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Innovative Food Science and Bioprocesses''



CHARACTERISATION OF PROTEINS AND SURFACTANTS AT FLUID INTERFACES RELEVANT FOR FOAMS AND EMULSIONS IN FOOD

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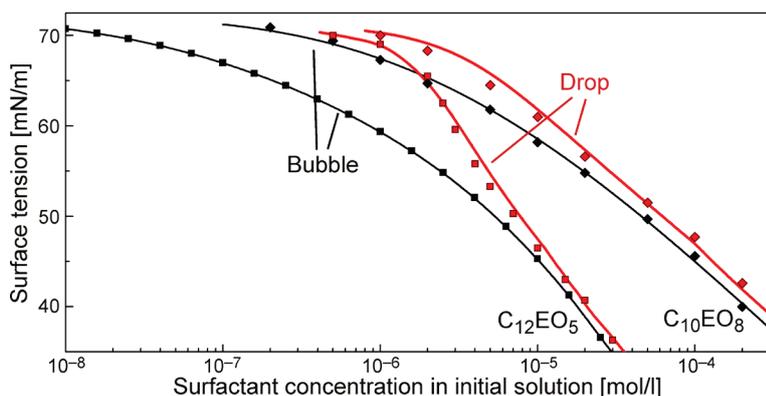
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Drop and bubble profile analysis tensiometry (PAT) is the most advanced method for measuring the equilibrium and dynamic surface/interfacial tensions of surfactant and protein solutions at water/air and water/oil interfaces. The methodology provides not only important information about the self-assembling of surfactants/proteins at interfaces, but also the mechanical properties of the formed interfacial layers. By slow harmonic interfacial perturbations, the interfacial relaxation processes and hence the dilational visco-elasticity of the adsorbed layers can be determined.

The values, however, measured via the analysis of drop and bubble profiles, respectively, can deviate from each other significantly. This difference can be of the order of 10 mN/m or even more, depending on the bulk concentration and surface activity of the studied surfactant/protein. The differences are caused by a depletion of molecules from the bulk of single drops due to their adsorption at the drop surface. For experiments with bubbles formed in the solution under study, the depletion is 2 to 3 orders of magnitude less and therefore in most cases negligible.

The depletion due to adsorption can lead to a final concentration in single drops after reaching the adsorption equilibrium which is much lower than the initial concentration. If ignored, these effects lead to incorrect surface tension isotherms and also dilational visco-elasticity of the interfacial layers. Thus, using data from drop profile analysis tensiometry without the correction of possible depletion effects can lead to huge errors in the estimated adsorption parameters.



Surface tension isotherms of C₁₀EO₈ and C₁₂EO₅ with different surface activities, as measured by bubble profile tensiometry (black) and drop profile tensiometry (red); from: T. Kairaliyeva *et al.*, Surface tension and adsorption studies by drop profile analysis tensiometry,



J. Surfactants Detergents, **20** (2017) 1225.

The presentation provides a detailed information about the drop/bubble profile methodology, a characterisation of the mentioned depletion effects, and a number of practical data for selected surfactant and protein solutions.

Keywords: proteins, surfactants, foams



BIOPOLYMERS AS VEHICLES FOR DELIVERING INNOVATIVE FUNCTIONALITIES IN FOOD SYSTEMS

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Biopolymers have been exploited for many years in the food industry as hydrocolloid ingredients for preparing food products with improved quality, functionality and shelf-life. Broadly speaking, they are used to perform a number of functions including thickening and gelling aqueous dispersions, water binding, fat replacing, flavor masking, stabilizing emulsions and foams, as well as acting as coating and encapsulation agents of bioactives and other small molecules for effective protection and target release delivery. Lately, a range of health benefits has been also attributed to the consumption of certain food hydrocolloids, e.g. fibers, which include lowering risk factors for cardiovascular diseases, control of glycemic responses, improvement of colonic health and immune function. As a result, legislative authorities around the world have approved the use of specific health or nutritional claims for some hydrocolloids, opening the potential for new markets of food products formulated with these versatile polymeric ingredients. All these characteristics and beneficial effects of food biopolymers are related to their structural features (both chemical and physical constitution) and to modifications they may undergo as a result of various processing treatments (mechanical-thermal and/or chemical-enzymic).

This review paper will highlight some aspects on the use of certain food biopolymers as functional ingredients in food matrices with an attempt to relate structure and interactions to functionality issues and challenges in formulating products, with an emphasis on cereal fiber components. In this context, structure-physical property relations of cereal soluble fibers will be discussed, in both model or real food products fortified with these polysaccharides, as well as the impact of processing and matrix effects on fiber functionality.

Keywords: biopolymers, hydrocolloids, functionality, food products

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PROCESS SIMULATION IS AN ESSENTIAL TOOL IN PRODUCT DEVELOPMENT

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The potential to meet the future protein demand through increased production of animal and plant products is unsustainable because of the growing demand for agricultural land. Alternative sources of nutritional products that are safe, non-GMO, and of non-animal origin are needed to meet projected dietary needs. Current lifestyle trends, such as the increased consumption of vegan, sustainable food products, and health-promoting nutraceuticals has brought both leafy plants and microalgae at the forefront of alternative food sources. In this presentation, we will demonstrate how technical and commercial barriers during the development of microalgae nutraceuticals have been addressed with the help from process simulation. Specifically, we have assessed the feasibility of an integrated algal source platform for carotenoid and protein production from *Chlorella vulgaris*. Process simulation with SuperPro Designer® software revealed that the co-production of high-value carotenoids would significantly improve the economic outcome of algae-based protein hydrolysates and concentrates, whereas these protein products alone would not be sustainable or economically possible. Another take home message from the process simulation was that achieving an integrated, cost-effective bioprocess required a careful tradeoff between desirable traits (e.g. color, taste, and nutritional quality), specific unit operations, and operating cost. For example, the more expensive enzymatic extraction and conversion of native algal protein to protein hydrolysates when followed by membrane filtration eliminated the undesirable green color of extracted protein and increased the protein monetary value. On the other hand, non-hydrolyzed “green” protein concentrates were less expensive and had better emulsification properties than the hydrolysates, but the latter were of superior biological and nutraceutical value compared to the concentrates.

Keywords: proteins, product development, process simulation



INNOVATING WITH BIOACTIVES FROM BRASSICA BYPRODUCTS - THE SPIN-OFF EXPERIENCE

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The integrated studies “from farm to health” at CEBAS-CSIC (the Spanish National Research Council Institute in Murcia, SE Spain), developing healthier foods enriched in bioactive phytochemicals, is uncovering new possibilities for the use of cruciferous foods and ingredients with added value of high-density in chemopreventive glucosinolates, biotechnological ingredients for healthy foods, and new cosmetics, patented ingredients for culinary and industrial products. The harvest remains, non-commercial plant parts, and other bioburden of the intensive agricultural production, have only been minimally used as animal feed or as sources of glucosinolate standards. However, the boosted crop productions for the growing international trade to Europe and Asia in recent years makes unbearable to manage such amount of byproducts generated every year. Therefore, novel opportunities and strategies for economic activities in the EU-arena were envisaged: The development of a Spin-off company active on R&D to offer a pipeline of scientifically-based products for commercialization – Aquaporins & Ingredients SL. Current developments on bioactive compounds obtained from agri-food waste recovery open new opportunities for the effective transfer of technology from public research to industry applications in this global era of producing healthy plants for healthy foods for health and wellbeing.

Keywords: Brassica, agrowaste, bioactive compounds, glucosinolates, functional foods

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FROM FOOD TO SAFE FOOD: PROBLEMS AND CHALLENGES IN NEW PRODUCT DEVELOPMENT

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Food safety is integral part of food science including technology and processing but also needs social and cultural sciences to ensure effective food safety management. We need to protect the consumer while also preserving food heritage. If we consider development as a permanent challenge in the food supply chain, we can notice several ongoing changes at different levels, from the farm to the fork: 1. At the level of substrates - inclusion of many additives, which did not use to be common in traditional food processes; 2. At the technical level - adoption of new processing equipment with very specific features; 3. At the processing and organizational level – new information technology for management of processes; 4. At the level of packing - reduction of packaging sizes for one person portions; 5. At the level of trade - absolute domination of global trading networks; 6. At the level of food preparation - more convenience foods over traditional food preparation; 7. At the level of nutrition - invention of new eating habits focused towards exclusion of family and reduction/elimination of social life during eating.

What do all the aforementioned challenges have in common? Exclusion or at least limitation of the human factor and its impact on the food supply chain. In these trends, safety has an important place in daily practice, when we start to get uncomfortable and we start to worry about a particular issue regarding food, its origin, composition, impact on our health or environment, and so on. In the last century, food processing and preservation methods have been traditionally associated with an array of fundamental concepts. Consequently, new discoveries in sciences and engineering fields enabled fast developments of several non-thermal techniques such as High-Pressure Processing (HPP), Pulsed electric fields (PEF) and others. Food research and development found them very appropriate and useful. Usage of non-terminal techniques during processing can achieve microbial inactivation and reduce detrimental changes in physical or sensory properties of foods. Since these techniques are effective, the result are healthier and more natural foods. Additional impact has been made by the emergence of artificial intelligence and robotics, which has tremendously changed food production and distribution streams. The use of AI has made some complex techniques easier to implement, guide and maintain. Additionally we face both E-commerce in the food industry and also virtual restaurants and so on. Innovation connected to ingredients using old knowledge based on health benefits of native foods has opened the path for the growth of native foods. On the soft side of development within food production and supply chain we face the rise of food safety culture. When foodborne illness starts due to contamination with food it requires very specific behaviour and reactions of those responsible in the food supply chains towards consumers. This is why education is an ongoing and key factor that must accompany development and research of food and eating habits.

Keywords: food, processing, technology, research, development, safety



APPLICATION OF SUPERCRITICAL CARBON DIOXIDE FOR DRYING OF FRUITS AND VEGETABLES – SENSORY AND FOOD SAFETY PERSPECTIVES

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Consumers have always been concerned about the safety and quality of the foods they eat expressing growing awareness for high-quality and fresh-like minimally processed foods of uncompromised safety. Super-critical carbon dioxide technology (scCO₂) foods has gained a particular scientific and industrial interest in the last decades owing to CO₂ being non-carcinogenic, nontoxic, non-mutagenic, non-flammable, colorless, odorless, thermodynamically stable and relatively inexpensive. In recent years, the attention has been particularly focused on application of scCO₂ in food pasteurization and drying. To achieve dehydrated fruit or vegetable commodities of high quality, with minimal loss of nutrients, volatiles and flavors, and with acceptable/desirable changes in color and texture, dehydration must occur fairly rapidly. During supercritical drying the water is not removed by vaporization or sublimation but is dissolved in the scCO₂. Due to these gas-liquid properties of scCO₂, the structure of the dried plant material may remain preserved at high extent. Another important advantage is that drying process takes place at low temperatures (e.g. 40 °C) which is also beneficial for the sensory and nutritional profile of final product.

ScCO₂-drying has not found its commercial industrial application yet. The company FeyeCon Development and Implementation BV (Weesp, NL) patented pilot scale equipment for industrial scCO₂-drying at the European Patent Office and a series of research studies have been performed within the H2020 scientific project 'Future-Food' in the period 2015-2018. The sensory evaluation trials performed on basil, bell pepper, beetroot, and apple showed that scCO₂-drying can bring and retain the same (or even higher in some cases) sensory quality of the dried plant material as compared to usual freeze-drying. ScCO₂-drying treatments resulted in 'very good' texture quality of the dried fruit/vegetables snack products that were characterized by low level of shape and surface deformations, pronounced brittleness and crispiness, and good chewiness and rehydration during mastication, with the typical color and flavor preserved. From the microbiological safety and quality point of view, the scCO₂ treatment at 40 °C showed potent antimicrobial effect against vegetative bacteria, yeasts and molds on all tested herbs, fruits and vegetables. Naturally present microbial flora appeared to be particularly sensitive indicating great potential to extend shelf-life and minimize risks to food safety. These aspects endorse use of scCO₂-drying as a promising 'green' technology than



combined drying and pasteurization in a single step. The spores of inoculated mycotoxin producing *Aspergillus flavus* and *Fusarium graminearum* in pistachios showed different level of resistance, with *A. flavus* being significantly more resistant. Natural mesophilic and psychrotrophic bacteria were strongly reduced (up to 4 log CFU/g). Bacterial spores (*Bacillus cereus* spores in the first place) were very resilient to this inactivation process, suggesting the need of additional treatments combination of hurdles to prevent spore germination and multiplication, and possible subsequent toxin production. Inoculated pathogenic *Escherichia coli*, *Salmonella enterica*, *Staphylococcus aureus*, and *Listeria monocytogenes* were reduced below the limit of detection after 360 min of the drying (6-7 log units of reduction), with inactivation kinetics being dependent on the type of food matrix. Bacterial toxins and mycotoxins were only partially (or not) inactivated under the conditions tested. In conclusion, sensory evaluation and microbial safety trials showed promising potential of scCO₂-drying technology to be used as an alternative drying method in the production of dried fruits and vegetables. Trials on industrial scale, as well as economic assessment are required for final commercial use.

Keywords: *supercritical CO₂ drying, fruits and vegetables, sensory quality, microbiological safety*

Acknowledgements: *A series of research studies encompassed by this overview have received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 635759: Faster Upcoming Technology Uptake Relevant for the Environment in FOODs Drying ("FUTURE-FOOD"). Concrete data presented in this presentation are result of joint efforts of Ghent University, University of Padova, University of Belgrade and FeyeCon Carbon Dioxide Technologies.*



CHALLENGES IN SUGAR PRODUCTION - FOOD SAFETY MANAGEMENT

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Sugar industry is one of the largest among the food industry. One plant processes around 8,000 tons of sugar beet daily and produces 1,150 tons of sugar. It can be said that sugar is a strategic product of Serbia, and the wastes and by-products are valuable as well, since they are raw materials for other technological processes which are also important. One of them is betaine from sugar beet molasses, which is essential for living. Through this paper the products of the sugar industry will be introduced in one hand, and the high product safety requirement in the other hand.

Due to unambiguous impact of globalization on modern business, one of the requirements set in front of manufacturers is standardization and certification of food safety and quality systems. This paper provides an overview of the GFSI Standards - *Global Food Safety Initiative* and key factors which collaboratively drive continuous improvement in food safety management systems around the world with a vision „Safe food for consumers everywhere”. Further, it provides an overview of the impact of the globalization process on sugar industry and applied GFSI approved schemes. The objective of this paper is individual analysis of the GFSI approved schemes (FSSC 22000 - *Food Safety System Certification*, BRC, IFS) with focus on their geographical distribution and the number of users.

Keywords: sugar, molasses, betaine, food safety, GFSI, FSSC 22000, IFS, BRC

GINGER ESSENTIAL OIL IMPROVES QUALITY OF COOKED PORK

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The purpose of this study was to evaluate the effect of ginger essential oil (GEO) on the quality of cooked pork sausages during 30 days of refrigerated storage. Instrumental parameters of color (CIEL*, CIEa* and CIEb*), Thiobarbituric acid-reactive substance (TBARS) values and sensory properties of flavour have been determined. Cooked pork sausages with different concentrations of GEO (0.1, 0.5 and 1.0 µl/g) and control (without GEO) were prepared. All three concentrations of GEO resulted in significantly (P<0.05) increase of CIEa* values. The addition of GEO at 0.5 and 1.0 µL/g affected (P<0.05) to reduction of TBARS values. The flavour of sausages produced with the addition of 0.1 and 0.5 µL/g GEO was slightly/moderately and significantly (P<0.05) different from the control. This study demonstrates that the ginger essential oil could be used as natural antioxidant and colour enhancer in processing of cooked sausages. Further studies should be performed in order to compare its effectiveness in other food products, as well as potential replacement of synthetic food additives.

Keywords: ginger essential oil, cooked pork sausage, quality

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EFFECT OF WILD OREGANO ESSENTIAL OIL ON LIPID OXIDATION IN MARINATED PORK CHOPS

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Lipid oxidation is one of the primary forms of deterioration affecting nutritional and sensory quality of meat and meat products. Cooked meat is more prone to lipid oxidation than raw meat. The use of antioxidants is an effective way to prevent or minimize lipid oxidation in meat products. However, majority of antioxidants added to these products are of synthetic origin. Due to consumers growing interest for products that are clean label, natural antioxidants are increasingly involved in food processing. Plant materials showed to be quite successful in retarding lipid oxidation. The essential oil of wild oregano (*O. minutiflorum*) has antioxidant and antibacterial effects, which can be attributed to its high content of phenolic compounds. Thus, the aim of this work was to evaluate the antioxidative effect of wild oregano essential oil on lipid oxidation in cooked pork chops. Freshly cut porcine meat samples (*M. longissimus thoracis et lumborum*) were divided into two groups regarding marinade composition. Group O was with added wild oregano essential oil (WOEO) at concentration of 30 µl/100 g meat, while group C served as control. Prepared samples were dipped into proper marinade for 20 hours, afterwards they were cooked in convection oven at 163 °C until reached 72 °C in the center of the sample. Lipid oxidation was measured by the concentration of thiobarbituric acid-reactive substances (TBARS) in marinated raw and cooked samples. Sensory evaluation (overall acceptance) was carried out on cooked pork chop samples. The TBARS value for marinated raw samples containing WOEO (0.04 mg MDA/kg) was significantly lower ($P < 0.05$) than that found for the control samples (0.10 mg MDA/kg), with inhibition of about 58.69% compared to control samples. Cooking significantly ($P < 0.05$) induced increase of TBARS in both samples, but in the C samples (1.21 mg MDA/kg) the oxidation was significantly ($P < 0.05$) more intense than in the O samples (0.62 mg MDA/kg). The percent inhibition against lipid oxidation by WOEO for O samples of cooked pork chops was 48.99% comparing to control. The C samples presented a lower sensory score (7.43) for overall acceptance ($P < 0.05$) compared to O samples (8.57). The addition of WOEO showed to be protective against lipid oxidation in raw and cooked pork chops. Moreover, the addition of aforementioned essential oil had no negative impact on overall acceptance.

Keywords: *wild oregano, TBARS, pork chops*

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THE EFFECT OF GENOTYPE (G), GROWING ENVIRONMENT (E), AND THEIR INTERACTION (G x E) ON THE CHEMICAL COMPOSITION OF WHEAT BRAN

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Wheat bran is a rich source of dietary fibres, some of essential fatty acids and contains important quantities of starch, protein, vitamins, minerals and phytic acid. It is also well known for its powerful anticarcinogenic and antioxidant properties. Field experiments with a collection of 23 wheat genotypes were conducted during two growing seasons in order to evaluate the effects of genotype (G), growing environment (E) and their interaction (G x E) on the nutrient composition of wheat bran. The samples of the bran portion of wheat grain were obtained as by-products of milling on the MLU 202 laboratory flour mill. Oil content was obtained by exhaustively extracting samples in a soxhlet apparatus using petroleum ether as the extractant. Tocopherols were determined by using extraction with hexane and normal-phase high-performance liquid chromatography with detection in fluorescent region. The mobile phase consisted a mixture of hexane:ethyl acetate (70:30, v/v). The results have shown that all sources of variation significantly affected chemical composition of wheat bran fraction. Total oil contents of wheat bran varied between 3.03-5.37% in the first and between 2.68-4.96% in the second growing season for investigated genotypes. Gas chromatography analyses of wheat bran oil showed major fatty acids to be linoleic, oleic and linolenic acids with wider range of variation expressed in the second growing season. Total tocopherols content in wheat bran extracts ranged from 16.3 to 77.6 mg/kg in the first and from 28.08 to 107.49 mg/kg in the second growing season. Significant differences in oil content and its chemical composition were found among the genotypes, indicating the great genetic potential for improvement. Also, the effect of the environment on all investigated compounds was highly significant, indicating the possibility to produce wheat with high levels of selected health components and antioxidant activities in a specific growing environment.

Keywords: tocopherols, wheat, wheat bran, HPLC, fluorescence detector

Acknowledgements: This study was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (the project no. TR 31066).



EXPERIMENTAL PRODUCTION OF HEAT-ACID COAGULATING CHEESE WITH ADDITION OF PLUM

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Cheesemaking is one of the most complex technology in dairy and cheese as a product of this art enjoys a special status among dairy products thanks to its high – value ingredients, so every type of cheese has its own story. Facts about rich cheese culture and tradition affect the modern consumer society, occupied by brands and consumers, as result, are no longer satisfied only with good product. So, cheese is ideal product for modern consumers. Upon this the idea of joining cheese and traditional Bosnian product – the prune was born. Prunes are well known as high valuable, nutritively rich food which are consumed as delight for centuries throughout the Balkan region. In this research the experimental preparation of thermally - acidic coagulating cheese with the addition of dry plum is presented with the assumption that this cheese is acceptable for consumers. The cheese was tested for chemical composition and the quality parameters and it is shown that the quality parameter values correspond to heat – acid coagulating cheese type. In order to examine the shelf life of the product cheese was stored for 5 and 10 days and the changes of that samples occurred were noted. After 10 days of storing physical and chemical analysis were repeated with the aim to note differences in composition between fresh and stored product. The conclusion was that chemical composition changed, but very slight. As a fundamental part of the research to determine the acceptability of the product sensory analysis were performed by the expert panel of evaluators and by consumers and the results were statistically processed. The research showed that the experimental product was 95% acceptable by consumers and 100% acceptable by panel of expert evaluators. The cheese cannot be stored for a long time and it is proper to be consumed in a fresh state.

Keywords: technology of cheese, prune, sensory analysis

LEAD CONTENT IN THE MEAT OF SAANEN GOAT MALE KIDS FROM VOJVODINA (NORTHERN SERBIA)

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Lead toxicity is an important environmental disease and its effects on the human body are devastating. There is almost no function in the human body which is not affected by lead toxicity. Lead occurs primarily in the inorganic form in the environment. Human exposure is mainly via food and water, with some via air, dust and soil. In average adult consumers, lead dietary exposure ranges from 0.36 to 1.24, up to 2.43 µg/kg body weight (bw) per day in high consumers in Europe. Exposure of infants ranges from 0.21 to 0.94 µg/kg bw per day and in children from 0.80 to 3.10 (average consumers), up to 5.51 (high consumers) µg/kg bw per day. Lead is absorbed more in children than in adults and accumulates in soft tissues and, over time, in bones. Half-lives of lead in blood and bone are approximately 30 days and 10-30 years, respectively, and excretion is primarily in urine and faeces. Joint Food and Agriculture Organization/World Health Organization Expert Committee on Food Additives established a Provisional Tolerable Weekly Intake of 25 µg/kg bw for lead.

The aim of this study was to investigate the influence of the anatomical location of muscles on their lead content. Lead content was determined in four muscles (*M. psoas major* - PM, *M. longissimus dorsi* - LD, *M. semimembranosus* - SM and *M. triceps brachii* - TB) of Saanen goat male kids (n = 20), using inductively coupled plasma optical emission spectrometry after dry ashing mineralization. Content of lead (mg/kg wet weight) was significantly affected by muscle type. The PM muscles had significantly ($P < 0.001$) higher lead content (0.039 mg/kg), comparing to LD (0.017 mg/kg), SM (0.019 mg/kg) and TB (0.014 mg/kg) muscles. Average lead content in meat from Saanen goat male kids from Vojvodina was 0.022 mg/kg. The maximum lead content found (0.050 mg/kg) in the present study was below maximum level (0.10 mg/kg) set by EU and Serbian legislation. Overall, 100% of meat samples had lead levels below or equal to the half of the maximum level. In conclusion, the results of the present study indicate lead availability in the local agricultural environment in Vojvodina.

Keywords: lead, meat, Saanen goat male kids, Vojvodina

ANTIOXIDANT POTENTIAL AND SENSORY QUALITY OF NEW GREEN WALNUT LIQUEUR

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Alcoholic beverages made from herbs and fruit extracts, that contribute to product functionality, are not only purposed for enjoyment, but also for prevention and treatment of various diseases. The selection of raw material for their preparation is strongly influenced by local tradition and preserved ethnopharmacological knowledge. Green walnut liqueur (GWL) is often served as an aperitif or used as a tonic and digestive aid with beneficial effects against stomach trouble and digestion inconveniences. The high content of phenolic compounds deriving from unripe walnut fruit results is strongly related to strong antioxidant potential, as well as to desirable sensory attributes of GWL, such as astringency and bitterness.

The main aim of this study was to prepare GWL with a unique composition of natural ingredients and to evaluate its total phenolic content (TPC), by Folin-Ciocalteu method and antioxidant potential, by DPPH and FRAP. The GWL was prepared by maceration of unripe walnut fruits, collected in Serbia, chopped into the quarters, together with sugar, honey, dark chocolate, aromatic herbs and spices (vanilla, cinnamon, nutmeg, clove), lemon, orange, cranberry, dried plums and dried figs, in the water-ethanol mixture for 60 days, exposed to the sun. The final liqueur had 25±1,5% vol of alcohol. Furthermore, the sensory quality of GWL was evaluated by the panel of experts, using a model of positive ranking and compared with selected similar commercial spirits, obtained from the local market.

Remarkable values were observed for TPC and AO activity, which were strongly correlated and were significantly higher in comparison to commercial green walnut tincture, used for immune system stimulation, and several herbal bitter spirits popular on the Serbian market. Sensory scores of the analyzed GWL and commercial spirits ranged from 16.43 to 18.66 points (of max. 20), whereas GWL was in the second place in terms of overall sensory quality (18.52 points). Therefore, GWL possessing high AO potential and satisfactory sensory quality may be a promising candidate for the potential commercialization on the Serbian market.

Keywords: Green walnut liqueur, sensory quality, antioxidant potential

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ORGANOCHLORINE PESTICIDES IN RAW MEAT ON THE SERBIAN MARKET

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The aim of this study was to determine the level of organochlorine (OC) pesticides in 30 samples of raw meat collected from supermarkets in Serbia. OC pesticides α -HCH, β -HCH, δ -HCH, dichlorodiphenyltrichloroethane (DDT), DDE, DDD, dieldrin, endosulfane I, endosulfane II, endosulfan sulfate, endrin, endrin ketone, heptachlor, heptachlor epoxide, lindane, aldrin, metoxichlor, cis-chlordane and trans-chlordane were determined using a GC-MS method with QuEChERS method of samples preparation. The highest concentrations ($\mu\text{g kg}^{-1}$, arithmetic means) in raw meat were for δ -HCH (40.6 ± 37.0), p, p'-DDT (35.0 ± 15.2), sum of endosulfans (45 ± 35.2). Measured values of all other pesticides were below the limit of detection of $5 \mu\text{g kg}^{-1}$.

Because of their toxicity and bioaccumulation, levels of OCP contaminants in raw meat is important to monitor as well as to try to find a solution to reduce human exposure and other living organisms to these compounds.

Keywords: organochlorine pesticides, raw meat, GC-MS, QuEChERS



DEVELOPMENT OF NEW IN VITRO COLORIMETRIC MTT ASSAY FOR HIGH-THROUGHPUT SCREENING OF ANTI-YEAST ACTIVITY OF PLANT-DERIVED PRESERVATIVE CANDIDATES IN AN ORANGE JUICE FOOD MODEL

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Yeasts are the usual contaminants in fruit juices and responsible for decreasing the quality and of such products. Preservatives are principally added to fruit juices to enhance their shelf life. However, aside from their advantages, some of the artificial preservatives may possess life-threatening side effects. Plant-derived compounds and their mixtures are perspective materials for the development of new preservative agents. For this reason, the antifungal activity of plant extracts and essential has been studied intensively, however the conventionally used methods are time and material consuming. Therefore, we developed simple in vitro method suitable for high-throughput screening of anti-yeast potential of plant-derived compounds in orange juices using standard 96-well microtiter plates and the tetrazolium-based colorimetric assay - MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide]. Minimum inhibitory concentrations (MICs) of benzoic acid, citric acid, curcumin, gallic acid, potassium sorbate, pterostilbene, sodium metabisulfite, sorbic acid and tannic acid were determined against standards strain of *Saccharomyces cerevisiae*. The most effective agents were pterostilbene and sorbic acid with the lowest MICs 64 and 128 µg/mL, respectively. This novel colorimetric assay for the determination of growth-inhibitory effect of plant compounds in orange juice using MTT proved to be more accurate and timesaving than conventional counting method. This method can be used in research of new eco-preservatives development for shelf life extension in fruit beverages, which could substitute commonly used additives.

Keywords: colorimetric assay, preservative agents, yeast inhibition, plant compounds, antifungal activity

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DISCRIMINANT ANALYSIS OF CHEMICAL AND ANTIOXIDANT PROPERTIES OF *Capsicum annuum* CULTIVATED UNDER DIFFERENT CONDITIONS

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Red pepper (*Capsicum annuum*) is one of the most important vegetables that has wide range of application in food preparation and it is rich in bioactive phytochemicals. The group of 18 samples of red pepper was cultivated and their chemical and antioxidant properties have been analyzed. The total phenols content (TPC), determined according to the Folin-Ciocalteu method, ranged from 414.75 to 718.61 mg/100 g DM, while total flavonoids content (TFC), determined by aluminum chloride colorimetric assay, varied from 258.26 to 561.96 mg/100 g DM. The antioxidant activity was assessed by 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging capacity and ranged from 1.66 to 3.53 mg/mL. In order to discriminate the samples regarding their cultivation conditions (with or without irrigation), the discriminant analysis (DA) has been applied using Minitab 16.1.1 software. The main goal of the discriminant analysis is to define discriminant function. In this study, two groups of red pepper samples have been analyzed: 9 samples cultivated without irrigation (group A) and 9 samples cultivated with irrigation (group B). The statistical performance of the established linear and quadratic functions was verified using cross-validation (CV). The obtained results indicate that there is 78% chance (applying linear function and using CV) and 72% chance (applying quadratic function and using CV) of correct estimation whether a sample of red pepper was cultivated with or without irrigation on the basis of their chemical and antioxidant properties. This indicate a certain correlation between the cultivation conditions and chemical and antioxidant properties of red pepper.

