions during maize growth, post-harvest handling, and the specific fungal species present. Consequently, obtaining a representative sample that accurately reflects the mycotoxin content of the entire lot is exceedingly difficult. The critical nature of this challenge is underscored by literature data indicating that up to 80% of the total error in mycotoxin analysis can be attributed to sampling. Sampling errors occur because even minor deviations in sample homogeneity can lead to significant variations in analytical results. Such variations contribute to inaccuracies and complicate data interpretation, potentially resulting in erroneous determinations or misrepresentations of mycotoxin concentrations. Thus, ensuring sample homogeneity is a crucial aspect of mycotoxin analysis, requiring well-defined sampling protocols and strategies to reduce inherent variability and associated errors. The study aimed to analyze 16 mycotoxins, including regulated, unregulated, and emerging types, in maize samples using high-performance liquid chromatography with tandem mass spectrometry. Sample homogeneity was maximized by adhering to European Regulation 2023/2782, which outlines the methods of sampling and analysis for controlling mycotoxin levels in food. The analysis confirmed the presence of aflatoxins B1 and B2, fumonisins B1 and B2, and moniliformin in all samples. Furthermore, alternariol, alternariol-monomethylether, and ochratoxin A were detected in some of the analyzed samples. Despite following the regulation, the analysis revealed heterogeneity in mycotoxin distribution within the samples. This uneven distribution leads to significant variations in the final results, making it challenging to accurately determine the true concentration of mycotoxins in naturally contaminated maize. This inherent variability underscores the complexity of accurately assessing mycotoxin levels and the need for rigorous sampling protocols to minimize errors in analytical outcomes.

**Keywords:** mycotoxins, sampling, homogeneity

**Acknowledgements:** The study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Contract number 451-03-66/2024-03/200143).

## INFLUENCE OF DISINFECTANTS ON ADHESION OF BACTERIA *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* AND *Listeria monocytogenes* TO STAINLESS STEEL SURFACES

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Bacterial adhesion to contact surfaces with direct contact to food presents a serious problem because it may lead to their contamination. Hence in our study we evaluated the potential of bacteria *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Listeria monocytogenes* to adhere to stainless steel discs (type AISI 304) with different degrees of surface roughness (Ra = 25.20 – 961.90 nm), a material commonly used in the food industry for processing equipment, which is regularly exposed to cleaning procedures. Consequently, the investigation included the commercial disinfectants hydrogen peroxide/peracetic acid and sodium hypochlorite which were evaluated for their antibacterial and anti-adhesion activity. The adhesion was assessed by the standard plate count method, while the broth microdilution method CLSI M07-A10 was used to determine the minimum inhibitory concentration (MIC) of disinfectants. Our results revealed that all bacteria were able to adhere to stainless steel surfaces, although differences were found for strains and surface roughness. The lowest adhesion rate of each strain was recorded on the roughest stainless steel disc at a Ra of 961.90 nm. Based on the MIC values, both disinfectants were effective in inhibiting the growth of bacterial strains. It was found that the MICs for hydrogen peroxide/peracetic acid and sodium hypochlorite for all tested bacteria were 250 µg ml<sup>-1</sup> and 500 µg ml<sup>-1</sup>, respectively. Further, at a concentration of 1 MIC, the disinfectant sodium hypochlorite reduced the initial bacterial adhesion to stainless steel surfaces to a greater extent than the disinfectant hydrogen peroxide/peracetic. These findings indicate the great applicability of the tested disinfectants for the control of bacterial adhesion in the food industry.

**Keywords:** adhesion, bacteria, stainless steel surfaces, disinfectants

**Acknowledgements:** Ružica Tomičić thank Provincial secretariat for higher education and scientific research, Autonomous Province of Vojvodina, Republic of Serbia (project no. 142-451-3498/2023-01/01).

## CONTENT OF ACRYLAMIDE AND HMF IN WHEAT-BASED COOKIES DEPENDING ON THE TYPE OF CARBONYL SOURCE

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The Maillard reaction (MR) and caramelization generate a multiplicity of sensory active volatiles and nonvolatiles that are the key compounds responsible for the development of aromas, colors and flavors and hence improved food palatability. On the other hand, these chemical reactions can induce the formation of potentially hazardous compounds such as acrylamide and 5-hydroxymethylfurfural (HMF). Free asparagine (Asn) alone may be converted thermally into acrylamide through decarboxylation and deamination reactions but the yield of acrylamide from Asn is much higher when a carbonyl source is present. In addition, HMF is produced by dehydration of hexose sugars. For this reason, the objective of this study was to evaluate the impact of different types of carbonyl sources on the formation of acrylamide and HMF in whole-grain wheat-based cookies. The recipes differed in the type of added sugar, while the total amount of sugar was kept constant. Sugars (sucrose, fructose, glucose, dextrose, brown cane sugar and coconut sugar, maltex), sweeteners (agave syrup), sweet sugar substitute (stevia), sugar alcohols (erythritol and xylitol), natural sweeteners (acacia honey, meadow honey) and jams (apricot jam, pumpkin jam) were used. Baking was performed at 18°C for 7, 10 and 13 min. According to the results, the type of carbonyl source has a notable effect on acrylamide formation. The yield of acrylamide was the highest in cookies with added fructose, then glucose, with added sweeteners and sugar additives that primarily consist of fructose and glucose, as well as with the added stevia as a sweet sugar substitute. Cookies with the addition of sucrose, dextrose and alcoholic sugars had the lowest acrylamide content. 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.

**Keywords:** whole-grain wheat-based cookies

**Acknowledgements:** This study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grant No. 1. 451-03-66/2024-03/200040), and the COST Action CA21149.

## THE INFLUENCE OF FILTER ON THE CONTENT OF POLYCYCLIC AROMATIC HYDROCARBONS IN SAUSAGE FROM TRADITIONAL SMOKING

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Polycyclic aromatic hydrocarbons (PAHs) are organic compounds that can cause many complications in human body, including increased risk of cancer. These compounds are formed during incomplete combustion of organic materials, during the smoking process, and can be transferred into meat products. Filters used during smoking in traditional conditions can reduce the content of PAHs compound in final products. Therefore, the aim of this research was to determine the content of 16 US EPA PAH (United States Environmental Protection Agency list) in sausages smoked in traditional conditions directly exposed to smoke (K1, K2 and K3 groups) and smoked with a gravel filter between the fire and the sausages (F1, F2 and F3 groups). The sausages were at a distance of 2 m from smoke source (fire) and smoking time was 2 hours (K1 and F1 groups), 3 hours (K2 and F2 groups), and 5 hours (K3 and F3 groups). Additionally, the impact of the smoking process on sensory and instrumental colour characteristics (CIE  $L^*a^*b^*$  system) was investigated. Smoking time of 5 hours had a positive effect on colour sensory evaluation and influenced on lower values of lightness ( $L^*$ ). Also, it was found that sausages from the F3 group had significantly ( $P<0.05$ ) lower 16 US EPA PAH content compared to sausages from the K3 group (316.81  $\mu\text{g}/\text{kg}$ ; 439.31  $\mu\text{g}/\text{kg}$ , respectively), as well as F1 compared to K1 (129.77  $\mu\text{g}/\text{kg}$ ; 233.38  $\mu\text{g}/\text{kg}$ ; respectively). According to the results obtained in this study, it is advisable to implement a smoking process with smoke purification filter under traditional conditions to obtain products with reduced levels of polycyclic aromatic compounds and desirable sensory characteristics.

**Keywords:** PAHs, gravel filter, traditional smoking

**Acknowledgements:** This study was part of Project No. 451-03-65/2024-03/200134 which is financially supported by the Ministry of Education, Science and Technological Development, Republic of Serbia.



## ANTIMICROBIAL PROPERTIES OF HONEY FROM SLOVENIA AGAINST BACTERIAL FOODBORNE PATHOGENS

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Honey has been used for nutritional and also therapeutic purposes since ancient times and has been found to have antimicrobial and healing properties. The antimicrobial properties of honey depend on several factors, including phenolic compounds, enzymatic production of hydrogen peroxide, high osmolarity and low pH level. Besides the botanical origin, classified into the nectar/floral (light) and mannin/forest (dark) honey, and geographical origin of honey, the antimicrobial activity also depends on the bees, environmental, seasonal, and climatic conditions that affect the composition of the honey, as well as the time of honey collection and subsequent handling. The spectrum of antimicrobial activity of 71 honey samples of defined botanical origin (meadow, linden, acacia, fir, chestnut and forest) from all Slovenian regions from the 2023 and 2024 seasons was assessed against Gram-positive (*Staphylococcus aureus*, *Listeria monocytogenes*, and *Bacillus cereus*) and Gram-negative (*Campylobacter jejuni*, *Escherichia coli*, and *Pseudomonas aeruginosa*) bacteria by determining the minimum Inhibitory (MIC) and minimum bactericidal concentration (MBC) using the microdilution method. *C. jejuni* and *L. monocytogenes* proved to be the most sensitive microbial targets, which is favourable given the fact that they are foodborne pathogens with the highest incidence and morbidity, respectively. When comparing dark and light honeys, the dark varieties proved to be more effective, especially against *C. jejuni*, *E. coli*, *S. aureus*, and *L. monocytogenes*. Chestnut honey exhibited the highest overall antimicrobial activity, while acacia honey demonstrated the lowest. Fir honey demonstrated strong antimicrobial properties comparable to chestnut honey, especially against *E. coli*, where both types of honey yielded similar results. The best antimicrobial effect was observed against *C. jejuni* while the weakest was against *B. cereus*. Due to high biodiversity, unique nature, and excellent beekeeping conditions, Slovenia is ideal for production of various types of high-quality honey, which has bioactive properties and wide range of applicability.

**Keywords:** honey, bioactivity, biodiversity, antibacterial, microdilution method

**Acknowledgements:** The results were achieved within the framework of the Program of measures in the field of beekeeping in the Republic of Slovenia in the years 2023-25, which was financed from the state budget and the budget of the European Union.



## AN INTEGRATED TOOLBOX FOR IMPROVED VERIFICATION AND PREVENTION OF ADULTERATIONS AND NON-COMPLIANCES IN ORGANIC AND GEOGRAPHICAL INDICATIONS FOOD SUPPLY CHAIN

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Farm to Fork Strategy and the Biodiversity Strategy have set a target of increasing by 25% the share of organic agriculture as well as significantly increasing organic aquaculture production, until 2030. However, given their undeniable value, such quality label food products are often prone to adulteration and fraud. THEROS is a Horizon Europe project that brings together a complementary consortium of 17 partners within 6 European countries, with the view to implement an integrated toolbox being capable to modernise the process of verifying organic and geographical indications food products and preventing adulterations and non-compliances, through the use of various technological innovations and data sources, while demonstrating enhanced security, transparency and interoperability in the quality labelled food supply chain.

THEROS approach consists of the following elements:

- Low-cost, digital and scalable solutions that rely on Earth Observation, photonics, Internet of Things and DNA authenticity methods being coupled with advanced analytics, machine learning and artificial intelligence in order to ensure efficient detection of fraudulent cases as well as monitoring of associated quality and sustainability elements.
- Blockchain enhanced traceability system and dynamic digital product passport for improved traceability, security and transparent data governance.
- Platforms and algorithms allowing management and harmonization of heterogenous data as well as their consolidation for the verification and validation of transactions across the supply chain.
- Interfaces to facilitate monitoring and inspections by competent authorities, informed decision making by supply chain actors and policy makers as well as business model driven approaches to support short supply chains of quality labelled food products.

THEROS toolbox components will be extensively evaluated in real life settings through four pilot demonstrations in four different countries (Serbia, Greece, Spain and Czech Republic), while covering diverse requirements, involvement of all relevant actors and different organic/geographical indication food products and fraud/adulteration cases.

**Keywords:** Organic food, Geographical Indications, Food safety, Adulteration detection, food quality

**Acknowledgements:** THEROS project has received funding under grant agreement No 101083579. It is funded by the European Union. Views and opinions expressed are, however, those of the author(s) only and do not necessarily reflect those of the European Union or the European Research Executive Agency (REA). Neither the European Union nor the granting authority can be held responsible for them.

## POLYPROPYLENE MICROPLASTICS IMPACTS THE DIGESTION OF COW'S MILK PROTEINS IN INFANTS

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Cow's milk forms an essential part of the diet of infants as a source of protein and other nutrients. Microplastics (MPs) have been found in milk products including infant formula. Despite overwhelming evidence that MPs are detrimental to human health, the impact of MPs on the digestion of proteins in infants has not been studied. To address this research gap, the current study investigated the *in vitro* digestion of cow's milk proteins in the presence of polypropylene MPs (PP-MPs) in simulated gastric fluids (SGF) using an infant digestion model (pH = 5.0; pepsin activity = 268 U/mL). The simulated *in vitro* digestion of skimmed (<1% fat) cow's milk proteins in infants in the presence of PP-MPs (63–180 µm in size; 20 mg/mL) was performed for 5, 30, and 120 min at 37 °C. For comparison purposes, the same experiments were performed using simulated gastrointestinal digestion conditions in adults (pH=3.0; pepsin activity=2000 U/mL). The final concentration of the cow's milk protein in the digestion mixture was 1 mg/mL. Soft corona and hard corona proteins binding to PP-MPs were extracted by centrifugation, and profiles from protein hydrolysis were analyzed using SDS-PAGE. Selected bands of proteins were then excised from the gels and analyzed using the LC-MS/MS technique. The results revealed that PP-MPs had a negative influence on the process of protein digestion in infants. In addition, binding of proteins that included β-casein, K-casein, α-S1, α-S2-casein, and β-lactoglobulin in the soft and hard corona was observed, and this differed with digestion time. These results indicate that PP-MPs could impede the absorption of proteins in infants, which could cause protein deficiency-related health problems.

**Keywords:** Cow's milk proteins, Polypropylene microplastics, Simulated gastric protein digestion, Soft and hard corona proteins

**Acknowledgments:** This study was supported by the European Union's Horizon Europe program, Microprot project, Grant Agreement number 101090313.

## KINETIC MODELING OF THE INHIBITION OF THE MINIMUM INHIBITORY CONCENTRATION BY CPE EXTRACTS OF HORNED MELON PEEL AGAINST SELECTED FUNGI

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The World Health Organization (WHO) and the Centers for Disease Control and Prevention (CDC) believe that seeking naturally occurring antimicrobial agents is imperative because microbial resistance threatens global development, health, and economy. Given the abundance of bioactive chemicals found in many plant parts, including leaves, roots, fruits, and peels, plants are well-known and have been utilized for a long time as sources of antimicrobial agents. Therefore, the goal of this study was to investigate antimicrobial potential of peel extract of horned melon prepared by innovative cloud point extraction method. To evaluate antimicrobial potential of prepared extract, the minimum inhibitory concentrations (MIC) was determined against *Aspergillus*, *Penicillium*, *Trichoderma*, *Geotrichum*, *Saccharomyces* and *Candida* species, followed by time-kill kinetic study. The results of kinetics profiles indicate the biocide effect for *Trichoderma*, *Penicillium* and *Aspergillus* after a contact time of 6, 48 and 60h for the MIC of 3.75, 7.5 and 30 mg/mL, respectively. Further, the biocide effect was observed against *Saccharomyces* after a contact time of 18h for the lowest determined MIC of 1.875 mg/mL. The reduction in number of viable cells at MIC of 60 mg/mL was observed against *Candida* and *Geotrichum* over the first 3 and 6h, while rise up of viable cells was noticed after 24 and 36h, respectively. Time-kill kinetic studies indicate that extracts obtained by cloud point extraction exhibited biocide and biostatic effect against selected fungi within MIC range between 1.875 and 60 mg/mL, suggesting its potential application as antifungal agents.

**Keywords:** horned melon, green chemistry, cloud point extraction, time-kill kinetic, minimal inhibitory concentration

**Acknowledgements:** This research study was funded by the Provincial Secretariat for Higher Education and Scientific Research of Autonomous Province Vojvodina (Serbia) within the project "New ecological phytopreparation for antifungal and antioxidant treatment of selected fruits and vegetables—EcoPhyt" (grant No. 142-451-2732/2023-01/02).

## ORGANIC MATTER AFFECTS *CAMPYLOBACTER JEJUNI* BIOFILM FORMATION ON MICROPLASTICS

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Microplastics persist in the environment, providing surfaces for organic and inorganic substances, as well as microorganisms, to attach and form biofilms. Consequently, they can transport and spread these contaminants, even along the food supply chain. Recent evidence suggests that microplastics are present in foods such as poultry meat and milk, which are among the main causes of campylobacteriosis in humans. *Campylobacter jejuni* is a fastidious microorganism, so it remains unclear how it persists through all stages, from farm to fork, and even into waste, ultimately posing a health risk to humans. The literature suggests that biofilms protect bacteria against unfavorable environmental conditions. This study examined the ability of *C. jejuni* to form biofilms on microplastic particles and assessed the role of organic compounds in facilitating this process. Plastic disks (5 mm) made from food packaging foil were used as substrates, coated with chicken exudate or milk, while others were left uncoated. Biofilm formation was measured by determining colony-forming units (CFU) at 24, 48, and 72-hour intervals. The findings revealed a significant increase up to 100-fold in biofilm formation on microplastics coated with chicken exudate after 24 hours, while milk-coated microplastics showed bacterial counts 10-fold lower than uncoated ones. These results suggest that chicken exudate provides a more favorable environment for biofilm development compared to milk. This study highlights the potential of food residues, such as chicken exudate, to enhance biofilm formation and facilitate the transmission of *C. jejuni* via microplastics. The insights gained will contribute to future research on microplastic-microorganism interactions and the development of strategies to quantify biofilm formation and assess the microbial transmission risks along the food supply chain.

**Keywords:** Microplastics, Food safety, Biofilm, *Campylobacter jejuni*, Foodborne pathogen transmission

**Acknowledgements:** This study was funded by the Slovenian Research Agency (MR 58148 (TŠ)) and J4-4548.

## THE QUALITY OF EGGS FROM DIFFERENT HOUSING SYSTEMS IN BELGRADE SUPERMARKETS, IN EXTREME TEMPERATURE CONDITIONS

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The aim of the research was to examine the supply and the quality of table eggs produced in different housing systems, i.e., cage, floor and free-range, in the conditions of extremely high summer temperatures. The supply was recorded in 10 Belgrade supermarkets and the analysis of the physical egg quality characteristics (external and internal) included grade M eggs, from 9 egg producers, up to 15 days old, i.e., laid between 3<sup>rd</sup> and 18<sup>th</sup> of July, during the heat wave in Serbia, with an average maximum air temperature of 34.43°C. According to the research results, it can be concluded the eggs from the cage system are still dominant in Belgrade supermarkets, which can be linked to the slow transition of producers to alternative housing systems. In terms of the external egg quality characteristics, statistically significant differences between housing systems were found for the egg shell color and cleanliness, while for the internal characteristics, statistically significant differences were recorded only for the yolk color. As measured by Haug's units, the quality of eggs was found to be below the valid standards, for all supermarkets, producers, and housing systems. The eggs from the free-range system had the highest Hog's units, with an average value of 60.67. The research's findings indicate that exceptionally high, prolonged summer temperatures had a very negative effect on the quality of eggs in Belgrade supermarkets. In order to improve the quality of eggs in the summer season, more research should be done to determine factors affecting egg handling on farms, transportation, storage in distribution centres and retail stores.

**Keywords:** egg quality, housing system, high air temperatures, Belgrade supermarkets

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovations, Republic of Serbia, contracts No. 451-03-66/2024-03/200045.

## *Food Authenticity and Regulation*

## HOW MANAGEMENT AND RESTAURANT CHARACTERISTICS INFLUENCE TRADITIONAL FOOD CONSUMPTION IN VOJVODINA'S TOURISM SECTOR (SERBIA)?

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The study investigated traditional food products (TFP) as critical elements in preserving tradition within the hospitality and tourism industries of Vojvodina (Republic of Serbia), focusing on the perspectives of 300 restaurant employees in managerial positions. It explored management's attitudes towards TFP and examined whether these attitudes varied depending on respondent and restaurant characteristics. Additionally, the study analyzed the procurement and consumption of TFP to provide further insights for operating within the hospitality and tourism sectors. The non-parametric Kruskal-Wallis test was employed to identify significant differences, with Dunn's post hoc test used to determine statistically significant differences between the observed groups. In the second part of the study, a binary logistic regression model was applied to identify which variables significantly influence respondents' decisions to purchase TFP within their field. The results indicated that educational attainment and educational field significantly impact views on TFP offerings in the hospitality and tourism industry, restaurant offerings, and the advancement of the business sector.