Keywords: *red pepper, discriminant analysis, antioxidant activity, phenols, flavonoids*

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EFFECT OF STARTER CULTURE (*STAPHYLOCOCCUS XYLOSUS*) ADDITION ON COLOUR CHARACTERISTICS OF DRY FERMENTED SAUSAGE

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The quality of food products, particularly meat and meat products, consumers often estimate based on colour. Therefore, the colour formation and colour stability are important indicators of quality of dry fermented sausages. Intense red colour is one of the essential features of *Petrovska klobasa*, which differentiate it from other products of the same type.

The aim of this study was to determine the effect of autochthonous starter culture (*Staphylococcus xylosus*) addition on colour characteristics of dry fermented sausage *Petrovska klobasa* produced in controlled conditions. Two groups of sausages were examined during drying period: K-sausages produced in traditional way without starter culture and S-sausages produced with addition of autochthonous starter culture.

Analysis of colour was performed on the fresh cut of sausages by adequate instrumental method using CIE L*a*b* system detection (lightness-*L**; redness-*a**; yellowness-*b**; hue angle-*h* and chroma-*C**). Additionally, pH value, moisture content and water activity in sausages were measured during the process of smoking, drying and ripening over the course of 0, 6, 12, 30 and 60 day of production. At the end of drying period (60 days of production) sensory colour characteristics of sausages were evaluated using scale from 0 to 5 (0-atypical colour; 5-optimal colour).

Results achieved in this study showed that addition of starter culture (*Staphylococcus xylosus*) to dry fermented sausages significantly ($P < 0.05$) increased redness (*a**) compared to sausages in control group (K-25.23; S-27.94) at the end of drying period. Also, addition of starter culture resulted in better sensory scores ($P < 0.05$) for colour (K-3.9; S-4.3), because sausages in this group had more intensive red colour compared to sausages in control group.

The addition of *Staphylococcus xylosus* had positive effect on redness and sensory characteristics of *Petrovska klobasa* colour.

Keywords: dry fermented sausage, Staphylococcus xylosus, CIE L*a*b* system, sensory characteristic

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THE EFFECT OF FREEZING AND FROZEN STORAGE ON THE CALORIMETRIC PROPERTIES OF LIQUID EGG PRODUCTS

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Eggs are used in the food industry because they have high nutrient value and also coagulating, foaming, emulsifying, colouring and flavouring properties. Processed egg products, such as liquid egg products are popular products in manufacturing. Liquid products have a shelf life of a few weeks, but it can increase up to 1 year by freezing. With this method, a large decrease in the number of microbes can be achieved while at the same time the functional properties are not greatly influenced. Sometimes, we can observe different unwanted phenomena, such as gelling of the yolk and protein denaturation. These processes are highly dependent on the conditions of freezing. Freezing in liquid nitrogen is a very fast freezing method. Freezing medium is boiling at -195.6 °C at atmospheric pressure, when it gets in contact with food material. Because of the large temperature difference between the sample and the cryogenic medium, a frozen layer appears on the food surface, which protects it from drying out.

In our experiment, liquid egg products (liquid whole egg, liquid egg white and liquid egg yolk) were dripped one after another into liquid nitrogen through a strainer. Frozen egg balls were taken out of liquid nitrogen after 60 seconds and then put in a polyethylene foil, sealed with a foil welder. Packages thus prepared were stored in a laboratory freezer at -18 °C for 5 months. The study investigated the effects of frozen storage, calorimetric properties were examined after 1, 2, 3, 4, and 5 months of storage. Samples were removed from the freezer and then thawed in tap water on measurement days. Tests were performed by Setaram MicroDSC III type differential scanning calorimeter, thereafter enthalpy of denaturation and denaturation temperature were evaluated by one-way ANOVA.

The results of the calorimetric studies show that frozen storage also had a significant effect on the denaturation enthalpy of liquid egg samples, as well as freezing, but the rate of changes was smaller. This means that less and less protein can be denatured in the samples during frozen storage. In order to investigate whether the thawing procedures affect this phenomenon, in our next experiment, we will examine different thawing methods.

Keywords: liquid egg, differential scanning calorimetry, freezing, frozen storage, liquid nitrogen

Acknowledgements: Our research is sponsored by the project „Egg white based dairy product analogs with probiotic effects for allergic people and development of low-fat and carbohydrate products for people on a protein-dense diet VALLALATI KFI_16”. This work was supported by Doctoral School of Food Sciences of Szent István University and Capriovus Ltd.

EFFECT OF HYDROCOLLOIDS ON THE RHEOLOGICAL BEHAVIOR OF YEAST RAISED DOUGH

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The economic loss resulting from baked goods' staling encourages research to control or retard staling process. Yeast-raised "bukta" and other yeast-raised or yeast-leavened baked goods, are prized for their soft texture and their fresh yeasty flavor characteristics. These baked goods have a relatively short shelf-life. In the baking industry hydrocolloids are of increasing importance as bread making improvers. They are widely used in baked goods to enhance dough handling properties, overall quality of fresh products, and to extend shelf-life of stored goods

The aim of the present study was to investigate the influence of the addition of selected hydrocolloids (xanthan, locust bean gum, inulin, potato starch, psyllium and arrow root respectively at 0.2, 3, 3, 5, 2 and 10% levels) individually and the combined use of those selected hydrocolloids with MAP (modified atmosphere packaging) stored in refrigerated room 6 °C; on the qualities of bukta and their potential use in retarding the staling process during 3 weeks. Moisture content, water activity and texture parameters of fresh and stored bukta were analyzed.

The results of this study indicated that, there is an increase in yield and softness of bukta crumb using xanthan gum, psyllium and locust bean gum that showed an antistaling effect, retarding the crumb hardening. Potato starch and xanthan gum were also able to reduce the loss of moisture content during bukta storage, reducing the dehydration rate of crumb. Hardness values indicated that the bukta containing xanthan gum up to 0.2% was softer even after 5 days of storage as compared to control under similar conditions and MAP.

To sum up some of the examined hydrocolloids positively affected crumb firmness and proved higher moisture content capacity. According to our results, xanthan gum is the best additive due to both its softening and retarding the firming of the baked goods crumb effects.

Keywords: hydrocolloids, yeast raised dough, shelf life, texture analyses

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EFFECT OF SUGAR REDUCTION AND SUBSTITUTION ON POUND CAKE

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In sweet baked goods one of the main ingredients is sugar. In these products sucrose delivers sweetness, influences the structural and textural properties of samples. Furthermore it may also have an impact on the viscosity, the color and the volume of the products.

The aim of this study was to investigate the effect of sugar reduction and sugar substitution on cake samples. The influence of sugar reduction (by 10 and 20%) and sugar replacement (by xylitol and stevia) on the texture properties, the viscosity and the sensory attribution of pound cakes were studied.

Based on our results both the sugar reduction and the substitution affect the different properties of the investigated samples. As expected the changes in sugar content as well as type influenced the texture of the cake samples. From the textural properties the changes in hardness were mostly pronounced.

In the case of viscosity measurement, the apparent viscosity values increased in proportion with the sugar reduction percentage. Xylitol and stevia also increased the viscosity comparing to the reference sample.

In the sensory tests the 10 and 20% of sugar reduction had a significant effect on the attribution of the samples. However, in the case of sugar replacers the sensory panelists found smaller differences between the control sample and the ones which were made with xylitol and stevia.

As a summary we could conclude that the sugar reduction and substitution have effect on the techno-functional properties of the cake samples. Further research can be done with higher reduction percentages or with other sugar replacers.

Keywords: sugar reduction, sugar substitution, pound cake, texture properties

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THE ANTIOXIDANT ACTIVITY AND COMPOSITION OF THE PHENOLICS FROM NON-GROUND NETTLE (*Urtica dioica* L.) SEEDS EXTRACTED BY DISTILLED AND TAP WATER

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In this study, the content, composition and antioxidant activity of the phenolics extracted from non-ground *Urtica dioica* L. seeds by using the distilled and tap water, were investigated. The content of phenolics was determined by using the spectrophotometric method with Folin-Ciocalteu reagent, and the antioxidant activity was evaluated as 2,2-diphenyl-1-picrylhydrazyl radical-DPPH scavenging capacity. The composition of phenolics was analysed by using two HPLC methods: by isocratic elution process for determining the composition of the phenolic acids, and by gradient elution for determining the composition of some phenolic compounds. The results showed that distilled water was more effective extractant than the tap water. The content of phenolics in the extract obtained by using the distilled water was 4317.71 µg gallic acid equivalent per g of dry seeds and it was 12% higher than the content achieved by using the tap water (3795.24 µg gallic acid equivalent/g). Although the extract obtained by tap water had lower content of phenolics, it had higher antioxidant activity (IC₅₀ was 0.104 mg dry matter of extract per mL) than the extract obtained by distilled water (IC₅₀ was 0.152 mg/mL), indicating that the higher content of compounds with no DPPH scavenging capacity probably were extracted by using the distilled than the tap water. The composition of phenolic acids obtained by isocratic elution showed that six acids were identified in both investigated extracts, and gallic and chlorogenic acid were the most abundant. The composition of phenolic compounds obtained by gradient elution showed that the kaempferol derivatives were the most abundant in both extracts. The results suggest that aqueous extracts of non-ground *Urtica dioica* L. seeds are good source of phenolics and have antioxidant potential for their practical application.

Keywords: Urtica dioica L., seeds, phenolics, antioxidant activity

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APPLICATION OF ARTIFICIAL NEURAL NETWORK FOR OPTIMIZATION OF MICROWAVE-ASSISTED EXTRACTION (MAE) OF DEFATTED WHEAT GERM PHENOLIC ANTIOXIDANTS

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Defatted wheat germ (DWG) is main by-product remaining after the process of the wheat germ oil extraction and represents suitable material for polyphenols isolation. In this work DWG obtained by supercritical CO₂ extraction was further utilized for recovery of polyphenols using microwave-assisted extraction (MAE). The main goal of this study was to evaluate influence of the operating parameters on the investigated responses and to optimize MAE in terms of the total yield (Y), total polyphenols content (TPC) and antioxidant activity (AA) of thus obtained DWG extracts. Input variables were ethanol concentration of solution (50, 70 and 90%), extraction time (5, 10 and 15 min), liquid-solid ratio (5, 15 and 25 mL/g) and irradiation power (400, 600 and 800 W). AA of DWG extracts was evaluated using three in vitro model systems (DPPH, ABTS and FRAP). Adequacy of the models obtained by ANN was suggested by high values of R² for all observed responses (0.9944, 0.9879, 0.9823, 0.9858 and 0.9798 for Y, TPC, DPPH, FRAP and ABTS, respectively). According to the sensitivity analysis operating parameters influenced responses in following order: ethanol concentration>liquid-solid ratio>extraction time>irradiation power. Optimization suggested that, in order to obtain maximum yield of extract with high antioxidant activity and polyphenols content, extraction conditions should be set as follows: ethanol concentration 50%, extraction time 15 min, liquid-solid ratio 25 mL/g and irradiation power 400 W. Predicted responses were: Y 37.71%, TPC 679.94 mg GAE/100 g, DPPH 14.54 μM TE/g, FRAP 11.83 μM Fe²⁺/g and ABTS 38.99 μM TE/g. Additional experiment was conducted in order to validate predicted results and obtained values for responses were: Y 38.37 ± 0.22%, TPC 658.13 ± 41.55 mg GAE/100 g, DPPH 13.82 ± 0.26 μM TE/g, FRAP 13.71 ± 0.38 μM Fe²⁺/g and ABTS 37.17 ± 0.85 μM TE/g. Results of the research suggested that MAE could be successfully employed for polyphenol recovery from DWG and that ANN represents adequate mathematical method for such extraction process modeling and optimization.

Keywords: *defatted wheat germ, microwave-assisted extraction, optimization, artificial neural network*

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RHEOLOGICAL, TEXTURAL AND SENSORY PROPERTIES OF HUMMUS SPREAD DEPENDING ON THE TECHNOLOGICAL PARAMETERS

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Chickpeas (*Cicer arietinum* L.) is one of the oldest legumes that contains carbohydrates, proteins, vegetable oil, cellulose, vitamins of B group, minerals, essential amino acids, carotenoids, flavonoids, phenols and antioxidants. The most commercial use of chickpeas is in the production of an increasingly popular humus spread. In this work, the influences of technological factors on rheological, textural and sensory properties of hummus spreads made by certain recipes were observed, with the aim to achieve the maximum quality of final product. The hummus spreads contain boiled chickpeas 55.3%, water, sesame paste 14%, rapeseed oil 4.9%, spices 1.5%, salt, citric acid, sodium bicarbonate and potassium sorbate. The water phase was in a form of water/ice and in varying amount (24%, 28%, 32% and 36%) and also the frozen boiled chickpeas were used. The way of preparation of raw materials and content of water phase were observed with the aim to remove the specific characteristic of the spreads to crack on the surface after the pasteurization phase and during cooling.

Rheological analyzes of spreads have shown that they are spreadable viscoelastic systems with thixotropic type of flow and relatively same ratio of elastic and viscous components. The increase in water content has greatly affected the textural properties of the spreads and decrease in their values. The functions of optimizing the sensory properties revealed a spread with sensory properties closest to the optimal properties. Also, relatively high correlation between the sensory and instrumentally determined texture parameters was pointed.

It can be concluded that in the production of hummus, frozen cooked chickpeas can be used to increase production efficiency. The amount of aqueous phase in the spread should be 28% in order to achieve adequate quality of the final product after all stages of production. Thus, the nutritionally and biologically very valuable functional food product with the desired sensory properties is obtained.

Keywords: hummus spread, technological parameters, rheology, texture, sensory properties

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SUSCEPTIBILITY OF REFERENCE MICROBIAL STRAINS TO CHOKEBERRY POMACE EXTRACT

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Black chokeberry (*Aronia melanocarpa* L.) has large, purple-black fruits with dry and acid taste, usually used for production of juice and wine, and potent natural food colorant. By-products of plant food processing, represent a major disposal problem for the industry concerned, even though they are promising sources of compounds (vitamins, phenolics, anthocyanins etc.) which may be used for their high nutritional and technological properties. The aim of this study was determination of antimicrobial potential of chokeberry pomace extract. Tested reference strains were bacteria: *Staphylococcus aureus*, *Bacillus cereus*, *Listeria monocytogenes*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella Typhimurium*, yeasts: *Saccharomyces cerevisiae*, *Candida albicans*, and moulds: *Aspergillus brasiliensis* and *Penicillium aurantiogriseum*. Screening of antimicrobial activity was determined by disc diffusion method and agar-well diffusion method. Minimal inhibitory (MIC), minimal bactericidal (MBC) and minimal fungicidal concentration (MFC) were determined by microdilution method. Chokeberry pomace extract showed antibacterial activity against all tested bacteria. Gram-positive bacteria were more susceptible than Gram-negative bacteria, because clear zones around the wells appeared, which indicate on bactericidal activity. MIC were in the range of 1.56-100 mg/ml, and MBC in the range of 3.125->100 mg/ml. The most susceptible strain was *Bacillus cereus* (MBC=3,125 mg/ml). The most resistant strain was *Salmonella Typhimurium*, because MBC was not found in tested concentration range. There was no any activity against yeasts and moulds. It could be concluded that chokeberry pomace extract possess high antibacterial potential, which indicate that waste material is a promising source of bioactive compounds and could be used as functional food ingredient.

Keywords: chokeberry, pomace, antimicrobial activity, MIC, MBC

COMPUTER VISION SYSTEM AS RAPID TOOL FOR VOLUME INSPECTION OF POGÁCSEA

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With increasing consumption of bakery products worldwide, the need for developing smart quality control methods is growing. One of these emerging methods is "computer vision system (CVS)" which has recently gained remarkable attention in the field of quality assessment of food products.

This study was carried out to examine the potential of image processing and assess the changes in volume of pogácsa (a Hungarian yeast-leavened bakery product) during production stages. 8 sample groups were prepared according to a standard industrial recipe, with different temperatures and times for proofing and baking. Fifteen pieces of each group were randomly selected and their volume was quantified using CVS and with the help of Octave software. After acquisition, the volume of the sample was calculated on the basis of the segmented area.

The results showed that the increase in temperature of proofing (from 35 to 45 °C) was associated with significant increase in the volume of pogácsa. However, change of proofing time (from 15 to 25 min) was not found to be significant on the volume of the products. On the contrary to the proofing, increase of baking temperature induced significant reduction in the volume of the final product, whereas, changes in baking time had no effect.

Overall, the results of the present study indicated that image processing technique is a powerful tool and might be considered as a rapid and non-destructive in-line technique for estimating and monitoring volume of pogácsa and to control the proofing and baking process.

Keywords: *computer vision system, volume, baking, proofing*

EVALUATION OF IN VITRO GROWTH-INHIBITORY EFFECT OF SPICE ESSENTIAL OILS AND SUPERCRITICAL CARBON DIOXIDE EXTRACTS ON FOOD PATHOGENIC BACTERIA IN LIQUID AND VAPOR PHASE USING BROTH MICRODILUTION VOLATILIZATION METHOD

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Foodborne microbial diseases still constitute a major health concern and therefore it is necessary to preserve food products in order to prolong their shelf life and safety. There are many ways of food preservation, whereas one of the options is application of volatile natural agents exhibiting antimicrobial activity into modified atmosphere in food packaging. Commonly used spices and condiments containing volatile oils could potentially be used. However, currently used methods for determination of antimicrobial activity in vapor phase are not designed for a high-throughput screening and are not yet verified for development of food industry applications. The antimicrobial activity of essential oils and supercritical carbon dioxide extracts isolated from spice samples *Allium sativum*, *Azadirachta indica*, *Cinnamomum cassia* (bark and flowers), *Cinnamomum verum*, *Elettaria cardamomum* and *Syzygium aromaticum* was evaluated using broth-microdilution volatilization method recently developed by our team (1) against common foodborne pathogens, namely *Bacillus cereus*, *Escherichia coli*, *Listeria monocytogenes* and *Salmonella enterica* Typhimurium. Essential oils were isolated by hydrodistillation using Clevenger-type apparatus and simultaneously, supercritical CO₂ extracts were obtained using Spe-ed SFE Helix system (Applied Separations, Allentown, PA, USA) under constant pressure and temperature. Results have shown that essential oils and supercritical CO₂ extracts from *C. cassia* and *C. verum* were the most effective agents against all tested microorganisms with minimum inhibitory concentration values ranging from 256 to 512 µg/ml in liquid and from 256 to 1024 µg/ml in vapor phase. Obtained results suggest that both *Cinnamomum* spp. could be potentially used in food industry as natural food preservatives. In addition, our findings have shown that broth microdilution volatilization method could be used for development of food applications based on volatile antimicrobial agents such as modified atmosphere packaging.

Keywords: *Essential oils, carbon dioxide extracts, antimicrobial activity, vapor phase, modified atmosphere packaging*

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THE POSSIBILITY OF USING EXTRUDED SUGAR BEET PULP FOR THE PRODUCTION OF COOKIES, FROM THE ASPECT OF MICROBIOLOGICAL PROFILE

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Using by-products is the growing trend in food industry. It may reduce overall waste and improve nutritional value of new products. Sugar beet pulp is a by-product of sugar extraction process. Each year, 120 million tons of beet pulp is produced around the globe. Physical and chemical properties of this by-product suggest its possible utilization as dietary fibers in human nutrition. Due to its high moisture content and chemical composition, this by-product is susceptible to microbiological degradation. The aim of this work was to investigate the possibility of using extruded sugar beet pulp (the ratio of corn grit and sugar beet pulp in extrudate was 70:30 and 85:15) on microbiological profile of cookies. Extruded sugar beet pulp was used for replacement of wheat flour during cookies preparation in the amount of 5, 10 and 15%. Microbiological analysis included determination of aerobic mesophilic bacteria, aerobic mesophilic sporogenic bacteria, Enterobacteriaceae, *E. coli*, *Clostridium* spp., lipolytic bacteria, yeasts and molds. In the sugar beet pulp extrudates total aerobic mesophilic bacteria and aerobic mesophilic sporogenic bacteria count was under 10^5 cfu/g and 10^3 cfu/g, while other microorganisms have not been detected. In cookies, extrudates addition has increased the total aerobic mesophilic bacteria and aerobic mesophilic sporogenic bacteria count, compared to control cookie sample, but that number did not exceed 10^3 cfu/g and 10^2 cfu/g, respectively.

Keywords: by-product utilization, sugar beet pulp, cookies, microbial profile

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QUALITY OF FERMENTED MILK PRODUCTS OBTAINED BY CONCENTRATED KOMBUCHA INOCULUM

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Two different kombucha inoculum: concentrated by microfiltration (15%) and evaporation (3.0%) were applied in fermented milk beverage manufacture. Samples were produced from milk of 0.9% fat content. Rheological properties improvement of low fat fermented milk products by transglutaminase (TG) addition were achieved. Physico-chemical and textural characteristics, as well as sensory properties of kombucha fermented milk products with TG addition were analysed after production. Milk fermentation lasted from 10 to 12 hours. Energy value of fermented milk products was lower in sample produced by microfiltrated inoculum than in samples obtained by concentrated inoculum. Application of transglutaminase in concentration of 0.02% positively effects textural and sensory characteristics of low fat fermented milk products obtained by concentrated kombucha inoculum.

Keywords: fermented milk, concentrated kombucha inoculum, quality

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STABILITY OF CAROTENOIDS IN PUMPKIN DURING THE DIFFERENT HEAT TREATMENTS

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Pumpkin contains high content of carotenoids, the majority of which are alpha-carotene, beta-carotene, alpha-cryptoxanthin, lutein/zeaxanthin and violaxanthin. The main beneficial effect of carotenoids derives from their antioxidant activity, i.e. protecting cells against the harmful effects of free radicals. The aim of this study was to determine the content of total pumpkin carotenoids during cooking, baking in oven and microwave oven to examine how different heat treatments affect the stability of the carotenoids. The standard spectrophotometric method was used to determine the content of total carotenoids, measuring the absorbance of extracted samples at a wavelength of 445 nm. Samples were extracted by petrolether. According to the obtained results, the highest content of carotenoids was in raw pumpkin (172.93 µg/g dry matter), then in an oven baked pumpkin (66.63 µg/g dry matter), then in a microwave oven baked pumpkin (65.97 µg/g dry matter) and the lowest content of total carotenoids was determined in cooked pumpkin (54.42 µg/g dry matter). These results indicated that different heat treatments significantly affected the stability of carotenoids in pumpkin. The higher losses were during cooking, while the losses of baking in the oven and microwave oven were similar.

Keywords: pumpkin, heat treatment, total carotenoids, spectrophotometry

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TECHNOLOGY AND QUALITY OF FRESH CHEESE PRODUCED BY KOMBUCHA INOCULUM

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The effect of kombucha inoculum and traditional culture on fresh cheese quality were investigated in this study. Fresh cheese samples were produced from milk with 2.8% milk fat content. Physico-chemical characteristics and textural properties, as well as sensory characteristics of fresh cheese samples were analyzed after production. Fermentation process was faster in the kombucha fresh cheese sample compared to sample produced by traditional culture. The obtained results showed significant differences in quality among cheeses after production. Kombucha fresh cheese had higher content of dry matter and higher value of textural properties than sample manufactured by traditional culture. Fresh cheese sample manufactured by kombucha showed specific sensory, nutritive and textural characteristics. It could be a new functional dairy product intended for all consumers' categories.

Keywords: fresh cheese, kombucha inoculum, technology, quality

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THE QUALITY OF SUNFLOWER OIL AND PALM OLEIN DURING THE PRODUCTION OF FRENCH FRIES

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The refined sunflower and palm oil are used in the food industry for the production of French fries. Literary data have been shown that the palm oil had less tendency to degradation than sunflower oil, due to its fatty acid composition. However, palm olein is a palm oil fraction and therefore has a different composition of fatty acids. The aim of this study was to investigate quality of the refined palm olein in relation to the refined sunflower oil during the production of French fries. The oil samples were used for multiple frying during seven days (40 minutes per day at a temperature of 165 °C). The peroxide number and free fatty acids content (acid number) were determined by standard analytical methods. The results showed that the peroxide number in sunflower oil and palm olein increased by 75.0% and 77.8%, while the acid number increased by 50.0% and 26.8%, respectively, in relation to their initial values in fresh oil samples. Based on these results it can be concluded that the palm olein was more suitable for frying. However, this finding cannot be indicated with certainty because the quality of the oil depends on many more parameters, not only of analyzed in this paper.

Keywords: sunflower oil, palm olein, peroxide number, acid number

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IMMOBILISATION OF LACTOBACILLUS RHAMNOSUS IN COMBINED POLYVINYL ALCOHOL/CALCIUM ALGINATE MATRIX FOR L-(+)-LACTIC ACID FERMENTATION

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Lactic acid (LA) is the most important hydrocarboxylic acid widely distributed in nature, with wide range of application in food, pharmaceutical, textile, leather, and chemical industries. LA bacteria represent one of the most promising microorganisms for application in extensive bioconversion of various substrates into value added products. There is a considerable scientific and industrial interest in applications of immobilised cells in various food and biofuel-related fermentation processes, due to the numerous advantages they offer compared to free cell processes. Sodium alginate (Na-alginate) is composed of polysaccharide backbone with two repeating monosaccharide units (guluronic acid, and manuronic acid), with hydroxyl and carboxyl groups in its structure that enable further structure modifications. Poly(vinyl alcohol) (PVA) is a biodegradable semi-crystalline synthetic polymer that is water soluble, nontoxic and transparent with good chemical resistance and physical attributes.

In this study immobilisation of *Lactobacillus rhamnosus* ATCC 7469 in poly(vinyl alcohol)/calcium alginate (PVA/Ca-alginate) matrix using “freezing-thawing” technique for application in lactic acid (LA) fermentation was investigated. PVA/Ca-alginate beads were made from sterile and non-sterile PVA and Na-alginate solutions. Obtained PVA/Ca-alginate beads were applied in batch L-(+)-LA fermentations on MRS broth. *L. rhamnosus* cells survived well, rather intense immobilisation procedure and significant cell proliferation was observed in fermentation achieving high cell viability (up to 10.6 log CFU/g) in sterile beads. In LA fermentation, the immobilised biocatalyst was superior to free cell fermentation system (by 37%) achieving the highest LA yield and volumetric productivity of 94.8% and 0.74 g/l h⁻¹, respectively.

Keywords: lactic acid fermentation; polyvinyl alcohol; sodium alginate; immobilisation

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TRITICALE AS POTENTIAL BREWING RAW MATERIAL

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Beer is an alcoholic beverage obtained from water, barley malt, hops, and yeast. The world beer market is extremely competitive, so many breweries have to expand their offers by developing innovative products to meet consumer demand. They also have to produce consistently high quality beer at lower cost. As a result, brewers have increasingly replaced malt with various adjuncts other than barley malt, which are sources of extract, which brings to increased beer variety, sensory modification of the beer and a lower price of beer production. The brewers need to ensure that wort prepared from mixed grists of malt and adjuncts does not diminish the traditionally high quality standards. Triticale is a hybrid of wheat and rye, which can be used as a partial substitute for barley malt. Typically, adjuncts only contribute starch without enzyme activity, but triticale is an exception, because some triticale varieties already contain high levels of amylolytic activity even in the unmalted form. Another great characteristic of triticale is a low gelatinization temperature, which indicates its capability of degrading its own starch with efficiencies equal to those of barley malt.

The objective of this study was to evaluate the possibility of triticale application as partial substitute for barley malt in wort production, based on the triticale analyses and analyses of the produced malts. Content of β -glucan and pentosans during the malting process were also monitored, due to their characteristic to increase wort viscosity. During micromalting, both examined triticale varieties NS Paun and Odisej, had similar β -glucan content in native grain and obtained low values in the malt, while both varieties showed increase in pentosan content during micromalting. α -amylase activity in produced malt was higher in variety NS Paun (329.49U/g) than in variety Odisej. Produced malt showed very high extract content (85.48 and 84.65% dry matter) and diastatic power of 810.43°WK and 682°WK. Overall, our research showed that both triticale varieties had higher malt extract, higher diastatic power, and higher α - and β -amylase activity than typical values of barley malt, which indicated that they could be used as a partial substitute for barley malt in wort production.

Keywords: triticale, malt, micromalting, adjuncts

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VERY HIGH GRAVITY FERMENTATION OF SUGAR BEET THICK JUICE BY *SACCHAROMYCES CEREVISIAE* IMMOBILIZED IN COMBINED ALGINATE-MAIZE STEM TISSUE BEADS

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Bioethanol produced by alcoholic fermentation from raw materials of biological origins is receiving growing worldwide interest as a sustainable and renewable bioenergy source, which represents alternative to fossil fuels. Very high gravity (VHG) fermentation is an attractive option to improve the ethanol production process and enable savings in water, energy and capital costs investments. This study investigates continuous VHG ethanol fermentation of sugar beet thick juice, by *Saccharomyces cerevisiae* entrapped in novel immobilization carrier prepared of Ca-alginate with the addition of grinded maize stem tissue. Continuous bioethanol production from substrate based on sugar beet thick juice with initial sugar concentration of 300 g/L was conducted in a laboratory cylinder glass bioreactor during 15 days at temperature 30 °C and pH 5. Firstly, fermentation was conducted at dilution rate (D) of 0,028 h⁻¹ during five days, next five days it was 0,023⁻¹, and afterwards 0,019⁻¹. At dilution rate of 0.028 h⁻¹, 0.023 h⁻¹ and 0.019 h⁻¹, maximum sugar utilization was 74.46%, 87.23% and 86.54%, while obtained ethanol concentration was 103.6 gL⁻¹, 112.7 gL⁻¹ and 112.4 gL⁻¹, respectively. The volumetric productivity of ethanol ranged from 3.29 gL⁻¹h⁻¹ to 4.66 gL⁻¹h⁻¹. Reduction of the dilution rate from 0.028 h⁻¹ to 0.019 h⁻¹ reduces the supply of sugar and dissolved oxygen into the bioreactor, and consequently the specific ethanol productivity of yeast decreased from 0.17 gg⁻¹h⁻¹ to 0.12 gg⁻¹h⁻¹. The investigated fermentation system was stable and efficient for continuous VHG fermentation during 15 days.

Keywords: *bioethanol, fermentation, immobilized yeast, alginate*

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TEXTURE CHARACTERISTICS OF DRY FERMENTED SAUSAGE AFFECTED BY DRYING PROCESS CONDITIONS

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Petrovska klobasa is a dry-fermented sausage traditionally manufactured in micro-processing plants within village households during winter, undergoing slow drying and ripening processes. In order to meet higher market demands for this type of sausage production should be shift from households to industrial conditions, and in that way the drying rate can be sped up. Processing parameters affect final texture of fermented sausages. As it is one of the most important components of sausage quality it is important to create a product of textural attributes accepted by the consumer.

The aim of this study was to determine the effects of traditional (atmospheric conditions; $t = 2-18$ °C; RH = 62-95% - sample T) and industrial (precisely defined conditions; $t=10$ °C; RH = 84% - sample I) processing conditions on texture characteristics of *Petrovska klobasa*. The sausages were produced from minced lean pork meat, pork fat and seasonings in traditional manner, and were stuffed in collagen casings. Production in traditional conditions lasted for 90, and in industrial for 60 days. Samples for texture analyses were taken after 2, 9, 15, 30, 60 and 90 days, and for sensory analyses after 60 and 90 days of processing. After 2 and 9 days of processing differences in hardness were not significant between groups. Texture parameters were significantly affected by the processing conditions from days 15 and till the end of draying period. In this processing period hardness was significantly higher for sausages processed in industrial conditions. Considering the results for hardness at the end of drying period (moisture content <35%), i.e. 90th day for T sausage, and 60th day for I sausage the difference still was significant, but the values were higher for T sausage. Also, sensory evaluated texture at the end of drying process had significantly higher score for T sausage. According to results obtained for texture characteristic further investigation should be made in order to harmonize the process parameters and final product quality.

Keywords: dry fermented sausage, texture, production conditions

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POYPHENOL PROFILE OF HONEYDEW AND PLOLYFLORAL HONEY FROM NORTHER PART OF MONTENEGRO

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Introduction: Honey is a high energetic sweet food composed, among other components, of bioactive compounds that attribute therapeutic properties of this product. As antioxidants, polyphenols are recognized as the major constituents in honey responsible for its health promoting properties. Bees produce a floral honey from the collected nectar of flowers, and honeydew honey from collected excrement of insects or plants. Considering that Montenegro is a country with traditional beekeeping, this is the first time to publish the results of its honeydew and polyfloral honey samples.

Experimental: Analyzed honey samples, honeydew and polyfloral, were collected from three locations in norther part of Montenegro, Berane, Bijelo Polje and Pljevlja. Polyphenol profile of six honey samples was determined by identifying polyphenols on UHPLC-LTQ OrbiTrap MS, and by their quantifications on UHPLC-DAD MS/MS.

Results: Of the 32 identified polyphenol compounds, 24 were present in all samples. Appearing the other eight polyphenol compounds has no constant diversity between the samples. The similarity between them was confirmed by their quantifications. The highest proportion in total polyphenols has a quercetin with a mean value of 3.82 mg/kg, followed by p-hydroxybenzoic acid and p-coumaric acid. Slightly higher content of chrysin, pinocembrin, p-protocatechuic and vanillic acid can be noticed. The quantifications of the target compounds include 33 polyphenols, and only 16 of them were previously identified. This indicates the occurrence of certain dissimilarities between two used techniques, which are the consequences of similar spectra of certain polyphenols and their derivatives.

Conclusion: This is the detailed study of polyphenols in Montenegrin honeys. Based on the obtained results, it can be seen that the investigated honey samples are very rich in polyphenols, with no significant differences between honeydew and polyfloral samples. Considering small areas that cover these three regions of norther part of Montenegro, it can be assumed that small diversity in plant species causes overlapping in honeybee's pastures and similarity of samples.

Keywords: *honey, honeydew, polyfloral honey, polyphenols*

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ELECTROCHEMICAL STUDY OF VITAMIN B₁ BY MEANS OF ADSORPTIVE STRIPPING CHRONOPOTENTIOMETRY

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The electrochemical behavior of vitamin B₁ was studied by chronopotentiometric stripping analysis, based on the specific adsorptive accumulation as the main effect in a quiescent solution, using the mercury film electrode. The most important experimental parameters of the analysis (type and concentration of the supporting electrolyte, pH, deposition potential, electrolysis time and oxidation current, as well as the thickness of the mercury film electrode), were investigated and optimized. At defined experimental conditions, the analytical signal of vitamin B₁ appeared as a well-defined oxidation wave at about -430 mV vs. Ag/Ag/Cl (3.5 mol/l KCl) electrode, in a citrate buffer solution of pH 6 (0.2 mol/l). Linear response of vitamin B₁ was obtained in the content range of 5 – 50 mg/l. The achieved limit of detection was 1.64 mg/l, whereas the limit of quantitation was 4.97 mg/l. The proposed method was applied for the analyses of the various multivitamin supplements. The accuracy of the developed method was tested and confirmed by the parallel HPLC analyses.

Keywords: vitamin B₁, adsorptive stripping chronopotentiometry, multivitamin supplements

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INSTANT COFFEE ENRICHED WITH DIETARY FIBRE

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Dietary fibre comprises non-digestible carbohydrates including resistant starch and lignin, and is an important constituent of a healthy diet. The aim of the study was to investigate the possibility of enrichment of coffee with soluble dietary fibre and to evaluate sensory acceptance of these coffee drinks by consumers. Panel consisted of 30 students in Ljubljana (Slovenia) and 31 students in Zagreb (Croatia), respectively. The nine-point hedonic scale was used for assessment of eight sensory parameters. Samples were prepared just before the sensory evaluation. Control sample was prepared with 2.0 g of decaffeinated instant coffee and 7.7 g of full fat powdered milk dissolved in 150 mL of hot water. Enriched samples of coffee were prepared in the same way with addition of 1.5 g of Nutriose™ for the first enriched coffee sample and 1.5 g of gum arabic for second enriched coffee sample. To prevent influence of the sample temperature on sensory perception all samples were evaluated at room temperature. Results show no differences in hedonic scores for sensory parameters between control sample of coffee and sample enriched with Nutriose™, while coffee sample enriched with gum arabic received lower hedonic scores by consumers. Our study shows possibility of instant coffee enrichment with soluble dietary fibre, without negative effect on sensory acceptance among consumers. Developed model coffee drink also qualifies for nutrition claim “source of fibre”. Our study demonstrates possibility of product development and enrichment as a mean to produce functional food alternatives to already widely consumed products.

Keywords: dietary fibre, enriched coffee, Nutriose™, gum arabic

EFFECT OF NITROGEN SOURCES ON BIOBACTERICIDE PRODUCTION BY *Bacillus subtilis*

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Bacillus species continue to be dominant antagonists in biocontrol of herbal bacterioses. The *B. subtilis* strains are one of the most important since they produce numerous antibacterial compounds. The yield of desired metabolites can be increased by changing the medium composition. So, the aim of this study was to examine the effects of variation of organic (yeast extract, soybean flour and peptone) and inorganic nitrogen sources (NaNO_2 , KNO_3 and $(\text{NH}_4)_2\text{HPO}_4$) in medium on biobactericide production by *B. subtilis* ATCC 6633. The bioprocess was carried out under aerobic conditions at 28 °C and 150 rpm for 96 h after which samples of cultivation broth, cell-free cultivation broth and thermally treated cell-free cultivation broth were analyzed. The antibacterial activity of samples against *X. campestris* ATCC 13951 was determined *in vitro* by disc-diffusion method. The obtained results indicate that thermally treated samples did not cause positive response and, according to *p*-value of 0.000112, cultivation broth showed significantly higher antibacterial activity compared to cell-free samples. The following statistical analysis pointed that both nitrogen sources have statistically significant effect on biobactericides production in applied conditions whereby the influence of inorganic nitrogen is more pronounced. The results of Duncan's test suggested that the highest biosynthesis of antibacterial compounds is achieved in both media, with or without yeast extract, when NaNO_2 is used as inorganic nitrogen source. The results from this study can be used in further research to improve biobactericide production by *B. subtilis*.

Keywords: *biobactericide production, Bacillus subtilis, nitrogen source, Xanthomonas campestris*

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ANTILISTERIAL EFFECTS OF AUTOCHTHONOUS LACTOCOCCI IN CHEESES FROM ULTRAFILTERED MILK

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Autochthonous strains of lactic acid bacteria isolated from traditional products have a high potential for application, most often as a starter cultures or potential probiotic cultures. However, there are some of them who exhibit antimicrobial effects on pathogens. This aspect of the application of autochthonous lactic acid bacteria (LAB) has not yet been sufficiently explored. The aim of this study is to examine the inhibitory effect of autochthonous LAB, on the growth of *Listeria monocytogenes* in cheese from UF milk. Six variants of cheese were produced with a starter culture CHN11 (Christian Hansen, Denmark). Three variants were inoculated with *L.monocytogenes* ATCC 19111 at concentrations of 10^3 and 10^4 cfug⁻¹, and as protective cultures the autochthonous *Lactococcus lactis* ssp. *cremoris* 565 and *Lactococcus lactis* ssp. *lactis* biovar. *diacetylactis* BG BU1-4 were used, which are individually added to each concentration, so that the two variants remained as control without protective cultures. *L.monocytogenes* were enumerated by the standard method ISO 11290-1, and autochthonous lactococci by dilution method using M17 agar with the addition of 0.5% glucose, during 56 days. Obtained results showed that the number of *L. monocytogenes* at concentration of 10^3 cfug⁻¹ in variants with autochthonous lactococci, decreased to the level of 2 log cfug⁻¹ after 21 days of storage, while in cheese with CHN 11 number remained at the same level. After 56 days, number of *L.monocytogenes* reduced to the level of 1 log cfug⁻¹ in cheeses with autochthonous lactococci, while in cheese with CHN 11, number was slightly higher (1,8 log cfug⁻¹). In all cheese variants at concentration of 10^4 cfug⁻¹ *L.monocytogenes*, the number decreased for 1-1,8 log cfug⁻¹ after 21 days in variants with lactococci, while at the end of storage the number was significantly reduced, to the level of 1 log cfug⁻¹. In variant with CHN 11 the number of *L. monocytogenes* decreased to the level of 2 log cfug⁻¹. It can be concluded that autochthonous lactococci showed significant antilisterial activity in cheeses made from UF milk during 56 days of storage, comparing to commercial starter culture which demonstrate lower inhibitory activity against *L. monocytogenes*.

Keywords: antilisterial effect, autochthonous lactic acid bacteria, cheese from UF milk

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PRODUCTION OF TEXTURIZED ROUND-BEEF SNACK USING FREEZE AND MICROWAVE PRE-DRYING METHODS

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Pretreatments are important for improving quality and saving time in many food processes essentially. In drying process, microbial hazards cannot be easily eliminated because of high moisture content and water activity of food materials and mild drying temperatures. However, with the support of pre-drying methods up to main drying process, the water activity and moisture content are decreased and also losing of valuable compounds such as vitamins, antioxidants can be prevented. Thanks to pre-drying methods throughout the main drying general quality loses are mostly restricted.

In this study, production of texturized meat slices from beef by puff drying combined with freeze-drying and microwave drying as pre-drying steps were investigated. Meat samples were cutted with three different shapes as parallelepiped (3x1x2cm), thick slice (3x1x0.5cm) and thin slice (3x1x0.2cm). Then they were marinated with a solution containing 1.5 % acetic acid and 1% salt to decrease the pH and improve the textural quality of samples. Marinated meat samples were pre-dried with freeze-drying method (0.1mbar, -55 °C) and microwave drying (540W) until reaching of 50% moisture content (wb) prior to texturizing process. Then different shaped and pre-dried products were texturized in the puffdrier (at 100 °C for 10 minutes puffing, at 80 °C for 2, 2.5- or 3-hours vacuum drying depending on the shape). Water activity, moisture content and textural properties of samples were measured. Thin shaped products showed the minimum hardness and maximum crispness value slightly different than thick shaped samples. Water activities and moisture contents of freeze-dried samples were less than microwaved ones. Freeze-drying was found to be successful as pre-drying method to obtain crispy and porous texturized meat samples.

Keywords: *round beef, puff drying, freeze-drying, microwave-drying, textural properties*

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***IN VITRO* BIOLOGICAL ACTIVITY OF OILSEED MEAL PROTEIN HYDROLYSATES**

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Residues of edible oil production, especially oilseed meals, possess a large potential in the biotechnological field. In this context, less than 10% of produced flaxseed and hempseed meals have been processed as raw material, although they are very rich in protein content and can be used for the preparation of protein isolates and hydrolysates. Obtained hydrolysates could be used as a protein source in culture media for optimal cell growth and productivity. Furthermore, proteins as well as other compounds from flaxseed and hempseed meal possess different biological activities with potential biomedical application.

Flaxseed and hempseed cakes obtained by cold pressing during oil production were used as a starting raw material for production of protein hydrolysates by microbial enzymes. The biological potential of hydrolyzed proteins was assessed with the emphasis on its antioxidant and anticancer properties. Antioxidant activity was determined by ORAC method while the effects on cell growth were studied *in vitro* on human tumor cell lines (HeLa and MCF7) using MTS cell proliferation assay. Protective effects of hydrolysates against oxidative stress were assessed by Muse[®] Cell Analyzer while evaluation of cell death was done by flowcytometric analysis using Annexin V & Dead Cell Kit.

Comparison of ORAC values revealed that prepared flaxseed and hempseed protein hydrolysates possess a significantly higher antioxidant capacity than the related protein isolates. This was also confirmed by cell-based assay which showed that hydrolysates could protect cells against induced oxidative stress. When comparing antiproliferative activities of hempseed and flaxseed protein hydrolysates, higher inhibition of cell growth was observed for hempseed protein which was related to cell death by the necrosis. Based on presented results, hempseed and flaxseed hydrolysates have potential for functional food and/or medicinal application.

Keywords: antioxidant activity, cell growth, oilseed meal, protein hydrolysates

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ANTIOXIDATIVE AND ANTICANCER EFFECT OF FLAXSEED PROTEIN HYDROLYSATES

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In recent years, the production of peptides/hydrolysates with potential application in food and nutraceutical industry has gained importance. Protein hydrolysates or bioactive peptides derived from food proteins have been reported to exhibit a wide range of bioactivity including immunomodulatory, anticancer, antihypertensive, antioxidant, osteoprotective and antimicrobial effects. Flax (*Linum usitatissimum*) belonging to family Lineaceae, is a blue flowering annual herb that produces small flat seeds that has emerged as a potential functional food being good source of high quality protein.

The aim of this study was to prepare flaxseed protein hydrolysate from flaxseed protein isolate by three commercial enzymes: Alcalase[®] 2.4L, Neutrase and Protamex. Biological potential of prepared protein hydrolysates was measured by ORAC method, while their effect on cell viability and molecular changes in normal and tumor cells was determined by *in vitro* methods. Prepared flaxseed protein hydrolysates showed strong antioxidant activity, according to measured ORAC values. Higher antioxidant capacity of protein hydrolysates in comparison to non-hydrolysed protein isolate indicates releasing of antioxidant peptides during hydrolysis process. Tested hydrolysates showed cytotoxic activities toward tumor HeLa cells with no effects on normal HaCaT cells, what can be related to its possible antitumor activity. Observed effects of flaxseed protein hydrolysates on tumor cells indicates relationship between cytotoxicity and antioxidant activity, as well as their potential as functional food ingredient in anticancer therapy.

Keywords: animal cell culture, antioxidant activity, antitumor activity, flaxseed, protein hydrolysate

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GREEN BIOREFINERY FOR THE ENHANCEMENT OF BIOACTIVITY OF PECTIN FROM SUGAR BEET EXTRACTION WASTE

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Mostly recognized as polysaccharide with high galacturonic content, pectin is actually one of the most complex cell wall components abundant in dicotyledons. Branched and complex structure of pectin is the source of its physicochemical properties suitable for wide area of application. Even more, pectin can be modified by many different enzymes with the aim to modulate its properties or to produce compounds with functional and nutritional interests.

The main objective of this study was the enhancement of bioactivity of pectin by its enzymatic modification following the principles of green chemistry. The concept of biorefinery was realized by using sugar beet extraction waste as the source of pectin.

Raw material was subjected to acid extraction and pectin was concentrated and partially purified by ultrafiltration. Enzymatic hydrolysis of pectin was performed by commercial endo-polygalacturonase and concentration of liberating reducing sugars and total antioxidant activity were analysed as responses. The parameters regarding concentration of enzyme, substrate and reaction time were evaluated. Products of enzymatic hydrolysis performed under conditions determined to enable the highest responses were fractionated by ultrafiltration and assayed.

Results showed that fraction of pectin higher than 10 kDa enhanced antioxidant activity for approx. 90% in comparison to unmodified pectin while fractions lower than 5 kDa and 1 kDa improved it for more than 70%. Achieved considerable enhancement of total antioxidant capacity of pectin by the action of endo-polygalacturonase indicated great potential of applied concept of green biorefinery for the production of high-value hydrolysates of pectin from sugar beet extraction waste.

Keywords: pectin, endo-polygalacturonase, biorefinery

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PHYSICAL AND NUTRITIONAL PROPERTIES OF EXTRUDED SNACKS WITH BREWER'S PROCESSING BY-PRODUCT ADDITION

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Snacks are defined as light repasts consumed between the meals. Since snack products are mainly made of cereal flours they have poor nutritional value. There is a great interest in improvement of nutritional quality of extruded snack products. Brewer's spent grain is a by-product of brewing industry rich in protein and fibre and its disposal is big environmental problem.

Two blends containing cornmeal and brewer's spent grain (100% - 0% and 80% - 20%) were extruded on a Bühler co-rotating twin-screw extruder (model BTKS-30, Bühler, Uzwil, Switzerland) with L/D ratio of 28:1. Extruding conditions were the same for both samples: screw speed of 800 rpm, feed rate of 35 kg/h, moisture content of the material in the barrel of 25%, and temperature regime 100/120 °C. Physical properties and chemical composition of extrudates were examined.

From nutritional aspect, the sample containing 20% of brewer's spent grain had higher quality, having significantly ($p < 0.05$) higher protein and fat content than control sample (100% of cornmeal). Furthermore, these snack products had significantly ($p < 0.05$) lower bulk density, higher length and lower density of extrudates, while the difference of expansion index between the samples was insignificant ($p > 0.05$). Moreover, addition of brewer's spent grain had significant ($p < 0.05$) impact on texture of samples, i.e. lower ($p < 0.05$) values of hardness and firmness.

Incorporation of brewer's spent grain resulted in snack products with better nutritional and physical characteristics. Thus, it can be concluded that brewer's spent grain represents a promising low-cost ingredient to be used in human nutrition.

Keywords: extrusion, brewer's spent grain, snack products, by-product utilization

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USE OF SAGE EXTRACT FOR GROWTH CONTROL OF *E. coli* IN MINCED PORK

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Sage (*Salvia officinalis*) is a perennial branched herb, known for the healing properties. In addition to its medicinal properties, sage is used in modern research due to its antimicrobial effects. The aim of this work was to monitor the effect of sage extract obtained by supercritical CO₂ extraction to the growth of *E. coli* in minced pork meat. In order to determine the antimicrobial effect of sage extract, minced pork was inoculated with *E. coli* in the initial number of 3.5 log CFU/g, and sage extract was added in concentrations of 0.4 µl/ml, 0.6 µl/ml and 1 µl/ml. The samples were stored at +4 °C temperature for 14 days, and the sampling was carried out in a period of 2 days. The number of *E. coli* was determined at violet red bile dextrose agar plates. Results showed that the sage extract in the concentrations of 0.4 µl/ml and 0.6 µl/ml had a low antimicrobial effect, while the concentration of 1 µl/ml had an effect on reducing the number of *E. coli* by 0.9 log CFU/g. Results also showed that the lowest number of *E. coli* cells was recorded after 8 days of storage. Based on the results, it can be concluded that sage extract obtained by supercritical CO₂ extraction can be used to control growth in minced pork during storage at +4 °C for 8 days.

Keywords: supercritical CO₂ extraction, sage, minced pork, E. coli

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EFFECTS OF DIFFERENT MARINATION PROCESSES ON DRYING OF TURKEY BREAST MEAT

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Pretreatments such as salting, curing, fermentation, smoking or marination can be applied before drying of the meat to increase the shelf life and improve the aroma and texture. An important aspect of marination is the increase of yield of the raw meat, which can provide benefits to the producer and the consumer. Beneficial effects of marination on meat texture also include a juicier texture and enhancing water loss during drying.

In this study, it was primarily based on textural properties of the turkey breast meat samples with marination. Marination process was carried out in three different formulations obtained by mixing the fixed ratios of phosphate, citric acid and acetic acid (1%, w/v) with constant salt concentration (2%, w/w). In this context, moisture content, salt, pH and texture of samples were analyzed in order to investigate the effect of marination solution. In general, it was suggested that the marination process provides more reliable products due to microbiologically away from isoelectric pH. Also, it was noticed that hardness of the product was reduced significantly by marination process. Moreover, in order to investigate the textural effect of the marination process on the dried product, the hot air drying (75 °C, 1.8 m/s air velocity), freeze drying (55 °C, 0.15 mbar vacuum pressure) and microwave drying (540 Watt) process were applied to the samples. It was thought that the marination process was effective on removing the water from the product throughout the drying process by improving the product texture.

Keywords: marination process, freeze drying, hot air drying, microwave drying, turkey breast

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EFFECTS OF MICROWAVE DRYING PRIOR TO PUFF DRYING ON THE PHYSICAL QUALITY CHARACTERISTICS OF DRIED REDUCED- FAT WHITE CHEESE

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Recently, because of the fast-living conditions and busy workdays, snack foods rich in carbohydrate and fat contents are widely consumed instead of main meals. However, due to the growing trend towards healthy eating, consumers have been trying to stay away from such snack products. Cheese is a good food that can be used as a healthy snack because of its high protein content and nutritive properties. Drying is a method used for the production of cheese snacks as well as for the production of other snack foods.

The aim of this study was to investigate the effect of microwave drying as pre-drying method prior to puff drying on the physical quality of reduced-fat white dried cheeses. For the production of dried cheeses by puff drying process, microwave drying (180-360-540 W microwave power) to reduce the moisture content (60-45 %, wb) of cheese samples was applied as pre-drying treatment before the puff drying. After the pre-drying step, the samples were placed into the puff drier (Rapid Gıda Teknolojileri, Izmir, Turkey), heated to 100 °C under approximately 200 kPa pressure and dried for 10 minutes, and then dried under a continuous vacuum of 18 kPa at 80 °C for 1 hour and then dried at 60 °C until reaching the final moisture content. Pre-drying effects on the physical quality of dried cheese samples were investigated by analyzing of pH value, water activity, color properties, weight loss, bulk density, expansion rate, rehydration ratio and textural properties. When the effect of pre-drying conditions was examined, 60 % moisture content and 180 W and 10.5 s process gave the best results in terms of color properties and rehydration ratio.

Keywords: cheese, puff drying, microwave drying, textural properties, expansion rate

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FACTORS THAT INFLUENCE DETECTION OF HUMAN NOROVIRUS IN RASPBERRIES

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Raspberries are very rich sources of bioactive compounds such as phenolics, anthocyanins, organic acids, minerals, etc. As such, the tendency for their growing shows permanent increase worldwide. Nevertheless, raspberries are very often implicated in human norovirus outbreaks and have lately caused several outbreaks in Europe. In the EU RASFF database alert notifications which involve norovirus in fruits and vegetables, raspberries was the fruit most often cited while countries most often cited as the countries of origin were Serbia and Poland. Therefore, the high number of foodborne norovirus outbreaks linked to frozen raspberries indicates essential accurate detection of human norovirus in raspberries. Because viral contamination levels in foods are usually low and the presence of only a few virus particles can lead to disease, highly sensitive detection methods are needed. One of the challenges of virus detection from raspberries is the presence of bioactive compounds that are able to inhibit used molecular assays. The objective of this study was to investigate different methods for detecting human norovirus GI and GII in raspberries by real-time RT-PCR assays with broadly-reactive degenerate MON primers. In scope of this work different volumes and state of samples were tested, as well as level of norovirus inoculation, effectiveness of various methods for their eluting from fruit and virus concentration using polyethylene glycol (PEG). From the 9 different tested methods, the optimized method that enabled improved results was as follows: frozen raspberries thawed to room temperature were spiked with 200 µl of norovirus GI and GII diluted stool samples containing 10⁴ genome copies. After 10 min incubation, 0.05 M glycine/ 0.14 M NaCl pH 9.0 was added and shaken vigorously for 2 min. Samples pulled over sterile cheesecloth into the sterile beaker were centrifuged at 3 500 rpm for 10 min. The top layer was treated with PEG 8000 in two steps, to 6% and to 12%. Final pellet was resuspended in Tris/Tween pH 8.0. RNA extraction was carried out using the TRIzol™ method. Tenfold serial dilution of the RNA extracts was analyzed by real-time RT-PCR kit. Internal amplification and external control were included. All the undiluted extracted RNA samples from raspberries failed to amplify both virus target and IAC by real-time RT-PCR, indicating the strong influence of inhibitors on molecular virus detection. Tenfold dilution of the extracted RNA reduced the level of PCR inhibitors and enabled amplification.

Keywords: raspberry, human norovirus, detection

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PREPARATION OF ZEIN NANOPARTICLES AND SELF-STANDING FILM

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Zein is a plant protein and it is major protein of corn kernel. It is non-toxic, biodegradable and biocompatible natural compound, so it is safe for use in human nutrition. It is water insoluble protein, and it can be dissolved in different organic solvents or binary solvent mixtures, i.e. aqueous ethanol. Using the right methods, zein forms different structures, such as micro- and nanoparticles, continuous films, gels, fibers, etc. In this paper, zein nanoparticles were prepared by antisolvent precipitation of zein from aqueous ethanol solutions, in water. Influence of different parameters on preparation process and prepared nanoparticles was tested. Also, self-standing films were casted from nanoparticle suspensions, with and without plasticizer. Thickness and surface roughness of different films was obtained. Results showed that ethanol share, concentration of zein stock solution and zein solution/water ratio affect the size of prepared nanoparticles. It was also found that concentration of suspensions has little influence on pH of suspension. Further, it was shown that size of nanoparticles and additions of plasticizer do not affect film thickness, but have a big influence on its surface roughness. Overall results showed that properties of zein nanoparticles and self-standing films can be controlled by variation of different preparation parameters.

Keywords: zein, protein, nanoparticles, self-standing films

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THE RELATION OF SHAPE AND MOISTURE CONTENT OF KASHAR CHEESE PIECES WITH PRE-DRYING METHODS FOR THE PRODUCTION OF DRIED CHEESE

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In recent years, consumers tend to have foods with high protein content, but low fat and carbohydrates content. Reduced fat cheeses and derived products, such as dried cheese, are important food groups to meet this demand for the food industry. The shape of the cheese plays a decisive role in the consumer preferences and the effectiveness of the drying process.

In this project, the light fresh kashar cheese pieces, which is pasta-fileta type and unique to Turkey, in the shape of cubes (3×0.1×3 cm) and thin slices (1×1×1 cm) were pre-dried using hot air drying (50 °C, 60 °C and 1.8 m/s air velocity) microwave drying (180 W, 360 W, 540 W microwave power) and freeze-drying (-55 °C and 0.15 mbar air pressure) and then puff dried. The changes in shape and color, and the degree of puffing of cheese samples were evaluated. In addition, the moisture content of cheese samples was determined at regular intervals to evaluate the effectiveness of drying. As microwave power increased in the microwave drying process, the browning of cheese increased. The shape was protected in thin slice samples for all pre-drying processes, while the cube samples did not retain their shape, especially in microwave drying. Moreover, the moisture of thin slice samples moved away faster than that of cube samples for all pre-drying methods. In hot air drying, removal of moisture from cube samples was more difficult because of incrustation on the surface. It was also determined that the moisture of freeze-dried samples was important for protecting the shape and obtaining the desired specifications such as porous and crispy structure during puff drying, especially in cube samples.

As a conclusion, the initial moisture content and the shape of cheese pieces are effective for the final product depending on the used pre-drying method.