**Keywords:** Traditional food, restaurant management, procurement, consumption, tourism

**Acknowledgements:** This research was funded by the Provincial Secretariat for Higher Education and Scientific Research under the project titled "Improving the sustainability of the production of traditional food products of Vojvodina through an innovative approach to placement in the hospitality and tourism offer," Grant No. 142-451-3503/2023-02. The authors express their gratitude for this support. Additionally, the authors are grateful to the Ministry of Science, Technological Development, and Innovation of the Republic of Serbia for supporting this work (Grants No. 451-03-66/2024-03/200125 & 451-03-65/2024-03/200125).

## CHARACTERIZATION OF HONEY FROM THE REGION OF RTANJ MOUNTAIN

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To ensure the protection of honey's geographical origin, it is crucial to identify chemical markers that pinpoint the specific region of production. This paper presents a comprehensive analysis of 76 Rtanj honey samples collected from various locations in the Rtanj Mountain region between 2019 and 2021. The study involved melissopalynological analysis and a thorough assessment of physicochemical properties, including moisture, pH, electrical conductivity, free acidity, glucose, fructose, hydroxymethylfurfural (HMF), and color parameters, as well as evaluation of polyphenol profile and sensory properties of the honey. The melissopalynological analysis of Rtanj honey identified 106 types of pollen, with the *Rosaceae* family, particularly the *Sorbus* group, contributing most significantly. The moisture content of Rtanj honey ranged from  $13.6 \pm 0.23\%$  to  $19.2 \pm 0.07\%$ , and all samples were within the Codex Alimentarius Commission's recommended maximum limit of 20%. Free acidity levels varied from  $23.2 \pm 0.14$  to  $65.6 \pm 0.38$  meq/kg, with only one sample exceeding the 50 meq/kg limit. The pH values of the honey ranged from  $3.42 \pm 0.10$  to  $5.54 \pm 0.22$ . Electrical conductivity ranged between 220 and 1150  $\mu\text{S}/\text{cm}$ . All honey samples showed freshness, as indicated by hydroxymethylfurfural (HMF) levels, which were less than 10 mg/kg. Rtanj honey was found to predominantly contain K at levels exceeding 1000 mg/kg, along with notable amounts of Mg (50 mg/kg), Ca (90 mg/kg), Na (200 mg/kg), Mn (2.5 mg/kg), Fe (3 mg/kg), and other minerals. The phenolic profile of the honey featured several compounds, including epicatechin, *p*-coumaric acid, ferulic acid, naringenin, luteolin, kaempferol, and apigenin, with naringenin being the most prevalent phenolic compound across all samples. The colour of Rtanj honey varied from extra white to light amber. Markers for the geographical origin of Rtanj honey include a relatively low colour parameter  $L^*$  value ( $22.7 \pm 0.19$  to  $59.8 \pm 1.75$ ), a positive  $a^*$  value ( $1.55 \pm 0.07$  to  $22.3 \pm 1.36$ ), and a high K content ( $996 \pm 376$  mg/kg). These markers were validated through principal component analysis (PCA) as effective for authenticating the geographical origin of Rtanj honey. In 2022, Rtanj honey was awarded a geographical indication certificate at the national level.

**Keywords:** honey, Rtanj Mountain, characterization, geographical origin

**Acknowledgements:** This work was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).



## DIFFERENTIATION OF FABA BEAN AND GRASS PEA BASED ON GC-MS ANALYSIS OF AMINO ACIDS

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Food authenticity has become one of the most popular topics in the world, especially in Europe. Due to their richness in proteins, fiber, carbohydrates, minerals and vitamins, legumes are very important for human diet. However, legume authenticity is still an unexplored area. The aim of this work was to differentiate faba bean and grass pea samples based on free amino acid composition. Free amino acids were extracted from the legume samples and derivatized using ethyl chloroformate (ECF). A GC-MS analysis (gas chromatography coupled to mass spectrometry) of the prepared samples was performed and the obtained chromatograms were processed in terms to identify eluting amino acids. Peak surface areas of the identified compounds were collected into a data matrix. The data matrix was created for further data processing. Hierarchical cluster analysis (HCA) and principal component analysis (PCA), as a multivariate statistic tools, were applied to the data matrix to differentiate the legume samples. Dendrogram with the similarity level of 0.5, obtained by hierarchical cluster analysis, demonstrates clear separation of faba bean and grass pea samples into two clusters. Similarity level of faba bean cluster was about 0.67, while similarity level of grass pea cluster was about 0.71. Principal component analysis was employed to create a 3D PCA score plot that enabled distinguishing of the analysed legume samples. Grass pea samples were clustered together in the second octant of the 3D PCA score plot, completely separated from faba bean samples. Considering the obtained results, proposed method for differentiation of faba bean and grass pea shows a great potential in legume authentication based on free amino acid composition.

**Keywords:** food authenticity, legumes, GC-MS, amino acid, multivariate statistics

## SUGAR BASED LEGUME AUTHENTICATION METOD: SNAP BEAN VS FABA BEAN

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Legumes have a great importance in human diet due to their richness in proteins, fibers, carbohydrates and minerals. Moreover, legumes do not contain gluten, so their application is very wide lately. The aim of this paper was to develop a legume authentication method based on sugar composition. Two legume species were included in this research: snap bean (4 samples, 1-4) and faba bean (8 samples, 5-12). Before sugar extraction, lipids needed to be removed from the samples using solvent n-hexane for triple extraction of liposoluble compounds. Further, methanol was used as a solvent for extraction of sugars from the samples. Derivatization of the sugar compounds was performed using reagent trimethylsilyl-imidazole (TMSI). A GC-MS (gas chromatography coupled to mass spectrometry) analysis was performed and the obtained chromatograms were processed in term to identify the eluting compounds. Peak surface areas of identified sugar compounds were collected into a data matrix for further multivariate analysis. By performing hierarchical cluster analysis (HCA), a similarity dendrogram was obtained. Faba bean samples are grouped together in the same cluster with the similarity level of about 0.7. On the other hand, snap bean samples 1-3 are clustered together with the similarity level of also about 0.7, while snap bean sample 4 is an outlier showing the least similarity to all other analyzed samples. This sugar composition based method exhibits a strong potential in developing procedures for legume authentication.

**Keywords:** GC-MS, multivariate statistics, legume authentication, snap bean, faba bean

## BLOCKCHAIN BASED FOOD SUPPLY CHAINS: THE ALLIANCE APPROACH

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ALLIANCE is a European Horizon 2020 initiative aimed at improving seven Food Supply Chains (FSCs) that support the production and distribution of Italian PDO/PGI Extra Virgin Olive Oil, Greek PDO Feta Cheese, French Organic Honey, PGI Asturian Faba Bean, Croatian PGI Lika Potatoes, Italian Organic Pasta, and Serbian PDO Arijle Raspberries. ALLIANCE has created an architecture that provides a comprehensive implementation strategy covering the entire spectrum from data acquisition to data utilization. IoT networks facilitate data collecting through a modular design, allowing for the dynamic integration of various sensor devices. The acquired data are analyzed and saved prior to consumption by the applications that facilitate the FSC management. Blockchain is the fundamental technology employed for secure and immutable data storage in a commercially feasible manner. All stakeholders engaged in an FSC contribute to the Blockchain network and are assured that no individual can anticipate illegal advantages from altering data. However, data storage on a Blockchain network, while safeguarding against illicit tampering, has performance challenges. Efficient data storage necessitates the proper management of data, separating the information designated for Blockchain from those retained in a supplementary database known as off-chain. The concurrent data storage in both Blockchain and off-chain systems is complex, as consistency between the two data repositories is essential, necessitating the removal of data from one database if the other experiences a failure. In ALLIANCE, we iteratively communicated with stakeholders associated with each FSC to develop its corresponding data model and identify the essential data to be maintained on the Blockchain, while the remainder is stored on off-chain. Furthermore, we employed the GS1 EPCIS protocol to provide the data of each FSC in a common way, facilitating in this way the interoperability among the FSCs.

**Keywords:** Blockchain, Food supply chain

**Acknowledgements:** This project has received funding from the European Union's HE research and innovation programme under grant agreement No 101084188 (ALLIANCE). Views and opinions expressed are, however, those of the authors only and do not necessarily reflect those of the European Union or the European Research Executive Agency (REA). Neither the European Union nor the granting authority can be held responsible for any use that may be made of the information the document contains.

## TRADITIONAL MILK PRODUCTS IN THE AUTONOMOUS PROVINCE OF VOJVODINA: CONSUMER ATTITUDES AND PREFERENCES

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This study aimed to assess the position of traditional dairy products in Vojvodina and to analyse which dairy products are most frequently chosen by consumers. A sample of 540 consumers from the Autonomous Province of Vojvodina participated in an online questionnaire survey divided in two parts, socio-demographic characteristics and consumer attitudes and preferences toward traditional food products and position of dairy products in it. In the Autonomous Province of Vojvodina, traditional food products are widely consumed, with milk and dairy products on the significant second place, having a consumption rate of 65.1%, while meat and meat products being the most prevalent having a consumption rate of 85.4%. The most preferred dairy product among consumers in Vojvodina is cheese (58.7%), followed by kajmak (25%). Within the cheeses local varieties are most preferred (44.4%), in addition to *Pirotski sir* (10.2%), *Sjenički sir* (9.26%), *Zlatarski sir* (5.56%) and *Zlatiborski sir* (5.56%). Moreover, other varieties such as *Crnogorski* and *Sremski sir* (3.70%), *Mokrinski* and *Šumadijski sir* (2.78%) are also consumed, but in smaller amounts. The results obtained in this study highlight the significant role of traditional dairy products among the dietary preferences of consumers in Vojvodina, with a particular emphasis on cheese as the most preferred dairy item. The findings also indicate a strong preference for local varieties with unique characteristics what emphasized importance of promoting these products to enhance their availability, which would influence support for local producers and local economy.

**Keywords:** traditional food products, traditional dairy products, homemade cheese, kajmak

**Acknowledgements:** This research was funded by the Provincial Secretariat for Higher Education and Scientific Research under the project titled "Improving the sustainability of the production of traditional food products of Vojvodina through an innovative approach to placement in the hospitality and tourism offer," Grant No. 142-451-3503/2023-02.

## UNTARGETED APPROACHES FOR FOOD AUTHENTICITY

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Food authentication is a process of paramount importance because food fraud negatively affects public confidence in food producers and regulators worldwide, resulting in huge food industry losses every year. Fighting food fraud is a very complex area requiring several intervention strategies. The possibility to detect fraudulent activities is closely related to the type of fraud, ranging from altered composition and labelling, geographical origin, mixtures and mimicking supplements, to claims of sustainable production or unethical actions. The different types of analytical methods used to detect food fraud can be classified in two main categories, *i.e.* targeted and untargeted methods. Although many conventional targeted methods, based on the analysis of specific marker compounds, are commonly used to detect fraud, they are time consuming, expensive and require skilled personnel. For this reason, in the last decade, untargeted methods have gained great interest in this field thanks to their rapidity and high potential for the authentication processes. Untargeted methods applied to food authenticity issues are mainly based on spectrometric or spectroscopic techniques in combination with chemometric analysis, which allow the detection of a huge number of metabolites and rapid identification of numerous deviations from the expected food composition, providing a comprehensive characterization of complex food matrices. Although many studies have already demonstrated the feasibility of using untargeted approaches for food authenticity purposes, their uptake and implementation into routine analysis is still limited. Indeed, there is a lack of harmonized and standardized untargeted methods to be used for routine analysis and official control. Potential and limitations of untargeted methods for food authentication will be presented. Furthermore, an overview of rapid untargeted methods that we have recently developed and validated, based on NMR, DART-MS, MS-nose and IR (using both benchtop and portable/pocket instruments) technologies, to assess food authenticity will also be provided.

**Keywords:** food authenticity, fraud, geographical origin, untargeted methods, chemometrics

## *Packaging and shelf life*

## THE INFLUENCE OF BLENDING AND LAMINATION OF DIFFERENT OILSEED CAKES ON THE PROPERTIES OF COMPOSITE BIOPOLYMER FILMS

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The use of food processing industry waste to obtain new biopolymer materials has been on the rise in recent years. The main residues of oil industry processing are oilseed cakes - so far underutilized. The aim of this paper was to produce biopolymer films based on two oilseed cakes – pumpkin oil cake (PuOC) and Camelina Sativa oil cake (CSoC). The films were produced by blending and laminating film-forming suspensions obtained from both oilseed cakes in various ratios (25%-75%, 50%-50%, and 75%-25%). Plain biopolymer films served as controls. The aim was to characterize the obtained 8 film samples regarding mechanical, physico-chemical, and barrier properties and to check if, and how, each blended/laminated sample differs from controls. The obtained results pointed to better mechanical properties of the films obtained from PuOC and all samples with a higher ratio of this oil cake in its composition. On the other hand, CSoC transferred better barrier properties against water vapor to the samples with a higher ratio of applied CSoC. Both mechanisms – lamination and blending are effective methods for improving the mechanical and barrier properties of biopolymer films. Still, lamination turned out to be a more effective technique if strong mechanical strength and good barrier qualities were essential. Blended films may be the better option in applications where biodegradability and ease of processing are more crucial.

**Keywords:** oilseed cakes, blending, lamination, biopolymer films, properties

**Acknowledgments:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant number 451-03-66/2024-03/200134 and 451-03-65/2024-03/ 200134.

## VALORISATION OF SHELL OF INVASIVE CRAYFISH FROM DANUBE RIVER (*FAXONIUS LIMOSUS*): PROTEIN EXTRACTION AND CHARACTERIZATION

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In order to deal with invasive crayfish *Faxonius Limosus* impact on the native crayfish, as well as fish biodiversity in the Danube River, possible solution would be to find and adopt mechanisms for its utilizing for novel valuable products production. Apart from utilizing edible part for novel food products, shell can be also considered as a source of valuable compounds. Complex structure of shell is mainly composed of three basic compounds: chitin, protein and minerals-mainly calcium carbonate. In this paper, shell proteins were extracted using three extraction routs. The first route was to use naturally present enzymes (proteases and lipases) in crayfish wastes and recover proteins using autolysis process. To accelerate the process, UV radiation was used. Remaining two extraction routs were alkaline extraction of proteins, where in one rout alkaline extraction was applied directly to the shell and in the other rout alkaline extraction followed the step of acidic demineralization of the shell. Obtained protein concentrates were analyzed for yield, crude protein content, DPPH radical scavenging ability and structure using FTIR spectroscopy. Results have shown that similar percent of protein content was obtained by all three methods: 67-68%, but extraction yield was considerably different. Alkaline deproteinization with or without the step of demineralization resulted in 10 % yield, while UV radiation accelerated autolysis resulted in only 3,41 % yield. Although proteins extraction without using exogenous enzymes or chemicals is very interesting approach, drawback of this approach is low process yield. FTIR spectroscopy revealed secondary structure that was similar in all three concentrates, according to the peak deconvolution, whit autolitic concentrate differing in a lesser extent, having slightly higher share of sheet structures. DPPH assay revealed high antioxidant activity of the concentrates (72-88 %), probably originating from active peptides derived from proteins and residues of carotenoides led by astaxanthin.

**Keywords:** crayfish, shell, protein, FTIR, DPPH

**Acknowledgments:** The research was supported by the Science Fund of the Republic of Serbia, 7417, Reducing the negative impact of invasive crayfish *Faxonius limosus* in the Danube by smart exploitation of their meat and shells – DANUBEcare, as well as Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Registration number: 451-03-66/2024-03/200134 and Registration number: 451-03-65/2024-03/200134).



## THE PRIORITIES TO REDUCE FOOD LOSS AND WASTE IN THE FOODWASTOP COST ACTION

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Food loss and waste (FLW) is a global challenge recognised by international governments and organisations. Reducing FLW is key to sustainably ensure nutritional food security for an increasing world population. It is a target of the Sustainable Development Goals of the United Nations, and the Farm to Fork Strategy of the European Green Deal. The FoodWaStop COST project addresses these challenges and aims to: (i) build an interdisciplinary and multi-actor European Network that is also connected with non-EU Mediterranean countries, to promote knowledge on FLW beyond the state of the art; (ii) determine incidence of FLW in the critical points of the fruit and vegetable value chain; (iii) foster technological innovations and sustainable management strategies to reduce and prevent FLW; and (iv) valorise agrofood waste to promote a circular bio-economy. The experience of the Coordinators and Participants gained from other related projects (e.g., PRIMA, H2020, HE, other COST Actions), the background from diverse EU and extra-EU countries, and the involvement of stakeholders and industry partners contribute to increase awareness of this problem, to determine its incidence, to seek strategies for its management through exploitation of the potential of innovative technologies, and to define good practices to prevent FLW. The FoodWaStop Network provide benefits to various stakeholders and end-users, including all actors in the agrofood value chain, from farmers (Farm) to consumers (Fork). FoodWaStop COST CA22134 Action started on 21 September 2023 and currently involves over 500 working group (WG) members from over Countries, joining one or more of the 6 WGs. Interested people are welcome to contribute sharing their knowledge applying for WGs at the link [FoodWaStop](#). The first plenary meeting was hold in Ancona on 24-25 January 2024 (see <https://stopmedwaste.net/#postharvestancona2024>) and opportunity for young students were planned, with over 20 Grantee of short term scientific missions (STSM), 26 participants to a Training school on quantification of food loss and waste organized in Cranfield on 9-11 July, and further activities in progress to be open. All information is mainly published on Facebook FoodWaStop page (<https://www.facebook.com/profile.php?id=61551787798541>), as well as on X (<https://twitter.com/CostFoodwastop>) and Instagram (<https://www.instagram.com/foodwastopcostaction2023/>) accounts, and on LinkedIn profile (<https://www.linkedin.com/company/foodwastop-cost-action-ca22134/?viewAsMember=true&success=true>). The video of the first plenary meeting are available on the YouTube channel ([https://www.youtube.com/channel/UCZ8PLeRL\\_OK9PYMVv1Dou\\_A](https://www.youtube.com/channel/UCZ8PLeRL_OK9PYMVv1Dou_A)). Any contribution is welcome, and the FoodWaStop COST Action aims to create a knowledge platform that will promote innovation, deliver guidelines, and favor dialogue with policymakers, to focus their attention on the social and economic implications of FLW.

**Keywords:** Agrofood waste, Euro-Mediterranean knowledge hub, Sustainable food management, Circular bio-economy, Socio economic empowerment of smallholders

## THE USE OF ESSENTIAL OILS AND ESSENTIAL OIL NANOEMULSIONS IN THE DEVELOPMENT OF EDIBLE PECTIN-BASED COATINGS

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The increased demand for food that is nutritionally rich and safe for consumption affects the use of different methods and materials to extend its shelf life. Today's use of plastic and similar packaging results in large amounts of waste that later creates environmental problems, therefore the increasing use of natural and biodegradable coatings and films is being examined today extensively. The edible coatings have long been used to provide natural casing materials for meat products, to delay respiration rate and moisture loss of fruits and vegetables, and to prevent diffusion of moisture and oxygen into nuts. These coatings can be enriched with other additives, which affect the improvement of aroma, colour, taste, smell and appearance of food. Moreover, they have more functional properties than classic packaging because they can contain different bioactive substances, so they can have antimicrobial and antioxidant properties, as well as probiotic activity. An important source of biologically active components are essential oils, which are increasingly used in the food industry, especially because of their antimicrobial effects, and as a natural preservative. Pectin is a natural thickener, stabilizer and gelling agent, and due to its extremely good water solubility and gelling ability, it belongs to the most commonly used hydrocolloid in the production of edible coatings. Pectin coatings have good barrier properties for gases, are environmentally friendly, biodegradable, available and cheap. Incorporation of essential oil into the pectin-based coatings, can create active packaging systems, which provide more efficient preservation and shelf life of fresh and processed food items. The aim of this paper is the review of the possible use of essential oils in development of pectin-based edible coatings. Moreover, the paper will also refer to the application of essential oil nanoemulsions as a new field of study for the development of edible coatings and films which can extend the shelf life of food.