Keywords: *dried cheese, explosive-puff drying, pasta-filata cheese, freeze drying, high protein snack*

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EFFECTS OF EXPLOSIVE PUFF DRYING AND FREEZE DRYING ON QUALITY CHARACTERISTICS OF HIGH NUTRITIVE PUMPKIN CHIPS

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There has been a tendency towards the products with high nutritional properties, with the increase of healthy consumption awareness of people throughout the world. It is well known that pumpkin is an important source of dietary fiber, β -carotene, lutein, vitamins A, B6, K and C and minerals such as potassium, phosphorus, magnesium, iron and selenium and phenolic compounds. However, due to high moisture content and water activity there has been seeing quality losses in the pumpkin during storage. Drying of pumpkin improves the quality characteristics and increases the storage time. Generally, dried pumpkin can be used in pasta formulations, cakes, bread doughs or in yoghurts as powder form.

In this study, the pumpkin was brought into the form of snack food with freeze-drying and puff drying processes and the effect of different drying processes on the quality aspects of the pumpkin snacks has been compared. In freeze drying process, pumpkin slices ($3 \times 2 \times 0.3 \text{ cm}^3$) which were frozen at -18°C in deep freezer (Blomberg A⁺) were dried in the freeze dryer (Telstar lyoquest -55 plus eco). The parameters of freeze-drying for vacuum pressure and condenser temperature were set at 0.015mbar and -55°C , respectively. For the production of pumpkin snacks by puff drying process, pre-drying treatment was applied by freeze-drying (0,015mbar, -55°C) to reduce the moisture content of 45% (wb) in 1.5 hours. After pre-drying step, the pumpkin slices were placed into the puff drier (Rapid Gıda Teknolojileri, Izmir, Turkey) and heated to 100°C under approximately 200 kPa pressure and dried for 10 minutes, and then dried under a continuous vacuum of 18 kPa at 70°C for 3 hours until reaching the final moisture content of 4% (wb). Effects of freeze-drying and puff drying processes on the quality of pumpkin snacks were investigated by determining the moisture content, water activity, antioxidant activity, color properties, expansion rate, textural and sensory properties. Compared with only freeze-dried and freeze and puff dried pumpkin snacks, the best textural properties and the lowest color differences were obtained for the samples with freeze and puff dried.

Keywords: pumpkin, freeze drying, puff drying, quality, snack foods

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ENCAPSULATION EFFICIENCY OF CARVACROL IN ZEIN/ROSIN NANOPARTICLES

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This paper investigates the potential of zein/rosin nanoparticles as carriers for water-insoluble carvacrol oil. Zein is a major corn protein. It is inexpensive, abundant, biodegradable, and biocompatible. Rosin is one of the natural gums obtained as a resinous constituent of the oleoresin exuded from various species of pine. Both carriers are natural and suitable for nanoparticles formation. Zein/rosin nanoparticles with different carrier compositions were synthesized by liquid-liquid dispersion and carvacrol was encapsulated by antisolvent precipitation. Carvacrol is an essential oil used as a volatile oil component, as flavouring agent and antimicrobial agent. Its encapsulation efficiency is determined by a HPLC method developed for this compound. The HPLC system consisted of Zorbax eclipse plus C₁₈ Rapid resolution column, 4.6×100mm, 3.5µm, whereas the mobile phase used was a binary mixture methanol and water on 25 °C. The chromatogram was recorded at 275nm and standard curve was developed using a 0.01-1 mg/mL solution of carvacrol. Different ratios of the carriers were used. It was found that nanoparticles with 80:20 zein/rosin ratio had a better possibility of encapsulation than those with 60:40 ratio.

Keywords: *nanoparticles, encapsulation efficiency, High performance liquid chromatography, carvacrol*

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THE INFLUENCE OF THE FREEZING RATE ON THE PHYSICO-CHEMICAL PROPERTIES OF PORK MEAT (*M. Longissimus dorsi*)

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The meat freezing, as a method of preservation, aims to maximize the storage period with preserving the quality and nutritional value of the product. However, freezing causes certain changes in the foodstuff. These changes are the result of ice formation and lead to a change in meat quality after thawing. The aim of this paper was to examine the effect of freezing rates on the physico-chemical properties of pork meat (*M. Longissimus dorsi*). The meat samples were frozen at 10 different rates and after 24 hours of storage at -18 °C, thawed at +4 °C. Samples frozen at the lowest freezing rate (0.23 cm/h) had the lowest water content (72.02%), while for the samples frozen at the highest rate (1.43 cm/h), significantly higher ($p < 0.05$) water content recorded (73.85%). The protein content ranged from 23.34% (rate 0.50 cm/h) to 23.76% (rate 1.43 cm/h). The increase in freezing rates from 0.23 cm/h to 1.43 cm/h statistically significant ($p < 0.05$) affects the increase in the pH value (from 5.41 to 5.72). By measuring water holding capacity (WHC) at least amount of released liquid (2.27 cm² and 2.23 cm²) were recorded at higher freezing rates (1.00 cm/h and 1.43 cm/h, respectively), while in pork samples frozen at slow rate (0.23 cm/h), the highest mass loss after thawing is determined (3.61%).

Keywords: pork meat, freezing rate, physico-chemical properties

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RHEOLOGICAL PROPERTIES OF VEGETABLE CREAMS INCLUDING DIFFERENT TEXTURIZING AGENTS

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During the last decade urbanization, industrialization, technology development, economic development, and market globalization have led to rapid changes in diet and lifestyle. Consumption of sufficient amounts of fruits and vegetables is recommended as part of a healthy diet. Vegetables are typically lower-calorie, nutrient dense foods, and are considered to be vital elements of healthy diets. In general, improving fruit and vegetable consumption among children and adolescents is an important public health issue. Spreadable vegetable cream is a good possibility to increase vegetable consumption day-by-day.

During this work vegetable creams were made using different texturizing agents, as inulin, rice flour and starch. Inulin and oligofructose belong to a class of carbohydrates known as fructans. Inulin is used for a variety of purposes, including as a replacement for fat and sugar a low-caloric bulking agent and a texturizing agent. Rice flour is an important commercial commodity today which is fairly widely traded and used for preparation of various products, especially in industrialized countries. Starch is the most common carbohydrate in the human diet, it can be used for improving texture.

Since texture is one of the most important determining factors for the acceptability of food several rheological methods are developed for direct evaluation of food texture with high reproducibility. Spreadable vegetable creams were tested by oscillatory tests using amplitude sweep method (Anton Paar, Physica MCR 51 rheometer). Oscillatory techniques are useful for determining both the viscous and elastic properties (storage and loss modulus, (G'_{0} ; G''_{0}) respectively) of viscoelastic materials (ie. gels, semi-solid and solid materials) and can be used for testing several emulsion or gel food product. Amplitude sweeps are useful in practice to describe the behavior of different gels, pastas etc.

Based on the rheograms the following parameters were determined: G'_{0} , (Pa): initial storage modulus; G''_{0} (Pa): initial loss modulus; DF: damping factor, the ratio of initial loss modulus to initial storage modulus; τ_{LVE} (Pa): shear stress at the end of linear viscoelastic range which is the strain at storage modulus decrease to 95% of its initial value; CO (Pa): shear stress at crossover point of G' and G'' curves which indicates the turning point between viscoelastic solid and liquid.

Vegetable cream samples containing different texturizing agents (inulin, rice flour and potato starch) had different initial G' and G'' values indicating different gel forming properties compared to each other. Organoleptic texture properties of vegetable creams prepared with rice flour proved to be slightly better than that of the inulin and starch. Based on our results, rice flour seems to be suitable to use as a natural additive in spreadable vegetable creams.



Keywords: *rheology, vegetable, texture*

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CREATION OF INNOVATIVE MEAT PRODUCTS BY RE-USE OF SPENT BARLEY

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The fermentation industry, especially related to beer production, in Poland is a significant producer of side stream materials, including mashed malt, used hop, spent grains, germ flour and yeast. These by-products are valuable biological materials (still containing large amount of proteins, lipids, minerals) and can be used in other types of industries, increasing innovation and the profitability of production, as well as reducing the negative impact on the environment. The aim of the project covered valorization of the mashed spent barley grains collected after brewery processing and its further utilization for creating innovative meat products with improved functionality and preserved or enhanced sensory qualities. Brewing spent grains, i.e. insoluble parts of malt remaining after mashing and filtration of beer worth, collected from a craft brewery, after grinding and drying, were characterized by high content of dry matter (over 95%), proteins (17.8% - 22.9%), fibers (11.3% - 15.9%) and 570-1830 mg/kg of potassium and 50-100 mg/kg of sodium, a high content of Ca and Mg and the presence of Zn, Fe, Mn and Cu. Model homogenized sausages and pâtés were prepared from poultry or pork meat and pork fat with the addition of 1.5%, 2.5%, 3.5%, 5.0%, 6.5%, 7.5% and 10.0% of dried brewery's spent barley. The addition of spent barley had a positive effect on the inhibition of lipids oxidation (TBARS), both in sausages and pâtés. The used additive material did not deteriorate the sensory quality of sausages and poultry pâtés. The addition of more than 2.5% increased the hardness, chewiness and gumminess of the products and worsened the objectively measured (Konica Minolta, Lab) color (the product is darker in the gray-brown shade). It is reasonable to introduce spent barley to the production of meat products in an amount not exceeding 2.5%. The brewery by-products like spent barley can be an alternative source of protein, fat and fiber in meat processing.

Keywords: spent barley, meat products

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OIL AND PROTEIN CONTENTS IN RAPESEED (*BRASSICA NAPUS* L.) AS A FUNCTION OF ENVIRONMENTAL CONDITIONS DURING SEED FILLING PERIOD

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Rapeseed (*Brassica napus* L.) is the third most important oilseed crop in the world, after palm and soybean, and is an excellent raw material for edible oil and biodiesel production. It is one of the most important plants of *Brassicaceae* family which its seed contains more than 40% oil and its meal is rich in protein. Oil belongs to a group of semi-drying oil with iodine value 95-120. It is used in nutrition and for technical purposes. Meal from rapeseed after oil extraction provides a protein rich animal feed. Many studies reported that the quality of rapeseed greatly influenced by the water stress occurred from flowering up to the ripening of seeds and under water stress condition the varieties that are capable of maintaining much more water would have more seed yield and consequently more oil content. In this study we used a multi-year results (2007-2018) the quality of rapeseed as a raw material for the further processing, obtained from the company Victoriaoil, Šid. In order to determine the association between seasons and meteorological indicators, precipitation, temperature (minimum, maximum and mean) were analyzed during each growing season. Based on these data for each meteorological parameter were calculated three variables corresponding to the months of the vegetation period of the rapeseed. PCA analysis was done on a set of meteorological indicators and the content of oil and protein in grain, after which a biplot from the first two PCA axes was constructed in order to visualize the association between the season, grain quality and meteorological indicators. According biplot, there was a close relationship between oil content and precipitation during the month of April. Also, the precipitation in May is in a positive association with the oil content, while the rainfall in June was not related to this trait. On the other hand, there was no clear association between the temperatures in May and the protein content, which is probably the result of low temperature variations in May during the four studied years. Then again, the association of protein content with meteorological indicators was analyzed during the last 4 years (2014-2017), with the first PCA axis explaining 45.82% and the other PCA axis 40.93% of the variability. Weather conditions had a significant impact on the content of oil and protein in grain during the growing rapeseed season, especially temperature and precipitation.

Keywords: oil and protein content, rapeseed, environmental conditions

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PRODUCTION AND EXTRACTION OF ANTIMICROBIAL COMPOUNDS EFFECTIVE AGAINST PHYTOPATHOGENIC *Xanthomonas* spp.

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Biological control of plant diseases has been an acceptable alternative in plant disease management in recent years, in comparison to standard techniques requiring high amount of chemical pesticides. In this study, *Bacillus velezensis*, previously identified using 16S rRNA sequencing, has been employed as the producing microorganism for biosynthesis of antimicrobial compounds effective against two phytopathogenic isolates: *Xanthomonas campestris* Mn 7-2, isolated from cabbage with black rot symptoms, and *Xanthomonas euvesicatoria* PAP LIST 1, isolated from pepper with symptoms of bacterial spot. Production of antimicrobial compounds was carried out in a laboratory-scale bioreactor with working volume of 2 L using previously optimized cultivation medium based on raw glycerol from biodiesel production containing 10 g/L of glycerol, 3 g/L of K₂HPO₄ and 0.3 g/L of MgSO₄·7H₂O. Cultivation parameters in a laboratory-scale bioreactor were: temperature 28 °C, agitation rate 250 rpm and aeration rate 1 vvm. After 96 hours of cultivation, cultivation broth was centrifuged (10000 rpm, 10 min) to separate supernatant from biomass of producing microorganism. Antimicrobial compounds were extracted from supernatant using three folds higher volume of several organic solvents: methanol, isopropanol, butanol, ethyl acetate and chloroform. Crude extracts were evaporated using rotary vacuum evaporator to remove organic solvents. The resulting extracted antimicrobial compounds were diluted in sterile distilled water (5 mL) and used for antimicrobial activity testing by diffusion-disc method. Antimicrobial activity of the supernatant was also tested. Negative controls were pure solvents used for extraction of antimicrobial compounds. The results of this study have showed the ability of *Bacillus velezensis* to synthesize value-added antimicrobial compounds effective against the tested black rot and bacterial spot causers using the raw glycerol – the major byproduct of biodiesel industry. The best results were obtained using methanol and ethyl acetate for extraction of antimicrobial compounds with inhibition zone diameters of 25.15 mm and 31.65 mm, respectively, whereas these two solvents haven't showed antimicrobial activity when tested as negative controls. Considering the inhibition zone diameter of 14.67 mm obtained during testing of supernatant's antimicrobial activity, the extraction and concentrating of antimicrobial compounds using the proposed protocol represents a simple method for separation of antimicrobial compounds intended for utilization in agricultural practice.

Keywords: *Bacillus velezensis*, *Xanthomonas* spp., bioreactor, raw glycerol, antimicrobial compounds

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SENSORY AND SOME CHEMICAL CHARACTERISTICS OF OLIVE OILS PRODUCED IN LYBIA

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Growing olive trees (*Olea europaea* L.) is a widespread activity throughout the Mediterranean Basin, as well as other regions. Olive oil is produced from fresh and healthy olive fruits by cold pressing or by other physical processes under low thermal conditions that do not lead to alteration in the oil. Consumption of virgin olive oil is highly recommended due to its great health benefits which have been known since antiquity. Libya is one of the olive oil producers in North Africa. Using olive oil in Libya is part of the culture and a large number of individual manufacturers produce oil applying the process of cold pressing.

The aim of this work was to determine the sensory profile and some chemical characteristics, as: free fatty acids, peroxide value, total phenols content, iodine value and induction period of olive oils obtained from *Roghiani*, *Hammudi* and *Endori* olive cultivars in five different regions of northern Libya in the crop year 2015. Sensory characteristics were evaluated by panel of five skilled members, while chemical parameters were determined applying standard methods. The content of total phenolic compounds was estimated according to the Folin–Ciocalteu spectrophotometric method.

Obtained results have shown that cold pressed olive oil samples were characterized by different sensory profile and chemical characteristics, due to peculiar effect of growing areas of olive fruits. On the basis of investigated attributes, some oils were of excellent quality, but other were under. The free fatty acid contents varied between 0.73 to 2,78 % of oleic acid, peroxide values ranged from 6.40 to 13.65 mmol/kg and induction period from 4.89 to 18.33 h. The content of total phenols, as especially important compound for health benefit of olive oil, was significantly various also, from 56.0 to 238.3 mg GAE/kg. It can be concluded that the oil from Gharyan production region was quite different from the other Libyans' olive oils. This oil had the best sensory profile, basic chemical quality, oxidative stability and the highest phenolic content.

Keywords: olive oil, sensory quality, acidity, peroxide value, oxidative stability



SIMULTANEOUS ULTRASONICATION AND ENZYMATIC SACCHARIFICATION FOR FERMENTABLE SUGAR PRODUCTION USING *CHLORELLA VULGARIS*

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Identification and bioconversion of new available biomass are advantageous to enhance economic and environmental benefits. From the biotechnological point of view, microalgae are potential candidate of renewable and sustainable biomass resources for production of bioethanol and enzymes due to their high levels of carbohydrates and low levels of lignin. The main challenge of bioconversion of biomass is to select suitable pretreatment process. However, there still remains significant room for improving the production of fermentable sugars from biomass in a cost-effective way that raises enzymatic saccharification efficiency. To best of our knowledge no work has been reported on usage of ultrasonication as an extraction technique for the bioconversion of microalgae. The objective of the present study was to determine the effect of simultaneous ultrasonication and enzymatic saccharification (SUES) on production of fermentable sugars. The ultrasonication pretreatments were carried out in a glass bottle containing 0.3 g of dry microalgae in 87 ml of dilute acid solution. Based on preliminary experiments, the ranges in the pretreatment were selected for the variables of sulphuric acid (H₂SO₄) concentration (1, 2 and 3% (v/v)), amplitude (70, 80 and 90), and pretreatment time (15, 22.5 and 30 min). Then, enzymatic saccharification (cellulase, amylase and Viscozyme L at 50°C and 120 rpm for specific time intervals (24, 48 and 72 h) were employed. Amplitude and acid concentration had significant effects ($p < 0.05$) on fermentable sugars production. The total fermentable sugar production (TFSP) from various pretreated samples ranged at 442.90-593.21 mg/g dry weight microalgae. The highest TFSP (593.21 mg/g) was obtained from simultaneous ultrasonication and enzymatic saccharification of microalgae at a 80% amplitude, 1% H₂SO₄ and 30 min after 24 h. Thus, the SUES process appeared to be a successful alternative method when compared to the conventional pretreatment methods.

Keywords: fermentable sugar, ultrasonication, optimization, enzymatic saccharification

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SEED SURFACE DISINFECTION METHODS: CURRENT APPLICATIONS AND NEW DIRECTIONS

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Agricultural pesticide use in the world has made it possible for farmers to produce a great volume of food. Chemical pesticides (herbicides, insecticides, fungicides, rodenticides, etc.) have allowed farmers to reduce human labor costs in production, and remain competitive in an increasingly global marketplace. However, this success in productivity has often been to the detriment of wildlife and the environment in many countries of the world. Many areas, for example, have experienced pesticide contamination of surface and ground waters.

In addition to environmental problems, human health problems also arise from agricultural pesticide usage. Farmworkers and farmers have experienced chronic, long-term health problems from exposure to agricultural chemicals, and there are been numerous cases of acute, or emergency health problems resulting from pesticide exposure. Additionally, consumers have long been concerned about the presence of pesticide residues in their foods. Thus, reducing the use of pesticides is primarily a social problem.

Different approaches such as hot water treatment, ionizing radiation, ozone, and cold plasma were used for surface disinfection of different seeds with regards to seed vigour. Effectiveness of each treatment varied depending on the disease agent and the type of the seed. Recent efforts were made with pulsed electric field (PEF) treatment of vegetables as well as cereal seeds. Constructed PEF system with parallel plate electrodes with different frequencies provided inactivation of fungi and bacteria. While reasonable amount of inactivation was obtained in endogenous microbial load, seeds germination was faster for PEF treated winter barley with better and stronger root development. Inactivation studies were more deepened with red cabbage seeds revealed that germination rate was better with PEF treatment. It was seen that both bench and pilot scale are viable option for surface disinfection of seed surface.

Keywords: pulsed electric fields, wheat, seed, seed germination, vegetable seeds

BACTERIAL ADHESION ON KITCHEN SURFACES

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The bacterial contamination of food contact materials is a common reason for foodborne disease outbreaks. Understanding the relationship between surface conditions and microbial adhesion, strategies can be developed to inhibit the attachment of bacteria and spores on the kitchen surface. Among the different parameters characterizing a surface, its roughness and its surface free energy are believed to play also an important role in its hygienic behaviour. In our study, the surfaces of commonly used kitchen materials were characterized and tested for the bacterial adhesion. The bacterial adherence on kitchen surfaces was determined by scanning electron microscopy and spectrophotometric method. The results showed that bacterial adhesion and therefore the environmental persistence is not only strain-dependent but is also greatly conditioned by the surface itself. The experimental results can be linked to the food safety issues and quality of the food item in the food supply chain.

Keywords: bacterial adhesion, *E. coli*, *P. aeruginosa*, *C. jejuni*, kitchen surfaces

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MICROWAVE-ASSISTED DILUTE ACID PRETREATMENT FOR IMPROVING PRESSURIZED FERMENTABLE SUGAR EXTRACTION FROM HAZELNUT SHELLS

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Identification and bioconversion of new locally available agro-wastes are advantageous for both economic and environmental benefits. From the biotechnological point of view, bioconversion of agro-wastes to fuels, food enzymes and chemicals is a vital step. Conventional pretreatment using acid or alkali at high temperature and pressure is limited due to high energy input. So there is a need for alternative heating techniques which not only reduce the energy input but increases the total process efficiency. Microwave (MW) pretreatment may be a good alternative as it can reduce the pretreatment time at a higher temperature. This study aimed to determine and optimize the effect of microwave-assisted treatment with dilute acid and enzymatic saccharification on the production of fermentable sugars from hazelnut shells. Pretreatment was carried out at three different levels each thus: dilute acid concentration (X_1 ; 0.5, 1.25, and 2.0%, v/v), pressure (X_2 ; 5, 10 and 15 bar), temperature (X_3 ; 120,140 and 160 °C) and pretreatment time (X_4 ; 5, 15 and 25 min) in order to obtain hydrolysates used for the fermentable sugar extraction (FSE). H_2SO_4 concentration (X_1), pressure (X_2), temperature (X_3) and pretreatment time (X_4) showed significant effects ($p < 0.05$) on FSE from the MW assisted dilute acid process. An optimal fermentable sugar extraction was found as 317.9 mg/g pretreated biomass with %1.2 of acid concentration, 8 bar at 160 °C for 25 min using Box-Behnken response surface methodology. During operational conditions, severity parameter was also calculated. It was ranged from 1.35 to 3.13. The MW-assisted processing appeared to be a successful method when compared to the conventional pretreatment method.

Keywords: lignocellulosic waste, microwave, pretreatment, reducing sugar, severity parameter

FATTY ACIDS PROFILE OF RED RASPBERRY (*RUBUS IDAEUS* L.) SEED OIL: OPTIMIZATION OF SUPERCRITICAL FLUID EXTRACTION

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Red raspberry (*Rubus idaeus* L.) seeds are by-products in food industry, and also a potential valuable source of nutrients. It is consisted mainly of dietetic fibers (66%), lipids (18%), proteins (7%), and carbohydrates (3.5%). Raspberry seed oil is rich in tocopherols (290 mg/100 g), with γ -tocopherol content up to 180 mg/100 g. Also, it contains more than 80% of polyunsaturated fatty acids (PUFA), having more than 30% of essential ω -3 fatty acid. The aim of this work was optimization of supercritical fluid extraction (SFE) of raspberry seed oil in order to maximize extraction yield of ω -3 and ω -6 fatty acids. In order to optimize process, three levels and three variables Box-Behnken experimental design in combination with response surface methodology was applied. Independent variables were pressure (250-350 bar), temperature (40-60 °C) and CO₂ flow rate (0.2-0.4 kg/h), while ω -3, ω -6 fatty acids extraction yield and PUFA content were response variables. Experimental results were fitted to a second-order polynomial model with multiple regression, while analysis of variance (ANOVA) was employed in order to assess model fitness and determine optimal conditions. The range of experimentally obtained ω -3 yield was from 2.32 to 6.37 g fatty acid/100 g of seeds, while ω -6 yield was from 3.37 to 9.03 g fatty acid/100 g of seeds. The optimal conditions due to highest yield, total PUFA content and yields of ω -3 and ω -6 fatty acids were at pressure of 350 bar, temperature of 50 °C, and solvent flow rate at 0.4 kg CO₂/h. Total PUFA content in raspberry oil was relatively high and in the range from 83.04 to 84.56%. Therefore, raspberry seed oil could be used as alternative source of essential ω -3 fatty acid which has proven health benefits in preventing heart disease, cancer, hypertension, and autoimmune disorders.

Keywords: raspberry seeds, oil, fatty acids, supercritical fluid extraction, optimization

Acknowledgements: This work is the result of the research within the project III 46005, financed by Ministry of Education, Science and Technological Development, Republic of Serbia.

COMBINATIONS OF HIGH PRESSURE PROCESSING AND HEAT TREATMENT: SAFETY AND PROTEIN STRUCTURE OF LIQUID EGG WHITE

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High hydrostatic pressure (HHP) technology is one of the most promising nonthermal processes for food processing. Several food products like sliced ham, or fruit juices are some of the products currently available on the market processed by HHP. In our study the different combinations of HHP and heat treatments were examined.

Samples prepared from homogenized liquid egg white (LEW) were treated in different combinations of high hydrostatic pressure (HHP) and heat treatments. Single HHP treatments at 350 MPa and 450 MPa, 5 min or combined treatments: first: pasteurization: 57 °C, 7 min, or long-term heat treatment: 53 °C, 6 hours followed by HHP treatment: 350 MPa, 5 min. were applied.

Aerobic mesophilic microorganisms were enumerated immediately after treatments. Protein structure of LEW was examined by differential scanning calorimetry (DSC, MicroDSC III).

Results showed that single HHP treatments were effective in reducing initial microbial counts, but heat treatment combined with HHP showed higher effectivity. Combined treatments gave satisfying results in aspects of food safety. DSC method pointed out that infinitesimal reducing of all denaturatable protein can be found in case of every treatment. However, the quantity of nondenaturated ovalbumin decreased.

Our results pointed out that required food safety aspects can be fulfilled with minimal changes of protein structure by application of HHP and mild heat treatments.

Keywords: egg white, HHP, minimal processing, DSC, food safety

Acknowledgements: *Our research is sponsored by the projects „Egg white based dairy product analogs with probiotic effects for allergic people and development of low-fat and carbohydrate products for people on a protein-dense diet” and „ EFOP-3.6.3-VEKOP-16-2017-00005”. We are very thankful for that.*

MOLECULAR IDENTIFICATION OF AFLATOXIN BIOSYNTHESIS GENES IN *ASPERGILLUS* SPECIES

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Molecular methods have been widely applied in the identification of *Aspergillus* species. *Aspergillus* species are one of the most important producers of aflatoxin that can contaminate wide range of agricultural and food commodities at any stage of production. The most important species of *Aspergillus* able to produce the aflatoxins are *Aspergillus flavus* and *Aspergillus parasiticus*.

One of the goals of this study was to characterise and identify the isolates of 6 standard *Aspergillus* species. *Aspergillus* isolates were identified in the level of species by using morphological and molecular methods. PCR products obtained after amplification with primers pairs (ITS1/ITS4 and Bt2a/Bt2b), allowed us to identify *Aspergillus* species including *Aspergillus flavus*, *A. ochraceus*, *A. nidulans*, *A. versicolor*, *A. candidas* and *A. tamari*.

Aspergillus species identified in this study were further analyzed for the presence of two regulatory (*aflS* and *aflR*) and two structural (*aflD* and *aflQ*) genes involved in aflatoxin biosynthesis pathway. Genomic DNAs from all tested *Aspergillus* species were extracted. The presence of *aflS*, *aflR*, *aflD* and *aflQ* genes were evaluated by PCR using 4 pairs of primers. Amplified PCR products were analyzed by 1% agarose gel electrophoresis. Specific PCR products corresponding to expected molecular size of *aflS* (684bp), *aflR* (1079bp), *aflD* (852bp) and *aflQ* (757bp) were detected only in *Aspergillus flavus* species.

The presence of structural genes (*aflD* and *aflQ*) and regulatory genes (*aflS* and *aflR*) could be considered as an early indicator of aflatoxin production, but many other aflatoxin biosynthesis genes and environmental factors affect for the aflatoxigenicity in *Aspergillus* species. Our investigation regarding the influence of abiotic factors (water activity and temperature) on the expression of aflatoxin biosynthesis genes in *Aspergillus flavus* - is in progress. This type of study is required to clarify the crucial role of environmental factors in the activation of the aflatoxin biosynthesis genes.

Keywords: Aflatoxin biosynthesis genes, *Aspergillus flavus*, PCR

Acknowledgements: The research was financed by the Ministry of Education, Science and Technological development of the Republic of Serbia (Project No. III 46005 - "New products based on cereals and pseudocereals from organic production").

XENOBIOTICS IN *LACTUCA SATIVA* L.

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Controlling the contamination of crop plants with pollutants has raised increasing interest in recent years, especially in developed countries. Among the investigated pollutants, heavy metals and polycyclic aromatic hydrocarbons (PAHs) are of particular interest because some are dangerous for their toxicity and/ or carcinogenicity. Lettuce (*Lactuca sativa* L.) is an important leafy vegetable crop, used mainly as a fresh vegetable in salads, but also cooked. Exposing a relatively high surface area of leaves, this plant can be easily contaminated with chemicals from environment, which enter the food chain and can influence the consumer's health. The major aim of this research is to establish the degree of contamination with the xenobiotics lead, cadmium, copper, zinc and 16 Environmental Protection Agency priority PAHs for experimental cultures of lettuce carried out during three years in three locations with different pollution patterns: a reference field situated in the west of Jucu de Jos (experimental field of USAMV Cluj-Napoca), a contaminated surface from diffuse sources (an intensely circulating area located in Cluj-Napoca) and a site with historical contamination caused by SC Sometra SA Copșa Mică, located in Șeica Mare. PAH determinations were performed by high performance liquid chromatography (HPLC) on an Agilent 1100 system, using an Envirosep PP column, with acetonitrile:water as mobile phase (45:55 v/v), while the heavy metals' content was determined by atomic absorption spectrometry using a Shimadzu AA-6300 double beam atomic absorption spectrophotometer with both flame and graphite furnace, after microwave-assisted digestion. The recorded concentrations for heavy metals showed maximum values for samples originating from the historical pollution site (1.41 µg/ kg Pb, 0.09 µg/ kg Cd, 2.96 mg/ kg Cu and 5.09 mg/ kg Zn), while the PAH's concentrations were highest in samples from urban area (8 µg/ kg total PAH's), the average PAHs' content ranging from 0.08 µg/ kg for benzo(g,h,i)perylene to 3.27 µg/ kg for naphthalene, in the contamination mechanism predominantly intervening the atmospheric deposition loaded with combustion products (car traffic). The obtained results revealed the higher share of low molecular weight PAHs, mainly naphthalene, fluorene and acenaphthene, high molecular weight PAH contamination being due to benzo(a, h)anthracene, benzo(b)fluoranthene and dibenzo(a, h)anthracene. Overall, the obtained results revealed a low concentration of the studied xenobiotics in lettuce, the main contributors being soil pollution and car traffic.