**Keywords:** edible coatings, essential oils, food preservation, pectin

## FOOD WASTE IN SERBIA: CHALLENGES AND STRATEGIES FOR SUSTAINABLE MANAGEMENT AND REDISTRIBUTION

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The Serbian tradition of welcoming guests with abundant meals signifies good hospitality. However, Serbia lacks an established food waste management system, resulting in significant surplus food generation across all social groups without a mechanism to redirect it to those in need. Annually, approximately 770,000 tons of food are wasted in Serbia, an untenable luxury for a country with an average monthly salary below 500 Euros. The absence of accurate data on food production and waste exacerbates the issue. In Belgrade, which produces around 550,000 tons of municipal waste annually, it is estimated that 30% is food waste, totalling about 165,000 tons. This situation highlights a critical gap in food waste management and redistribution, affecting vulnerable groups such as the urban poor and the elderly. Current habits, such as purchasing more food than necessary and restaurants increasing portion sizes, significantly contribute to food waste. Experts estimate that about 90% of total waste in Serbia ends up in landfills, leading to substantial greenhouse gas emissions. Efforts are underway in Belgrade to introduce a voluntary scheme for sustainable food waste management, starting with mapping the largest food waste generators and creating a digital platform for data consolidation. This initiative aims to minimize surplus food generation and redirect edible food to social groups in need, improving food security and reducing environmental impact. Effective food redistribution systems could notably benefit vulnerable populations, including women victims of violence, by providing safe and reliable food sources. Adopting such models aligns with the global shift towards a circular economy and supports the forthcoming Waste Management Strategy and new Law on Waste Management in Serbia.

**Keywords:** Food waste management, Serbia, Sustainable practices

**Acknowledgments:** The study was funded by the Serbian Ministry of Science, Technological Development and Innovation (Contract No 451-03-66/2024-03/200030).

## ANTIOXIDATIVE AND FUNCTIONAL PROPERTIES OF PLUM OIL CAKE PROTEIN ISOLATE PREPARED BY DIFFERENT DRYING METHODS

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The global demand for proteins is constantly increasing, resulting in the need for science and industry has to explore novel raw materials for protein extraction. Plum oil cake, which is obtained after plum oil cold pressing, has great potential as a nutritious, low-cost material. The high protein content (up to 50%) of this by-product is ideal for valuable protein-rich ingredients extraction. Protein isolates from plum oil cake (PPI) were prepared using different drying methods- thermal drying (PPIT) and freeze-drying (PPIF). Obtained isolates were compared in terms of their antioxidative properties and techno-functional properties. Protein content and process yield were also examined, resulted in high protein content (over 96%), with no influence of drying method. Light color of PPIF would be more appealing for consumers and more suitable for incorporation in food systems. Functional properties were not significantly affected different drying methods, except for protein solubility. Both PIs exhibited minimum protein solubility at pH 5.0 and maximum solubility at pH 10.0, while PPIF was much more soluble compared to PPIT. The freeze-drying method led to a much higher antioxidant activity of PPIF. Overall, protein isolates from plum cake obtained from different processing methods differed in appearance, solubility and antioxidant capacity, but yield, protein content and other functional properties were similar. This information will be useful to optimise the production of this protein isolate and benefit its applications.

**Keywords:** plum oil cake protein isolate, drying, functional properties, antioxidative activity

**Acknowledgements:** Research within this work was financed by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, program number: 451-03-65/2024-03/ 200134 and 451-03-66/2024-03/ 200134.

## PLUM OIL CAKE PROTEIN ISOLATE-BASED BIOFILMS: INFLUENCE OF PROCESSING PARAMETERS ON MECHANICAL PROPERTIES

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As food consumption is expected to increase in the coming years, the harmful effects of the food system must be minimised. In addition, packaging materials - especially plastics - generate significant amounts of waste, which, due to their slow degradability, have serious disadvantages for flora and fauna. Furthermore, petroleum-derived plastics are derived from non-renewable sources, which is why there is an urgent need to replace them. Modern technologies are becoming more efficient, consumer awareness is increasing and science is focussing heavily on reducing food waste. In view of the need to valorize food waste and obtain environmentally friendly packaging materials, biopolymers offer solutions. Proteins from renewable sources are commonly used to make biodegradable films for food packaging. A high protein content in by-products is advantageous for the extraction of proteins. In plum oil cake (POC), the protein content is up to 40%, which is considered a favourable, reliable and cost-effective alternative for the development of biofilms. Plum oil cake protein isolate (POCPI) was used to obtain biofilms, by varying the process parameters: Glycerol content (10%, 20%, 30%) pH (8, 10, 12) and temperature (60 and 90 °C). POCPI films were characterized by examining the mechanical (thickness, tensile strength and elongation at break) properties. It was found that the glycerol content had the greatest influence on the mechanical properties. Films with 30% of glycerol exhibited the maximum mechanical properties. Tensile strength values were up to 11.52 MPa and elongation at break values were up to 149%. As the POCPI films exhibited good mechanical properties, they can be considered as a potential biomaterial for food packaging.

**Keywords:** plum oil cake, protein-based biofilm, processing parameters, mechanical properties

**Acknowledgements:** Research within this work was financed by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, program number: 451-03-65/2024-03/ 200134 and 451-03-66/2024-03/ 200134.

## COMPARATIVE ANALYSIS OF MECHANICAL PROPERTIES OF CHITOSAN AND COMPOSITE CHITOSAN-STARCH FILMS WITH ANTHOCYANIN EXTRACTS

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The mechanical properties of pure and modified chitosan films are the main focus of this study. Four groups of chitosan films were prepared: control (CH-C), modified with starch (CH-ST-C) anthocyanin incorporated chitosan (CH-E), and anthocyanin incorporated chitosan-starch (CH-ST-E). The anthocyanins were obtained from purple cabbage, using solvent extraction. The puncture test and tensile test were performed in order to evaluate Young's modulus, tensile strength, breaking strength, deformation, and elasticity using texture analyser (MicroStable systems). The results indicated that CH-C films had the highest breaking strength (2.405 MPa) and deformation (9.840%), demonstrating superior resistance to applied force compared to other films. On the other hand, CH-ST-E films exhibited the lowest breaking strength (0.199 MPa) and a deformation of 8.355%, suggesting significant weakening as the extract is added. CH-E films had a slight improvement in tensile strength (11.094 kPa) over CH-C films (10.082 kPa), whereas CH-ST-E films had the lowest tensile strength (1.226 kPa). Elasticity was highest in CH-ST-E films (14.222%), indicating enhanced flexibility with the addition of anthocyanin, while CH-C films had the highest Young's modulus (148.225 kPa), reflecting their stiffness. The incorporation of anthocyanin extract generally reduced the breaking strength and deformation, but improved the elasticity, especially in modified chitosan-starch films. These results are significant because they highlight the potential use of these films in flexible packaging systems. The inclusion of anthocyanin is a promising alternative for various packaging applications, as visual freshness or spoilage indicator of food products.

**Keywords:** chitosan, chitosan-starch, anthocyanin, mechanical properties

## COMPARISON OF PROPERTIES BETWEEN STARCH-GELATIN FILMS WITH AND WITHOUT ANTHOCYANIN EXTRACT DERIVED FROM PURPLE CABBAGE

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This study is focused on characterization of composite starch-gelatin films with and without the addition of anthocyanin derived from purple cabbage, aiming at developing intelligent packaging system that can be used to monitor food quality. The comparison is made between two films: control composite starch-gelatin films (ST-GEL-C) and anthocyanin incorporated composite starch-gelatin films (ST-GEL-A). A comprehensive analysis was conducted to determine the thickness of the films, moisture content, optical properties, mechanical properties, degree of swelling, solubility and WVP. The results demonstrated that the incorporation of anthocyanins significantly affected the properties of the starch-gelatin films. The inclusion of anthocyanin extract resulted in altered transparency, absorbance and transmission spectra, which highlights the potential use of these films as visual spoilage indicators. The changed color of the films at different pH (2-12) was also monitored and photographed and it showed that this film can be successfully used as intelligent packaging. Additionally, the mechanical properties and water vapor permeability of the films were assessed to determine their suitability for food packaging applications. ST-GEL-A films displayed higher tensile strength and elasticity, as well as thickness. The findings suggest that anthocyanin-enriched films not only provide enhanced functional properties but also offer a promising approach for intelligent packaging solutions.

**Keywords:** starch-gelatin, intelligent packaging, anthocyanin

## PRESERVING TRADITIONAL CULTIVAR "DUGA BELA" PEPPER: THE EFFICACY OF HOT WATER DIPPING

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The "Duga bela" pepper (*Capsicum annuum* L.), a traditional Serbian cultivar, is highly valued for its unique flavor and cultural significance but faces challenges in maintaining quality during postharvest storage. This study investigates the efficacy of hot water dipping (HWD) as a postharvest treatment to enhance the shelf life and preserve the quality of "Duga bela" peppers. Peppers were subjected to three different storage conditions: recommended storage at 10 °C, cold storage at 4 °C, and cold storage at 4 °C following HWD at 55 °C for 1 minute. The results indicated that HWD, combined with 4 °C storage, significantly reduced chilling injury and decay, maintained better color intensity (ASTA units), and preserved quinic and succinic acid content compared to the other storage conditions. Notably, HWD-treated peppers displayed the best overall quality and marketability after 21 days of storage, followed by an additional 3 days at shelf-life conditions. The findings underscored the potential of HWD as an effective postharvest treatment for traditional pepper cultivars, supporting both quality preservation and the extension of marketability. This approach may offer a practical solution for the postharvest management of culturally significant agricultural products.

**Keywords:** *Capsicum annuum*, quality, postharvest treatments, shelf life

**Acknowledgements:** This study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (under Grants: 451-03-66/2024-03/200222 and 451-03-66/2024-03/200007).



## MANUFACTURING OF ANTIMICROBIAL BIODEGRADABLE FOOD PACKAGING

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Antimicrobial biodegradable packaging represents a dual-pronged strategy for addressing microbial contamination and environmental pollution resulting from plastic waste generation in modern society. Although many efforts have been conducted on the development of antimicrobial biodegradable films in the last decade, there are few commercial products currently available due to the non-universality of the machine and poor performance of the film. In our recent researches, starch/poly(butylene adipate-co-terephthalate) (PBAT) blown films have demonstrated the good potential for loading antimicrobial agents and a combined advantage of the low cost of starch (ca. 1 \$/kg) and the excellent processability and mechanical properties of PBAT (ca. 6 \$/kg). Notably, starch is proved as a smart, affordable, and eco-friendly gatekeeper for the controlled release of the antimicrobials from the biodegradable packaging films. Furthermore, our findings revealed that quaternary ammonium salts (QAS) represent an effective alternative to antimicrobial peptides in starch/PBAT films, which not only enhances the film performance but also exhibits antimicrobial properties, thereby extending the shelf life of beef. Overall, we successfully developed strategies for low-cost and high-throughput preparation of antimicrobial biodegradable packaging.

**Keywords:** food packaging, biodegradable film, extrusion blowing, starch

**Acknowledgments:** This research received funding from the Taishan Industrial Experts Program, National Natural Science Foundation of China (32102015), Great Science and Technology Innovation Project of Shandong Province (2023CXGC010707), Natural Science Foundation of Shandong Province (ZR2020QC222), China Postdoctoral Science Foundation (2022M711965), Young and Innovative Talents Introduction and Education Program of Universities in Shandong Province.

## EFFECT OF N, Ca, MAP, AND 1-MCP TREATMENTS ON ETHYLENE- I<sub>ad</sub> INDEX INTERACTIONS DURING APRICOT STORAGE AND SHELF LIFE

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Aim of this study is to investigate the interrelation between ethylene production, DA index (IAD), and postharvest treatments in four apricot (*Prunus armeniaca* L.) cultivars: 'Buda,' 'NS Kasnocvetna,' 'NS Rodna,' and 'NS6.' Apricots were harvested with IAD values between 0.41–0.80, stored at 1°C for 15 days, followed by 3 days of shelf life at room temperature. Ethylene production and IAD were measured at harvest, after cold storage, and after shelf life. Preharvest treatments included nitrogen (N) and calcium (Ca), while postharvest treatments included modified atmosphere packaging (MAP) and 1-methylcyclopropene (MCP). Results showed cultivar-specific ethylene responses and IAD, with 'Buda' exhibited the most rapid decline of IAD and highest ethylene levels, and 'NS6' showing a lower decrease of IAD, especially under MAP and Ca treatments. Postharvest treatments effectively reduced a decrease of IAD for all tested cultivars. The findings highlight the need for cultivar-specific calibration of the IAD index to optimize the management of apricots at a given ripeness stage, thereby maintaining their overall quality and storage capability.

**Keywords:** stone fruit, postharvest, DA meter, preharvest treatments

**Acknowledgements:** The research was financed by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, Faculty of Agriculture, University of Novi Sad, number 451-03-66/2024-03/ 200117 and Institute of Food Technology in Novi Sad, University of Novi Sad, number: 451-03-66/2024-03/200222.

## *Sensory and Consumer Science*

## MULTI-MODAL TASTE PERCEPTION ANALYSIS OF WHITE AND BLACK GARLIC BREAD: CHEMICAL PROFILE, TASTE RECEPTORS, AND PERCEPTUAL EFFECTS

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This study investigates how adding white and black garlic to white and whole wheat breads affects their sensory and textural properties. The results indicate that the control group of white bread (KBE) had the lowest hardness value ( $1114.37 \pm 50.98$ ), which significantly increased with the addition of garlic, especially at a 1% concentration (BE-%1SS:  $1214.95 \pm 142.06$ ). A similar increase was observed in whole wheat bread, where the initial hardness ( $1738.01 \pm 235.42$ ) reached its maximum with the addition of 7% garlic (TBE-%7SS:  $3079.15 \pm 229.61$ ). The stickiness analysis showed that white bread's stickiness values increased with garlic addition, while whole wheat bread generally exhibited lower stickiness values. White breads displayed the highest values in springiness, whereas whole wheat bread showed lower springiness. The addition of black garlic decreased the springiness values of white breads. Adding black garlic slightly darkened the color of the breads, with color values ranging from 72.67 to 75.21 at 1% concentration and dropping to 51.14 to 52.78 at 7%. White garlic also had a similar effect, with color values varying from 79.32 to 80.33 at 1% and between 74.99 and 78.73 at 7%. Moreover, the addition of white garlic caused a noticeable decrease in the overall volume of the breads, while their weights remained similar to the control breads. Sensory analysis results indicated that white bread was rated more positively by panelists. Notably, the 1% white garlic-added bread received high scores due to the pleasant contribution of garlic aroma. Although whole wheat bread is preferred for its natural and fiber-rich structure, some panelists found its taste and appearance lacking. In conclusion, the addition of black garlic significantly impacts the textural properties and consumer perception of both white and whole wheat breads. The addition of white garlic enhances the flavor and visual appeal of the breads, providing a positive contribution from a marketing perspective. These findings should be considered in product development and marketing strategies, emphasizing garlic-infused white bread while improving the taste and appearance of whole wheat bread.

**Keywords:** Garlic, Bread, Chemical profile, Sensory analysis, Color analysis

**Acknowledgements:** We would like to express our sincere gratitude to all the panelists who participated in this study. They provided valuable insights and feedback on the sensory characteristics of garlic bread, which was essential to the success of this research. We would also like to extend our heartfelt thanks to the Field Crops Central Research Institute for its support and resources, which facilitated this research. Your commitment to advancing agricultural science and innovation has been instrumental in our work. This work is part of the activities of the COST Action FLAVOURsome, CA22161, supported by COST (European Cooperation in Science and Technology).

## SENSORY PROFILE OF WHOLEGRAIN WHEAT SOURDOUGH STARTER ODOUR

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The wholegrain wheat sourdough starter was developed from a spontaneously fermented mixture of flour and water, containing naturally occurring lactic acid bacteria (LAB) and yeasts. During fermentation, the odour of the sourdough starter evolves due to various chemical reactions, which can provide insights into the fermentation process's progression or potential issues. To monitor the development of sourdough starter odour over time, the Temporal Dominance of Sensations (TDS) method was employed. Twelve experienced sensory panellists evaluated the dominant odour attributes (including bran, flour, dough, yeast, acetic acid, lactic acid, sour milk, yogurt, cheese, and fruity) of six sourdough starters collected every hour over a six-hour activation period. TDS was performed in three replicates, with data collected using Excel and analysed with XLSTAT software. The results were presented as curves showing the dominance rate for each 0.2-second interval during a 1-minute period. The odour profile of the sourdough starter varied throughout the fermentation process. In the early stages, the odour was simple and subtle, primarily featuring flour, bran, and dough notes. As yeast activity increased, the odour became more fruity. With further fermentation, the profile grew more complex and pronounced, incorporating sharp notes reminiscent of sour milk, cheese, yogurt, and acetic acid. These changes in the odour profile highlight the intricate biochemical processes occurring during sourdough starter fermentation.

**Keywords:** sensory analysis, sensory profiling, dominant sensations, odour, sourdough starter

**Acknowledgements:** The research presented in this abstract was financially supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (No. 451-03-66/2024-03/200222).

## **SENSORY ANALYSIS OF MILK CHOCOLATE WITH INULIN AS A SUGAR SUBSTITUTE**

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Overconsumption of sugars in diets is linked to various health issues, including dental diseases, diabetes, and obesity. Despite this, eliminating sugar from products like chocolate remains a challenge for manufacturers. The objective of this research was to examine the impact of inulin on the sensory characteristics of milk chocolate. Two types of milk chocolate with partial (50 % sugar: 50 % inulin – 50S – 50I) and total (100 % inulin – 100I) substitution of sugar with inulin were compared to its conventional counterpart (100 % sugar – 100S), all produced on the laboratory. Chocolate products were subjected to a consumer test using a 10 cm VAS structured scale for overall acceptability, while the intensity of different sensory attributes were examined on 3-point JAR "just about right" scale. The survey involved 72 healthy random participants, who recognized the terminology "alternative sweeteners", but only half of them were aware about their health benefits. When buying the product 80.56% of judges will choose the product according to the taste, texture and appearance and much less will select a product beneficial to health. Newly developed chocolates containing reduced amount or no sugar at all were well accepted in the consumers test with no significant difference compared to the chocolate produced by the ordinary formulation. The intensity of butter and cocoa flavor was less pronounced in inulin containing chocolates and the sweetness was described as not enough. On the other hand, acidity, gloss on the surface, snap and mouthfeel were perceived in optimal range by highest percentage of consumers than the control sample.

**Keywords:** chocolate, functional product, inulin, sensory acceptance, JAR test

**Acknowledgements:** The authors would like to acknowledge University's Student Parliament for funding this project.

## SENSORY EVALUATION AND ORAL PROCESSING OF BUTTER BISCUITS ENRICHED WITH GRAPE POMACE FLOUR

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In the last decade the food loss and food waste became a major concern due to the fact that almost one third of the food produced globally for human consumption is lost or wasted. Food loss usually refers to any food that is discarded, incinerated or otherwise disposed of along the food supply chain from harvest/slaughter/catch up to, but excluding, the retail level, and is not used for any other productive use, such as animal feed or seed. Globally, around 13 percent of food produced is lost between harvest and retail. Grape pomace represents food loss/waste produced from the wine industries after the pressing and fermentation process. This solid waste causes pollution, difficulties in its disposal and huge economic loss. The content of the grape pomace is around 1/3 crushed grape mass rich in value-added products like unfermented sugars, polyphenols, pigments, alcohol, tannins, and lignocellulosic compounds and could be used as a promising feedstock for the production of renewable energy. The aim of this research was to use the pomace from red grapes that is discarded from the biggest winery in the region, to obtain grape pomace flour that can be used to develop new products such as bakery products. Repurposing the food loss from the grape pomace meant drying the wet pomace at 70°C, milling it to obtain grape pomace flour and fractioning it into fractions with different size (0.25; 0.5 and 1 mm). Afterwards butter biscuits were prepared according standard recipe with different concentration of pomace flour (2.5; 5; 7.5 and 10 %) as substitution of the wheat flour. Developing new product usually acquires detailed sensory analysis to evaluate the acceptance of the product among the consumers. Sensory analysis consisted of several tests including olfactory test, descriptive test and just about right test. A panel of 25 participants evaluated the cookies in two separate sessions. For the olfactory test the panellists were presented with brown bottles filled with crushed cookies to evaluate the aroma without tasting. For the sensory analysis they were served 6 g of cookies randomly coded on white plastic plate with glass of water and apple slices to neutralize the taste between different samples. A separate sheet was offered for registering primary sensation. In the questionnaire, panellists were asked to describe the samples and to group them according to the aroma, taste and texture and not the color. Furthermore, oral processing of the biscuits was also performed to investigate the effect of the pomace flour (fractions size and concentration) on the chewing and swallowing of the biscuits. The panellists were given around 3 g of cookies and were instructed to bite the cookie with the front incisors and to chew the sample to the point of swallowing to obtain a bolus. Time of chewing was recorded as well as number of bites. The bolus was characterized for saliva uptake, color, texture and morphology. The results proved that the dominant aroma came from the butter and masked the other volatile compounds from the grape pomace regarding the sensory acceptance. On the other hand the concentration and the size of the pomace flour affected the oral processing of the cookies. In conclusion, the red grape pomace is a promising source of value-added components that can be successfully used in bakery products.

**Keywords:** sensory evaluation, oral processing, biscuits, grape pomace flour

## UNVEILING SENSORY INSIGHTS INTO GLUTEN-FREE COOKIES WITH SORGHUM FLOUR

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This study evaluated the sensory profiles of gluten-free cookies with 20% and 40% sorghum flour substitutions using the Rate all that apply (RATA) method with 15 assessors. The low-tannin sorghum flour was produced at the Institute of Field and Vegetable Crops. The aim was to assess how these ingredient variations affect sensory attributes and compare these profiles with commercially available products (Comm\_1 and Comm\_2). The basic ingredient composition of the developed cookie formulations included a commercial gluten-free flour blend, virgin coconut oil, cane sugar, and baking powder. Sensory attributes collected from the literature were selected through RATA applied to five cookies. Assessors were instructed to select from a list of descriptors all sensations that described the samples and to rate their intensity on a 5-point scale. A list of 52 sensory attributes was compiled for the characterization of individual cookie samples. ANOVA revealed significant differences ( $p < 0.05$ ) in texture, odour, and flavour among the cookie samples. The addition of 20% sorghum flour resulted in balanced improvements in both texture and odour compared to the control. In contrast, cookies with 40% sorghum flour generally performed well but exhibited a slight decrease in sweetness. Conversely, cookies with notable visual and textural defects (Comm\_1 and Comm\_2) showed poorer performance in sensory attributes. PCA results ( $p < 0.05$ ) showed that the first principal component (PC1) explained 51.53% of the variance, while the second principal component (PC2) accounted for 28.92%, together capturing 80.45% of the total variance. PCA revealed that shape and surface irregularities were associated with poorer visual quality, whereas sweet odour and crispness were linked to better sensory attributes. Notably, control sample achieved the optimal balance of visual quality, texture, and flavour, setting a high benchmark for product excellence. The findings indicated that incorporating sorghum flour significantly enhances the sensory profile of gluten-free cookies, addressing common deficiencies in commercial products. The RATA methodology effectively captured detailed sensory differences, providing valuable insights for refining production techniques and adjusting product formulations to better align with consumer preferences.

**Keywords:** gluten-free cookies, sorghum flour, Rate all that apply (RATA) technique

**Acknowledgements:** The authors wish to express their sincere gratitude to the Ministry of Science, Technological Development and Innovation of the Republic of Serbia for its financial support (grant numbers: 451-03-66/2024-03/200222; 451-03- 47/2023-01/200032, 451-03-68/2022-14/200116).



## CONSUMER ACCEPTANCE OF NEWLY DEVELOPED FUNCTIONAL SNACK PRODUCT

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Two newly developed extruded snack products composed of corn meal (80.7%), brewer's spent grain (14.8%), mechanically deboned poultry meat (3%), chicken liver (1%) and salt (0.5%) with pizza and cheese flavour were subjected to consumer test. Snacks were produced using co-rotating twin screw extruder and seasoned with and clean label pizza and cheese flavour seasoning, using palm oil as binder. During the consumer test, 64 consumers had the opportunity to try and rate developed snacks as well as their commercial alternatives of similar flavours. Respondents first provided sociodemographic information and eating habits, and then evaluate the following characteristics: overall likability, appearance, taste, texture, hardness, crunchiness, and adhesiveness of the product. They also expressed their willingness to purchase these products. The respondents were mostly younger, educated females who were employed, mindful of their diet, but not yet required to adhere to strict diets or avoid fatty and spicy foods, and who enjoyed consuming snacks. Consumer rates of all four rated samples in terms of overall likability, taste, texture, hardness, and crunchiness were not significantly different ( $p>0.05$ ). The control commercial samples were rated significantly higher ( $p<0.05$ ) in terms of appearance, due to the more creative product shapes, as well as the lighter colour of the products. Significantly higher ( $p<0.05$ ) ratings for adhesiveness were given to the nutritionally improved snack products. Pizza-flavoured snack was rated higher in terms of all characteristics compared to cheese-flavoured snack product. Since, the likability ratings of control commercial samples, available on the market for many years, are almost identical to those for the nutritionally improved snacks, it can be concluded that there is place in the snack product market for this type of product as well. Furthermore, 80% of consumers preferred to purchase functional snack products, when were informed about their nutritional content.

**Keywords:** Consumer test, Functional food, Snack products, Brewer's spent grain

**Acknowledgements:** This study was supported by the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Agreement no.: 451-03-66/2024-03/200222.).

## **BARRIERS TO INCREASING FIBRE INTAKE AND STRATEGIES TO ACHIEVE HIGHER FIBRE INTAKES FROM BEHAVIOURAL TO REFORMULATION**

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The benefits of increasing fibre intake have been promoted by public health campaigns across Europe. Simultaneously, the food industry has implemented reformulation and innovation efforts to increase fibre in products with varying degrees of success. Despite these efforts, fibre intakes have remained low in many European countries including the UK and remain below the current recommendations. There are excellent examples of public health industry partnerships which aim to increase fibre intake. For example, the Danish Wholegrain partnership (DWP) aimed to make it easier for Danes to consume wholegrain by increasing availability and make wholegrain a natural part of the daily diet. The DWP was successful in increasing products available carrying a wholegrain label, almost doubling fibre intake of the population as a whole. Here we consider learnings from the DWP and how to apply this to the UK (and by implication other European countries). Differential effects were reported according to SES in Denmark and in other countries people from low SES groups tend to have the lowest fibre intakes and poorest diet quality. They are also most vulnerable to diet related disease. Targeting health behaviour change interventions at low SES groups could reduce dietary-related health inequalities. However, some dietary interventions may increase inequalities by disproportionately benefiting less disadvantaged groups – known as 'intervention-generated inequalities'. This highlights the need to tailor nutritional interventions for low SES groups as a method to reduce health inequalities. Strategies to improve nutritional intake and reduce health inequalities need to take into account the agency of the target population and the resources required to achieve a healthy diet which have been significantly reduced by the cost of living crisis. This presentation will consider barriers to changing diets specifically to increase fibre consumption - especially in low income consumers and the various strategies that can be employed ranging from reformulation to exposure in anchor institutions such as schools via school breakfast programmes. Health by stealth approaches which consider reformulation to increase fibre in familiar foods/products could be one of the most effective methods but requires technical innovation in terms of taste, texture, acceptability and affordability.

**Keywords:** Fibre, reformulation, behaviour change, low income intervention strategies

**Acknowledgements:** L.D. is partly funded by the UKRI-SPF 'Transforming UK food system' programme-funded H3 project (Grant No: BB/V004719/1).

## NAVIGATING FOOD LABELS: A FOCUS GROUPS INVESTIGATION OF SLOVENIAN CONSUMERS' FOOD SAFETY THROUGH PERCEPTION AND USAGE OF FOOD LABELS

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Food labels are of great importance for the consumers as they contain important information (such as food storage and preparation instructions) that can help consumers ensure appropriate food safety of their food. Consumer food safety is often overlooked, but considering that most foodborne outbreaks in the European Union occur in the home environment (EFSA, 2016-2021), the importance of consumer food safety should not be underestimated. Introducing positive changes in consumer food handling requires a good foundation of in-depth understanding of consumer food safety - especially the aspects of consumers' knowledge, attitude and practice related to food safety. The aim of this study was to investigate consumer food safety in Slovenia through a series of focus group discussions about a wide range of food handling activities from shopping to cooking - including the use of food labels. The focus groups were conducted both in person and online and had altogether 40 participants - consumers in Slovenia with different demographic characteristics gender (female, male, other), age (21-80 years old) and education (secondary school, university). There were on average four participants in each focus group with each person only participating in one focus group. Focus groups discussions were structured using 19 questions. The duration of each focus group conversation was approximately 1 to 1.5 hours. The results covered in the presentation include highlights on the topic of food labelling with an emphasis on: 1.) consumers' perceptions and opinions regarding the clarity, usefulness and font size on the labels and 2.) consumers' experiences in using food labels. The diverse examples underline: 1.) possible issues in food labelling that could be informative for the food industry and regulatory authorities to improve food labels and make them more consumer friendly and 2.) challenges in consumer food handling related to following the food label information, which could be important for different interventions aiming to improve consumer food safety.

**Keywords:** consumer food safety, focus groups, food labels, consumers' perception

**Acknowledgements:** The authors acknowledge the financial support from the Slovenian Research and Innovation Agency ("Young Researchers" program - Maja Bensa, research core funding No. P3-0388 and P1-0005).

## A SENSORY PERSPECTIVE ON SUNFLOWER OLEOGEL AND BASIL ESSENTIAL OIL IN LIVER PÂTÉ FORMULATIONS

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This study explored the replacement of animal fat with sunflower oleogel in liver pâté and assessed the impact of basil (*Ocimum basilicum* L.) essential oil additives on its sensory attributes. Eight formulations were evaluated, incorporating oleogel at 20% and 40% substitution levels, and essential oil at 0.075 µl/g and 0.150 µl/g. In addition to oleogel and essential oil, the formulations included various combinations of pork backfat, sodium caseinate, hot broth, lean pork meat, chopped liver, salt, spice mix (comprising pepper, dried onion, and marjoram) to determine their effects on sensory qualities. Sensory evaluation was performed using a trained sensory panel of 10 assessors, who rated the pâté on colour, homogeneity, spreadability, odor, taste, mouthfeel, flavour, and overall quality. ANOVA revealed significant differences ( $p < 0.05$ ) in taste and flavour between different essential oil concentrations, with higher levels (0.150 µl/g) notably enhancing these attributes. Oleogel substitution also significantly affected spreadability and mouthfeel, with 40% oleogel improving spreadability but altering mouthfeel compared to the control. PCA further clarified these findings by showing that the first principal component (F1), accounting for 48.37% of the variability, was strongly associated with taste (0.926) and flavour (0.942), indicating their critical role in overall quality. The second principal component (F2), explaining 22.12% of the variability, was linked to spreadability (0.851) and homogeneity (0.702), highlighting the effects of oleogel on these attributes. The third component (F3), accounting for 14.55% of the variability, was associated with mouthfeel (0.577) and overall quality (0.953). The findings revealed that oleogel is a highly effective substitute for animal fat in liver pâté, while the addition of essential oils notably enhances its sensory attributes. The study confirmed that oleogel not only replaces animal fat but can also improve various quality aspects of the pâté. Furthermore, essential oils, particularly at higher concentrations, significantly enriched the sensory profile, contributing to a more appealing and flavorful product. This approach provides a healthier and higher-quality alternative to traditional liver pâté formulations, ensuring both superior sensory characteristics and enhanced nutritional benefits.

**Keywords:** liver pâté, sunflower oleogel, basil essential oil, sensory evaluation

**Acknowledgements:** The authors wish to express their sincere gratitude to the Ministry of Science, Technological Development and Innovation of the Republic of Serbia for its financial support (Agreement no.: 451-03-66/2024-03/200222).

## CONSUMER ACCEPTANCE AND ATTITUDES TOWARDS JUICES ENRICHED WITH SPIRULINA POWDER

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Sour cherry (*Prunus cerasus* L.) and tomato (*Solanum lycopersicum* L.) are popular cultures in Serbia for juice production. These juices, besides the health benefits that they confer alone, can be fortified with various food ingredients to improve their nutritional properties. Spirulina (*Arthrospira platensis*) is a cyanobacteria biomass used in human nutrition. It is considered a healthy and sustainable food due to its high protein and micronutrient content, as well as its easy cultivation. Spirulina powder is often purified to remove its unpleasant sensory properties, resulting in a blue powder. The presented study aimed to analyse consumer acceptance and attitudes towards juices enriched with spirulina blue powder. Blue spirulina powder was added to sour cherry and tomato juices at two different concentrations (0.8% and 1.6% w/w) per portion (250 ml) to meet the recommended daily intake of 2-4 g. The sensory acceptability of juices (colour, odour, texture, taste, overall) was assessed by consumers (n = 110) using a seven-point hedonic scale (ranging from 1=extremely dislike to 7=extremely like). A survey questionnaire was created to examine the attitude of consumers (n = 137) towards the consumption of juices enriched with blue microalgae. The snowball method was used to distribute the survey among the population. The results of consumers' test showed that juices with the addition of blue spirulina powder were acceptable (values on scale greater than 4). Juices with higher concentration of spirulina received a slightly lower rating for overall acceptability. The majority of respondents who filled out the questionnaire were female (76.6%), university educated (88.3%), employed (83.1%) with an average monthly income above the average (58.4%). Their knowledge about algae is excellent, but most of them have not tried algae or would know how to prepare a dish with it. As the main motives for consuming microalgae, respondents cited a high level of protein and polyunsaturated fatty acids, as well as other positive health effects.

**Keywords:** tomato juice, sour cherry juice, spirulina, consumer acceptance, consumer attitudes

**Acknowledgements:** The Provincial Secretariat for Higher Education and Scientific Research, Autonomous Province of Vojvodina, Republic of Serbia (Project No. 142-451-3144/2023-01/01), the Ministry of Science, Technological Development and Innovation, Republic of Serbia, contract no. 451-03-66/2024-03/200222.

## APPROACHING THE SENSORY ATTRIBUTES OF ARILJE RASPBERRIES WITH PROTECTED DESIGNATION OF ORIGIN

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This study focused on the sensory evaluation of fresh raspberries, specifically the Arilje varieties, which are cultivated in the Arilje region and have a Protected Designation of Origin (PDO). The objective was to approximate the definition of the sensory attributes of *Arilje raspberries* produced within the PDO region. Six raspberry varieties from different locations and altitudes were evaluated, including Vilamet, Fertodi, Tulamin, Enrosadira, Sky, and Miker, with Vilamet and Tulamin being replicated to verify panel performance. A total of 8 samples of fresh raspberries were assessed by a panel of 15 assessors from the "Arilje Raspberry Association" using the Rate All That Apply (RATA) methodology. Sensory attributes were selected through RATA and applied to these raspberry samples, encompassing 47 descriptors grouped into dimensions such as appearance, odour, taste, flavour, texture, aftertaste, and trigeminal effects. The evaluation was conducted with attributes rated on a 5-point scale, and data were analyzed using a two-way ANOVA model and Tukey's post hoc test. ANOVA revealed significant differences in attributes, with Vilamet and Fertodi showing high values for flavour ( $F = 29.826$ ,  $p < 0.0001$ ) and taste ( $F = 9.722$ ,  $p < 0.0001$ ). Fruit size ( $F = 6.865$ ,  $p < 0.0001$ ) and colour ( $F = 13.243$ ,  $p < 0.0001$ ) were significant, especially for Vilamet and Enrosadira. Principal Component Analysis (PCA) identified three key factors: freshness and shape (PC1, 37.078% variance), colour (PC2, 22.459% variance), and flavour (PC3, 16.466% variance). Vilamet and Fertodi correlated with freshness and shape, while Tulamin and Enrosadira exhibited different characteristics. The study showed higher consistency in evaluating Vilamet compared to Tulamin and Enrosadira, suggesting variability in evaluation across attributes and samples. High repeatability in Vilamet and Fertodi may indicate better standardization, whereas Tulamin's lower repeatability could reflect greater quality differences. *Arilje raspberries* were noted for their intense red colour, medium to large size, uniform shape, rich fruity aroma with possible floral hints, and a well-balanced flavour between sweetness and mild acidity. Their juicy texture and long-lasting pleasant aftertaste made them distinctive. These attributes reflected the unique growing conditions in Arilje and supported PDO protection strategies.

**Keywords:** fresh raspberry, Protected Designation of Origin (PDO), Rate all that apply (RATA) method

**Acknowledgements:** This work was financially supported by the ALLIANCE project, funded through Grant Agreement ID 101084188 under the Horizon Europe Programme, as well as by the Ministry of Education, Science, and Technological Development of the Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## CONSUMER ACCEPTANCE OF CROPDIVA SPREADS COMPARED TO COMMERCIAL SAMPLES

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This study assessed consumer acceptance of two novel spreads, based on faba bean and lupine, developed by FINS (Institute of Food Technology, Novi Sad, Serbia) as part of the CROPDIVA project, benchmarking them against three commercially available chickpea-based spreads. The study, conducted with 100 participants during the 2023 "Researchers' Night Festival" in Novi Sad, Serbia, focused on sensory attributes including colour, homogeneity, odour, saltiness, acidity, flavour, spreadability, grittiness, and overall acceptability. Participants rated each spread on a scale from 1 (least acceptable) to 5 (most acceptable). The spreads were presented in randomized order to minimize bias, and data were analysed using ANOVA and post hoc Tukey tests. Significant differences were observed ( $p < 0.05$ ), with a CROPDIVA spread achieving a high overall acceptability score of 4.2, surpassing two commercial spreads (3.8 and 3.6). Commercial spreads excelled in colour and homogeneity, scoring 4.5 and 4.3, respectively, while CROPDIVA spreads were superior in flavour and spreadability. Notably, Sample 857 (Comm2) achieved top scores in colour (4.13), homogeneity (4.54), and spreadability (4.71), with an overall acceptability of 4.01. Sample 426 (faba bean-based spread) showed lower scores in colour (3.85) and homogeneity (3.96) but excelled in spreadability (4.31), with an overall acceptability of 3.79. Sample 149 (lupine-based spread, FINS) had high scores for colour (4.08) and spreadability (4.62), with an overall acceptability of 3.76. In contrast, Sample 762 (Comm 1) received lower scores in colour (3.46), homogeneity (2.99), and spreadability (3.07), leading to an overall acceptability of 3.30. Sample 314 (Comm 3) had moderate scores across attributes, with an overall acceptability of 3.15. The results highlight the strong potential of CROPDIVA spreads in flavour and spreadability but also reveal the need for improvements in visual appeal and consistency. Insights from this research are vital for refining product development and market strategy.

**Keywords:** faba bean and lupine-based spreads, chickpea-based spread, consumer acceptance

**Acknowledgements:** This work was financially supported by the project financed by the H2020-EU.3.2.1. "Climate Resilient Orphan croPs for increased DIVERsity in Agriculture – CROPDIVA" (ID: 101000847) and Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).



## SENSORY ANALYSIS OF FROZEN RASPBERRY VARIETIES

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This study evaluates the quality of frozen raspberries using the RATA (Rate All That Apply) methodology, focusing on sensory analysis by a panel of 19 assessors. Eight samples cultivated in different locations within the Republic of Serbia were analyzed, including three raspberry varieties from Arilje (Miker, Vilamet, and Enrosadira), Miker from Bačka Palanka, Tulamin from Đurđevo, Polana from Bečej, and two commercial samples without specific variety labeling, Premia (Sample 1) and Frikom (Sample 2). Sensory attributes were selected from the literature and used to characterize the samples in a controlled environment at the Institute of Food Technology in Novi Sad, employing 52 descriptors rated on a 5-point scale. ANOVA results revealed significant differences in sensory attributes among the raspberry varieties, with notable variations in fruit size, shape consistency, and flavor characteristics. The analysis indicated that the Arilje varieties generally differed from the other samples in key sensory attributes, highlighting their distinct profiles. Principal component analysis (PCA) further clarified these differences, with the first three principal components (PC1, PC2, and PC3) accounting for 36.42%, 19.23%, and 14.75% of the variance, respectively, and a cumulative variance of 70.40%. PC1 was associated with attributes such as small fruit size and metallic off-flavor, distinguishing samples like Polana, which had high positive scores for these attributes, from Miker (Arilje), which displayed more balanced characteristics. PC2 related to fruit shape and consistency after thawing, with Tulamin (Đurđevo) showing high negative loadings for inconsistent shape and size, in contrast to Vilamet (Arilje), which exhibited better visual consistency. PC3 captured differences in overall flavor and aroma, with Enrosadira (Arilje) scoring high for sweet aroma and juiciness, while Sample 1 (Premia) showed lower scores in these attributes. Overall, the RATA methodology, supported by ANOVA and PCA, effectively differentiates between raspberry varieties, providing detailed sensory profiles that reveal significant quality differences. This approach offers valuable insights for producers and distributors, aiding in the enhancement of product standards and alignment with market preferences.