Keywords: heavy metals, polycyclic aromatic hydrocarbons, lettuce, Lactuca sativa L.

INFLUENCE OF pH VALUE OF CEMENT-BASED SUBSTRATES ON VIABILITY OF BIOCALCIFYING BACTERIA *SPOROSARCINA PASTEURII* DSM 33

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In the recent decades, extensive studies have been conducted on engineered processes of self-healing effect of cementitious materials. Appearance of cracks strongly influence on service life and durability of cement-based constructions. The cement-based materials have ability of self-healing through hydration of residual cement and carbonation processes. These natural phenomena cannot be in charge of a complete healing of cement matrix and development of engineered processes can lead to more durable materials. The healing effect of cement-based materials depends on many factors including pH value, dissolved inorganic carbon, nucleation sites and presence of free calcium ions, etc. One of ecofriendly engineering approaches of healing is microbiologically induced calcium carbonate precipitation (MICP) which occurs naturally in presence of active ureolytic microorganisms.

The aim of this study was to determine influence of pH value of different cement-based substrates on viability of biocalcifying bacteria *Sporosarcina pasteurii* DSM 33. Three types of cement-based materials are used in this study: historical mortar from Bođani monastery, limestone models and cement prisms. The viability of bacteria and the changes of pH value of the substrates were investigated monthly during 150 days (aerobic incubation at 30 °C, 100% relative humidity).

Based on the obtained results, the change of pH value of the examined substrates over time can be the result of MICP activity, which is followed by the increase of number of viable bacteria in the samples.

Keywords: Sporosarcina pasteurii, ureolytic bacteria, cement-based substrates, autonomic healing

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STUDY THE EFFECT OF DEEP FRYING IN OIL CONTENT CHANGES OF FATTY ACIDS FOR SUNFLOWER OIL AND COTTON

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Growing olive trees (*Olea europaea* L.) is a widespread activity throughout the Mediterranean. The chemical composition of oil, especially its content of polyunsaturated fatty acids, plays the most important role in determining the validity of oil for frying. In this research, cotton oil containing linoleic acid was used in a lower amount of 8.37% and oleic acid more 8.86% compared with sunflower oil in frying. Provide a good degree of stability in two ways of frying without adding fresh oil for frying. In one of the places where falafel was produced and sold, it was frying for four hours a day, for four days, in two ways: the first is not to cover and filter the oil after each mixture, and the second to cover and filter the oil after each mixture. The second method gave better results in both oils. The amount of linoleic acid decreased in the cotton oil by 4.70% and 2.04% in the first method and about 3.42% and 1.03% in the second way respectively. The decrease and increase in linoleic acid and oleic acid in the sunflower oil by frying methods was 11.60% and 3.75% using first method and about 8.64% and 5.42% in the second one, respectively. The second method gave preference to the quality of frying oil in both ways. The deterioration of the sunflower oil was greater compared with the cotton oil. This reflected the validity of the oil for frying during the four quintiles over the four days.

Keywords: palmitic acid; oleic acid; linoleic acid, sunflower oil, cotton oil, frying

SUPERCRITICAL CARBON DIOXIDE EXTRACTION OF CAROTENOIDS FROM *PHAEODACTYLUM TRICORNUTUM*

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Microalgae are considered as alternative feedstocks for production of valuable bioproducts, with potential applications in pharmaceuticals, nutraceuticals and cosmetics. Due to its high polyunsaturated fatty acids and carotenoid content the pennate diatom *Phaeodactylum tricornutum* is a promising natural source for these high-added value products. Nowadays, besides the economic cost biorefinery approaches focus on the utilization of sustainable extraction techniques with small environmental impact. Supercritical carbon dioxide extraction (SFE-CO₂) is considered as green sustainable extraction technique, for isolation of functional products from various sources including microalgae.

The aim of this research was the optimization of SFE-CO₂ conditions for recovery of carotenoids and polyunsaturated fatty acids from *P. tricornutum* biomass. Towards this end, response surface methodology and central composite design were employed to establish the optimal extraction conditions. Extraction parameters such as temperature, pressure and the amount of ethanol as a co-solvent were optimized for the total lipophilic product yield and carotenoid content chosen as response factors. Total lipophilic product yield varied from 4.8 to 11.7 g/100g DW, whereas carotenoid content ranged between 14.6 and 40.6 mg/g of extract. Under optimal conditions (30 MPa, 30 °C and 9.2% ethanol) the yield obtained was 9.8 g/100g DW and total carotenoid content was 35.1 mg/g of extract. The fatty acid profile of the extracted oil was analyzed by means of GC-FID. Moreover, extracts and solid residues after SFE-CO₂ were evaluated for their in vitro radical scavenging capacity and the total phenolic content. No significant changes in the antioxidant capacity of *P. tricornutum* biomass was observed prior and after non-polar fraction removal; thus, indicating that the solid residues after SFE-CO₂ could be further utilized for recovery of antioxidant rich fractions.

Keywords: microalgae, Phaeodactylum tricornutum, supercritical carbon dioxide extraction, response surface methodology, carotenoids

EFFECT OF FAT CONTENT ON AROMA RELEASE OF FLAVOURED PUDDINGS

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There is an increasing demand for low fat foods nowadays. Depending on consumer demands, puddings are produced with different fat content. Low fat content affects the flavour of puddings and acceptance of the consumer. In this study, the aim was to determine the effect of fat reduction on sensory properties of flavoured puddings. Flour, granulated sugar and corn starch were mixed with milk to prepare the puddings. Aroma was added and mixture was cooked. Then cream was added to puddings. Puddings flavoured with 3 different oil-based and water-based aromas (strawberry, banana, vanilla) and 5 different percentages of cream (0%, 5%, 10%, 20%, 30%) were prepared and sensory analyses were performed in order to determine flavour, hardness, oiliness and overall liking. In sensory analysis, ranking test was applied. Flavoured pudding samples with oil-based and water-based aroma, also with different fat ratios, were found to vary in flavour, hardness, oiliness and overall liking, with statistical significance. It was observed that flavour of pudding with oil-based aroma increased, hardness perception decreased and oiliness increased with the increase of fat ratio. In addition, it was determined that flavour of pudding with water-based aroma generally increased, hardness perception generally decreased whereas oiliness perception increased with the increase of fat ratio.

Keywords: aroma release, oil-based aroma, pudding, water-based aroma



USE OF LEGUMES IN GLUTEN-FREE PASTA PRODUCTION

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Coeliac disease is an immune-mediated enteropathy triggered by the ingestion of gluten and related cereal proteins in genetically susceptible individuals. The Coeliac disease is one of the most common food induced disorders worldwide with a mean prevalence of 1% of the total population. The only effective treatment for individuals suffering the Coeliac disease is adherence to a gluten-free diet throughout their lifetime. Gluten-free diet results in an increased demand of gluten-free products namely pasta, bread and cookies. Pasta contains gluten proteins, mainly composed of gliadins and glutenins that coagulate and form a strong viscoelastic protein network, also trapping starch material inside. Legumes are gluten-free ingredients that could be used to produce gluten-free pasta with high nutritional quality. Legumes are rich in proteins, dietary fibers and resistant starch. The *in-vitro* and *in-vivo* glycemic index could be reduced or maintained at its low value in legume enriched pasta in comparison to their no-legume counterparts. Some legumes such as bean, buckwheat, yellow pea, chick pea, lentil and maize were used in gluten-free pasta production. The aim of this study is to provide an overview about use of legumes in gluten-free pasta production.

Keywords: coeliac disease, gluten-free, legumes, pasta

THE INFLUENCE OF ROASTING TEMPERATURE ON THE PHYSICAL PROPERTIES OF ARABICA AND ROBUSTA COFFEE

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Coffee quality depends of numerous factors of harvest and postharvest processing conditions. During the roasting process, coffee beans change their physical, chemical and sensory properties. Physical properties (colour, bean size, weight etc.) are usually the first criteria for evaluation of coffee quality. The aim of this research was to investigate the influence of different roasting temperatures on physical properties of Arabica and Robusta coffee.

Arabica (Rio Minas) of two quality classes and Robusta coffee samples were used for the research. The green coffee samples were roasted at 167, 171 and 175 °C. Physical properties of roasted coffee samples such as bulk density, total weight loss, water activity, texture and colour were determined. Colour was analysed by colorimeter in CIELab system (L*, a*, b*) and by sensory analysis.

Total weight loss increased, and bulk density of Arabica and Robusta coffee beans decreased with increasing of roasting temperature. Total weight loss was the highest in Robusta and the lowest in Arabica 1st class coffee samples. The highest bulk density had Robusta and the lowest Arabica 2nd class coffee samples. The highest braking force values were showed by Arabica 1st class coffee samples roasted at 167 °C and by Arabica 2nd class coffee samples roasted at 171 and 175 °C, compared to other samples roasted at the same temperatures. Arabica 1st class roasted at 167 °C had significantly higher ($P < 0.05$) braking force than same sample roasted at 171 and 175 °C. Although braking force values of Robusta and Arabica 2nd class beans decreased as the roasting temperature increased, these changes were not statistically significant ($P > 0.05$). Water activity values were significantly lower ($P < 0.05$) for coffee samples roasted at 175 °C than for samples roasted at 167 °C. L*, a* and b* colour parameters decreased as the roasting temperature and browning increased.

This research showed that physical properties of coffee depend on the roasting temperature and on the type and class of coffee as well. Parameters such as colour and weight loss can be used as parameters for determination of roasting degree in industrial conditions of coffee production, but they must be defined for each type and class of coffee in particular. Sensory analysis of coffee beverage colour can be used as a good tool for a detailed description of this property, as an important indicator of quality.

Keywords: Arabica, Robusta, roasting, physical properties

THE TECHNOLOGICAL QUALITY OF SUGAR BEET IN VOJVODINA DURING 2016-2018

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The technological parameters of the sugar beet are qualitative descriptors which affect the efficiency of the entire sugar production. Regarding the technological quality of sugar beet, the most important requirements are high concentrations of sucrose and low concentrations of non-sucrose compounds. The aim of the presented study was to determine the technological quality of 1752 sugar beet samples collected from three different regions in Vojvodina during the period of three years (2016-2018). Additionally, the influence of weather conditions prevailed during sugar beet growing seasons on the quality parameters was investigated. The highest average concentration of sucrose (16.08, 16.13, 15.87%) was recorded in samples from all three regions harvested in 2017. The lowest average concentrations of sodium (1.01 mmol/100g) and α -amino nitrogen (1.92 mmol/100g), as well as relatively low concentrations of potassium (2.48 mmol/100g) were detected in sugar beet samples collected during 2017. High monthly average of air temperatures (18.6 °C) and the moderate sum of precipitation (341.74 mm) for period of growing seasons (from March to September) were recorded in 2017. Two other observed years (2016 and 2018) were characterized by lower average temperatures and much higher amount of rainfall (483.87 mm and 445.30 mm, respectively) resulting in lower yield of sugar beet. It was noticed that weather conditions during the sugar beet growth and harvesting period can significantly affect the technological quality of sugar beet.

Keywords: *sugar beet, technological quality, weather conditions*

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PROPERTIES OF BIOPOLYMER FILM WITH ESSENTIAL OILS

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The ability to decompose in environment, as well as fact that they are isolated from natural sources, make biopolymers one of the most interesting topics for researchers in packaging industry. Biopolymer materials are usually synthesized from proteins, polysaccharides and lipids. In recent years, due to their chemical composition, agro-industrial by-products present a real potential in the production of environmentally friendly plastic. Pumpkin oil cake, by-product obtained after cold-pressing oil extraction from pumpkin seed, is a rich source of proteins, polysaccharides and lipids, and because of its composition, represent an interesting substrate for the production of biopolymer films. However, application of biopolymer films is limited by the high sensitivity to moisture and water-vapor permeability due to their hydrophilic nature, and addition of different hydrophobic additives can improve this property. Meanwhile, these additives can also have unwanted influence on some characteristics of films, therefore it is very important to examine effect of appropriate additives and find their optimal concentration for obtaining films with optimal performance for packaging. Recently, use of essential oils, as alternative to synthetic chemical products, has been studied. Essential oils exhibit antimicrobial and antioxidant properties, and most of them are classified as Generally Recognized as Safe. This makes them as an interesting additive in food industry. Essential oils can be added directly to the food, or incorporated into food packaging. Research has shown many advantages of incorporating additives into films compared to their direct addition into food.

The objective of this study was to examine influence of winter savory and basil essential oil on composite pumpkin oil cake film properties. Essential oils were added in concentration of 1% into film casting solution and mechanical, physico-chemical and barrier properties of obtained films were evaluated. Pumpkin oil cake-based film without essential oil was used as a control. According to the obtained results addition of essential oils into pumpkin oil cake composite film has shown the highest impact on reducing moisture sensitivity and water permeability. However, effect on mechanical properties was not observed.

Obtained results propose that with the addition of hydrophobic components in specific concentrations to biopolymer films, their sensitivity to moisture and water permeability can be reduced without major impact on mechanical properties.

Keywords: biopolymer films, pumpkin oil cake, essential oils, properties

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MECHANICAL PROPERTIES OF PUMPKIN OIL CAKE BASED COMPOSITE BIOPOLYMER FILMS

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With the increase in the number of population and resource constraints, as well as with the growing environmental threat, the use of biopolymers for the production of biodegradable materials is intensively explored. Biopolymer films are also studied for the food packaging application, where their potential is particularly pronounced, thanks to relatively good mechanical and excellent barrier properties for gases. Some disadvantages that limit the scope of the use of biopolymer food packaging films can be significantly reduced by the formation of composite films based on different biopolymers or biopolymers and lipids.

In this work, pumpkin seed oil cake that remains after process of cold pressed oil extraction, was used for the production of composite biopolymer-based packaging films. The whole pumpkin oil cake is used for the production of films, without isolating and purifying individual components, thus saving additional technological procedures that would contribute to increasing the ecological footprint of such materials production. During the production of films, four degrees of filtration were applied in order to produce composite films using different particle size, which can lead to higher yields of the process and additional reduction in the amount of residual waste. The obtained composite biopolymer films were analyzed for mechanical properties.

Obtained results showed that different filtration procedures affected both tested mechanical properties of obtained films, tensile strength and elongation at break. With increasing particle size and increasing yield, increase in tensile strength (from 2.32 MPa to 4.30 MPa), followed by decrease in elongation at break (from 150% to under 40%) was observed. Analyzing tensile properties of films in two perpendicular directions, no differences were observed, leading to conclusion that there were no specific orientation of biopolymer molecules during the film forming process. Considering that some of possible applications of these films might imply refrigerated storage, this factor was also tested and results showed that refrigerated conditions affected tensile properties of films, leading to higher elongation at break and lower tensile strength of the films in two of four filtration fractions, the finest and the roughest fraction. One additional parameter that affected mechanical properties of films was mass of casted film forming solution. With increasing mass of casted film forming solution, both tested mechanical property parameters increased.

Keywords: biopolymer film, pumpkin oil cake, composite, mechanical properties

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QUALITY CHARACTERISTICS OF STRAWBERRY COATED WITH QUINCE SEED MUCILAGE AS EDIBLE COATING

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In recent years, living conditions are rapidly changed and working hours are prolonged so consumers prefer food products that are practically prepared, ready to eat and have a long shelf life. Therefore, global demand for consuming fresh fruits and vegetables containing important nutrients such as proteins, vitamins, minerals and dietary fiber for human health and nutrition is increasing day by day. Production and consumption of minimum processed fruits and vegetables are gained popularity. However, post-harvest metabolic activities such as respiration, transpiration and small-scale processes such as cutting, slicing or peeling increase the quality losses in minimum processed fruits and vegetables during storage. Edible film and coatings can be used to improve sensory characteristics, prevent losses of nutritional value, color and weight and reduce physical disorders of minimally processed fruits and vegetables. The aim of this study was to investigate the suitability of quince (*Cydonia oblonga*) seed mucilage as an edible coating on the shelf-life extension of strawberry fruit in ready form for consumption. For this purpose, the seeds of quince were obtained manually and extracted without waiting. The mucilage obtained from the quince seeds extract were used for covering the strawberries after the fruits were washed and leaves of strawberries removed. Strawberries were dipped in film solution and dried at room temperature (~23 °C), and stored at 4 °C for 10 days. At 0, 2, 4, 7 and 10 days, color, firmness, weight loss measurements and sensory analysis were performed for fruits coated with edible film and uncoated fruit as control. Coating significantly delayed softening, reduced weight loss and maintained color values of fruits. Sensory characteristics of coated strawberry such as color, taste and texture were much better preserved. The results of this study indicated that usage of edible coatings will be effective for strawberry fruit preserving and quality characteristics of strawberry fruits maintained longer by edible coating.

Keywords: edible coating, seed mucilage, strawberry

IMPACT OF AFLATOXIN B₁ ON THE QUALITY OF STORED SPELT WHEAT

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Aspergillus flavus might become a serious danger to the worldwide grain industry due to harmful effect of aflatoxins on human and animal health, as well as the potential loss of yield and reduction of end-use quality. This paper presents the first assessment of the impact of aflatoxin B₁ (AFB₁) on the trade and technological quality of the stored spelt wheat. Different AFB₁ contamination levels (25, 50, 100 and 250 µg/kg) of spelt grains, artificially inoculated with aflatoxin-producing *A. flavus* isolate from common wheat grains, were compared with the control (uncontaminated) sample. The results showed that aflatoxin contamination significantly reduced both trade and technological quality of spelt. Protein content (16.10%) in the grain sample with the highest level of AFB₁ (250 µg/kg) was significantly higher than the protein content in the samples contaminated at lower levels, as well as uncontaminated sample (15.6%). There was a decrease in absolute grain mass (39.75 g and 40.20 g) for the high-level contaminated samples (100 and 250 µg/kg, respectively) in comparison to control and samples contaminated at the levels of AFB₁ from 25 to 100 µg/kg. Increasing the concentration of AFB₁ in spelt samples led to an increase in the water absorption capacity of spelt flour, as well as an increase in dough development time. The stability of the dough showed a tendency of decline with an increase in the level of AFB₁ contamination. The results have shown that some parameters of spelt quality can be disturbed only at high levels of AFB₁ contamination. Therefore, there is the concern with respect to consumer health risks which AFB₁ can cause at much lower concentrations in grains. In terms of food safety, continuous monitoring of aflatoxin contamination must be of high priority, in both field and storage facilities of cereal grains.

Keywords: aflatoxin B₁, spelt wheat, quality

Acknowledgements: *This paper is the result of the research within the project III 46005 “New products based on cereals and pseudocereals from organic production”, financed by the Ministry of Education, Science and Technological Development, Republic of Serbia.*

BETAINE CONTENT IN BUCKWHEAT ENRICHED WHOLEGRAIN WHEAT PASTA

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This study was conducted in order to investigate the effect of the addition of buckwheat flour, as well as the influence of buckwheat grain pretreatment, on betaine content in wholegrain wheat pasta. The buckwheat is traditionally used in the human diet for centuries due to its nutritional benefits and positive effect on health. From the other side, betaine is a bioactive compound important for many biochemical processes in the human organism. The single screw extruder was used for the production of wholegrain wheat pasta as a control sample and pasta in which wholegrain wheat flour was substituted with one of two types of wholegrain buckwheat flour (autoclaved wholegrain buckwheat flour and flour without treatment), with a level of substitution from 10 to 30%. High-performance liquid chromatography with ELSD was used to analyze betaine contents in different pasta samples. Betaine content in wholegrain wheat pasta was 30.07 mg/100g and the highest content of betaine in pasta enriched with wholegrain buckwheat flour was detected in sample with 10% flour substitution (21.63 mg/100g). Following the presented results, pretreatment of buckwheat grain did not show important differences in betaine content in comparison with pasta enriched with untreated buckwheat flour.

Keywords: *pasta, buckwheat flour, betaine, HPLC, ELSD*

Acknowledgements: *These results are part of the research carried out within the project of the Ministry of Science, Education and Technological Development of the Republic of Serbia, III 46005.*

OPTIMIZATION OF MICROWAVE ASSISTED ENZYMATIC EXTRACTION OF STEVIOL GLYCOSIDES AND PHENOLIC COMPOUNDS FROM STEVIA LEAF

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Stevia (Stevia rebaudiana Berton) has gained great interests due to its non-caloric and health related properties with its steviol glycosides and phytochemical constituents. Currently, it is being utilized as a natural sweetener in many food formulations such as soft drinks, jams, dairy products, chocolates, etc. Green extraction techniques using non-hazardous solvents and yield-enhancing strategies have been subject matters of various disciplines such as food technology, chemistry and pharmacology. Stevia powders are industrially produced by aqueous-extraction with further purification and drying steps. There has been no any report handling combined microwave and enzyme assisted technique on the extraction of steviol glycosides and antioxidant compounds to date. The objective of this study was to determine optimum processing parameters of microwave assisted enzymatic extraction of steviol glycosides and phenolic compounds by investigating the effects of independent variables. The plant was collected from a stevia garden and its leaves were detached from flower part, dried and milled into powder form before commencement of the treatments. Operations were performed using a microwave extraction system equipped with a digital control system for irradiation time, temperature and microwave power. Viscozyme L which is a widely used cellulolytic enzyme cocktail was used in this study. The effects of enzyme concentration (1.2 – 60 FBG unit/g), temperature (25 – 60 °C) and time (1 – 20 min) were evaluated using five-level rotatable central composite design by response surface methodology (RSM). The validities of models were assessed by ANOVA outputs which were determination coefficient (R^2), lack of fit and Fisher's test value (F-value). The maximized yields of stevioside, rebaudioside A and total phenolic compounds were determined as 62.5, 25.6 and 20.7 mg/g, respectively at optimum processing conditions which were estimated as 10.9 FBG unit/g, 53 °C and 16 min. In addition, compounds present in stevia leaf extract were tentatively identified using liquid chromatography/quadrupole-time of flight/mass spectrometry (LC/Q-TOF/MS) analysis.

Keywords: stevia, enzymatic extraction, microwave, steviol glycosides, phenolic compounds

Acknowledgements: Authors respectfully thank Mr. Selahattin Güvenç (Burhaniye Stevia Garden, Balıkesir, Turkey) for providing the plant material for this study.

HYPOGLYCAEMIC AND ANTIHYPERGLYCAEMIC ACTIVITY OF NEW DRUG FORMULATION OF BASIL EXTRACTS

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New drug carrier systems, as well as their formulations, allow the transport of the drug exactly to the site of action, and have significant effects on its efficacy. Microencapsulation is a process that allows the prolongation of drug release and reduction of adverse effects. Experiments were carried out on laboratory Wistar rats. Animals were treated with water extract of *Ocimum basilicum* in microcapsules and with combination of basil extracts and monoketocholic acid in microcapsules for 7 days. Alloxan was used to induce hyperglycemia. The effects of samples on glycemia were evaluated by measuring blood glucose levels in alloxan-induced diabetic rats. Microcapsules were prepared by the method of microencapsulation and alginate matrix produced microcapsules. The dose of basil extract that was orally administered in rats was 200 mg/kg and the dose of monoketocholic acid was 4 mg/kg. A seven-day treatment with basil aqueous extract, as well as a combination of basil and monocetocholic acid extract in the pharmaceutical formulation, led to a statistically significant reduction in the blood glucose concentration of animals with alloxan-induced hyperglycemia compared to pre-treatment values ($p < 0.05$ and $p < 0.01$), which indicates that basil has hypoglycaemic and antihyperglycaemic effects. Microcapsules, as a pharmaceutical-technological formulation, substantially enhance the hypolipidemic action of basil and extract monocetocholic acid extracts.

Keywords: basil, microencapsulation, glycaemia

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PHENOLICS CONTENT AND ANTIOXIDANT CAPACITY OF BERRY NECTARS

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Among the different types of fruits, berries attract growing attention regarding the many health-promoting effects. The outstanding biological value of berries is based on the presence of different phytonutrients, such as flavonoids, especially anthocyanins, other polyphenols, as well as carotenoids, vitamins, and minerals. Many studies have revealed antioxidant, anti-inflammatory, anticancer, antimicrobial, and neuroprotective properties of these biologically active compounds receiving advice to increase consumption of berries as part of a balanced healthy diet. Berry products, like juices and nectars, are considered as functional beverages, due to high polyphenols content. Besides the type of fruit, there is evidence that extraction and applied processing significantly effects the composition and antioxidant properties of these products. This study was conducted to evaluate and compare the phenolic content and antioxidant activities of commercial berry-based nectars consumed in Serbia. A total of 15 berry nectars were purchased from local supermarkets. The total phenolic content (TPC) and total flavonoid content (TFC) were determined by spectrophotometric methods. Evaluation of the antioxidant capacity of berry nectars was made based on the results of ABTS, DPPH, FRAP and CUPRAC assays and comparing values of the calculated antioxidant composite index (ACI). The TPC and TFC of commercial berry nectars were in the range of 170.60-379.27 and 3.77-191.10 mg GAE/100 mL, respectively. The values of antioxidant activity of berry nectars evaluated by CUPRAC assay were higher than those obtained by DPPH and ABTS, as FRAP assays too. A significant correlation between TPC and TFC and antioxidant activities were observed. In term of antioxidant capacity, the highest value of ACI was determined for the nectar containing aronia and blueberry mixture (98.6 %), while the lowest value was obtained for the strawberry nectar (19.3%). Despite observed differences among analyzed products, berry nectars are significant sources of phenolic compounds and their regular consumption may be a useful intervention strategy to increase antioxidant dietary intake.

Keywords: *juices, berries, polyphenols, antioxidants*

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APPLICATION OF CORRELATIVE ANALYTICAL TECHNIQUES FOR THE DETECTION OF HEAT TREATMENT OF HONEY

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Honeys from various botanical origin can present in different crystallized forms, depending mainly on sugar composition and water/sugar ratio. Crystallized honey is not preferred by consumers and makes its handling difficult for beekeepers. Therefore heat treatment is applied for honey but heating could result in quality degradation. The techniques used for detection of heating, like quantification of hydroxymethyl-furfural (HMF) and diastase or invertase enzyme activity are not sensitive enough to detect changes caused by lower heat treatment levels (below 50 °C), even though composition of honey could also be affected below this level. Therefore, there is a need for techniques with higher sensitivity, such as the electronic tongue (ET) and near infrared spectroscopy (NIRS) which have been successfully used for monitoring quality of honey previously. The aim of this study was to determine the applicability of ET and NIRS in detection of heat treatment in honey.

Heat treatment was applied at 40 °C, 60 °C, 80 °C and 100 °C for 60, 120, 180, 240 minutes on sunflower, acacia and bastard indigo honeys. HMF content, pH, electrical conductivity and moisture content were determined as quality parameters. CIE L*a*b* colorimetric method was used for the determination of colour of honey. ABTS radical scavenging was applied for the quantification of antioxidant capacity of honey. As correlative techniques, ET and NIRS were applied. Statistical evaluation of the data was performed in R-studio software. The individual quality parameters were evaluated with one way ANOVA to determine significant differences between levels of heat treatment. Principal component analysis (PCA) and linear discriminant analysis (LDA) was used for the evaluation of results of ET and NIRS.

Results of HMF content did not show significant increase ($p < 0.05$) at lower heat treatment levels, while antioxidant capacity of heated samples increased compared to control honeys. LDA classification models showed good classification accuracy of control honeys from treated samples especially at 60 °C and above.

Results obtained by NIR and ET methods showed that changes in composition can be detected even for samples heated at 40 °C, while HMF and ABTS methods were not sensitive enough for this aim. Therefore, the correlative techniques could be promising methods in detection of heat treatment of honey.

Keywords: honey, heat, NIR, electronic tongue



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SUGAR BEET MOLASSES QUALITY ENHANCEMENT THROUGH TREATMENT WITH MODIFIED SUGAR BEET PULP

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The significant sugar beet molasses amount generation (4–5% on beet) in the sugar production process and consequently sucrose loss represent every year struggle for the sugar producers. Over the past years, different processes were established in order to tackle the problem of sucrose recovery by molasses desugarization. The possibilities for further enhancement of the corresponding processes are enabled through non-sugars separation by adsorption based pre-treatments under appropriate conditions. In this respect, industrial by-products utilization as adsorbents gained attention due to the low-cost, easy handling and favourable sorption properties. Considering successful application of sugar beet pulp in metals and dyes adsorption, the presented study aims to investigate the adsorption performance of chemically modified sugar beet pulp in molasses purification. Box-Behnken experimental design was applied in order to assess the influence of 3 independent parameters: pH (3–7), molasses dry substance (30–50°Brix) and modified sugar beet pulp concentration (9–21 g/L) on molasses colour, turbidity and sucrose content. The applied parameters significance was determined by response surface methodology. Among the examined parameters, pH was established as a parameter of great significance regarding changes in molasses colour, turbidity and sucrose content. Applied neutral conditions (pH 7) were adequate for reaching maximal molasses colour and turbidity reduction alongside with minimal changes in the molasses sucrose content. The influence of other independent parameters on molasses quality was negligible. According to the obtained results, molasses pre-treatment with modified sugar beet pulp could improve the molasses desugarization process and contribute to the increase in sucrose yield. Furthermore, sugar beet pulp utilisation in this respect would contribute to the development of sustainable waste managing method inside the sugar factories.

Keywords: *molasses, non-sugars, decolourisation, lignocellulosic adsorbent, modified sugar beet pulp*

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PASTING PROPERTIES OF SNACK PRODUCTS FROM SPELT WHOLEGRAIN FLOUR WITH ADDED BETAININE

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The pasting characteristics of extrudate are used to obtain information of its functional behavior during heating and cooling periods, which is common during the processing of starchy products. Viscosity was measured on Haake Mars rheometer (Thermo Scientific, Germany) equipped with Z43S measuring cup and FL2B propeller shaped rotor with 2 blades. The pasting curves of untreated spelt wholegrain flour with and without betaine were compared with the curve of optimal snack samples (with and without betaine), as well as snack sample with maximum expansion ratio and snack sample with minimum hardness. All samples were obtained during the processing of betaine enriched spelt flour based extrudates. An increase in initial viscosity at low temperatures was observed in all extruded samples (0.831-3.138 Pas) compared to untreated samples (0.145-0.148 Pas) what resulted from a higher amount of the present soluble substances that appeared after thermal degradation of starch. The highest values of maximum viscosity 5.66 and 6.06 Pas and gelatinization temperatures of 78.76 °C and 80.68 °C were recorded for untreated samples with and without betaine, respectively. In comparison to the untreated samples, in all extruded samples a decrease in maximum viscosity at 95 °C (0.78-2.46 Pas) was recorded, what was the result of partial gelatinization of starch during thermal treatment, i.e., the decrease in the number of granules whose swelling results in an increase in viscosity of the paste. The decrease in viscosity while holding the sample at high temperature (95 °C) is due to the leaching of starch granules, resulting in linear orientation of the polymer, what reduces the viscosity of the paste, and it was observed in all samples, except in the optimal sample with added betaine. Since this optimal extruded sample contained betaine, it can be assumed that during extrusion there was a formation of bonds between starch and betaine, what caused that the mentioned sample did not exhibit a decrease in the viscosity during holding the sample at high temperature.

Keywords: betaine, pasting properties, snack, spelt

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PREFERENCES TOWARDS SWEETS AND SALTY SNACK CONSUMPTION AMONG YOUNG POPULATION IN RELATION TO BMI

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Dietary habits reorientation and increased consumption of so-called “unhealthy food” become common in adolescence period. Excessive consumption of sweets and fatty, salty snacks alongside with reduced consumption of fruits and vegetables represent potential health risks associated with caries, obesity, high blood pressure, and diabetes. Nevertheless, recent studies indicate that chocolate intake could contribute to the cardiovascular diseases prevention and potential role of chocolate in benefits linked to lower body mass index (BMI). The presented study aims to examine adolescents’ habits regarding chocolate, sweets and snacks consumption as well as corresponding products intake frequency and relate them with BMI. In this regard, a questionnaire survey was applied to 525 individuals attending elementary and high schools in the province of Vojvodina, Serbia (57.5% females and 42.5% males). The participants’ nutritional status and risk for overweight were addressed through BMI assessment. Normal range nutritional status (70.5%) was determined among the majority of participants followed by overweight (19.8%) and obese (7.6%). Compared to females, increased number of overweight and obese nutritional statuses was recorded among male participants. Regardless of the participants’ nutritional status, fruits were most frequently consumed between meals, followed by salty snacks and sweets. The highest share of sweets consumption between meals was noticed among participants with obese nutritional status (15%). Furthermore, more than 40% of participants with overweight and normal range nutritional status do not consume chocolate at all, while 2.5% of participants with obese nutritional status stated that they consume chocolate daily during the entire week.

Keywords: adolescents, food choices, body mass index, intake frequency, chocolate, salty snack

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SPECTROSCOPY AS A RAPID METHOD TO DETECT PAPRIKA POWDER ADULTERATION WITH CORN FLOUR

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Paprika is a spice of important culinary and nutritional benefits, especially in its semi-processed or powdered form. Paprika powder has become famous for its ease of use, access and storage but has also equally become a target for adulteration for financial gains and greed. Although diverse types of conventional methods exist to track various forms of food adulteration, they are relatively slow and sometimes require a high technical expertise or sample destruction for successful analysis. Emerging methods such as near infrared spectroscopy (NIRS) operates on the non-destructive principle of light interaction with the sample and has shown promising tendencies as a rapid alternative for the conventional methods in detecting adulteration. This study aimed to apply two NIRS instruments (bench: metri NIR and Handheld: DLP NIRSCAN NANO) to detect artificial adulteration of red paprika powder with corn flour at adulterant concentrations of 5%,10%,15%,20%,25%, 30% and 40%. Discriminant analysis (LDA) was used for classification of samples of different adulteration levels. Partial least square regression (PLSR) was used to build models to predict the adulteration. LDA showed a recognition ability of 100% and 90% for metri NIR and NIRSCAN NANO respectively. Prediction accuracy in LDA was 97.8% and 88.9% for metri NIR and NIRSCAN NANO respectively. Both instruments showed high prediction accuracies in PLSR with coefficients of determination (R²CV) greater than 0.9 and Root Mean Squared Error Calibration (RMSEC) less than 2g/100g of paprika powder. The NIRS instruments exhibited discriminatory and predictive abilities capable of tracking paprika adulteration with corn flour and could be applied in further studies to test lower adulterant concentrations.

Keywords: *spice, transreflectance, spectroscopy, chemometrics*

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THE USE OF ADVANCED CONTROL SYSTEMS IN FOOD PROCESSING OPERATIONS

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In the food industry due to the scarce sources of foods, it is much more important to provide processes that have high yields, low production cost and high product quality. Considering this fact and the dynamic nature of food process operations control systems are very important. The dynamic nature of food processes is a result of variation in raw material composition and time-dependent change in the basic structure of the food processing operations. For these reasons, control of these processes is gaining high importance. There are basic and developed control structures like model-based controllers, fuzzy logic controllers, neural network-based controllers and several combinations of these controllers.

Due to the mentioned characteristics and complexities of food process operations, there is a demand for integration of numerical methods, control engineering principles, new embedded network control structures and communication technologies based on the internet of things. The use of advanced control structures will provide an effective solution for high-performance technological processes.

In several control applications basic or first principle models should be used, however, for most of the control problems they are not available for food process operations. For this reason, food process operations should be identified as developing dynamic models for these applications. The aim of this study is to provide a review of the development and applications of control systems in the food processing industry.

Keywords: process control, advanced control, food industry

THE USE OF SPRAY DRYING AND AGGLOMERATION METHODS TO OBTAIN SPINACH JUICE POWDER AND AGGLOMERATES

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Spray drying is the most commonly used drying technique due to its high efficiency and high product quality. Agglomeration can be defined as the size enlargement process in which the fine particles such as spray dried powders binding with one another, resulting in an aggregate porous structure much larger in size than the original material, and the primary particles can still be identified. The aims of this study were to obtain spray dried spinach (*Spinacea oleracea*) juice powders and to apply agglomeration process in order to improve the powder properties. Spinach was washed and kept in cold conditions (refrigerator, + 4 °C) before drying. The juice was prepared by a home type food processor and filtered with crude filter paper. The spray drying was carried out at a feed rate of 1.00 ± 0.20 kg/h, at the air inlet/outlet temperature of 160/100 °C; agglomeration was carried out in a fluid bed dryer at 60 °C. 1.6 m/sec (with water, maltodextrin, whey protein isolate and gum arabic as drying agents). The dried powders and agglomerates were analyzed for moisture content, water activity, wettability, solubility, flowability and hygroscopicity behavior. The moisture content and water activity values of the spinach juice agglomerates were; 6.44% -11.68% and 0.254-0.412 respectively. The agglomeration process showed that wettability and solubility times decreased, and stickiness, flowability and hygroscopicity were improved in comparison to the powder products. The friability values of spinach agglomerates were 5.02-95.4%. As a result of the study, it was found that spinach juice powders obtained by spray dryer can be successfully agglomerated in fluidized bed.

Keywords: spinach, spray drying, agglomeration, fluidized bed, powder properties

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PRODUCTION OF INSTANT LIQUORICE (*GLYCYRRHIZA GLABRA*) ROOT SHERBET

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Liquorice (*Glycyrrhiza glabra*) root sherbet is consumed widely in the southeast of Turkey. However, its shelf life is too short as it exhibits high moisture content, water activity and microbial load. Moreover, preparation of liquorice sherbet is not also practice. It is not known exactly in the other parts of Turkey due to these reasons. However, instant products such as instant coffee, tea have not these problems. Therefore, we produced instant liquorice root sherbet (powder) using maltodextrin and gum Arabic as wall material with ratio of core to wall (1:3) by spray drying technology. The product was safe against microbial spoilage as it exhibited low moisture content (1.47-2.09%) and water activity (below 0.3). Powder also showed good solubility (98.69-99.50%) but not flow ability (Hausner ratio: 1.43-1.53). Microencapsulation yield, microencapsulation efficiency, and color parameters of powder were also analyzed.

Keywords: instant, liquorice root sherbet, maltodextrin, gam arabic, spray drying

Acknowledgements: This research was funded by the Scientific Research Project Council of Harran University (HUBAK- 18135).

PHENOLIC FRACTIONS, ANTIOXIDANT AND ANTIDIABETIC POTENTIALS OF *QUERCUS INFECTORIA* GALL

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The *Quercus infectoria*, belongs to the family *Fagaceae* is valuable source of bioactive compounds, especially tannins. However, to the best of our knowledge, no researches have been carried out exactly on all properties of the plant in the literature. Therefore, the objectives of this study were to evaluate phenolic fractions, antioxidant and enzyme inhibitory activity of *Quercus infectoria* Gall ethanolic extracts. Phenolic fractions were performed Liquid chromatography electrospray ionization tandem mass spectroscopy (LC-ESI-MS/MS). Ellagic acid (28.16 g/kg) was predominant phenolics in *Quercus infectoria* Gall. The antioxidant activity of the *Quercus infectoria* Gall was determined by four different methods: DPPH, ABTS, FRAP and CUPRAC. DPPH, ABTS, FRAP and CUPRAC were determined in *Quercus infectoria* Gall at 2.29, 1.65, 1.52, and 1.98 mmol Trolox equivalent antioxidant capacity (TEAC) per g of sample, respectively. The enzyme inhibitory activity was investigated against alpha-amylase and alpha-glucosidase. The results showed that ethanolic extract of *Quercus infectoria* Gall was inadequate on preventing alpha-amylase but displayed better the inhibitory activity against alpha-glucosidase with an IC₅₀ value of 0.002 mg/mL compared to acarbose with an IC₅₀ value of 20.28 mg/mL. Total phenolic content, total flavonoid content and hydrolyzable tannin content of *Quercus infectoria* Gall were 434.16 mg gallic acid equivalents (GAE), 14.13 mg catechin equivalents (CE), and 419.14 mg tannic acid equivalent (TAE) per g of sample, respectively. As a result, ellagic acid contents, antioxidant and antidiabetic potentials of *Quercus infectoria* Gall revealed its nutritional value. Therefore, *Quercus infectoria* Gall could be evaluated as reliable source for human diet and also alternative to synthetic materials and drugs for treatment some disease.

Keywords: *quercus infectoria gall, ellagic acid, antioxidant, antidiabetic*

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OXIDATIVE STABILIZATION OF COLD-PRESSED SUNFLOWER OIL BY CARAWAY (*CARUM CARVI*L.) ESSENTIAL OIL

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Essential oils isolated from aromatic and medicinal herbs have been recognized as natural antioxidants and they are widely used in food preservation. In this study, the influence of caraway (*Carum carvi* L.) essential oil (CEO) in various concentrations (250, 500 and 1000 ppm) on oxidative stability of cold-pressed sunflower oil was examined in accelerated conditions (63±2 °C for twelve days). Stabilization effects were compared to butylated hydroxytoluene (BHT) and sample without any antioxidant, as a control. In order to determine a level of oxidative changes, samples were analyzed for peroxide value (PV), anisidine value (AnV) and conjugated dienes (CD) every 4 days. In addition, oxidative stability was measured using differential scanning calorimetry (DSC) coupled with thermogravimetry (TG) technique and by Rancimat apparatus (100°C, air flow rate of 20 L/h). CEO was analysed by GC-FID and GC-MS techniques. The main components of essential oil were carvone (57,43%) and limonene (39,91%) representing 98,30% of the total oil. In accelerated oxidation test, measured parameters of lipid oxidation showed that CEO in all concentrations inhibited the formation of primary and secondary products better than BHT. However, oxidative stability determined by Rancimat test showed that only addition of essential oil in concentration of 1000 ppm have antioxidative activity. Compared to BHT, significantly lower stabilization effect obtained by this method can be ascribe to the nature of essential oil and volatility of compounds at high temperatures. Results obtained by DSC method showed that addition of essential oil in highest concentration caused slight increase of oxidative stability in comparison with a control sample. Based on the obtained results it can be concluded that CEO may be used as natural antioxidant with moderate efficacy which may have different values depending on the applied temperature and method.

Keywords: *carum carvi* L., essential oil, sunflower oil, oxidative stability

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ASSESSMENT OF ANTIMICROBIAL ACTIVITY OF POMEGRANATE PEEL AND DETERMINATION OF ITS INDIVIDUAL PHENOLIC COMPOUNDS BY USING LC-ESI-MS/MS

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In this study, antimicrobial activity and individual phenolic compounds of pomegranate peel were evaluated. The antimicrobial activity of the peel extracts was investigated in vitro against Gram-negative bacteria (*E. coli* and *P. aeruginosa*) and Gram-positive bacteria (*S. aureus* and *E. faecalis*), fungi (*C. albicans*) and mould (*A. brasiliensis*) using agar diffusion method. The extract showed inhibitory activity on all microorganisms except *A. brasiliensis*. Among Gram-negative, Gram-positive and fungi, *C. albicans* was the most resistant microorganisms to pomegranate peel extract with MIC values of 200 mg/mL while the extracts had an antimicrobial activity to *P. aeruginosa* with MIC values of 12.5 mg/mL. Sixteen different phenolic compounds were determined in pomegranate peel by LC-ESI-MS/MS. Ellagic acid (1034.7 ppm) and catechin (1063.2 ppm) were principal phenolics in pomegranate peel, followed by phloridzin dihydrate and vanillic acid. This study provides direct data on antimicrobial activity and phenolic fractions of pomegranate peel. Results showed that pomegranate peel could be used as a functional ingredient in different sectors such as food, nutraceutical and pharmaceutical industries.

Keywords: *pomegranate peel, antimicrobial, lc-esi-ms/ms*

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THE EFFICACY OF CONTINUOUS SYSTEM ULTRAVIOLET LIGHT TREATMENT ON THE INHIBITION OF MOLDS INOCULATED TO SURFACES OF YOGHURT WITH SURFACE CREAM

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Ultraviolet light (254 nm) technology is one of the non-thermal processes that can be used for the surface decontamination of food products. The aim of this study was to investigate the germicidal effects of UV-C light treatments on moulds inoculated to yoghurt with surface cream. UV-C resistant moulds (*Cladosporium* spp. Y18, *Aspergillus* spp. YS21) isolated from commercial yoghurts and *Geotrichum candidum* RSKK 2036 were inoculated to surfaces of yoghurt samples at levels of 4-4.5 log CFU/g. Inoculated yoghurts with surface cream were subjected to continuous system UV-C light at two different distances (7.5 and 20 cm) and at six different conveyor belt speeds (0.2, 0.4, 0.8, 1.6, 2.4 and 3.2 m/min), corresponding to the treatment doses in the range of 0.86–26.73 kJ/m². Results show that UV-C doses of 0.86–26.73 kJ/m² resulted in 0.21-1.47 log CFU/g, 1.10-1.63 log CFU/g and 0.90-1.46 log CFU/g reductions of *Cladosporium* spp. Y18, *Aspergillus* spp. YS21 and *G. candidum* inoculated on the surface of yoghurt samples, respectively. The numbers of test cultures were significantly different when compared to untreated control samples for *Aspergillus* spp. YS21 and *G. candidum* cultures even at the lowest UV-C dose of 1.08 kJ/m². On the other hand, a further increase in the UV-C light doses did not result in any additional significant reductions on the numbers of *Aspergillus* spp. YS21 and *G. candidum*. *Cladosporium* spp. Y18 was the most resistant culture and UV-C dose of 4.43 kJ/m² was needed to achieve 1 log reduction. The results suggest that UV-C light could be an alternative technique for the decontamination of yoghurt surfaces to prevent post-contamination after the production.

Keywords: *ultraviolet light, surface decontamination, mould inhibition, yoghurt*

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GUAR-XANTHAN EFFECT ON STARCH BIOPOLYMER FILMS PROPERTIES

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Research in the field of biopolymer materials has been intensified in the recent years with the ultimate tendency for biopolymer materials to find application for the food and pharmaceutical products packaging. Starch is one of the most common biopolymers for this purpose. Starch biopolymer films do not adversely affect the packaged content; have excellent barrier properties to oxygen and carbon dioxide and moderate mechanical properties. Their main disadvantage is poor water vapour barrier property. There are numerous methods for starch material properties improvement and in this paper the influence of the guar xanthan mixture on the properties of starch films (mechanical and barrier) was examined. Guar gum and xanthan find application in the food industry as stabilizers, thickening agents, binders, because they form viscous solutions at very low concentrations.

Starch films were synthesized from a 1.5% aqueous starch solution, to which glycerol was added as a plasticizer and a guar-xanthan mixture at a concentration of 0.05%, 0.15% and 0.25% (calculated on the weight of the starch). A sample without added guar-xanthan was used as a control. Mechanical (tensile strength and elongation at break) and barrier characteristics (water vapor permeability) were examined.

The obtained starch films were transparent and firm, with uniform thicknesses. The addition of the guar-xanthan mixture affected the decrease in the tensile strength value and the increase in elongation at break. The highest value of the tensile strength was recorded at the minimum concentration of guar xanthan, while the highest values of elongation at break were recorded at the maximum concentrations of guar xanthan. Based on this, it can be concluded that the applied mixture has the effect of a plasticizer and that films with added higher concentrations of the guar-xanthan mixture have improved properties. Starch films have lower values of water vapor permeability than other polysaccharide and protein films, but still these values are very high. The reason for this is the hydrophilic nature of starch. The effect of the guar-xanthan supplement does not significantly change the water vapor permeability value in relation to the values measured for the control sample. The obtained results confirm the justification of the guar-xanthan mixture use in the starch biopolymer films synthesis.

Keywords: biopolymer films, starch, guar-xanthan, properties

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INNOVATIVE PROTOTYPE OF VACUUM DRYER FOR FRUIT DRYING

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It is well known that fruit consumption brings many benefits for human health, since it is rich in bioactive compounds. However, a problem of unavailability of fruit throughout the whole year exists. Thus, in order to prolong the fruit shelf life and its availability through the year, different preservation techniques have been applied. Among all preservation techniques, one of the most important and most commonly applied is drying technique.

Nowadays, the most common type of drying in the food industry is convective drying with hot air. Disadvantage of this type of drying is significant loss of bioactive compounds of fresh fruit during drying, since high temperatures and presence of oxygen are applied. Vacuum drying presents technique by which this problem could be successfully resolved. However, this technique has no wide application, since it requires high investments in equipment and high equipment maintenance costs. The main goal within this research was to apply vacuum drying in order to preserve fruit bioactive compounds during drying and to improve vacuum drying technique in terms of reducing investment and maintenance costs using a simple ejector vacuum system for achieving vacuum.

Within research, innovative vacuum drying prototype has been designed and constructed; preservation of fruit bioactive compounds during drying has been thoroughly analyzed and various statistical methods were applied in order to analyze the obtained results. The possibility of vacuum dryer with ejector system prototype application and also preservation of fruit bioactive compounds using this type of drying has been investigated. Temperature, pressure and drying time were applied as drying parameters, while bioactive compounds such as total phenolic, flavonoid and monomeric anthocyanins content and antioxidant activity were investigated to determine the extent of the preserved bioactive compounds in dried fruit. Beside these analyses, physical characteristics such as water activity, moisture content, total color change and texture were also analyzed in order to obtain the complete picture of the dried fruit characteristics. All the results of investigated analysis of dried fruit, obtained in this research, were processed using different statistical methods. Obtained and statistically analyzed results successfully approved set hypothesis such as the possibility of application of ejector system to the vacuum dryer and successful preservation of bioactive compounds of fruit dried with designed vacuum dryer.

Keywords: *innovative prototype, vacuum drying, fruit*

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PHYSICO-CHEMICAL PROPERTIES OF VACUUM DRIED APRICOT: INFLUENCE OF DIFFERENT PACKAGING MATERIALS

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The main goal of this research was to investigate influence of different packaging materials, in combination with modified atmosphere packaging (MAP), on physico-chemical properties of vacuum dried apricot samples. Fresh apricot was first dried on 55 °C and 20 mbar and then packaged with two different packaging materials. First one was commercial packaging material and the second one was biopolymer packaging material produced in a Laboratory for packaging at the Faculty of Technology Novi Sad, both in combination with MAP. Physico-chemical properties (moisture content, water activity, total phenolic and total flavonoid compounds) were analyzed and compared in fresh apricot sample, in dried apricot samples (before packaging) and in dried apricot samples after one, two and four month (after packaging).

Based on the results obtained for moisture content and water activity for the first and second packaging materials (10.6% and 10.5%; 0.396 and 0.400, respectively) it was noticed that there were no significant differences in dried apricot samples in terms of these two parameters after four months of packaging. The levels of total phenolic and total flavonoids compounds in dried apricot samples were analyzed and it was noticed that levels of these compounds after four months of packaging in the biopolymer packaging material (2.78 mg GAE/100 g and 1.00 mg CE/100 g) were slightly higher compared to levels of these compounds in dried apricot samples analyzed after four months of packaging in commercial packaging material (2.54 mg GAE/100 g and 0.66 mg CE/100 g). These results indicates on potentially slightly better application of biopolymer packaging material for vacuum dried apricot, compared to commercial one, in terms of total phenolic and total flavonoids content.

Keywords: vacuum dried apricot, physico-chemical properties, modified atmosphere packaging, biopolymer, commercial packaging

Acknowledgements: Ministry of Education and Science of the Republic of Serbia (Project TR 31044).

SENSORY CHARACTERISTICS OF FERMENTED CABBAGE OBTAINED ON DIFFERENT CONDITIONS OF FERMENTATION

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In the framework of this research cabbage fermentation was set in industrial conditions in order to compare sensory properties of fermented cabbage samples obtained under different conditions. Three varieties of cabbage (Futoški, Bravo and Tenisiti) were observed. In barrels with fresh cabbage samples were added: salt solution (3.3% NaCl), salt solution (3.3% NaCl) with addition of starter culture; and brine obtained from previous cabbage fermentation. Fermentation lasted 55 days at the temperature round 17 °C.

The main goal of the research was to compare obtained cabbage samples on these fermentation conditions in terms of their main sensory parameters: appearance, color, smell, sourness, salinity, bitterness, appearance of cross section, elasticity and taste. Based on all these parameters for nine fermented cabbage samples, the average grade was calculated. The sample with the highest average grade (4.7/5.0) in terms of sensory properties was cabbage sample fermented on the following conditions: variety Bravo fermented with addition of salt solution (3.3% NaCl); while the sample with the lowest average grade (2.1/5.0) was: variety Tenisiti fermented with addition of brine obtained from previous cabbage fermentation.

Keywords: *cabbage, fermentation, sensory properties*

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PREPARATION AND CHARACTERIZATION OF FIBROIN BASED pH SENSITIVE FILMS

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Bombyx mori silkworm cocoons consist of two main components: sericin and fibroin. Silk fibroin (SF) is protein that has very wide range of applications, because of its remarkable mechanical properties, biocompatibility and can be chemically modified. Aqueous solutions of SF can be formed by dissociation of intermolecular bonds, and without breaking the polypeptide chains. Aqueous solutions can be further processed into different material formats, like films, gels or powders.

In this work, films made of SF aqueous solution were prepared and characterized. SF was obtained from silkworm cocoons from which sericin was previously removed. SF was treated with 9.3M LiBr solvent. The resulting solution was dialyzed against water and films were made by casting the solution in silicone molds and drying them at 40 °C. In order to make films less brittle, plasticizer polyethylene glycol 200 (PEG 200) was added into aqueous solution of SF. SF films with added red cabbage dye were also made. Red cabbage dye is natural pigment used mainly as a food color. Anthocyanins are compounds that contribute to the color of this dye. The pH of red cabbage solution can affect both its color and intensity. In this work, the red cabbage dye was evaluated as a potential pH indicator that can be used for food packaging. The obtained films were characterized for FTIR analysis, water barrier and optical properties, solubility, and tested to color change on different concentrations of base, which imitates food spoilage.

Keywords: silk fibroin, films, red cabbage dye

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THE EFFECT OF REPLACING BEEF FAT WITH OLIVE OIL AND PRODUCTION METHODS ON OXIDATIVE CHANGES IN FERMENTED SAUSAGE (SUCUK)

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Sucuk is a dry fermented sausage produced in Turkey with beef and beef fat with the addition of red pepper, black pepper, cumin and garlic. Sucuk also is a meat product which is more prone to lipid oxidation due to its high fat content during the ripening and storage period. Oxidation of proteins also causes changes in quality of meat, involves the loss of essential amino acids and decreases protein digestibility which affects its nutritional value. α -aminoadipic (AAS) and γ -glutamic semialdehydes (GGS) are considered the main carbonyl products of oxidized proteins and play up as protein oxidation biomarkers in biological systems. Objective of this study was to investigate the effect of replacing beef fat with olive oil and production methods (fermented and heat treated) on lipid oxidation (peroxide value) and protein oxidation (carbonyl content, AAS, GGS) in sucuk (Turkish sausage).

Four different formulations of sausages, containing 4 kg meat each, were prepared. Each treatment was formulated to contain 20% total fat. Control (C) group was consisted of 100% beef fat. Olive oil was replaced with beef fat at levels of 15% (O15), 30% (O30) or 45% (O45). Fermented sausages produced with two different methods; fermented and heat treated.

The results of our study showed that fatty acid composition or type of fat and production method of sucuk have an effect lipid oxidation and protein oxidation and on the formation of specific protein oxidation markers in terms of α -aminoadipic semialdehydes (AAS) and γ -glutamic semialdehydes (GGS).

Keywords: sucuk, lipid oxidation, protein oxidation

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COLOR INVESTIGATION OF COLD PRESSED OILS OF THE LATEST CONFECTIONARY SUNFLOWER HYBRIDS

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The cold pressed oils color depends of the composition and content of pigments in the oil as well as the pressing conditions. The dominant pigments in cold pressed sunflower oils are carotenoids, mainly xanthophyll's (76 to 81%). In addition to carotenoids, sunflower oil also contains chlorophyll in small quantities. The aim of this study was to examine the color parameters using CIE L*a*b* color system as well as transparency of cold pressed oils from the latest confectionary sunflower hybrids. Sunflower hybrid seeds (NS-H-6304, NS-H-6791 and NS-H-6488) were grown in small plot area in 2017. L* values designates the lightness of the sample (100 white, 0 black), a* values indicates redness when positive, greenness when negative and b* values indicates yellowness when positive, blueness when negative. The L* value (lightness) ranged from 23.50±0.01 (sample NS-H-6791) to 25.55±0.01 (sample NS-H-6304). The highest positive b* value (yellowness) was determined for the sample NS-H-6304 and was 9.87±0.01 while the lowest b* value was 9.05±0.03 found in the sample NS-H-6791. B* value found in the sample NS-H-6488 was 9.59±0.03. The highest positive a* value (redness) was found in the sample NS-H-6791 and amounted to 2.87±0.02, while the negative a* value (greenness) was found in the sample NS-H-6304 and amounted to 1.50±0.02. The transparency ranged from 34.30±0.00% (sample NS-H-6791) to 59.43±0.05% (sample NS-H-6304). The a* and b* values determined by instrumental measurement of color are in accordance with the obtained transparency, i.e. expected values of the contents of carotenoids and chlorophyll of cold pressed sunflower oils.

Keywords: sunflower, cold pressed oils, color, CIE L*a*b*, transparency

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INHIBITION OF SALMONELLA ENTERITIDIS GROWTH AND STORAGE STABILITY IN CHICKEN MEAT TREATED WITH BASIL AND ROSEMARY ESSENTIAL OILS ALONE OR IN COMBINATION

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Pathogens from the genus *Salmonella* belong to the group of the most common food poisoning causative agents. The present paper investigated the effect of the basil and rosemary essential oils as well as their combination on the growth of *Salmonella enterica subspecies enterica serovar Enteritidis (Salmonella Enteritidis)* in chicken meat, together with their spoilage protective potential at the two storage temperatures.