**Keywords:** frozen raspberries, variety comparison, Rate all that apply (RATA) methodology

**Acknowledgements:** This work was financially supported by the ALLIANCE project, funded through Grant Agreement ID 101084188 under the Horizon Europe Programme, as well as by the Ministry of Education, Science, and Technological Development of the Republic of Serbia (contract No. 451-03-66/2024-03/200222).



## SENSORY PROPERTIES OF EMULSIFIED CHICKEN SAUSAGES WITH FAT SUBSTITUTION DURING STORAGE

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This study assessed how different fat formulations affect the sensory properties of emulsified chicken sausages, including colour, texture, and acceptability, during 90 days of storage at 4 °C. Six formulations were tested: F1 (100% chicken skin), F2 (100% chicken abdominal fat), F3 (100% hydrogenated vegetable fat), F4 (50% chicken skin + 50% chicken abdominal fat), F5 (50% chicken skin + 50% hydrogenated vegetable fat), and F6 (50% chicken abdominal fat + 50% hydrogenated vegetable fat). A panel of 14 trained assessors evaluated the samples based on colour intensity, firmness, and overall acceptability using a ranking method. Data were analyzed using the Friedman test and LSD method, comparing F-values to critical values at  $\alpha = 0.05$  to identify statistically significant differences and provide a statistical representation of general trends and pairwise comparisons. The findings revealed that F1 was the least intense in colour and achieved the highest ranking across all sensory attributes. F2 and F4 also demonstrated superior colour stability by maintaining a lighter hue throughout storage. Conversely, F3 and F6 exhibited darker colours and softer textures, which were less desirable from a sensory perspective. Notably, F4 exhibited the highest firmness, indicating a firmer texture, whereas F3 and F6 were notably softer. In summary, F1 and F4 were rated highest for colour, texture, and overall acceptability. The study highlighted that substituting traditional fats with chicken skin or its combinations significantly influenced the sensory properties of sausages, emphasizing that prioritizing the selection of appropriate fats, particularly chicken skin or its blends, is beneficial for improving sensory appeal and maintaining quality over time. Future studies should investigate various fat types and combinations to further refine the sensory attributes of emulsified chicken sausages and better align with consumer preferences.

**Keywords:** emulsified chicken sausages, chicken skin, chicken abdominal fat, sensory properties

## EXPLORING CONSUMER PERCEPTIONS AND ACCEPTANCE OF SPROUTS AND INDUSTRIAL HEMP PRODUCTS IN SERBIA: AN ONLINE SURVEY

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Growing interest in the health benefits of food, beyond basic nutrition, has significantly increased the popularity of functional foods. Industrial hemp, especially its sprouts, serves as a potent source of bioactive and antioxidant compounds such as polyphenols, tocopherols, and fatty acids. These compounds can substantially enrich the bioactive profile of products in which they are incorporated, offering the potential for value-added foods that not only satisfy nutritional requirements but also deliver significant health benefits to consumers. Therefore, the aim of this study was to explore attitudes and perceptions among consumers in Serbia towards the consumption of hemp sprouts, with a particular focus on those derived from industrial hemp. Additionally, the study aimed to evaluate their potential acceptance and marketability. An extended model of the Theory of Planned Behaviour was used to analyse consumers' knowledge, attitudes, subjective norms, perceived behavioural control, intentions and behaviour. Conducted in May 2024, the study gathered data from 420 participants across Serbia using an online questionnaire and a snowball sampling methodology. The findings reveal a significant lack of awareness among Serbian consumers regarding the consumption of industrial hemp sprouts, with more than 60% of respondents having never tried them. Over 90% of Serbian participants are unfamiliar with the taste of hemp sprouts, emphasizing a gap in consumer knowledge. Despite this, more than half of the respondents expressed a willingness to try them. Furthermore, over 60% of respondents believe that the public is not adequately informed about the potential for consuming industrial hemp and its sprouts, and similarly, over 60% expressed a desire for more information about these products. These results suggest that increasing consumer education and awareness through targeted campaigns could significantly enhance the acceptance and marketability of hemp sprout-based products. Future activities should focus on promoting the nutritional and health benefits of these sprouts, offering taste-testing opportunities, and providing accessible information to better inform the public.

**Keywords:** industrial hemp, sprouts, consumer, survey, theory of planned behaviour

**Acknowledgements:** The authors thank the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, project number 451-03-66/2024-03/200222 for financial support.

## SENSORY QUALITY OF FRESH AND COOKED EGGS OBTAINED FROM HENS FED A SPECIALLY DESIGNED FEED MIXTURE

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This study aimed to assess the influence of dietary inclusion of flaxseed, camelina seed, and hempseed in hens' diets on the sensory attributes of both fresh and cooked eggs. Two hundred and forty Lohmann Brown laying hens were divided into eight groups (two controls, K1 and K2, and six experimental treatments, L1, L2, C1, C2, H1, and H2) and fed for four weeks. The control groups were fed a corn-soybean meal based diet. The experimental groups received co-extrudates: flax-corn meal (13.5% - L1, 22.5% - L2), camelina-corn meal (16.6% - C1, 27.6% - C2), and hemp-corn meal (18.4% - H1, 30.7% - H2). K1 contained up to 3% fat without pigments, while K2 contained up to 5% fat and synthetic pigments. All experimental treatments included natural pigments (1% carrot and 0.5% paprika). In the sensory evaluation of fresh egg yolks, experimental treatments (L1, L2, C1, C2, H1, H2) with 1% carrot and 0.5% paprika had similar ( $p>0.05$ ) RYCF (Roche Yolk Color Fan) values ranging from 12.20 to 12.80. Control K1 (no synthetic pigments) had the lowest RYCF value (8.00), while K2 (with synthetic pigments) had the highest RYCF value (14.20), both significantly different from the experimental treatments ( $p<0.05$ ). Color acceptability and yolk color shade were similar across experimental treatments but were lowest in the control groups (K1 and K2). Cooked eggs in K1 and K2 also had lower color acceptability and yolk color shade compared to the experimental treatments. The addition of co-extrudates did not affect yolk color, smell, or taste, except for flaxseed co-extrudates (L1 and L2), which negatively impacted yolk taste ( $p<0.05$ ).

**Keywords:** sensory quality, egg yolk, natural pigments, flaxseed, camelina seed, hemp seed

**Acknowledgements:** This study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia No. 451-03-66/2024-03/200222.

## **SENSORY ACCEPTABILITY AND STALING OF GLUTEN AND GLUTEN-FREE FLATBREAD WITH ALTERNATIVE INGREDIENTS**

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Flatbreads (FB), a traditional staple food, make an excellent medium for nutritional enhancement with alternative ingredients but their acceptance among consumers needs to be investigated. We have developed two gluten-free (GF) in which 10% (w/w) of the rice and corn flour was replaced with the blend of millet bran and flour and carob flour milled from carob seeds or carob seeds and pods. In the gluten FB, 30% (w/w) of wheat flour was replaced with oat or barley flour that was either native or pretreated with a pulsed-electric field (PEF) (150 Hz, 2 $\mu$ s, 12 kV/cm, 9 min) to prevent the degradation and improve extractability of the  $\beta$ -glucans. The FB were tested for their sensory acceptability (ISO 6658:2017), and staling rate by texture profile analysis for 96 h or 72 h for gluten and GF FB, respectively. In addition, the staleness of GF FB during 72 h was sensory tested (AACC method 74-30). The hardness of both bread types decreased after 24 h of storage, while resilience and cohesiveness showed a constant decline. The hardening during the 96 h of storage of barley FB was well-fitted to the Avrami model, which was not the case for oat FB or GF FB. The hardening of gluten FB from 24 to 96 h was small and further delayed by PEF pretreatment of oat and barley flour. The use of carob seed flour reduced the hardening of GF FB (by 24%). Both GF FB were slightly stale after 24 and 48 h of storage as indicated by sensory analysis. The enhanced breads were liked moderately to very much (oats 7.7, barley 7, and GF 6.5) which did not significantly differ from control breads made with conventional ingredients (corn and rice or wheat flour).

**Keywords:** Avrami model, texture profile analysis, hardness, shelf life

**Acknowledgements:** This work is supported by the PRIMA program under grant agreement No 2031 project "FLAT BREAD MINE.". This project is part of the PRIMA programme, which is an Art.185 initiative supported and funded under Horizon 2020, the European Union's Framework Programme for Research and Innovation.

## THE INFLUENCE OF DIGITAL MEDIA AND INFORMATION TECHNOLOGIES ON HEALTHY EATING PRACTICES

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Digital media and information technologies have transformed various aspects of daily life, including people's awareness and behaviour related to healthy eating practices. This study explored the influence of these technologies on healthy eating habits among the population of Vojvodina (Serbia) before VUCA (Volatility, Uncertainty, Complexity, Ambiguity) world, prior to 2018. The research also highlighted the changing significance of these information channels within the post-VUCA world, which is crucial for developing effective strategies to promote healthier eating behaviors and improve nutritional outcomes. After in-depth interviews with 20 participants, an online survey *The consumer awareness of healthy eating* questionnaire was designed using the SurveyMonkey<sup>®</sup> platform and distributed to 628 respondents via email and social networks, utilizing a snowball sampling technique. Data collected before VUCA world were analysed using descriptive methods, revealing that digital media and the internet play a crucial role in promoting healthy eating by sharing information and educating consumers. More specifically, before the VUCA period, digital media had already become dominant, with the internet serving as the primary information channel for over 99% of daily users. High levels of daily engagement were observed on platforms such as Facebook (>96%), YouTube (>95%), and Instagram (>60%). Although usage of traditional media (television, radio, newspapers, magazines, and books) was declining, trust in these sources remained higher compared to modern media. Notably, 73.3% of respondents searched for healthy eating information online, while 52.9% used social media platforms for this purpose. Most recent reports indicate that since the onset of the VUCA era, the digital media landscape has continued to change. While Facebook and YouTube remain significant, platforms such as TikTok and Instagram have gained popularity. Daily use of the internet and smartphones remains high. In contrast, trust has increasingly shifted toward digital media, despite ongoing concerns about misinformation. Trust and engagement with traditional media have further declined. In conclusion, the VUCA era has intensified the influence of digital media on healthy eating practices, with social media emerging as a key source of information despite the associated risks of misinformation.

**Keywords:** consumers, healthy eating, information technologies, digital media, VUCA

**Acknowledgements:** This study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Agreement no.: 451-03-66/2024-03/200222).

## DETECTION OF ADULTERATION IN ACACIA HONEY USING CONSUMER-BASED SENSORY PROFILING

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This study focused on detecting adulteration in acacia honey from the Tuzla region of Bosnia and Herzegovina (B&H) by utilizing consumer-based sensory profiling. The rapid Pivot Profile (PP) technique was employed to compare the sensory attributes of a standard reference product (pivot product, PVT) against acacia honey adulterated with 20%, 40%, 60%, and 80% fructose-glucose syrup. Sensory evaluation was conducted with a consumer panel consisting of 72 participants (25 women and 47 men), all of whom were acacia honey producers aged between 20 and 55 years. The chi-square test ( $\chi^2 = 3032.37$ ,  $p < 0.001$ ) demonstrated that consumers could effectively distinguish between pure and adulterated honey samples. A chi-square test per cell was used to analyze variations within the data, identifying sensory descriptors that deviated significantly from the PVT. The study identified 48 sensory attributes (5 for appearance, 14 for odor, 4 for basic tastes, 3 for aftertastes, 16 for flavor, 2 for trigeminal effects, and 4 for texture). Correspondence Analysis (CA) was used to visually map sensory changes in honey samples based on adulteration levels, explaining nearly 60% of the variability across the first two dimensions and illustrating how these changes align with consumer perceptions. The results revealed a decline in flavour and appearance attributes, alongside the emergence of sensory defects such as off-flavors, unpleasant trigeminal effects, and altered viscosity. The PP technique, applied with consumer input, offered a detailed and insightful analysis of how adulterated samples compared to the PVT, providing a modern approach to sensory evaluation that complements traditional expert assessments. This method shows significant potential for further research into sensory vocabulary development and adulteration detection, applicable not only to acacia honey but also to other food products at risk of adulteration.

**Keywords:** adulteration detection, acacia honey, consumer-based sensory profiling, pivot profile technique

**Acknowledgements:** This work was financially supported by MULTILAB Ltd., Tuzla, Bosnia and Herzegovina; the Chamber of Economy of Tuzla Canton, Bosnia and Herzegovina; the Association of Beekeepers from Tuzla Canton, Tuzla, Bosnia and Herzegovina; as well as by the Ministry of Science, Technological Development and Innovation, Serbia (Contract No. 451-03-66/2024-03/200222).

## CHANGES IN THE SENSORY CHARACTERISTICS OF CRAFT BEER DURING STORAGE

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Beer is a drink of distinctive, characteristic flavour and bitterness. It has gained popularity for centuries and is considered one of the oldest fermented beverages in the world, and is now widespread globally. Following trends and developing a growing number of craft breweries, over 100 different beer styles have been developed. Today, the imperative of good beer is not only its quality as a fresh product but also the preservation of its flavour and stability over time. In order to discover factors that affect changes in the aromatic profile of beer and their suppression and longer preservation of flavour, sensory testing is being conducted more and more. Many classes of compounds have been shown to play an important role in the development of a distinctive beer profile. This significantly affects its taste and sensory properties, affecting market performance as well. Despite intensive research aimed at uncovering the precise mechanism and regulation of beer flavour creation, existing knowledge is not yet complete. This paper shows current knowledge of beer biochemistry and discusses factors that influence its formation. The main goal of the study is to change the sensory characteristics of four different beer styles over a 30-day period. The research tracked the impact of different packaging materials and temperatures on beer flavour. Changes were followed in cans, glass bottles and PET packaging at 40 °C, resulting in forced ageing of 2 years. Quantitative Descriptive Analysis (QDA) was used to examine the changing sensory characteristics of the product. From the aspect of sensory evaluation, it was determined that unfiltered, unpasteurized beer stored at a temperature of 40 °C, shows significant deviations in the sensory profile after ten days of storage. For lager craft beer, the most significant deviations in the following attributes were observed after ten days: sweet, lime, rancid, diacetyl, honey and sulphur.

**Keywords:** beer, sensory analysis, ageing beer, aromatic profile

**Acknowledgements:** This research was funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia, University of Belgrade—Faculty of Agriculture (Agreements No. 451-03-47/2023-01/200134, 451-03-47/2023-01/200116, and 451-03-1202/2021-09).



## TRADITIONAL SERBIAN LIQUEURS: UNLOCKING THE POTENTIAL FOR INDUSTRIAL PRODUCTION

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Since ages, various raw materials have been used for centuries as a source of aromatic compounds in liqueurs. The aim of this research is to provide an overview of the various materials used for homemade liqueur production in the Republic of Serbia investigating online sources. In addition, the selected liqueurs produced with the most frequently used material for flavoring are sensory characterized using model of positive ranking. The common quality parameters were evaluated: clearness (max 1 point); color (max 1 point); distinction (max 2 points); odor (max 6 points); taste (max 10 points). In this evaluation, a liqueur sample may have a maximal score of 20 points. Obtained results showed that more than 25 ingredients are used for flavoring liqueurs produced in artisanal, small-scale production. Among the liqueurs as most frequently made, walnut liqueur, sour cherry and raspberry liqueurs were selected for sensory evaluation. All tested samples were evaluated with a total score higher than 18 points, and their sensory characteristic was acceptable for sensory evaluators. Although the potential of the traditional drink is undisputed, a more in-depth study of its chemical and sensory characterization is crucial to exploit its full potential and production at the industrial level. Homemade liqueurs can serve as inspiration for industrial products as they have an unlimited number of variations.

**Keywords:** liqueurs, aromatic materials, sensory characteristics

**Acknowledgments:** This research is supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (No. 451-03-65/2024- 03/200375, and 337-00-3/2024-05/145) and the Slovak Research and Development Agency under the Contract no. SK-SRB-23-0015.



*Sustainability, decarbonisation and food chains*

## BILBERRY POMACE POWDER AND ETHANOLIC EXTRACT AS ACTIVE PAPER COLOURANTS: PROPERTIES AND COLOUR STABILITY

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Bilberry pomace is a fruit processing by-product that is rich in lignocellulosic fibres and polyphenols, especially anthocyanins. Anthocyanins are natural pigments whose colour changes with the pH of the environment, making them a promising pH-responsive colourant. Our study investigated bilberry pomace powder and its ethanolic extract as potential paper colourants/pH indicators by evaluating their mechanical, optical, and colour-changing properties. For paper production, the bilberry pomace powder and the acidified ethanolic extract were added to pulp mixtures in three different concentrations. The mechanical and optical properties of the produced papers were measured according to ISO standards. Colour stability was evaluated during a 6-week storage period under daylight and dark conditions, and colour change was tested using a range of buffers (pH 3.0-10.0). All formulations with pomace extract showed better mechanical properties than those with pomace powder; however, both additions resulted in poorer performance compared to the commercial paper. The samples with added powder had a darker and more intense colour, but were also more susceptible to change during storage. For all samples, the overall colour changes ( $\Delta E$ ) were more pronounced in daylight than in the dark. The pH response was only recognisable in an acidic environment, which is probably due to the basic nature of the paper. Nevertheless, the colour change in the acidified samples was reversible, so that alkaline conditions could also be detected. Overall, all prepared materials are suitable for use as specialty papers, and their appealing colour and reactivity to different pH values enable rapid monitoring of various liquid products. To improve the mechanical and colour stability, optimisation of the colourant ratio and the addition of stabilising additives/coatings will be investigated.

**Keywords:** bilberry pomace, anthocyanins, paper colourants, pH indicators, by-product utilisation

**Acknowledgements:** This study was financially supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grant No. 451-03- 65/2024-03/200116) and the national research program Chemical Reaction Engineering (P2-1052, Slovenian Research and Innovation Agency).

## OPTIMIZING PARAMETERS RELATED TO THE POLYPHENOL AND FLAVONOID CONTENTS OF *Salvia officinalis* L. WASTE POWDER

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The current study had two objectives: (1) the determination of total phenol content (TPC), total flavonoid content (TFC), and *in vitro* DPPH radical scavenging activity ( $RSA_{DPPH}$ ) of waste sage (*Salvia officinalis*) powder prepared using microwave-assisted extraction (MAE) and (2) optimization of the extraction process parameters *via* varying solvent concentration (ethyl alcohol 30% (v/v), 50% (v/v), and 70% (v/v)). All experimental runs were conducted in the SR-15 rotor segment of the Milestone Ethos X microwave extractor. The temperature of the extraction was 100 °C, time of extraction was 2 minutes, and solid-to-solvent ratio was defined as 1:12.5. The total polyphenols were extracted most effectively by ethyl alcohol (70%, v/v) – approximately 5.02 mg GAE (gallic acid equivalents)/g dry weight. On the other hand, total flavonoids ranged from 10.27 to 21.76 mg CE (catechin equivalents)/g dry weight in respect to the concentration of the solvent. The strongest  $RSA_{DPPH}$  was measured in the sample where 70% ethyl alcohol was used as a solvent – about 65.98% at concentration of the extract of 1.76 mg/ml. Generally, for all tested analytes, it was observed that concentration of ethyl alcohol of 70% (v/v) was the most effective for extracting bioactive compounds from waste sage powder. Our study opens the possibilities for the production of polyphenol-rich extract of the waste sage which might be used in a different food and pharmaceutical products.

**Keywords:** waste sage, microwave-assisted extraction, optimization, polyphenols

**Acknowledgements:** The authors acknowledge their gratitude to the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, contract number 451-03-66/2024-03/200003.