Food model experiments included investigation of their effect to accompanying microbial flora on fresh meat, while anti-salmonella activity was evaluated on artificially inoculated raw and thermally processed meat. The tests were performed in two storage conditions, +4 °C (usual meat storage temperature) and +18 °C (room temperature, which favors the spoilage of investigated meat samples and development of food pathogens). Beside evaluation of microbiological status, physico chemical tests relevant as meat quality indicators (pH, color, texture, thiobarbituric acid, cooking loss), as well as sensory evaluation of the studied meat samples were performed. Changes in normal flora pointed to significant effect of both oils against microbial meat spoilage, where various groups were affected by different treatments. In addition, all the treatments reduced the number of salmonella cells in comparison to the control samples. Obtained results on the physico-chemical parameters highlight their applicative value since they showed either beneficial effect either did not caused any notable meat quality changes.

Keywords: essential oil, meat model, basil, rosemary, Salmonella Enteritidis

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TOTAL QUALITY INDEX APPROACH IN THE CULTIVATION OF OYSTER MUSHROOM (*PLEUROTUS OSTREATUS*) GROWN IN CELLULOSE PLANT WASTE

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Pleurotus ostreatus is the second most commonly cultivated and consumed mushroom in the world due to its specific mild taste of oyster in culinary processing. Its fruiting bodies can develop on different and specific cellulose plant wastes used as substrate without specific enrichment material. Advantages of using this type of waste are low costs of their production, usually without high technological investments. On the other side, productivity and quality of fruiting bodies, is affected by the chemical composition of the used substrate. In order to evaluate different quality parameters of fungii one of proposed methods is using total quality index (TQI).

The aim of this study was to investigate the influence of used plant waste on selected quality parameters of *Pleurotus ostreatus* fruiting bodies from the day of harvesting during the storage period of 7 and 14 days in refrigerating conditions (~4 °C).

Based on textural, color, antioxidative and biochemical quality parameters of *Pleurotus ostreatus* fruiting bodies, a mathematical model for calculating a single TQI has been proposed in order to identify optimal substrate concentration in growing selected variety of fungii.

Keywords: oyster mushroom, Pleurotus ostreatus, plant wastes, total quality index, storage period

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PERCEPTION OF SWEETNES INTENSITY AMONG STUDENTS FROM TWO COUNTRIES

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The preference to the sweet taste is innate; however, the perceived intensity of sweetness differs among individuals and was shown to be also related with their dietary patterns. Sucrose produced from sugar beet or sugar cane is the most widely consumed sugar, usually in the form of white granulated sugar. Interesting for the industry as well as for consumers are also brown sugar, invert syrup and lower energy sweeteners. The perception of sweetness intensity was tested with eight samples of different sugars and sweeteners, i.e. white granulated and powdered sugar, brown sugar from sugar beet, invert sugar syrup, high fructose syrup, sorbitol syrup, xylitol and stevia. The concentrations of their water solutions were prepared according to literature data for the relative sweetness to 10 g/100 mL sucrose. Two panels of students from University of Ljubljana and University of Novi Sad evaluated the intensity of sweetness of each sample solution on unstructured 10 cm scale, while the 9-point hedonic scale was employed to assess the level of liking. Results show similar relative differences in perceived sweetness intensity among different sweeteners' solutions, but not the same average intensities for sweetness of a certain sample perceived by the panels from the two countries. The Slovenian panel assessed the solution of high fructose syrup as the most sweet and the solution of brown sugar as the least, while for the Serbian panelists solution of powdered sugar was the most sweet and xylitol solution the least. Significant differences between the two panels were also observed in hedonic scores for the eight samples, where values of Serbian panelists were on average two scores higher than of Slovenians. Sorbitol syrup solution was found among most preferred samples for the both panels. Data of this pilot study suggest that there were differences in perceived sweetness intensity between the panels of students of similar age from the two countries, which may derive from the exposure to sugar in their diets and should be further investigated. Basing on that, appropriate measures in food reformulation may be developed to reduce sugar intake with food.

Keywords: *sweetness intensity, sensory analysis, sugar solutions, students, liking*

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THE INFLUENCE OF FUNCTIONAL INGREDIENTS ON THE TECHNOLOGICAL AND QUALITY PARAMETERS OF WHEAT BREAD

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Bread is a basic food and is part of everyday diet. It provides various nutrients, and is an excellent medium for the integration of functional nutrients. Growing consumer demand for healthy food and integrated nutrient sources is resulting in more and more research on the use of various natural sources as functional ingredients in bakery products. In spite of the positive effects, these components can affect technological properties and shelf-life of final product.

Functional ingredients sugar beet fibre (5%), carob flour (7.5%) and molasses (3%) were added as a partial replacement of flour. Rheological properties of dough were determined with farinograph test. After baking bread, sensory evaluation was carried out, total polyphenol content and microbiological stability were determined.

The addition of sugar beet fibre affected the greatest differences in dough rheological properties and bread quality. Sugar beet fiber increased water absorption capacity and dough development time, and the further decrease in volume and porosity in the baked bread. The total polyphenol content was most pronounced in carob flour, as was the most obvious positive effect on microbiological stability of bread, determined as the absence of visible mould growth. The latter was also affected by the molasses. Dietary fibre has an increased ability to bind water, which consequently increases the potential for microbiological spoilage, which needs to be further controlled. In bakery products, this could be prevented with natural antimicrobials, such as carob flour and molasses.

Keywords: *bread, carob flour, sugar beet fibre, molasses*

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HORSERADISH LYOPHILISATE AS BREAD INGREDIENT - EFFECT ON RHEOLOGICAL AND FERMENTATION PROPERTIES

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Horseradish is used in the culinary for preparation of various foods, to which it adds flavor while it also has as a protective role in preventing spoilage. It is also used in traditional medicine to treat various conditions. For both applications, efficiency is attributed to isothiocyanates - secondary plant metabolites typically found in Brassicaceae family - which have been found to be highly bioactive. In this study, horseradish lyophilisate was used as 2.5% and 5% substitutes for wheat flour for preparation of bread.

Proximate composition of used ingredients was determined. Rheological properties of the dough were evaluated with farinograph test, and fermentation properties with maturograph.

It was found, that horseradish lyophilisate was rich in sugars, proteins and ash when compared to wheat bread. Use of horseradish lyophilisate for dough preparation did not influence water absorption, dough development time and dough softening; however, it prolonged its stability. Additionally, fermentation process and baking time were prolonged. Yeast activity was most likely inhibited by bioactive compounds present in horseradish lyophilisate, which are known for their antimicrobial activity. This activity was also evident during the storage of bread, as a reduced visible growth of moulds. Use of horseradish improved tolerance of dough to mixing procedures, and indicated the influence on the microbiological stability of the bread, which must be further investigated.

Keywords: horseradish, bread, rheology, fermentation

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PHYSICAL AND MECHANICAL PROPERTIES OF DIFFERENT PROTEIN-BASED EDIBLE FILMS

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Films and coatings formulated from various protein sources are receiving great attention in the last decade due to the demand for environmentally-friendly, renewable alternatives for petroleum-based polymeric materials and plastics. The objective of this work was to compare the film-forming ability of potato protein isolate to the ones of well known protein sources, such as whey and soy protein.

To address this objective, films were manufactured at protein concentration of 7% w/w, protein : glycerol ratio of 1 : 0.6 and at temperatures of 90 °C. Films were characterized with respect to thickness, moisture content, film solubility in water, swelling degree and mechanical properties.

Results obtained indicated that potato protein films were characterized with the highest darkness and the most intensive red tones. In comparison to whey and soy protein formulated films, potato protein films exhibited the lowest solubility in water. All the films were characterized with moisture content between 20% and 30%. Potato and soy protein films were thicker which resulted in more than two times higher breaking strengths compared to whey protein films.

Results obtained confirmed that potato protein can be considered a very efficient film-forming material with good mechanical properties.

Keywords: protein isolate, edible films, mechanical properties

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pH VALUE, TEMPERATURE AND COLOR PARAMETERS L*, a*, b* EVALUATION IN RAW PORK DURING 72 HOURS POST-MORTEM

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The quality of raw pork depends from many internal and external factors. The internal factors include genotype and sex of the animal. The most important external factors are type of breeding, feeding and handling with animals. The external factors may include also the effects of various technological processes, such as a slaughtering procedure and chilling systems. These external factors with changes in temperature of the meat may have an effect on the pH value and color of raw meat, two most important factors for determination of the final quality of raw meat. The aim of this study was to investigate changes in temperature, pH and color parameters from the time of slaughter until 72 hours post mortem. One of the goals of the paper is to present the critical points, the time with the most intense changes in the pH and color of the pork in the first 72 hours after slaughter. The paper presents the changes of temperature, pH and color parameters L*, a*, b* of raw pork in the period from 1 to 72 hours *post mortem* muscle *M. longissimus dorsi* and *M. semimembranosus*. Measurements were made in intervals of 1, 2, 3, 24, 48 and 72 hours post mortem. The study comprised over 200 carcasses (Yorkshire Landrace crossbreeds). Slaughtering and bleeding of animals was carried out in the usual way, which is carried out in the slaughter industry. After processing, hot carcasses were refrigerated 24 hours until reached a temperature of +5 °C and lower. Cooling was carried out in chambers with cooled air. Research has shown that the most intense change in pH was during 4 hours *post-mortem* (from 6.25 to 5.71) for *M. longissimus dorsi*. Temperature of muscle was 35.3 °C after one hour *post mortem*, and after 4 hours temperature was 20.5 °C. The pork became darker after the first four hours (L* 45.36-39.11) and after four hours the value of parameter L* increased up to 51.29 (72 hours *post-mortem*).

Keywords: quality of raw pork meat, pH value, meat color



EXTRACTION OF BIOPOLYSACCHARIDES FROM MARINE BIOMASS USING ALTERNATIVE SOLVENTS

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Nowadays, the isolation of valuable compounds from marine sources is highly investigated for different potential applications. The use of marine wastes as a cost-effective abundant source for the extraction of bioactive polymers is highly important due to its economic and environmental benefits. Biopolymers such as chitin (CH), hyaluronic acid (HA) and chondroitin sulfate (CS) have been widely used in different fields including medicine, biotechnology and cosmetics. Thus, it is highly essential to extract them using low-cost and green extraction processes, while maintaining their quality and high purity to perform optimum exploitation of marine wastes. Ionic Liquids (IL) and Deep Eutectic Systems (DES) are investigated as alternative solvents to isolate natural biopolymers. IL are salts in liquid state below 100 °C and DES are mixtures with a lower melting point than the individual components.

Here we present alternative extraction processes that can replace the time-consuming and expensive conventional techniques. Marine by-products including crabs, codfish bones, mussels and tuna fish eyes are used to extract CH, HA and CS. The extraction conditions using alternative solvents are studied to obtain the optimum temperature, contact time and components ratio. The isolated biopolymers are quantified using HPLC and capillary electrophoresis.

Keywords: extraction, biopolymers, ionic liquids, deep eutectic systems, marine waste streams

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EXTRACTION OF POLYPHENOLS FROM OILSEED CAKES BY SUBCRITICAL WATER

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Pumpkin, flax and hemp seed oils have been recommended for human nutrition due to their high content of essential fatty acids. Oil extraction from oilseeds generates tonnes of processing wastes, referred to as oilseed cakes. The oil seed cakes are most often further processed into animal feed due to their high protein and energy contents.

However, these matrices represent low-cost renewable resources of high added-value compounds, such as phenolic acids and flavonoids. In this work, polyphenolic compounds from pumpkin, flax and hemp seed cakes were extracted by subcritical water in a homemade extractor. Operational parameters included the pressure of 20 bars, temperature of 160 °C and convective mass transfer defined by vibration frequency of the extraction vessels of 3 Hz. The sample to solvent ratio was 1 : 30. Total phenolic and total flavonoid contents were determined by spectrophotometric method.

Hemp seed cake extract showed the highest content of total phenolics (36.8±0.41 mg GAE/g dry weight) and total flavonoids (10.3 ±0.01 mg CE/g dry weight), while pumpkin seed cake extract had the lowest content (32.61±1.29 mg GAE/g dry weight and 4.55±0.04 mg CE/g dry weight) of total phenolics and flavonoids, respectively. Presented work demonstrates that subcritical water can be used as safe and green alternative for the extraction of polyphenolic compounds from oilseed cakes, applicable in food and pharmaceutical industries.

Keywords: *oil seed cakes, subcritical water, phenols, flavonoids*

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SUBCRITICAL WATER EXTRACTION OF PHENOLIC COMPOUNDS FROM COCOA BEAN HULLS

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Cocoa bean hull is considered to be an industrial by-product of cocoa processing remaining usually underutilized. This valuable biowaste is mainly used as a fuel, animal feed additive or for fertilizers preparation. Recently, the value of cocoa bean hull has received increasing attention due to high nutritional value and high content of phytochemicals and biocompounds, such as phenolic compounds, dietary fibers, lipids, sugars and proteins. Many researches demonstrate the use of this by-product as a food ingredient or other added-value applications. The aim of this work was to characterize the extracts of cocoa bean hull obtained by subcritical water in terms of polyphenolic content. The hull of a cocoa beans originating from Ghana, Togo, Ivory Coast and Grenada were extracted in a batch type house made extractor at the temperature of 150 °C and pressure of 30 bars for 30 min, maintaining sample to solvent ratio 1:30. Total phenolic and total flavonoid contents were calculated by spectrophotometric method.

The extracts of the cocoa hull from Grenada showed the highest content of total phenolics (45.46±1.88 mg GAE/g dry weight) and total flavonoids (21.82±0.38 mg CE/g dry weight). The lowest contents for total phenolics and flavonoids (37±1.07 mg GAE/g dry weight and 14.21±0.26 mg CE/g dry weight, respectively) were observed in the extracts of the cocoa bean hull from Togo. The findings of this study showed that subcritical water extraction can be used as an environmentally-friendly and safe technique for the valorization of biowaste such as cocoa bean hull.

Keywords: *cocoa bean hull, subcritical water, phenols, flavonoids*

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MICROBIOLOGICAL STABILITY OF COOKED SAUSAGES AS FUNCTION OF REPLACEMENT INORGANIC SALTS WITH NATURAL ADDITIVES

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The cooked sausages are complex colloidal disperse system composed of various dispersions of organic and inorganic substances (proteins, fats, salts, etc.). Special categories from this product group are sausages produced according to organic principles required by EU and USDA regulations. Basic requirements imposed by these regulations are the omission of the direct use of inorganic salts except the sodium chloride. Consequently, the aim of this paper was to replace the inorganic nitrite salt with the natural ones, and the conventional antioxidants and preservatives to replace by natural plant and fruit supplements.

As a substitute for nitrite salts, celery powder and juice mix composed of „1/3 of Kombuha ferment and 2/3 of leaves beet juice” were used. These naturally nitrate salts are reduced to nitrite ones by the nitro reductive starter culture *Staphylococcus carnosus*. As a substitute for antioxidants and preservatives were used acerola powder and fruit mix powder commercially „Superfruit”, as well as essential oil of Tea tree, Cloves, Oregano and Cinnamon. The tested models, after heat treatment, were stored in casings at +4 °C for a period of 45 days. Tests were carried out every 15 days. The total number of aerobic mesophilic microorganisms is determined on a standard agar (SPCA, Oxoid CM0463). The presence of Enterobacteriaceae was determined on purple-red bile-glucose agar (VRBG, Oxoid CM0485). *Escherichia coli* are detected on a violet bile agar medium (VRB), (MUG) (Oxoid CM0978). *Staphylococcus aureus* was detected at Baird-Parker agar (Oxoid CM0275). *Lactobacillus* spp. (LAB) are determined on the MRS agar (De Man, Rogosa, Sharpe) (Oxoid CM0361). Molds and yeasts determined on pink bengal chloramphenic agar (RBC, Oxoid CM0549).

In the tested models, the presence of pathogenic microorganisms was not detected. Essential oils of Oregano and Tea tree have been shown as the strongest inhibitors of the growth of other examined microorganisms. The models to which these essential oils are added have significant ($p \leq 0.001$) lower values of microorganisms, compared to the reference model. The total number of aerobic microorganisms in the reference model, after 45 days was (4.92 ± 0.28 logcfu/g), *Lactobacillus* spp. (3.78 ± 0.19 log cfu/g), and mold and yeasts (2.31 ± 0.13 logcfu/g). However, in the models with Oregano essential oil values were for total aerobes (3.15 ± 0.12 logcfu/g), *Lactobacillus* spp. (2.32 ± 0.08 logcfu/g), and mold and yeasts (1.29 ± 0.04 logcfu/g).

Keywords: cooked sausages, etheric oils, organic foods, food microbiology

INFLUENCE OF DIFFERENT FOOD BY-PRODUCTS ON RHEOLOGICAL BEHAVIOR OF WHOLEGRAIN WHEAT DOUGH

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Consumer awareness of the importance of dietary fiber in human diet influenced the continuing interest in providing dietary options whereby the fibers derived from by-products of plant food processing gained a special position. Bakery products represent a convenient medium for the provision of nutritional requirements for dietary fiber but the implementation of fiber caused some adverse effects on the physico-chemical and rheological properties of these products. Therefore, the objective of this study was to evaluate the effects of dietary fibers from different sources on rheological behavior of dough from wholegrain wheat flour using the Mixolab device.

For this purpose, brewers' spent grain, sugar beet pulp and apple pomace coextruded with corn grits in the ratio of 45:55 were used as a source of fibers. The dough was formed by supplementation of wholegrain wheat flour with selected by-product at level of 30%.

According to obtained results, it can be concluded that selected by-products modified the rheological properties of dough in the different way. Incorporation of by-products resulted in increase of water absorption where the highest value was recorded for dough containing sugar beet pulp. Among the investigated by-products, sugar beet pulp promoted the greatest effect on the mixing and pasting behaviour of the wholegrain wheat flour. Sugar beet pulps induced the highest increase of dough development time, highest value of minimum torque C2 and highest resistance of dough to applied mixing forces. Regarding the parameters which refer to the starch component of the dough system, the influence of by-products was expressed in lowering of maximum torque at point C3 as a consequence of competition for water between starch and fibers.

In addition, the presence of fibers caused the decrease of the gelatinization rate and decrease of starch retrogradation degree. These results indicated that incorporation of brewers' spent grain, sugar beet pulp and apple pomace in the form of coextrudates with corn grits, could have beneficial effect on bread staling.

Keywords: rheological properties, brewers' spent grain, sugar beet pulp, apple pomace, wholegrain wheat flour

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THE EFFECT OF DRYING TREATMENT ON THE RETENTION OF ANTIOXIDATIVE PROPERTIES OF STRAWBERRY

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Strawberries are very flavorful fruit and rich sources of phytonutrients and bioactive compounds. Being constituted of about 90% of humidity, the fresh strawberries are extremely perishable and have a rather limited shelf life in a fresh form. Thus, strawberry is candidate for further processing and preservation. Drying is largely used to preserve fruits and offers dehydrated products that can have an extended life of a year. The objective of this study was to evaluate the effect of different drying treatments on antioxidant properties, total phenolic and anthocyanins content, and vitamin C of the strawberry.

Whole strawberry fruits were dried by air-drying, freeze-drying and osmotically. The samples were analyzed for total phenolic content by Folin-Ciocalteu method and for total anthocyanins content by pH differential method. DPPH assay was applied for determining antioxidant activity of the fresh and dried strawberries and it was expressed as EC₅₀ value. The strawberry preserved by freeze-drying demonstrated significantly better retention of the antioxidant activity, vitamin C, total phenolic and anthocyanins content. The air-drying significantly influenced changes of vitamin C and total anthocyanins content while osmotic dehydration caused significant decrease of all the investigated parameters compared to the fresh strawberry. Generally, anthocyanins and vitamin C in the strawberry fruits have been found more sensitive to drying treatments than phenolic compounds. The results showed that strawberry retain high antioxidant activity after freeze-drying.

Keywords: drying, strawberry, antioxidant property, freeze-drying

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CHARACTERISATION OF FRUIT WINE PRODUCED FROM QUINCE (*CYDONIA OBLONGA*) CONCENTRATED JUICE

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Quince (*Cydonia oblonga*) is often grown fruit in Serbia. The fruits are used for table consumption and processing, where a significant amount of quince is processed into brandy, a distillate of quince fermented juice. The quince concentrated juice (65-70% dry matter) is mainly used in soft drinks industry and it represent a raw material available throughout all year. For this reason, the aim of this research was to evaluate its potential use as a substrate for fruit wine production.

The influence of different production parameters (fermentation temperature and pH) were investigated. The analyses of basic physico-chemical parameters were conducted during fermentation, as well as, in finished wines. The composition of aromatic compounds was also assessed. In order to get a better insight into the quality of produced wines, all obtained samples were subjected to the sensory analysis.

Keywords: quince, wine, concentrated juice

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DETERMINATION OF BISPHENOL A IN BABY BOTTLES AND DRINKING CONTAINERS BY HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY

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Bisphenol A (BPA, 4,4'-isopropylidenediphenol) is a monomer used primarily in the production of polycarbonate plastics and epoxy resins. Polycarbonate plastic is used in a wide variety of digital media products, electrical and electronic equipment, sport safety equipment, reusable food and drink containers, etc. Epoxy resins are used in engineering applications, in paints and adhesives, and in a variety of protective coatings in metal cans for foods, bottle tops, and water supply pipes.

The content of BPA was evaluated in 16 samples (6 baby bottles and 10 drinking containers - can) collecting during 2018/2019. During 2018 the three baby bottles and 8 cans and in 2019 a three baby bottles and 2 cans were collected and analyzed. A simple and economic high-performance liquid chromatography (HPLC-FLD) analytical method was validated for the quantitation of BPA from baby feeding bottles and cans. The separation was performed on a C18 column.

Good linearity was obtained over the concentration range of 0.3-6.0 µg/mL with the regression coefficient (R²) of 0.9998. The limit of detection (LOD) and limit of quantification (LOQ) were 0.01 and 0.03 µg/mL, respectively. The repeatability of the method (%RSD) was between 4 and 6%, while recovery ranged from 104.3 to 109.7%. The extraction of BBA was done in accordance with SRPS EN 13130-1:2008. The method was applied to determine BPA release from baby bottles, performing repeated procedures according to EU and National regulations (SRPS CEN/TS 13130-13 (2008)). The conformity was evaluated according to National and EU requirements (Off. Gazz. SFRJ 26/83 and EU/321/2011 - EU/10/2011). The results show that bisphenol A was not detectable in the all analysed samples.

Keywords: *bisphenol A, baby bottles, drink containers*

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NOVEL RAPID SENSORY PROFILING METHOD AS A TOOL FOR DIFFERENTIATION BETWEEN TRADITIONAL AND COMMERCIAL DRY FERMENTED SAUSAGES

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Traditional products are recognized and highly appreciated by consumers mostly due to their unique and specific sensory properties. Sensory profiling of these products has traditionally been performed by using conventional methods such as QDA[®]. Although conventional descriptive analysis provides detailed information of products with reliable results, it is time consuming, and when it is used for profiling of expensive traditional products it is very cost consuming, as well. Sensory science has developed several new methodologies of sensory profiling as alternatives to traditional sensory descriptive analysis methods. These methods are rapid: can provide quick results with respect to the end user and a reduction of resources; they are more flexible: can be used with semi trained or naïve assessors. Flash Profile (FP) is new rapid sensory descriptive method where each assessor produces individual product profile, by using his/her own list of descriptor for describing them and to evaluate the whole product set comparatively. For each descriptor, samples are ranked based on their intensities on an ordinal scale anchored from „lower” to „higher”.

According to our knowledge, there are a limiting number of articles referencing to sensory analysis of traditional products such as dry fermented sausages, by using Flash Profiling method. The objective of this work was to examine usefulness of FP method for comparison of sensory profiles of commercially available dry fermented sausages with those that are produced in more traditional manner. Panel of expert sensory assessors evaluated six samples, three of commercial and three of traditionally produced dry fermented sausages. Generalized Procrustes Analysis (GPA) method was used for data analysis in order to reduce the scale effects and to obtain a consensus configuration. The average assessors' list contained 12 attributes used for products profiles describing. The obtained results indicated that more than 74% of data variability was explained with first two principal components. On the correlation circle, differently produced samples were on the opposite sides. All three traditionally produced samples can be described with more intense red color, with more pronounced hot taste, smoke aroma and higher chewiness. Apart from these, commercially produced sausages were characterized with more uniform cross section appearance and profound acidic taste and odour on sour.

The obtained results from presented study indicate that Flash Profile method seems to be promising approach for efficient screening of sensory properties of dry fermented sausages and traditional products of meat.



Keywords: sensory profiling, flash profile, textural properties, color properties, traditional products

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TESTING 3D PRINTED STATIC MIXER WITH IMPROVED GEOMETRY

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Static mixers are precision-engineered devices widely used in the food industry. By inserting in tube, they can mix fluids without moving components. Energy used for mixing will result a higher pressure drop along the tube as the effect of friction between fluids and static mixer. Most common commercially available static mixers are helical mixers with clockwise and contra-clockwise mixing elements alternating each other along the length of the object. They have a double effect: radial mixing and flow division. This second effect is more important, especially for fluids with lower dynamic viscosity.

Our research started with using static mixer inside tubular membranes, due to its positive effect to membrane filtration. We were searching for new geometries, with reduced pressure drop along the membrane. Using Computational Fluid Dynamics (CFD) simulation, we noticed that most energy loss is generated on the flat surface perpendicular to flowing direction on the beginning of the mixing element. New geometries are developed, they are good for membrane filtration, however they lost their mixing capabilities, but in most cases, this is not an issue for filtration processes.

In this work 3D printing technology has been used to manufacture static mixers in new improved geometry with similar mixing properties but with reduced effect of pressure drop along the tube. FDM printer was used to make mixers in two most common 3D printing materials: PLA and PETG. For better strength and smoother surface, the mixer was coated with two-component epoxy resin. Frictional pressure drop effect was tested on cross-flow set-up membrane filtration unit with 6.8mm inner diameter tubular membrane.

New optimized geometry resulted a similar pressure drop on the first half of the tested velocity range (up to 1 m/s) but on higher flow rates, there was noticeable pressure drop reduction. 3D printing technology can be used for creating static mixers with improved geometry, but new shape is too complex for manufacturing metallic tools for mass production with injection molding.

Keywords: *3D printing, static mixer*

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PHYSICOCHEMICAL PROPERTIES OF OLIVE OIL OLEOGELS STRUCTURED WITH MONOGLYCERIDES

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Oleogels are semi-solid systems comprising of a liquid oil phase, structured with one or more lipophilic structurants. This structure is thermo-reversible and is achieved by the formation of supramolecular assemblies of oleogelators via non-covalent interactions. Olive oil oleogels could be used to replace saturated fats and add value to meat, dairy or dough products.

The aim of the study was to assess the properties of olive oil oleogels structured with monoglycerides (10%, 15% and 20% w/w) or monoglycerides and 5% phytosterols. Oleogels were stored at ambient (25 °C), refrigeration (5 °C) and freezing (-20 °C) temperatures and samples were analyzed at 0, 1, 3, 7 and 14 days after preparation. The effect of temperature and storage duration on oleogel mechanical properties (hardness, gel strength) was studied by texture analysis. Oleogel melting profile was assessed by differential scanning calorimetry and microstructure was studied by polarized light microscopy.

The 10% monoglycerides oleogel showed the lowest hardness and gel strength, while the 15% and 20% monoglycerides oleogels exhibited similar mechanical properties. The combination of 15% monoglycerides with 5% phytosterols produced gels with the greatest hardness and gel strength. The 20% monoglycerides oleogel had the highest melting point.

The hardness and gel strength of the oleogels decreased during storage time, especially in higher storage temperatures. This was due to the rearrangement of the crystalline structures over time, a finding that was supported by polarized microscopy analysis.

In conclusion, monoglycerides-structured olive oil has the potential to be successfully incorporated in various food systems, as the structured oleogels exhibited mechanical properties similar to animal or hydrogenated fats. This could allow for the substitution of animal fat, the reduction of trans and saturated fat and the production of healthier food products.

Keywords: *oleogel, olive oil, monoglycerides, phytosterols*

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IMPROVEMENT OF THE NUTRITIONAL PROFILE OF GREEK SAUSAGES BY ANIMAL FAT AND NaCl SUBSTITUTION

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Greek traditional sausages, as semi-dry meat products, are characterized by increased shelf life, microbiological safety and distinctive physicochemical and organoleptic properties. However, they are rich in saturated fat and cholesterol which is inconsistent with the guidelines of many health organizations. The use of structured vegetable oils in the form of oleogels, for the substitution of animal and/or saturated fat, is a promising innovative technique, but there are few applications in fermented sausages.

The objective of this study was to produce Greek traditional sausages (20% fat) with a healthier nutritional profile by substituting pork backfat with an olive oil oleogel, structured with 15% monoglycerides and 5% phytosterols. Thus, four treatments were produced: i) control, ii) 33% oleogel substitution, iii) 66% oleogel substitution and iv) 100% oleogel substitution. The samples were analyzed for weight loss, moisture, α_w , pH, total viable counts, lactic acid bacteria, *Micrococcaceae* and *Enterobacteriaceae* at days 0,1,4,7 and 14 after production.

The substitution of pork backfat by oleogel influenced significantly the rate of dehydration, affecting the weight loss, moisture and α_w of the samples, mainly at the 100% substitution level. The oleogel substituted samples had higher pH, moisture and α_w levels, due to lower dehydration during ripening. Total viable counts, lactic acid bacteria and *Enterobacteriaceae* were not affected, while there were different trends in the growth of *Micrococcaceae*.