## EVALUATING CURRENT AND FUTURE ENVIRONMENTAL IMPACTS OF DIETARY PATTERNS IN THE REPUBLIC OF IRELAND

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Dietary patterns are intrinsically linked to greenhouse (GHG) emissions, land use, and water use via agricultural practices, food production systems, and consumption habits. Therefore, analysing and comparing contemporary dietary patterns and their impact on the environment is crucial to identifying which dietary patterns should be promoted to make food systems more sustainable. A cross-sectional survey was conducted across Ireland to collate respondents' food consumption patterns with a representative sample size of 957 adult respondents. Subsequently, a farm-to-fork life cycle assessment (LCA) was employed in OpenLCA 2.0.4 using AGRIBALYSE® 3.0. to assess environmental impacts (global warming, land use, and water use) of dietary patterns in the population. Mean calculated dietary GHG emissions were 5.52 kgCO<sub>2</sub>eq person<sup>-1</sup> day<sup>-1</sup>, with the 'meat-focused' diet exhibiting the highest (6.62 kgCO<sub>2</sub>eq person<sup>-1</sup> day<sup>-1</sup>) and the vegan diet exhibiting the lowest (2.0 kgCO<sub>2</sub>eq person<sup>-1</sup> day<sup>-1</sup>). The 'meat-focused' diet was also associated with the highest per capita land use (8.62 m<sup>2</sup>a crop eq person<sup>-1</sup> day<sup>-1</sup>), compared to a calculated mean (i.e., whole population) of 7.06 m<sup>2</sup>a crop person<sup>-1</sup> day<sup>-1</sup>. Calculated mean water usage was 104 L person<sup>-1</sup> day<sup>-1</sup>, with the 'potato-focused' diet using the most water (121 L person<sup>-1</sup> day<sup>-1</sup>) and the pescatarian diet using the least (70 L person<sup>-1</sup> day<sup>-1</sup>). While 'red meat' was the greatest contributor to global warming, land use, and water use, relatively high water use was attributed to 'bananas, avocados, and citrus fruit', and comprised high proportions of the pescatarian (27.9%), 'seafood-focused' (24.1%), vegan (22%), 'vegetable-focused' (20.9%), and vegetarian (19.1%) diets. Future (2050) dietary scenarios compared to 'business-as-usual' patterns were projected; findings suggest that substantial environmental impact reductions can be achieved if segments of the Irish population transition to vegetarian, vegan, and 'vegetable-focused' diets.

**Keywords:** Dietary Patterns, Greenhouse Emissions, Life Cycle Analysis

**Acknowledgements:** This research was funded by the Environmental Protection Agency, Ireland under the Project ID: 2022-GCE-1119.

## FOOD WASTE IN IRELAND – ASSESSMENT, ENVIRONMENTAL & ECONOMIC BURDEN, AND MITIGATION STRATEGIES

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The Food and Agriculture Organisation (FAO) calculates that 1.3 billion tonnes of food is wasted globally per year, directly contributing to food shortages, water stress, biodiversity loss, up to 10% of greenhouse gas (GHG) emissions, and economic losses of €550 billion. Based on Ireland's commitment to the UN Sustainable Development Goals (SDGs), a 50% reduction in both food waste and emissions is required by 2030. Accordingly, it is crucial to accurately quantify volumes of food waste generated and in line with the National Waste Action Plan for a Circular Economy design mitigation strategies for converting waste-to-value and assess the environmental and economic implications of food waste. A total of 1180 participants from all over the Republic of Ireland contributed to this study's food waste survey. Statistical analysis identified that participants who did not cook their lunch and dinner wasted more food, similar to those who did not save or use leftovers. While brown/kitchen waste bin is the primary food waste disposal route it is closely followed by general waste bin. Regarding food waste quantities, a total of 435.06g/ cap/week is generated with fruits and vegetables being the most wasted commodities. Further a roadmap for converting household food waste into sustainable biobased products using the cascading approach of a biorefinery was developed. This included (a) functional ingredients biorefinery route (b) platform chemicals biorefinery route (c) bioenergy biorefinery route and (d) compost, biochar and feed pathway. These pathways can enable decision making regarding food waste valorisation into value added products. The research therefore contributes not just by quantifying the food waste generated but also by exploring the pathways and implications of food waste valorisation.

**Keywords:** Food Waste, Consumer Behaviour, Valorisation, Circular Bioeconomy, Life Cycle Analysis

**Acknowledgements:** This research was funded by the Irish Research Council under grant number COALESCE/2022/804.

## MISSING GAPS TO ESTABLISH SUSTAINABLE FOOD SYSTEMS

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To achieve a robust sustainable food system, it is essential to adopt a comprehensive approach that considers all aspects of food—production, storage, distribution, and consumption—ensuring that healthy, accessible food is available to everyone while simultaneously protecting biodiversity and combating food waste. A sustainable food system guarantees food security and nutrition for all, encompassing the entire journey of food from processing and packaging to transportation and consumption. However, several national and international reports indicate that current food systems are both inefficient and unsustainable; in 2022, approximately 9.2% of the world’s population faced chronic hunger, affecting about 735 million individuals—122 million more than in 2019. Moreover, unsustainable practices in food systems contribute significantly to the climate crisis, accounting for one-third of global greenhouse gas emissions and 70% of freshwater usage. To establish effective sustainable food systems, several critical gaps must be addressed, including the need for a clear understanding of resilience as it relates to sustainability, the identification of current and future shocks and stresses impacting food systems, an investigation of how diverse stakeholders perceive resilience, and the exploration of new and emerging solutions that enhance resilience. Key steps to address these gaps include integrating sustainability into resilience studies, exploring trade-offs and synergies among various strategies, developing robust metrics and indicators to evaluate resilience alongside sustainability, engaging all food system actors, and investigating innovative approaches that encompass ethical, social, and economic implications. By addressing these knowledge gaps and implementing these recommendations, future research on food system resilience can significantly contribute to creating a more sustainable and accessible food system for all.

**Keywords:** Food systems, processing technologies, climate change

**Acknowledgements:** European commission (GA: 101084437) and Research Ireland (SFI) funded BiOrbic – Bioeconomy Research Centre funded under the Science Foundation Ireland Research Centres Programme.

## HEALTH AND NUTRITION STANDARD MANAGEMENT SYSTEM FOR RUMINANTS IN THE RODOPI MOUNTAINS

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The rearing of small and large ruminants in the Rodopi Mountains is a main source of income for the inhabitants of the region but faces significant challenges. Hellenic Statistics Agency data show a significant decline in farms in Eastern Macedonia and Thrace in recent years. The challenges include inadequate nutrition, infectious factors and unsuitable stabling conditions, negatively affecting the profitability and quality of livestock products. The research project included questionnaire completion, field inspections and laboratory analysis of feed and milk from 12 farms, including 4 small and 8 large ruminants. Three milk samples were taken over a period of 5 months, collecting 36 milk samples and 45 feed samples. All samples were subjected to chemical analysis of their composition, antioxidant status and the milk samples were also subjected to determination of their fatty acids. The findings of the questionnaire showed variation in feeding and management methods between farms, with farms focusing on serving the local population. Analysis of milk samples showed that cow's milk contains  $3.26\% \pm 0.2$  protein and  $4.38\% \pm 0.37$  fat, while sheep's milk contains  $4.86\% \pm 0.37$  protein and  $6.33\% \pm 0.42$  fat. Finally, it was noted during the visits that the facilities were considered inadequate, and that ventilation did not meet modern practices. Inadequate vaccination and the absence of biosecurity measures contributed to health problems. The results underline the need to improve nutritional patterns, implement preventive measures and improve facilities in order to enhance animal health and the sustainability of farms in mountainous Rodopi. The project was financed by the "Operational Plan Sub-measure 16.1-16.2". Project Code: M16SYN-00231. Acronym 'RodopiFarms'.

**Keywords:** ruminants, milk, nutrition, health

## PURIFICATION OF PROTEINS FROM COMMON BEAN SEEDS EXTRACT ON ION-EXCHANGE RESIN AMBERLITE™ IRA 900 CL IN BATCH MODE

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In the era of rapid environmental degradation, we are confronting significant challenges related to water scarcity and contamination of freshwater supplies with diverse organic and inorganic pollutants. Turbidity is a critical parameter in the assessment of water and wastewater quality, and its reduction is commonly achieved through the use of chemical coagulants and flocculants, such as alum and iron salts, and acrylamide. However, these chemical agents pose potential risks to human health and the environment. Consequently, there has been a surge in research focusing on natural coagulants and flocculants. These biopolymer-based natural coagulants offer several benefits, but their primary limitation is the organic load they introduce into the treated water. This issue can be mitigated through appropriate purification processes. In this work, proteins from a crude extract obtained with 0.5 mol L<sup>-1</sup> NaCl from *Phaseolus vulgaris* seeds were first precipitated by ammonium sulfate and then redissolved and purified on the anion exchange resin Amberlite™ IRA 900 Cl in a batch mode. Prior to purification optimization of adsorption conditions was performed. The step elution was done using NaCl solutions with increasing concentrations. In obtained eluates protein contents were determined and they were tested in coagulation tests in mode water of initial turbidity 35 NTU and pH 9. The highest coagulation activity of 44.3% was achieved at the dose 1.67 mL/L of eluate obtained with 2 mol/L NaCl solution.

**Keywords:** natural coagulant, proteins, coagulation activity, water clarification, ion-exchange, organic load

**Acknowledgements:** This research was funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant numbers 451-03-66/2024-03/200134 and 451-03-65/2024-03/200134.



## RECOVERY OF PECTIN FROM BLACK CURRANT POMACE: PHYSIOCHEMICAL, STRUCTURAL ANALYSIS AND POTENTIAL APPLICATIONS

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The biorefinery of bio-products generated in the food processing industry into high-value products is an effective way to reduce the amount of waste sent to landfills, thereby contributing to the development of sustainability. Black currant pomace, which remains after juice and jam production, is rich in bioactive compounds, with polysaccharide pectin being the most important. Due to its unique physicochemical properties, pectin is one of the most widely used biopolymers across various industrial sectors. The aim of this study was to isolate pectin from black currant pomace using conventional (acid hydrolysis) and enzyme-assisted (1 % v/v of cellulase; activity 17.03 FTU) extraction methods, to determine the degree of methyl esterification (DM) of the obtained pectin samples using FTIR, and to analyze the preliminary composition of sugar units by NMR. The obtained results showed that the enzyme-assisted extraction method was more efficient compared to the conventional hydrolysis method, with pectin yields of 31.53 % and 19.43 %, respectively. The integrals of methyl and carboxyl functional groups from FT-IR spectra indicated the degree of esterification of the obtained pectin. The enzyme-assisted extraction method resulted in a high-methoxy (HM) degree of esterification (50.5%), while the conventional acid hydrolysis method produced a low-methoxy (LM) degree of esterification (32.2 %). The <sup>1</sup>H NMR spectra revealed a high amount of different types of sugar units in the pectin obtained by conventional acid hydrolysis, including galacturonic acid, rhamnose, galactose, xylose, arabinose, and D-fructose, whereas main carbohydrate units: galacturonic acid, rhamnose, arabinose and D-fructose were identified in the pectin obtained by the enzyme-assisted extraction method. The concentration of galacturonic acid determined by the UPLC/MS method showed that higher amounts of galacturonic acid were 61.9% and 73.9% in pectin isolated by conventional and enzyme-assisted extraction methods, respectively. The pectin extracted by conventional acid hydrolysis exhibited higher emulsifying ability (64.04%) and emulsion stability after 42 days at both 25°C and 4°C, while the pectin isolated by enzyme-assisted hydrolysis showed lower emulsifying ability. It is possible that the extraction buffer used in this method may have influenced the results. In conclusion, it can be stated that not only the yield of pectin but also its degree of esterification and structural composition depend on the extraction method and the conditions applied. Black currant pomace, when subjected to enzymatic extraction, shows great potential as a source of pectin with a high degree of methylation.

**Keywords:** pectin, black currant pomace, conventional acid hydrolysis, enzyme assisted hydrolysis, esterification

## THE POTENTIAL OF OLIVE POMACE COMPOST FOR USING AS A SOIL AMENDMENT

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In the Mediterranean basin, large quantities of olive-processing residues, such as olive pomace (OP) are generated during olive processing each year. Due to the phytotoxic nature of OP, its appropriate management remains one of the major issues faced by the olive oil-extraction industry. However, composting was recommended to recycle this waste aiming to produce organic fertilizers. This study was set up to evaluate the applicability of olive pomace compost (OPC) for agricultural use. Samples of OPC were taken from the surface (5-10 cm) and depth layers (40 cm) of the pile after 8 months of composting and analyzed for their physical and chemical characteristics, as well as colloidal properties and microbial community structure. The results revealed that pH was within limits from 7.8 to 8.6, while electrical conductivity (EC) between 770 and 1608 mS/cm. Further, levels of N, P and K varied from 1.5 to 27.2, 1.6 to 1.8, and 6.5 to 7.5 g/kg d.w., respectively. The contents of Cr, Cu, Ni, Pb and Zn which ranges from 22.4 to 28.9, 34.2 to 35.7, 13.5 to 16.4, 8.7 to 10.2 and 42 to 48 mg kg<sup>-1</sup> d.w., respectively fall well below the EU limits for soil improvers. The microbial structure follows the changes of the gradient from outer to the innermost layer with relatively low amounts of DNA. The gradient nature shows that it is needed to develop better strategies for composting surpassing the conventional approach. However, low amounts of total phenols and oil residues suggest that composting was efficient in their biodegradation. In addition, C/N ranging from 13 to 16 is an indication that OPC can be used as a soil amendment. According to this study it seems that composting may be a promising strategy for nutrient recovery and eco-friendly OP recycling.

**Keywords:** olive pomace compost, characterization, agricultural use, soil amendment

**Acknowledgements:** The Croatian Science Foundation, the program of Slovenian – Croatian research projects (IPS), is acknowledged for financial support of the study (contract No. IPS-2022-02-2099).

## EFFECT OF PEA SORTING BYPRODUCT AS PROTEIN SOURCE IN GOAT MILK DIETS, FOR REDUCED WASTE IN A CIRCULAR ECONOMY SYSTEM

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The aim of present paper was to study alternative inclusion of byproducts (broken beans) from sorting pea process (*Pisum sativum*) in lactating goat's diet. Experiment consists in three groups (control, pea and broken bean pea supplemented diet) each with 10 goats from Murciano Granadina breed. Base ration consisted in concentrated feeds and alfalfa hay ad libitum. During experiment were measured milk yield (in three different measurements), milk quality parameters, fatty acids content, and blood parameters for animal health. Group homogeneity was tested and all animals are on a similar age and stage of lactation. Statistic parameters were analysed using statistical packages from R. Milk production and quality in control and experimental groups are not different at any level of significance. Animal health parameters are similar for all groups, excepting glucose, who is slightly decreased from 61.66 mg/dL at control group, to 50 respectively 49 mg/dL at pea and broken pea experimental groups. Quality of milk fat was influenced as follows. Saturated fatty acids (SFA) are increased on experimental groups with 5% for pea experimental group and with 17% for broken pea experimental group. Total Mono-unsaturated fatty acids (MUFA) are slightly increased on pea experimental group, but with no significant differences between control and broken pea experimental group. Total Poly-unsaturated fatty acids (PUFA) content is decreased with 5% in pea group milk and with 19% in broken pea group versus control group. Regarding n6/n3 ratio, is decreased with 20% in pea group and to 15% in broken pea group, closer to optimum 4:1 ratio. Considering the results of present experiment, inclusion of pea sorting byproducts in lactating goats diet represent a viable alternative for classical protein sources.

**Keywords:** goats, milk yield, pea broken beans, animal health, milk fatty acids

**Acknowledgements:** Ministry of Research, Innovation and Digitalization, Core Program, Grant PN 23.20-0101.

## STRUCTURAL AND ANTIMICROBIAL PROPERTIES OF NANO-HYDROXYAPATITE/BIO CELLULOSE COMPOSITE MATERIAL

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To achieve new sustainable technologies and materials that impact greater environmental protection and reduce resource use, we developed new composite materials synthesized regarding green pathways. Bio cellulose hydrogel represents a highly pure polysaccharide biopolymer that various bacterial genera can produce. It has a similar chemical structure as in plants and with high porosity and specific surface area can be successfully used as carrier material. On the other hand, cellulose itself doesn't have any functional properties. Hydroxyapatite material represents one of the most used materials from the calcium phosphates since it has a high specific surface area and mild antimicrobial activity. The structural properties and chemical stability of hydroxyapatite enable good resistance to microbiological influences, changes in the pH of the environment, and deficient solubility products in physiological conditions. The main mission of this study was to develop functional polymer-ceramic material using green technologies. Obtained material was investigated by XRD method, to obtain phase and structural properties. The FTIR method determined functional groups related to bacterial cellulose and hydroxyapatite. The microstructural properties of the material were determined by the SEM-EDS method. The antibacterial activity of the obtained composite was tested against Gram-positive bacteria *Staphylococcus aureus*, *Listeria monocytogenes*, and Gram-negative bacteria *Pseudomonas aeruginosa*, *Acinetobacter baumannii*, by total plate count assay. The microstructural analyses revealed that nanoparticle hydroxyapatite was successfully incorporated into the cellulose hydrogel, resulting in the creation of a new functionalized polymer-ceramic based material. The FTIR results identified vibration groups belonging to cellulose and hydroxyapatite. Additionally, the results of the powder X-ray diffractograms showed broad peaks belonging to cellulose with incorporated hydroxyapatite. Microbiological analyses demonstrated moderate antimicrobial activity of the composite material obtained. After 24h, the reduction in the number of *S. aureus* cells was greater compared to the control (the control is only broth with bacteria without the presence of our material). In prolonged duration, the action of obtained composite material showed a significant impact on *S. aureus* after 14, and especially after 28 days. This effect could be successfully used in the potential application of hydroxyapatite material for the reduction of synthetic preservatives concentrations in products.

**Keywords:** hydroxyapatite, cellulose, microbiology, structure, green technologies

**Acknowledgments:** The research was funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia, registration number 451-03-66/2024-03/200017, number of research topic 1702407.

## PHIND DATABASE – A TOOL TO DEVELOP RESEARCH AND BUSINESS WITHIN GREEN AGENDA

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Access to high-quality and robust datasets is of crucial relevance for the development of any successful strategy (food waste reduction strategy), since datasets serve as a tool to identify opportunities, challenges and development paths for any research or business area. Food waste reduction is and will be one of the important challenges for humanity on the global level during the following decades. In fact, the United Nations set Sustainable Development Goal 12 (target 12.3) aiming to decrease global "food waste" per capita by half by 2030. Database such as PhInd may aid us in achieving aimed goals by identifying potential sources for polyphenol valorization from the wide range of agricultural and food waste sources. PhInd is the first comprehensive database on polyphenol quantity in plant-based side streams generated by the agriculture and food industry. It was constructed using in total of >450 scientific peer-reviewed manuscripts (2015–2021). The main database inclusion criteria were polyphenols identification and quantification by HPLC/UHPLC methods. PhInd is a user-friendly and interactive website and can be explored through several criteria divided into four subsections: (a) plant material properties and compounds; (b) source processing in industry; (c) source pre-treatment in the laboratory; and (d) extraction step of polyphenols. Data from PhInd may be used freely by everyone including industry and academia, and its unique feature to draw graphs for any set of search criteria allows us to have quick answers to many questions e.g. most prominent polyphenols in specific by-products?; most commonly used extraction techniques?; most studied plant matrices? - and much more.

**Keywords:** polyphenols, HPLC, food waste, extraction technologies

**Acknowledgements:** The authors would like to thank the Ministry of Science and Technological Development and Innovation of the Republic of Serbia, Grants No. 451-03-66/2024-03/200222 and the Science Fund of the Republic of Serbia, IDEJE, Grant No. 7750168, BioUtilize.

## OPTIMIZATION OF *Aspergillus awamori* BIOMASS PRODUCTION ON BREWER'S SPENT GRAIN

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Brewers' spent grain is a side product obtained in large quantities in the production of beer. It has a potential application in biotechnology as a raw material for obtaining a large number of different products. To enhance its nutritional value and obtain high-valued compounds, such as protein, brewers' spent grain can be used as raw material for different biotechnological processes. The aim of this study was optimization of cultivation medium based on Brewer's Spent Grain for the production of *Aspergillus awamori* microbial biomass and valorisation of this agro-industrial by-product through submerged fermentation. Biosynthesis of microbial biomass was performed on media prepared in accordance with Box-Behnken experimental design with three factors on three levels and three repetitions in central point where contents of yeast extract (5-15 g/L), (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> (10-20 g/L) and KH<sub>2</sub>PO<sub>4</sub> (4-12 g/L) were varied. Experimental factorial design and response surface methodology were employed to optimize medium composition and enhance protein content of the obtained fermentation broths. Results showed that the dry matter content can be increased by 2.5 times, while the protein content up to 7 times. The ANOVA analysis showed that the best fit model is a quadratic one with high values of the coefficient of determination (over 0.96) for both examined responses (protein and dry matter content). The most significant model term for both responses turned out to be (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, followed by yeast extract, and finally KH<sub>2</sub>PO<sub>4</sub> had a significant effect only on the dry matter content. Hence, the optimal KH<sub>2</sub>PO<sub>4</sub> concentration varied from 9.2, 10.4 and 11.6 g/L, depending on the chosen criteria, i.e., only maximal protein content, maximal protein and dry matter content and only maximal dry matter content, respectively. The obtained results represent the basis for the further development of the bioprocess for valorisation of brewers spent grain by its application as a raw material for the growth of *Aspergillus awamori* biomass.

**Keywords:** bioprocess, biomass, *Aspergillus awamori*, Brewer's spent grain, optimization

**Acknowledgements:** This work was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grant no. 45-03-66/2024-03/200222 and 45-03-65/2024-03/200134).

## BIOACCESSIBLE PHENOLIC ALKYL ESTERS OF WINE LEES DECREASE COX-2-CATALYZED LIPID MEDIATORS OF OXIDATIVE STRESS AND INFLAMMATION, IN A TIME-DEPENDENT MANNER

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Lipophenols are phenolic compounds esterified with fatty alcohols or fatty acids. These compounds, present in plant-based foods, have gained attention due to their higher contribution to the health benefits of these foods compared to unesterified (poly)phenols. The present study uncovers the capacity of gastrointestinal digestion, through the specific enzymes intervening in each phase (pepsin, pancreatin, and pancreatic lipase) to release alkyl gallates and *trans*-caffeates from wine lees. Their capacity to prevent oxidative stress (OS) and para-inflammation, *in vitro*, was also achieved. The analysis of wine lees by UHPLC-ESI-QqQ-MS/MS revealed ethyl gallate and ethyl *trans*-caffeate as the most prominent compounds (1.675 and 0.872  $\mu\text{g/g dw}$ , respectively). Beyond this, when determining their bioaccessibility, it was found that the concentration resulting from this process is modified depending on the specific structural traits of the alkyl chain. This allowed describing the *de novo* formation of alkyl gallates through both gastric and intestinal digestion phases but, to a large extent, during the latter, due to the capacity of intestinal enzymes (working alone or combined) to catalyse esterification reactions. The development of an *in vitro* model of OS and para-inflammation led to describing the capability of bioaccessible gallic and *trans*-caffeic acids' alkyl esters to lower cyclooxygenase-2 concentration and to modulate OS (8-iso-PGF<sub>2 $\alpha$</sub> ) and inflammation (PGF<sub>2 $\alpha$</sub>  and PGE<sub>2</sub>), in a time-dependent manner. In conclusion, the presence of gallic and *trans*-caffeic acids' alkyl esters in wine lees, and their formation during digestion, emphasise the value of this by-product as a source of antioxidant and anti-inflammatory compounds and encourage its consideration as an ingredient for the development of added-value, health-promoting, co-products.

**Keywords:** Winery by-products, lipophenols, simulated *in vitro* gastrointestinal digestion, digestive enzymes, prostanoids, anti-inflammatory activity

**Acknowledgements:** This work was funded by the project CNS2022-135588 MCIN/AEI/10.13039/501100011033 and European Union NextGenerationEU/PRTR and a research project granted by the Ministry of Science and Innovation with funding from the European Union-Next Generation.



## USE OF RECYCLED MATERIALS AS MID LAYER IN THREE LAYERED STRUCTURES-NEW POSSIBILITY IN DESIGN FOR RECYCLING FOR LDPE FLEXIBLE FILMS

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Use of recycled materials in food packaging application has been in focus of decision-making authorities in recent years. Challenges of using recycled polymer materials in food packaging are related both to the properties of the material and demanding safety issues for food contact materials. The aim of new regulation on recycled plastic materials and articles intended to come into contact with foods ((EU) 2022/1616) is increase in use of recycled materials in food contact with specific focus on decontamination techniques applied. At the same time new packaging and packaging waste regulation (PPWR) also requires obligatory recycled content of 10% for contact sensitive applications up to 2030. One of the possibilities to use recycled materials in food contact applications is the use of functional barriers, where the food is not in direct contact with the recycled content. The aim of this research was to detect possible risks using recycled post-consumer plastic waste in three-layered low-density polyethylene (LDPE) films produced with virgin outer layers and a mid-layer from recycled flexible LDPE material. Safety assessment on multiple samples with different recycled LDPE qualities and functional barriers (EVOH and G-polymer) was performed and tested. In the first part of this research risk assessment through migration study has been performed with two qualities of recycled LDPE available on the market. The results showed high potential in use of three-layered structures. However, the additional risk assessment was performed with low quality LDPE recycled material, and toxicology study was performed to detect any potential toxicological risk in use of these materials. The results showed that specific structures designed to protect the food substrate from any risk in use of recycled materials can be efficient with lower recycled content (10%), while the higher content (up to 50%) poses higher risks and need more attention. The optimal recycled content is crucial for the safe use of recycled content behind the functional barrier, as well as special barriers that can ensure the safety in use in food contact applications.

**Keywords:** functional barrier, recycled content, food packaging

**Acknowledgements:** This research was funded by the Research Council of Norway, grant number 320461, (RecyFoodPack Project) and COST Action 19124, Rethinking Packaging for Circular and Sustainable Food Supply Chains of the Future (CIRCUL-A-BILITY) supported by COST (European Cooperation in Science and Technology).



*Entrepreneurship, technology transfer, innovation  
in teaching and learning*

## BLUEPRINT FOR FINS OPEN INNOVATION ROADMAP: CO-CREATION FRAMEWORK AS A TOOL FOR NEW FOOD PRODUCT DEVELOPMENT

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Open innovation has been recognized as innovation model that encourages institutions to collaborate with external key players along the food chain, such as companies, research institutions, policy makers, and the citizens in order to generate and develop new ideas, technologies, and products. In that light, an open innovation roadmap is a strategic plan that outlines how an organization can implement innovative practices, fostering creativity, accelerating development, and guiding organizations towards market-oriented achievements. Therefore, the study aimed to create blueprint for FINS Open Innovation Roadmap, with a particular focus on the co-creation methodology. The roadmap phases were defined through a systematic review of relevant scientific literature (2020-2025) and available industry white papers. The main specificity of this roadmap is the co-creation approach in food technology. It emphasizes the integration of insights from citizens, especially consumers, alongside industry expertise and academic research. The proposed roadmap outlines a structured approach to foster collaboration, improve innovation processes, and achieve successful outcomes in the food technology sector. This approach transforms the innovation process by facilitating collaboration within a multi-stakeholder ecosystem, incorporating both internal and external ideas. The methodology will be implemented in forthcoming research initiatives to further refine and validate its effectiveness, offering a framework for developing sustainable, appealing, and innovative food products that meet the evolving consumer demands. By implementing co-creation methodology and engaging stakeholders directly in the ideation and prototyping phases, the roadmap aims to boost FINS's visibility and relevance in open innovation. This approach is designed to better align product development with market needs and preferences, thereby increasing product acceptability and market success.

**Keywords:** open innovation, co-creation, food technology, stakeholders

**Acknowledgements:** This project has received funding from the European Research Executive Agency (REA), under grant agreement number 101094435 - CoCOI FoodTech: Co-Creation of Open Innovation in Food Technology, REINFORCING Grantee.

## CREATING MINIMUM VIABLE PRODUCTS USING BIOWASTE TRANSFORMATION METHODS

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Waste generated during the processing and consumption of food is a global problem that negatively affects the environment, social status, and the economy. For sustainable development, it is essential to manage biowaste from the circular economy point of view. The application of the biorefinery concept is significant, considering that products with high added value, such as biochemicals and biofuels, can be obtained from the biowaste. Depending on the chemical composition and characteristics of the substances, biowaste can be valorized by the application of conventional and eco-friendly technologies for the extraction of biologically active substances, as well as thermochemical and biochemical conversion methods to produce biofuels and energy. The obtained bioproducts are used in the food and pharmaceutical industry, cosmetics, synthesis processes, and the treatment of wastewater from various sectors. This work involves the preparation and delivery of a methodological framework for research on how biowaste coming from 30 pre-selected SMEs and companies can be transformed into new Minimum Viable Products (MVP). The MVPs were developed as prototypes using the equipment at the Biohacking Lab in Skopje, with significant cross-collaboration and research with representatives of each of the pre-selected companies. The developed prototypes were characterized by the determination of the chemical composition, textural characteristics, color attributes, total number of bacteria and yeasts, as well as sensory characteristics. The research and development of new valuable products is the first and most important step for the implementation of all technological stages and operations for the transformation of the biowaste to a semi-final product and a final product.

**Keywords:** biowaste, valorization, high value-added products, prototype

**Acknowledgments:** Special thanks to UNDP in North Macedonia, the City of Skopje, and PE Landfill DRISLA.

## CO-CREATION OF SPREADS FROM ALTERNATIVE INGREDIENTS: A COLLABORATIVE APPROACH INVOLVING INDUSTRY, CONSUMERS AND ACADEMIA

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The objective of this study was to explore potential of co-creation methodology in the early stages of new product development, focusing on the case study of creating innovative, nutritionally valuable spreads made from alternative ingredients - underutilized plant and animal raw materials, such as legumes, seaweed and/or river whitefish species. The aim was to ensure that these products meet both market demands and consumer needs for healthy, additive-free, and environmentally sustainable food products. The concept consisted of three phases. Firstly, an online survey with 1000 participants was conducted in order to better understand market trends, consumer preferences and potential resistance to novel spreads (e.g. food neophobia). Secondly, 15 participants were engaged in two moderated co-creation sessions where they explored potential ingredients, prototyped products, and developed product narratives using Lego Serious Play and the SCAMPER methodology. The three most promising spread prototypes were evaluated by experts for further development and concept prototyping. Additionally, the involvement of key stakeholders in the product ideation process (industry professionals, scientists and consumers) improved the appeal of the final products, providing a model for future food innovations that effectively bridge the gap between industry, market needs and scientific research. During the process, participants prioritized on factors such as nutritional value, taste, texture, health benefits, and allergenicity of the selected ingredients, as the most important attributes for the spreads, and emphasized the importance of incorporating natural and sustainable ingredients into novel spreads. This approach adds valuable insights to the expanding research on alternative nutritionally valuable food innovations by demonstrating how co-creation can lead to the development of convenient, ready-to-eat, sustainable, and nutritious spreads. The findings suggested that co-creation is an effective strategy not only for creating spreads, but also for developing other food products in line with consumer expectations.

**Keywords:** co-creation, stakeholders, spreads, innovation

**Acknowledgements:** This study was funded by the Institute of Food Technology in Novi Sad, University of Novi Sad, under the Seed Research Grant Program for young scientists "SPREADify: Co-creating Healthy and Innovative Spreads through Multi-Stakeholder Engagement", through the financial support of the Serbia Accelerating Innovation and Entrepreneurship (SAIGE) Project, a joint investment by the Republic of Serbia, Ministry of Science, Technological Development and Innovation, the World Bank and the European Union.

**Disclaimer:** The statements, opinions and data contained in this publication are solely those of the individual author(s) and contributor(s) and not of the SAIGE Project. The SAIGE Project disclaims responsibility for any use that may be made of the information contained therein.

## **REINFORCING project and grants: RESPONSIBLE TERRITORIES AND INSTITUTIONS ENABLE AND FOSTER OPEN RESEARCH AND INCLUSIVE INNOVATION FOR TRANSITIONS GOVERNANCE**

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The REINFORCING project kicked off in January 2023, bringing together 11 European partners. This project's efforts are directed at supporting European organizations, institutions, and territories in their transition towards a future where responsibility and openness are the driving forces behind research and innovation. At the core of the project is the One-Stop Source platform designed to serve as the entry point to knowledge, training, and trainings on ORRI. The platform will select and curate the most interesting resources and centralize them in one single platform. Furthermore, the project provides unique networking opportunities for organizations, institutions and territories interested in becoming part of the European ecosystem that is transforming the R&I system by bringing it closer to society. The project acknowledges that facilitating lasting change requires also financial support mechanisms to ensure the successful implementation of ORRI practices. Thus, REINFORCING has allocated funding for 96 cascading grants spread in 7 open calls, ranging from 20,000 to 60,000 euros. This financial support is specifically earmarked to help organizations and institutions scale up their ORRI expertise and provide support to new territories venturing into this field for the first time. The combined endeavors of these initiatives are anticipated to result in more than 150 sustainable institutional changes and the broad adoption of exemplary, transparent, and responsible Research and Innovation practices across the European Research Area.

**Keywords:** responsibility, research, innovation, RRI, ORRI

**Acknowledgements:** Funded by the European Union, Grant agreement ID: 101094435. Views and opinions expressed are however those of the author(s) only and do not necessarily reflect those of the European Union or the REA. Neither the European Union nor the granting authority can be held responsible for them.

*H2020 CROPDIVA project*

## ASSESSMENT OF MYCOTOXINS LEVELS IN LUPIN AND FABA BEAN SPREADS

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Recent trends in the food market have shown an increasing demand for functional spreads made from plant seeds, as an alternative to traditional meat or liver pâté. For that purpose, spreads were made from two currently underused crops. Functional spreads were produced using faba beans (commercially available) and lupin seeds (generously provided by the Institute of Field and Vegetable Crops in Novi Sad). The other ingredients included water, sunflower oil, sesame seed paste, an acidity regulator (citric acid), salt, and spices (garlic, black pepper). In addition to functional requirements, these spreads also need to meet food safety requirements. This is particularly critical given that mycotoxins, which are toxic secondary metabolites produced by fungi, present a significant concern for food safety due to their proven and substantial toxicity to human health. Their presence in food products requires rigorous monitoring and control measures to ensure consumer safety, prevent potential health risks, and avoid economic losses. The aim of this research was to investigate the presence of 16 mycotoxins in faba bean and lupin spreads using the developed and validated liquid chromatography-tandem mass spectrometry (LC-MS/MS) method. The obtained results indicate that the frequency and levels of the analysed mycotoxins were low in the investigated spread samples. Specifically, only alternariol-methylether was detected in the faba bean spread, while alternariol-methylether and sterigmatocystin were found in the lupin spread, all at concentrations below 10 µg/kg. Both spreads were negative for all regulated mycotoxins, including aflatoxins, fumonisins, zearalenone, deoxynivalenol, ochratoxin A, and non-regulated mycotoxins such as T-2, HT-2, moniliformin, and tentoxin. Based on the results obtained in this study, it can be concluded that, with regard to mycotoxin content, spreads represent safe food products for consumers.

**Keywords:** mycotoxins, lupin, faba bean, spreads

**Acknowledgements:** This research was financially supported by the project financed by the H2020-EU.3.2.1. "Climate Resilient Orphan crops for increased DIVERSITY in Agriculture – CROPDIVA" (ID: 101000847) and the Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## MOLECULAR TOOLS FOR THE PROTECTION OF AGROBIODIVERSITY AND PROMOTION OF SUSTAINABLE AGRICULTURE

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The protection and conservation of grain agrobiodiversity represent milestones in modern and sustainable agriculture and food waste reduction. Molecular tools play a vital role in understanding grain traits, although their integration into agricultural practices remains limited. Our study, part of the CROPDIVA project, focuses on developing biochemical methods to assess grain quality and select high-performing varieties for different applications. Our research aimed at establishing a high-throughput screening pipeline for CROPDIVA varieties, enhancing agricultural diversity and promoting new supply chains. Diversified agricultural products contribute to soil enrichment and dietary improvement. We conducted comprehensive bromatological and phytochemical characterizations of CROPDIVA samples, evaluating protein, lipid, humidity and fiber content, enzymatic starch assays, and bioactive and nutritional features such as total polyphenols, anti-nutritional factors, and total antioxidant capacity. Furtherly, selected CROPDIVA grain varieties were evaluated by adopting an *in vitro* digestion models to assess dry matter and protein digestibility, and nutrient bioavailability of selected nutrient compound. In conclusion, integrating minor crops into feed enhances agricultural sustainability, benefits human nutrition, and supports ecosystem resilience. Our findings underscore the importance of agrobiodiversity in fostering resilient agricultural systems that meet global food security challenges.

**Keywords:** grain quality, seeds composition, molecular characterization, nutritional evaluation, agrobiodiversity

**Acknowledgements:** This project has received funding from the European Union’s Horizon 2020 research and innovation programme under grant agreement NO 101000847.



## THE ROLE OF YOUNG CONSUMERS IN SHAPING ATTITUDES TOWARD UNDERUTILIZED CROPS: INSIGHTS FROM FOCUS GROUP RESEARCH

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Underutilized crops like oats, buckwheat, faba beans, and barley have significant potential to support sustainable food systems and diversify diets. However, their consumption remains low, particularly among younger generations. To explore young consumers' perceptions and acceptance of products made from these crops (oat flakes, buckwheat pasta, faba beans, and barley groats), we conducted focus groups with 12 participants aged 18-25 as part of the Climate Resilient Orphan Crops for Increased Diversity in Agriculture (CROPDIVA) project. Discussions focused on factors such as knowledge, perspectives on quality, perceived health benefits, taste, consumption patterns and convenience of purchase, preparation and consumption to gather qualitative insights into their experiences, preferences, and barriers to adopting these products. Oat flakes were well perceived due to their ease of preparation and association with healthy eating habits, with participants appreciating their high fibre content, making them a favourable option for breakfast or snacks. Buckwheat pasta, valued for its gluten-free properties and unique taste, faced challenges due to the bitter taste noted by some participants as well as its limited availability in mainstream markets. However, its perceived health benefits were noted, particularly among those with dietary restrictions. Faba beans were recognized for their high protein content and potential as a meat alternative, aligning with the trend towards plant-based diets. Despite initial concerns about preparation complexity and availability, participants were willing to incorporate faba beans into their diets. Barley groats, although less familiar to participants, were acknowledged for their high fiber content, though barriers included a lack of awareness and limited culinary applications. Overall, the study underscores the importance of education and targeted marketing in promoting the acceptance of underutilized crops in young consumers' diets. By addressing sensory preferences, convenience, and health benefits, product innovation can enhance the appeal of these crops, supporting their role in more sustainable and diverse food systems.

**Keywords:** focus groups, oat flakes, buckwheat pasta, faba beans, barley groats

**Acknowledgements:** This work was financially supported by the project financed by the H2020-EU.3.2.1. "Climate Resilient Orphan crops for increased DIVERsity in Agriculture – CROPDIVA" (ID: 101000847) and Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## FABA BEAN FLOUR AND PROTEIN ISOLATE AS PARTIAL SUBSTITUTES IN WHEAT-TRITICALE BREAD: A TECHNOLOGICAL AND SENSORY PROPERTIES

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The incorporation of faba bean flour (FBF) and protein isolate (FBPI) into wheat-triticale flour blends is a potential promising strategy to enhance the nutritional and functional properties of bread. This study aimed to investigate the potential of these ingredients from both technological and sensory perspectives. Multiple analyses were conducted, including chemical composition, texture profile analysis, color, specific volume and sensory properties of breads made with a blend of wheat and triticale flours (70:30). Part of the blend was substituted with either 20% faba bean flour or 7.2% faba bean protein isolate to ensure an equal proportion of faba bean protein in the final formulations. The results showed a significant increase in protein content for both FBF and FBPI breads, with the faba bean protein isolate also increasing the ash content. Texture profile analysis (TPA) indicated that the substitutions affected bread properties by increasing hardness, gumminess, and chewiness, particularly in bread with FBPI. However, cohesiveness, springiness, and resilience decreased similarly in both cases. Additionally, the specific volume decreased with the addition of faba bean flour and protein, especially in FBPI bread. From the colour aspect, analysis revealed a darker hue and a more pronounced reddish nuance in the bread crust in FBF and FBPI breads. Sensory evaluation indicated a slightly lower overall likeability with faba bean addition, yet the breads remained acceptable to the panellists. Detailed sensory analysis supported the TPA findings and colour differentiation, showing higher values for hardness, crust compactness, and crumbliness. Texture and color were rated lower compared to the control sample. Moreover, both FBF and FBPI breads exhibited higher overall odour intensity. Specifically, bread with the protein isolate had less uniform pores and a more pronounced flour/cereal/bran odour. In contrast, bread with faba bean flour had more uniform pores but a noticeable bitterness. In conclusion, this study demonstrates that faba bean flour and protein isolate can be used as partial substitutes in wheat-triticale flour blends to produce bread with acceptable properties. This leaves room for future in-depth analysis and modifications to address the identified challenges.

**Keywords:** faba bean, protein isolate, triticale, bread

**Acknowledgements:** This work was financially supported by the project financed by the H2020-EU.3.2.1. "Climate Resilient Orphan croPs for increased DIiversity in Agriculture – CROPDIVA" (ID: 101000847) and Ministry of Science, Technological Development and Innovation, Republic of Serbia (Contract No. 451-03-66/2024-03/200222).

## SENSORY AND PROTEIN QUALITY EVALUATION OF A HYBRID BEEF BURGER WITH FAVA BEAN AND PEA PROTEINS

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FAO recommends digestible indispensable amino acid score (DIAAS) to assess the protein quality. Moreover, the sum of the IAAs per gram of protein in the recommended scoring pattern for the age group of 3 years and over is 291 mg, consequently 709 mg are dispensable amino acids (DAAs). In protein sources with DIAAS value above 100, the limiting factor for the optimal utilization of the amino acids could be DAAs. When IAAs are present in excess, some of these IAAs can be utilized as precursors for DAAs. The aim of this study was to evaluate the quality of a hybrid burger (HB) by combining beef with a texturized protein (TP) of fava:pea (20:80) with respect a beef burger (BB). HB and BB had the same protein content, but 50% of protein in HB came from fava:pea TP. Content and standardized ileal digestibility (SID) for each AA were obtained from literature for each protein source. DIAAS scores and the percentage of IAAs used for protein synthesis were calculated. A sensory analysis was performed by a panel of experts in meat products. HB had a specific smell and taste (not unpleasant). They showed a satisfactory binding and were scored with a similar juiciness than BB. Both HB and BB had excess of utilizable IAAs for protein synthesis and DAAs became the limiting factor. Two scenarios were considered: i) excess of IAAs are oxidized, ii) part of them are used as precursors of DAAs. HB showed lower DIAAS score (112%) than BB (131%). However, it showed higher percentage of IAAs utilizable for protein synthesis than BB, both in scenario i) (51.6% vs 46.6%) and in scenario ii) (61.2% vs 58.4%). In conclusion, HBs are a good alternative to BB burgers, with a specific taste and a similar nutritional quality.

**Keywords:** hybrid beef burger, fava bean protein, pea protein, sensory analysis, protein quality

**Acknowledgements:** This work was funded by the Horizon 2020 UE (CROPDIVA ref. 101000847) and CERCA programs.

## SUSTAINABILITY IN PROTEIN EXTRACTION: FROM SOURCE TO TECHNOLOGY – A CASE STUDY OF FABA BEAN PROTEIN

Tamara Dapčević Hadnađev<sup>1\*</sup>, Nikola Maravić<sup>1</sup>, Jelena Tomić<sup>1</sup>, Aleksandar Marić<sup>1</sup>, Milica Šalgo<sup>2</sup>,  
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Sustainability in protein extraction can be achieved either by using sustainable protein sources - such as waste materials, abundant or orphan crops, or bio-renewable resources - or through the application of green extraction techniques, process optimization in terms of resource usage and energy consumption and a biorefinery approach. This study explores sustainability in protein extraction by comparing three isolation techniques for extracting protein from faba bean, an orphan crop. The methods examined include alkaline extraction followed by isoelectric precipitation, NaCl extraction (micellization), and the use of deep eutectic solvents (DES), specifically choline chloride/urea and choline chloride/glycerol. The NaCl technique was found to best preserve the protein's natural state, while the DES method resulted in higher protein solubility, particularly near the isoelectric point, and improved foaming capacity and stability. Isoelectric precipitation yielded high water retention and oil absorption capacities. Despite all techniques producing high-purity proteins, the DES approach showed a lower yield, suggesting the need for optimization in terms of solvent water content and precipitation methods. The study highlights how different extraction techniques influence the functional properties of faba bean protein, offering insights into sustainable protein processing through innovative methods and resource efficiency.

**Keywords:** faba bean, protein extraction, green solvents, sustainability

**Acknowledgements:** This study is supported by H2020-EU.3.2.1 - project "Climate Resilient Orphan croPs for increased DIVERsity in Agriculture - CROPDIVA (GA No. 101000847).

## POTENTIAL TO ENHANCE CEREAL SEED QUALITY THROUGH INTERCROPPING WITH PEAS IN SERBIA'S PANNONIAN CLIMATE

Vesan Župunski<sup>1\*</sup>, Radivoje Jevtić<sup>1</sup>, Ljiljana Brbaklić<sup>1</sup>, Sanja Mikić<sup>1</sup>, Vladimir Aćin<sup>1</sup>, Anja Dolapčev Rakić<sup>1</sup>, Milosav Grčak<sup>2</sup>

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Mixed intercropping is regarded as a promising strategy for reducing nitrogen (N) usage in cultivation systems. The positive impact of cereal-legume intercropping on protein content in companion cereal crops has been observed across various agro-ecological zones. However, a limited understanding of how intercropping influences N use efficiency and the performance of individual crops within these systems is currently restricting the broader adoption of intercropping practices. To explore the potential of intercropping for improving seed quality in spring oats, hull-less barely and winter triticale under Serbia's agrp-ecological conditions, these cereal crops were cultivated alongside spring and winter peas in mixed intercropping systems in Novi Sad, Northern Serbia. Seeding rates of the three crops in mixed intercropping were set at 70% (pea): 30% (oats, hull-less barely) and 80% (pea): 50% (triticale) of the conventional seeding rate. We used principal component analysis with mixed data and general linear modelling to explore potentials to enhance cereal seed quality through intercropping with peas in Serbia's Pannonian climate. Our study showed that variations in yield, thousand-kernel weight (TKW), and crude protein content, as well as their interrelationships under intercropping cultivation, were cultivar-specific and differed among cereal crops exposed to the same agroecological conditions. In addition, the potential of intercropping to improve crude protein content and TKW in cereal crops has been demonstrated in field experiments. However, the extent of this improvement depends on the appropriate combination of cereal and pea varieties and is highly influenced by climatic conditions. As the relationship between TKW, yield, and protein content has been shown to be crop- or even variety-specific under the same climatic conditions, selecting appropriate varieties and using optimal sowing rates are essential for improving TKW and crude protein content in intercropping systems.

**Keywords:** triticale, oats, hull-less barely, crude protein, TKW

### Acknowledgements:

1. CROPDIVA - Climate resilient orphan crops for increased diversity in agriculture, the project funded from the European Union's Horizon 2020 research and innovation programme under grant agreement N°101000847
2. Ministry of Science, Technological Development and Innovation of the Republic of Serbia, grant number: 451-03-47/2023-01/200032"



# HROMATOGRAFIJA

thermo  
scientific

## GASNA HROMATOGRAFIJA

TRACE 1600 GC



Novu TRACE 1600 seriju gasnih hromatografa odlikuje jedinstveni modularni koncept i mogućnost zamene injektorskih i detektorskih modula od strane korisnika, te omogućava fleksibilnost konfiguracije. Stoga je pogodan kako za rutinske, tako i za najzahtevnije laboratorije. TRACE 1600 može da se nadoveže sa čitavom paletom MS detektora, gde se kontrola i akvizicija podataka vrši kroz Chromeleon softver.

## TEČNA HROMATOGRAFIJA

Thermo Scientific portfolio za tečnu hromatografiju čini Vanquish serija, koja obuhvata HPLC, UHPLC, nano LC sisteme:

- Vanquish Horizon UHPLC sistem sa ultravisokim pritiscima do 1500 bara
- Vanquish Flex UHPLC sistem sa pritiscima do 1000 bara i Vanquish Core HPLC do 700 bara
- Vanquish Duo UHPLC sistem – sistem dizajniran za istovremeno izvođenje dve aplikacije



Vanquish Core HPLC system

## JONSKA HROMATOGRAFIJA



Dionex ICS-6000 HPIC sistem

Bilo da imate samo nekoliko uzoraka ili veliki broj, jednostavan analitički zadatak ili pravi izazov, nudimo odgovarajuće rešenje za analizu katjona i anjona:

- Modularni jonski hromatografi za zahtevne IC aplikacije: Dionex ICS-6000 HPIC sistemi
- Integrirani jonski hromatografi za rutinske analize, RFIC sistemi i kapilarni: Dionex Integriion HPIC, Dionex Aquion, Dionex Easion sistemi i Dionex ICS-4000 kapilarni HPIC sistem

## ORBITRAP ASTRAL MASENI SPEKTROMETAR

Orbitrap Astral maseni spektrometar kombinuje visoku produktivnost, sveobuhvatnu pokrivenost i kvalitet podataka i veću osetljivost pružajući tačnu i preciznu kvantifikaciju u širokom dinamičkom opsegu. Kombinacija tri masena analizatora (maseni filter visokih rezolucija, Orbitrap™ maseni analizator i novi Astral maseni analizator) omogućava brzu akviziciju izvanrednih HRAM podataka sa visokom osetljivošću i dinamičkim opsegom. Performanse ovog instrumenta čine ga idealnim za tačnu i preciznu kvantifikaciju bez presedana, u analizi kako pojedinačnih ćelija tako i masovnih uzoraka.



## SOFTVER ZA HROMATOGRAFIJU CHROMELEON 7.3 I CHROMELEON CDS ENTERPRISE



Chromeleon 7.3 CDS softver

Chromeleon 7.3 CDS hromatografski softver obezbeđuje najbrži put od uzorka do rezultata. Podržava kontrolu masenog spektrometra i obradu podataka svih separacionih tehnika (LC, GC, IC). Softver je potpuno integrisan sa GC-MS/MS i LC-MS/MS instrumentima kao i hromatografskim sistemima drugih proizvođača opreme.

# MASENA SPEKTROMETRIJA

thermo  
scientific

## GC-MS/MS TRIPLE QUADRUPOLE SISTEMI

Najnovija generacija GC-MS/MS sistema TSQ 9610 u pogledu osetljivosti i robusnosti odgovora na najstrožije zahteve EU regulative za analizu dioksina i furana. Zahvaljujući novom jonskom izvoru (AEI) detekcioni limit instrumenta doseže ato opseg. NeverVent Technology omogućava da se bez narušavanja vakuuma promeni hromatografska kolona za svega 35 min, odnosno zameni jonski izvor za svega 5 min.

TSQ 9610 Triple  
Quadrupole GC-MS



## LC-MS/MS TRIPLE QUADRUPOLE SISTEMI



TSQ Quantis+

Nova generacija triple stage quadrupole masenih spektrometara (TSQ Fortis+, TSQ Quantis+, TSQ Altis+) je bez kompromisa u pogledu osetljivosti i robusnosti. Zahvaljujući jedinstvenoj active ion management (AIM+) tehnologiji, unapređenom jonskom izvoru i elektron multiplajeru, pružaju pouzdanu kvantifikaciju. Model TSQ Altis+ je najprestižniji u seriji i pruža nenadmašnu osetljivost za sve tipove molekula u različitim matriksima. TSQ maseni spektrometri se prodaju sa trogodišnjim fabričkom garancijom.

## ORBITRAP EXPLORIS LC-MS/MS SISTEMI

Orbitrap Exploris HRAM serija masenih spektrometara nudi izuzetnu osetljivost i selektivnost, te omogućava analize do najnižih nivoa sa visokom kvantitativnom tačnošću i preciznošću. U zavisnosti od kompleksnosti analita, možemo da ponudimo tri modela:

- Orbitrap Exploris 120: rezolucija 120.000 m/z, za male molekule
- Orbitrap Exploris 240: rezolucija 240.000 m/z, za male molekule i peptide
- Orbitrap Exploris 480: rezolucija



Orbitrap Exploris 240 MS

## HRAM ORBITRAP TRIBRID LC-MS/MS SISTEMI



Orbitrap Eclipse Tribid MS

Orbitrap Tribid sistemi su najnapredniji HRAM maseni spektrometri na svetu, koji kombinuju najbolje osobine kvadrupola, Orbitrapa i jonskih zamki i omogućavaju analizu najzahtevnijih uzoraka. Rezolucija i povećana brzina ciklusa čine ove uređaje pogodnim za rad na uzorcima visoke kompleksnosti koji se sreću u proteomici, metabolomici i drugim high-throughput tehnikama. Kroz veliki broj tehnoloških inovacija omogućena je identifikacija više jedinjenja, tačnija kvantifikacija i otkrivanje više strukturnih detalja.

## STELLAR MASENI SPEKTROMETAR

Thermo Scientific™ Stellar™ maseni spektrometar donosi inovacije u osetljivosti, specifičnosti i produktivnosti. Stellar maseni spektrometar kombinuje revolucionarne hardverske i softverske napretke sa robusnom kvantifikacijom. Povećajte pouzdanost rezultata zahvaljujući specifičnosti hiper-brze MSn akvizicije u punom skeniranju iz kvadrupolne-linearne ionske zamke i detektorskog sistema, koji omogućava osetljivost za pojedinačne jone.



Orbitrap Exploris 240 MS

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# MOLEKULSKA SPEKTROSKOPIJA

## INFRACRVENA SPEKTROSKOPIJA I MIKROSKOPIJA (FT-IR)

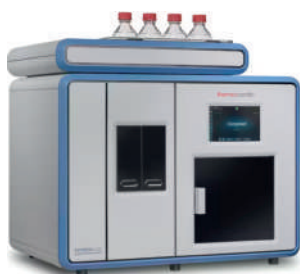


Nicolet iS50 FT-IR spektrometar sa  
Nicolet RaptIR FT-IR mikroskopom

Thermo Nicolet iS5 FT-IR spektrometar obezbeđuje napredne FT-IR performanse u kompaktnoj veličini po pristupačnoj ceni i predstavlja idealno rešenje za industrijske, forenzičke i akademske laboratorije. Model Nicolet iS50 pokriva VIS, NIR, MIR i FAR oblast. Preporučeni model FT-IR mikroskopa: Nicolet iN10 i novi Nicolet RaptIR pogodan za široki spektar industrija.

## EXTREVA AŠE AUTOMATIZOVANI EKSTRAKTOR RASTVARAČEM (ACCELERATED SOLVENT EXTRACTOR)

Automatizovano bez ikakve intervencije, izvršite ekstrakciju uzoraka, cleanup u ekstrakcionoj ćeliji i evaporaciju—sve u jednoj besprekornoj operaciji—skratite vreme manualnog rada sa sati na minute i omogućite sebi da se fokusirate na druge laboratorijske prioritete dok koristite do 50% manje rastvarača od drugih tehnika pripreme uzoraka. EKSTREVA ASE ubrzani ekstraktor rastvarača nudi sveobuhvatnu pripremu uzoraka: automatski ekstrahuje i koncentriše uzorke, eliminišući potrebu za ručnim premeštanjem ekstrakta uzoraka na drugi uređaj za isparavanje rastvarača.



## UV-VIS SPEKTROFOTOMETRIJA



Evolution One UV-Vis

Thermo Scientific nudi široku paletu UV-Vis spektrofotometara, od jednozračnih do dual i double beam UV-Vis spektrofotometara, koji zahvaljujući svojoj raznovrsnosti nalaze primenu u edukaciji, kontroli kvaliteta i istraživačkom radu.

## RAMANSKA SPEKTROMETRIJA

DXR RamanSmart spektrometar dizajniran je za rutinske i procesne analize u analitičkim i razvojnim laboratorijama. Zahvaljujući izuzetnim performansama, nalazi primenu i u istraživačkim, akademskim i forenzičkim laboratorijama. Omogućava jednostavno uzorkovanje i minimalnu pripremu uzorka, kao i snimanje kroz pakovni materijal.



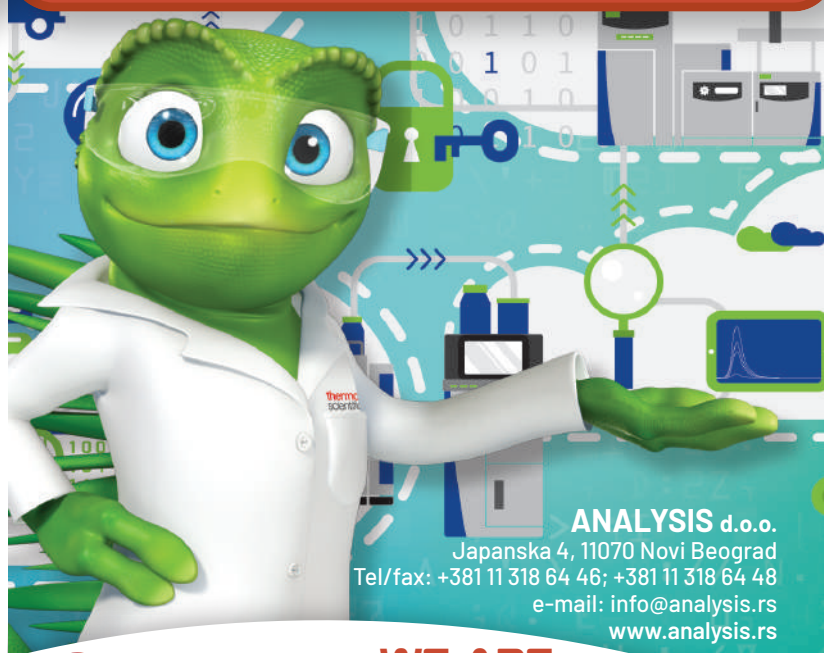
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Mikroskop

## KOLONE I POTROŠNI MATERIJAL ZA HROMATOGRAFIJU



Thermo Scientific nudi veliki izbor kolona za tečnu, gasnu i jonsku hromatografiju po povoljnim cenama, kao i veliki izbor potrošnog hromatografskog materijala za pripremu uzorka: špic filtere, Quecherse, SPE kertridže, viala, itd.

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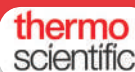
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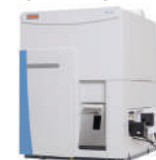
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TO WORK TOGETHER**

## ELEMENTALNA ANALIZA



### MASENA SPEKTROMETRIJA SA INDUKOVANO- SPREGNUTOM PLAZMOM (ICP-MS)

Thermo Scientific portfolio ICP-MS instrumenata sadži single quadrupole (ICP-MS SQ), triple quadrupole (ICP-MS TQ) kao i masene spektrometre visoke rezolucije (ICP-MS HR). Fleksibilnost i jednostavnost korišćenja u kombinaciji sa visokom osetljivošću i stabilnošću instrumenta čine ih idealnim za kvalitetne rutinske kao i za zahtevne analize.



iCAP TQ ICP-MS

### SPEKTROMETRIJA SA INDUKOVANO-SPREGNUTOM PLAZMOM (ICP-OES)



iCAP PRO XPS ICP-OES

Nova Thermo Scientific iCAP PRO serija ICP-OES obezbeđuje veoma brzu multielementalnu analizu tragova elemenata. Instrument karakteriše napredne performanse, visoka produktivnost, jednostavno korišćenje, pouzdani rezultati u skladu sa propisanim regulativama i standardima. Modeli: iCAP PRO i PRO X - osnovna konfiguracija; iCAP PRO XP - rutinske analize pogodne za QA/QC laboratorije i iCAP PRO XPS - na najzahtevnije analize, namenjene R&D laboratorijama.

### FLASH SMART CHNS/O, N-protein, TOC ANALIZATOR

FlashSmart analizator rešava više laboratorijskih zadataka, poboljšava i ubrzava laboratorijski rad i skraćuje vreme trajanja analize, zahvaljujući Dumas metodi koja omogućava visoku fleksibilnost i modularnost u radu, nudeći više od dvadeset konfiguracija u jednom instrumentu. Moćni softver podržava automatizaciju i precizno izveštavanje.



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CIP - Каталогизација у публикацији  
Библиотеке Матице српске, Нови Сад

663/664:658.562(048.3)  
614.31(048.3)

**INTERNATIONAL Congress "Food Technology, Quality and Safety" (5 ; 2024 ; Novi Sad)**

e-Abstract book [Elektronski izvor] / 5th International Congress "Food Technology, Quality and Safety", 16-18 October 2024, Novi Sad ; [main editor Ljubiša Šarić]. - Novi Sad : Institute of Food Technology, 2024

Način pristupa (URL): [https://fins.uns.ac.rs/publications/?\\_sft\\_publicationss=zbornik](https://fins.uns.ac.rs/publications/?_sft_publicationss=zbornik). - Opis zasnovan na stanju na dan 22.10.2024.

ISBN 978-86-7994-063-6

а) Животне намирнице -- Контрола квалитета -- Апстракти б) Животне намирнице -- Хигијена -- Апстракти

COBISS.SR-ID 155106313