In conclusion, the produced traditional sausages overall showed improved nutritional characteristics, having lower saturated fat and cholesterol. The olive oil oleogels were successfully incorporated into Greek traditional sausages allowing for the reduction of the pork backfat in the formulation.

Keywords: *Greek traditional sausages, pork back fat, olive oil oleogel*

Acknowledgements: *We acknowledge support of this work by the project “Research Infrastructure on Food Bioprocessing Development and Innovation Exploitation – Food Innovation RI” (MIS 5027222), which is implemented under the Action “Reinforcement of the Research and Innovation Infrastructure”, funded by the Operational Programme “Competitiveness, Entrepreneurship and Innovation” (NSRF 2014-2020) and co-financed by Greece and the European Union (European Regional Development Fund).*

SPOILAGE BACTERIA *Pseudomonas* – PRODUCTION OF HYDROLYTIC ENZYMES AND ABILITY TO GROW AT 5 °C

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Microbial growth and metabolism in food leads to organoleptic spoilage through altering colour, odour and texture of food, and slime or liquid production on the surface. In the dynamics of organoleptic spoilage, initial microbial contamination and storage temperature of food play an important role. It is generally accepted that the most numerous present bacteria in the spoiled food are the ones responsible for its spoilage. As such, psychrotrophic bacteria prevail in refrigerated food, and among them *Pseudomonas* are known as the most common spoilage bacteria. Due to abundance of spoilage *Pseudomonas* species, there are differences in growth dynamic, exploitation of nutrient and spoilage potential. Thus, the aim of our study was to evaluate the ability of selected *Pseudomonas* strains to grow at 5 °C in different food models and to evaluate their spoilage potential as the ability to form different hydrolytic enzymes at 5 °C. Of the four selected *Pseudomonas* strain (*P. fragi* CC151, *P. fragi* CC275, *P. psychrophila* CC291, *P. lactis* CC194), all were capable of producing lipolytic and proteolytic enzymes. In both *P. fragi* strains, lipolytic activity was observed only after 7 days of incubation at 5 °C, while for the other two strains it was visible already after 3 days. When monitoring *Pseudomonas* growth in different food models (minced meat, pasteurized milk, apples) as compared to broth, the best growth was achieved in minced meat model, and the lowest in apple model. Thereby, importance of the nutrients availability and the use of hydrolytic enzymes to exploit more complex nutrient molecules, especially proteins, was shown. Selected *Pseudomonas* strain are able to grow in a variety of food media, and have ability to form hydrolytic enzymes, confirming their universality as refrigerated food spoilers with high potential of persisting in the food storage environment.

Keywords: *food spoilage, Pseudomonas, psychrotrophic, lipolysis, proteolysis*

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Materials Design and Applications





BIOLOGICAL OILS AS A PLATFORM FOR SUSTAINABLE DEVELOPMENT OF POLYMERIC MATERIALS

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Natural oils and greases provide an excellent platform for new materials. They are produced by all living organisms (plants, animals, algae, fish, microbes etc). There are as many oils as living species. Oils are triglycerides i.e., esters of glycerin and fatty acids. The number of known fatty acids is large, but less than thousand. Oils are relatively inexpensive and competitive in price with many petrochemicals. Some advantages of vegetable oil polymers are: they are a renewable resource, have historically stable prices compared to petrochemicals, have high hydrophobicity – providing good water resistance, have better thermo-oxidative stability than corresponding petrochemical products, in many applications, and have excellent electrical properties. Generally, natural raw materials, such as oils, are heterogeneous in structure. The composition depends on the origin of the oil, but it may differ within the same type of oil, depending on the geographical region, climatic conditions, and genetic makeup of the plant. The source of heterogeneity in oils arises from the combination of fatty acids in a triglyceride; they are usually composed of several fatty acids which differ in length, presence of functional groups, number of double bonds, position of double bonds in fatty acids, geometry of double bonds (cis-trans). Heterogeneity requires a different approach to making polymers than working with more structurally uniform petrochemicals. Different strategies may be employed in converting oils to polymers. They include direct polymerization of oil (oxidation, sulfur, Diels Alder, cationic) to final products, functionalization of triglycerides (which are used as monomers for further processing), utilization of fatty acids rather than triglycerides, break-up of fatty acids to oleochemicals (preparation of monomers) and utilization of glycerin for new monomers and polymers. The type of polymer determines its processing and properties. Cross-linked polymers (networks) are usually two-component systems. Their properties depend on crosslink density and chemical structure. Applications for these types include cast resins, foams, adhesives, coatings, and elastomers. They are usually made by the functionalization of triglycerides. Linear polymers (thermoplastics and elastomers) are processable by injection molding and extrusion, as well as from solution. They are made from fatty acids, or their fragments (monomers), generated by different chemical procedures. A new trend in oils is “designer oils” with desired structures prepared by genetic modification of plants, algae etc. Designer oils may have fatty acids of specific lengths and number and type of functional groups, at specific positions, preferably terminal. This presentation discusses chemical methods and structures obtained from biological oils.

Keywords: Biological oils, Designer oils, Converting oils to polymers

POLYCARBONATE-BASED POLYURETHANES AND POLYURETHANE NANOCOMPOSITES

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The presentation is aimed on the preparation and multidisciplinary characterization of polycarbonate-based polyurethanes (PU) and PU nanocomposites. All-aliphatic PU systems were prepared either in bulk or in the form of waterborne PU dispersions. Some of PUs contain D,L-lactide based oligomer resulting in the formation of biodegradable materials. Nanofillers differing in size and shape were used, but the attention is in this case pointed on colloidal silica and montmorillonite particles. Multidisciplinary characterization spans from segmental up to macroscopic levels. For this purpose, the set of spectroscopic, scattering and microscopic techniques combined with characterization of thermal, mechanical and thermomechanical properties were used. PUs and PU nanocomposites prepared in bulk possess outstanding functional properties. Materials made from waterborne PU dispersion differ in functional properties: If dispersions are made from linear PU chains, then PU films can be dissolved in acetone, and thus, they are recyclable. The tensile properties are substantially worse compared to PUs prepared in bulk, however. If PU films are prepared from dispersions containing crosslinked PU chains, tensile properties are better; in some cases they are even comparable with properties of PUs made in bulk. All PUs feature high level of ordering issuing in spherulite formation of micrometer size (Fig. 1). Materials are thermally stable up to 200 °C in the minimum. Due to achievement of miscellaneous functional properties of PU films and PU nanocomposites, tailor-made materials can be prepared.

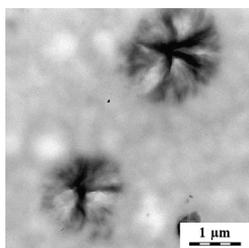


Fig. 1 TEM image of PU film made from waterborne PU dispersion.

Keywords: *Polyurethane, Waterborne dispersion, Nanocomposite, Film, Biodegradable material*

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FOAMING OF ALKALI ACTIVATED MATERIALS

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Alkali-activated materials (hereinafter: AAM), often also called geopolymers, are obtained by process where materials rich with glassy aluminosilicate are dissolved in alkali media. After the dissolution and transport of the components (Al, Si) then through poly-condensation of the Al and Si an aluminosilicate network is formed. Suitable precursors for AAM are different waste materials (fly ash, bottom ash, bio-based ashes, various slags, waste glass), but also thermally activated clay (metakaolin), and natural pozzolan (e.g. volcanic ash). Many products, which could replace traditional building products, such as concrete or ceramic, can be obtained by the alkali activation process. In recent years AAM foams have attracted much attention since they are inorganic, non-combustible material for whose production waste materials can be utilized, and the process takes place at relatively low temperature. Foams are formed by adding either preform organic foams and/or by adding of foaming agent which reacts with a slurry or it decomposes into a gas which remains trapped inside the structure.

To obtain a stable, uniformly porous structure many parameters play role in the process, and have been subject of our investigations. The most important one is type of precursors and the influence of their chemical compositions, part of amorphous phase and particle size on reactivity and mechanical parameters. In our research precursors like fly ash and slags have been used and activated by sodium water glass and NaOH, as well as potassium water glass and KOH. Optimal curing regime for our systems has been defined, i.e. heating at slightly elevated temperature (between 70 and 90 °C). To obtain foamed AAM foaming agents such as Al powders, H₂O₂ or NaOCl have been added and properties like porosity, pore size distribution and mechanical properties of so obtained foams have been studied in order to produce tailor made foams with densities down to 0.3 g/cm³.

Keywords: Alkali activated materials, Foaming agents, Curing, Density, Porosity, Mechanical properties

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RAMAN SPECTROSCOPY FOR CHARACTERISATION OF BRONZE OBJECTS IN DIFFERENT ENVIRONMENTS AND ITS PROTECTION SYSTEMS

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Bronze objects can show different types of patina formation depending on the environment of exposure and/or potential artists' patinations, which can be efficiently characterized by Raman spectroscopy. Furthermore, the method is useful also for characterisation of protection systems applied to bronze in order to prevent further corrosion development.

For the present study outdoor bronze monuments were selected in coastal and urban traffic and non-traffic environment and characterised by Raman spectroscopy, and correlations were made with the results on the laboratory samples containing artificially induced patina. Moreover, selected corrosion inhibitors, such as benzotriazole (BTA), 2-mercaptobenzimidazole (MBI) and 4-metil-1-(p-tolil) imidazole (TMI) were investigated.

The Raman investigation on Mary's pillar (at a highly traffic road in Ljubljana), Prešeren statue (non-traffic environment in Ljubljana) and Tartini statue (coastal area in Piran) revealed that sulphates in the acid rain have the main influence on bronze in Ljubljana, however, the local environment influences the formation of specific forms, while in Piran chlorides in addition to sulphates are the main influential factors due to the vicinity of the Adriatic sea. For all three statues Cu₂S was found on the dark brown areas, indicating that brown artist's patina was applied in the past, possibly already on the original.

Raman study on inhibitors applied to bronze and patinated bronze showed that BTA and MBI form chemical bonds, while TMI bonds only physically, which can influence the efficiency of corrosion inhibition when protecting outside exposed bronze objects.

Keywords: Bronze objects, Environmental exposure, Raman spectroscopy, Corrosion inhibition

Acknowledgements: *The presented research was partially supported by the Slovenian Research Agency (grant number: J7-9404 - Protection of bronze monuments in the changing environment, 2018 - 2021). The author would also like to thank to dr. Tadeja Kosec from Slovenian National Building and Civil Engineering Institute for the fruitful collaboration on the bronze objects characterisation and development of its protection materials.*

NANO-MODIFIED BIO-FIBRES FOR ADVANCED CEMENT MORTARS

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The growing interest in environmentally sustainable building materials, has brought back to the forefront the use of bio-fibres as a cement-based reinforcing agent. However, with the use of natural fibres, difficulties such as low elasticity, poor water absorption properties, biodegradation and variability of the properties of natural fibres occur. This research work examines the use of Posidonia Oceanica algae in 3 different lengths (1, 2 and 4 cm) in cement mortars. Additionally, the fibres were nano-modified by using nanosilica in order to improve the adhesion between the fibres and the paste. Prismatic samples of 40x40x160 mm were produced, and the bending and compressive strength, as well as the porosity -at 28 and 90 days from the mortar production- were measured. In addition, the shrinkage, capillary absorption, dynamic modulus of elasticity as well as the fracture energy of the specimens were calculated. Finally, a microscopic observation of the specimens was carried out with a stereoscope. Based on the results, it appears that the use of Posidonia Oceanica fibres is rather promising. The strength of the specimens in bending is significantly improved and their fracture energy is increased. The length of the fibre is an important parameter for the mortars' properties, especially since it is inversely proportional to the capillary phenomenon and the fracture energy. Adding nanosilica has a long-term positive role on the mortars' strength and fracture energy.

Keywords: Bio-fibres, Advanced cement mortars, Bending and compressive strength, Nanosilica

ZnO/TiO₂ AND Ag/ZnO/TiO₂ CATALYSTS FOR DECOLORIZATION OF MIXTURE OF ORGANIC DYES AND COMPARISON OF THEIR EFFICIENCY WITH TiO₂, ZnO, TiO₂+ZnO, Au/ZnO AND MnO₂

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In this contribution the preparation of ZnO/TiO₂ and Ag/ZnO/TiO₂ photocatalysts is described. The new catalysts were made by modification of anatase nanopowder (TiO₂) in two consecutive steps. First, an ethanolic suspension of TiO₂ was mixed with an ethanolic solution of Zn(NO₃)₂·6H₂O. After 30 minutes of mixing at room temperature, slow solvent evaporation and drying was performed. The received solid mass was grinded in a mortar and thereafter calcined at 500 °C. In the second step, modification of newly prepared ZnO/TiO₂ with silver was done by photoreduction of silver ions (from silver nitrate) at the catalyst surface in ethanolic suspension using natural solar light. Morphological and microchemical characterization of the new catalysts were done with the aid of scanning electron microscopy with energy dispersive spectrometry. The efficiency of new catalysts was compared to the efficiency of TiO₂, ZnO, TiO₂+ZnO (1:1, w/w), Au/ZnO (received earlier by direct current sputter coating of ZnO with gold) and MnO₂ in decolorization of mixture of organic dyes (safranin O, crystal violet and methylene blue) in non-buffered aqueous solution. Ag/ZnO/TiO₂ showed higher efficiency than TiO₂, ZnO/TiO₂ and MnO₂, but Au/ZnO, ZnO and TiO₂+ZnO were more efficient than the earlier mentioned catalysts. The behaviour of TiO₂+ZnO mixture was much closer to ZnO, than TiO₂. Decolorization power of Au/ZnO was additionally tested in presence of different anions.

Keywords: *Organic dyes, Decolorization, Ag/ZnO/TiO₂, ZnO/TiO₂*

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CRACK SELF-HEALING ABILITY OF BIO-MORTAR

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Bacteria can promote a self-healing phenomenon of the existing cracks in a concrete structure, but a relatively high pH value of a cement-based concrete affects negatively the bacteria living time and consequently the self-healing effect. Therefore, a method for encapsulation of bacteria healing agent in a polymer hydrogel system has been successfully established in order to protect the bacteria. The mortar specimens (160×40×40) mm were prepared, according to the EN 196-1 standard, in order to investigate the healing capacity of the employed bacterial colony (*Sporosarcina pasteurii* DSM33) and the effectiveness of the proposed encapsulation methods. The specimens were made systematically in three groups: standard reference mortar samples, mortar samples with addition of hydrogel of the same amount and species as in bio-mortars and bio-mortar samples prepared with bacteria species encapsulated in a hydrogel system surrounded with nutrition, also encapsulated in the hydrogel. After casting, they were cured in a climate chamber controlling the temperature and relative humidity. The prepared samples were characterized by X-ray diffraction (structure), X-ray fluorescence analysis (chemical content), mercury intrusion porosimetry (pore size distribution) as well as by scanning electron microscopy (surface morphology). Microstructure and textural analysis were performed before and after the healing treatment for the sake of comparison. The mortar specimens were notched at mid-span and then pre-cracked in 3-point bending in CMOD-control mode (Crack Mouth Opening Displacement); a target value of 200 µm was set for the crack opening. After unloading, the sorptivity test was performed and then the specimens were kept in a moisture room (90% RH). Their healing ability was measured by crack opening displacement change examined by optical microscopy at intervals of one week, two weeks, one month and two months. A cross-comparison analysis of the obtained results was provided.

Keywords: Self-healing, Mortar, Bacteria, Polymer, Material characterisation

Acknowledgements: The authors are grateful for the support of the Ministry of Education, Science and Technological Development of the Republic of Serbia (project number: III 45008) and for the support of COST Action CA 15202 "Self-healing as prevention repair of concrete structures - SARCOS".

NON-INVASIVE INVESTIGATION OF PAINTING TECHNOLOGY OF PROMINENT 20TH CENTURY PAINTER MIĆA POPOVIĆ

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Mića Popović was one of the most important Yugoslav painters of the 20th century, being a prominent artist of the Informel and the Scenery painting art movements. His pictorial artwork often employed use of non-traditional materials – wood, plastic, plaster and metal – forming three-dimensional elements. Because of his unusual painting technique, artistic importance and the fact that his work had never been subjected to scientific analytical investigation, it was decided to perform analysis on three of his paintings: “*Gvozdenov grafikon I*” (“Gvozden’s graph I”), 1970; “*Barka*” (“Barque”), 1957; and “*U čast Rembrantu IV*” (In Rembrant’s honour IV”), 1983-87. The main goal of the investigation was to determine the painting technique (pigments and binder media) in order to facilitate future restoration and conservation efforts. The analyses were performed in situ at the Central Institute for Conservation, Belgrade (where paintings had been moved for preliminary damage evaluation) using portable instruments from the Laboratory for Materials in Culture Heritage, Faculty of Technology Novi Sad. Analytical techniques used in the investigation were non-invasive: X-Ray Fluorescence Spectrometry (XRF), Fourier-Transformed Mid-Infrared Spectrometry (FTIR) and Optical Microscopy. The results showed that the painting technique varied significantly between the analyzed artworks. “*Gvozdenov grafikon I*” was done using alkyd resin as binder, and titanium white, lithopone, zinc white, carbon black, and iron-based pigment; “*Barka*” contained acrylic emulsion as binder for most part, except for one zone where alkyd resin was detected (in combination with zinc white), and the pigments used were iron-based pigment, lithopone, zinc white, azo β -naphthol red, ultramarine, earth-based pigments; finally, the analysis of “*U čast Rembrantu IV*” revealed combination of alkyd and acrylic binders, and titanium white, lithopone, zinc white, Prussian blue, azo β -naphthol red, iron-based pigment, lead white, and bone black. This approach once again proved the value of collaboration between research laboratory and conservators on revealing crucial facts essential for selection of restoration methodology in order to preserve valuable artworks for future generations.

Keywords: Mića Popović, Culture Heritage, Non-invasive, In situ, Painting

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SELF-CLEANING OF HISTORICAL AND MODERN MINERAL MATERIALS – LABORATORY AND IN SITU APPROACHES

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Aging and deterioration of building materials are specific challenges in maintenance of modern buildings and conservation of historic structures. One of the solutions is active surface protection based on the development of self-cleaning active materials harnessing natural phenomena. This approach could provide preventive and durable surface protection from various contaminants from environment. This paper presents results obtained using novel photocatalytic suspension designed in Laboratory for Materials in Cultural Heritage-Faculty of Technology and scaled up to industrial production (NANOFAS material, GP HGP, Serbia). This paper presents results of laboratory and in situ application and testing of novel self-cleaning material on different mineral facade materials including mortars, renders, bricks, colour layers and facade paints. Aiming to achieve high compatibility of the novel material with mineral surfaces, laboratory models were prepared based on the characterization of systems from real environment, including both historical and modern materials. After compatibility and efficiency proven in laboratory conditions, novel materials were applied and tested on several case study objects, from medieval fortresses (*Bač* and *Petrovaradin* in Serbia), baroque mansion (Dornava Manor in Slovenia), to modern murals freshly painted on residential buildings in the City of Novi Sad. The examination involved comparative chemical-mineralogical, textural, microstructural, microbiological, photocatalytic, durability and colorimetric tests performed before and after the treatment with the protective material, Nanofas, which is based on TiO₂-layer-double hydroxide (LDH). The results confirmed that Nanofas material is of high compatibility with the facade materials involved in this research. The efficiency monitoring was performed continually for 5 years and the obtained results suggest that the Nanofas suspension represents a functional system which allows both historical and modern structures long-term protection from natural contaminants.

Keywords: Self-cleaning, Cultural Heritage, Modern Facades, Nanofas

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SYNTHESIS OF EPOXY BASED COATINGS

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Polymer coatings are well known and effective protection of materials against corrosion. Coating protects construction materials by inhibition of internal or external causes of corrosion creating barrier against corrosion environment. Epoxy resins have a wide application as coatings, adhesives and polymer matrix for composite preparation. After curing, epoxy resins are forming films with excellent chemical resistance and mechanical properties. The use of nanofillers in epoxy resins has gained significant importance for preparation of the advanced composite coatings with improved mechanical properties, anticorrosion and wear resistance with the possibility of tailoring properties of epoxy coatings. The objective of this work was the preparation of reinforced epoxy coatings by addition of titanium nanofiller. For this purpose, pure epoxy coating was prepared by mixing EPODUR 531 epoxy resin and appropriate ratio of hardener (acid anhydride), and then curing at 110°C for 8 hours. Nanocomposite coatings were prepared by adding titanium nanofiller (1; 2 and 5 wt%, calculated on epoxy resin) to the reaction mixture, which was homogenized by treatment in the ultrasonic bath for 10 minutes and cross-linked at 110°C for 8 hours. FTIR spectra of prepared epoxy coatings confirmed molecular structure of obtained materials and quantitative crosslinking. The physico-mechanical properties such as hardness, adhesion and gloss of the epoxy films have also been determined considering the effect of filler content on coating properties. Obtained results illustrated that the addition of nanofiller leads to an increase in the hardness of the coating, with no negative effect on the gloss of the coating. Adhesion test has shown slight enhancement of adhesion with the increase of the filler loading in epoxy coatings. According to the results of physico-mechanical testing it was assessed that obtained epoxy nanocomposite coatings are suitable for applications.

Keywords: *Polymer coating, Epoxy resin, Nanomaterials*

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INTERDISCIPLINARY ASPECT OF EDUCATION AND MATERIALS SCIENCE

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Materials science is an interdisciplinary field based on different technologies and engineering. This field was formed by the integration of chemistry, solid state physics, polymer science, metallurgy and ceramics. Materials science focuses on the relationship between materials' atomic and molecular structures, properties (such as thermal stability, strength, elasticity, conductivity, or biocompatibility) and methods how these materials can be produced into a desired product, having in mind quality considerations and environment. This field is also paying particular attention on new areas such as biomaterials, nanotechnology, smart materials, electronics, energy savings and optical systems. The importance of mathematical modeling in materials science is highly valued. The goal of this work was to introduce the materials science research into high school mathematical teaching and learning. The combination of two disciplines is not common, this multidisciplinary field is not part of the middle school curriculum, even though it combines different aspects of science and offers important knowledge for everyday life. Polymers, adhesives, coatings, rubbers, wood composites, ceramics, solar cells are widely used every day, but they are rarely explored in classrooms. In order to promote interdisciplinary approaches in classrooms and connect mathematical theories to the recommended curriculum, materials science research results could be used as a base. In our proposed paper, we present examples and results from teaching practices and their applications in the classroom.

Keywords: *Kinetics, Modeling, Mathematics, Polymers, Composite materials*

Acknowledgements: *This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (Project III45022).*

THE INFLUENCE OF THE TYPE OF ACTIVATION OF THE MONTMORILLONITE ON THE HYDROLYTIC STABILITY OF UREA-FORMALDEHYDE NANOCOMPOSITE

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In this work the hydrolytic stability of nanocomposites based on urea-formaldehyde resin (UF) and montmorillonite (K10) as formaldehyde scavenger was investigated. Montmorillonite particles were activated by sulfuric acid with and without mechanical mixing using magnetic stirrer. The activation degree was determined using measurements of specific surface (*Sir's method*) and the cation-exchange capacity (CEC). The hydrolytic stability of prepared composites was determined by the mass loss estimation and via free formaldehyde and liberated formaldehyde concentration after composites acid hydrolysis. The amount of free and liberated formaldehyde was 0.06 % and 4.6 % for activated K10 without mixing and 0.12 % and 3.99 % for activated with mixing. The CEC values were 0.145 mol/kg and 0.162 mol/kg for activated K10 without and with mechanical mixing on a magnetic stirrer. The CEC value for inactivated K10 was 0.21 mol/kg. Specific surface measurement indicates that lower values were obtained for inactivated scavenger (74.2 m²/g) compared to the activated. Specific surface area for activated scavenger without mixing was 183 m²/g, but for activated scavenger with mixing it was 167 m²/g. It was assessed that the UF resins with activated formaldehyde scavenger have a smaller content of free formaldehyde (0.06 %) compared to resin with inactivated (0.3603 %). The higher resistance to acidic hydrolysis and lower released formaldehyde percent (1.2252 %) has urea-formaldehyde resin with inactivated montmorillonite powder.

Keywords: *Hydrolytic stability, Formaldehyde scavenger, Montmorillonite, Urea-formaldehyde resin, Free and liberated formaldehyde*

Acknowledgements: *Financial support for this study was granted by the Ministry of Science and Technological Development of the Republic of Serbia (Projects Numbers 45022).*

THERMAL PROPERTIES OF AMINE CURED EPOXY HYBRID MATERIALS WITH DIFFERENT CONTENT OF MONTMORILLONITE

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In this paper, thermal properties of hybrid materials based on epoxy resins obtained by the reaction of diglycidylether bisphenol A (DGEBA) and poly(oxypropylene)diamine (Jeffamine D-230), and with organically modified montmorillonite (Cloisite 15A) having a different mass ratio, were investigated. Five different samples were prepared: First one without montmorillonite (MMT) and the rest as follows with 1 wt. %, 3 wt. %, 5 wt. % and 10 wt. % of MMT content. The thermal decomposition was monitored by simultaneous thermogravimetry and differential scanning calorimetry in flowing air and nitrogen atmospheres. Thermal stability of the sample with 10 wt. % MMT is higher in comparison with the average thermal stability of the other samples. The decomposition in the air takes place in three well-separated ranges, while in flowing nitrogen all processes occur practically, in one step and a narrow temperature range. The decomposition reactions are exothermic in both atmospheres and for all samples. Obtained thermogravimetric data are very similar, except for the sample with 10 wt. % MMT; however, in the air the total enthalpy decomposition change shows a clear dependence on the mass ratio of MMT and with increasing MMT ratio decreases linearly with MMT content. The total exothermic change is significantly lower in flowing nitrogen than in the air. In order to describe the different decomposition mechanisms in nitrogen, an air atmosphere thermobalance coupled with mass spectrometer was employed and the evolved gases were analysed. At the beginning of the decomposition, the signal intensities are in the same range in both atmospheres. At higher temperatures in air, the intensities of the oxidized fragments, i.e. water and carbon(IV) oxide are significantly higher compared to the fragments of the organic matrix. From obtained data, it can be concluded that in the samples with the 10 wt. % of montmorillonite content, the oxidation reactions in the air, are constrained to a certain degree and montmorillonite stabilizes the organic matrix.

Keywords: Epoxy, Montmorillonite, Thermal decomposition, Polymers, Hybrid materials

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STRUCTURING OF ENVIRONMENTALLY FRIENDLY TYRES USING SBR ELASTOMER AND COMBINED ACTIVE NANOFILLERS

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The investigation is focused on the impact of two active fillers (carbon black and synthesized silica (50/0; 35/15; 25/25; 15/35; 0/50; phr)) on the structure and end-use properties (thermal, dynamic-mechanical and mechanical) of elastomeric nanocomposites based on styrene-butadiene rubber (SBR). Scanning Electron Microscopy detected the largest agglomerates in the structure of the elastomeric material filled with 25 phr of carbon black and 25 phr of SiO₂. Modulated Differential Scanning Calorimetry confirmed that the presence of combined active fillers had no influence on the glass transition temperature of prepared SBR nanocomposites. Obtained dynamic-mechanical data on "Payne" effect are important for predicting the tyres tread performances such as the slip resistance on the road on ice and in the wet conditions. It is revealed that the increasing silica loading caused a decrease of the dynamic mechanical loss factor of SBR nanocomposites in the temperature range from 40 to 80 °C which is very significant information for structuring of tyres with reduced rolling resistance and fuel consumption. The highest tensile strength is determined for SBR elastomer with the highest carbon black content.

Keywords: Eco-friendly tires, Styrene-butadiene rubber, Silica, Carbon black, Elastomeric nanocomposites reinforcement

Acknowledgements: The authors wish to thank to the Ministry of Education, Science and Technological Development of the Republic of Serbia (project III45022) for financial support.

THE USE OF MATHEMATICAL MODELING FOR DETERMINATION OF OPTIMAL PMMA/NANOOXIDE MATERIAL DESIGN

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Poly(methyl methacrylate) (PMMA) is an amorphous thermoplastic polymer which has good mechanical properties, high impact resistance, dimensional stability and transparency, and it has found various applications in the field of optics, microelectronics, food packaging, medicine, dentistry, cosmetics, and etc. However, the wide range of PMMA usage is influenced by low thermal stability under high-temperature conditions. In this work, PMMA hybrid materials were prepared to contain 1 vol % of silica, alumina or titania, in order to evaluate the influence of oxide nanoparticles on the thermal stability and degradation kinetics of the obtained samples. A detailed study of adequate material design and assessment of the high-temperature range for PMMA processing and application was performed by simultaneous thermogravimetry and differential scanning calorimetry. The proposed mathematical model that includes all three heating rates in one minimizing a function, well fitted TGA data, and enabled the calculation of thermal decomposition kinetic parameters of novel PMMA materials.

Keywords: *PMMA hybrid materials, Oxide nanoparticles, Mathematical modeling, Thermal stability and degradation*

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MEASURING THE DRYING SENSITIVITY OF CLAYS: A REVIEW

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It is well known that intrinsic parameters such as mineralogical composition, clay minerals content, grain distribution, packing, porosity, as well as forming method and raw material aging are affecting on the easiness of the drying process. Often many of previously mentioned parameters are inter-dependently related to each other. That is the reason why attempts to relate the drying sensitivity to one or more of the above mentioned intrinsic parameters have encountered limited success. A logical solution was to relate the drying crack with the term "drying sensitivity". By doing that the evaluation of the cracking tendency is linked with the easiness of the drying process in each proposed method. The main objective of this review was to present five methods (Bigot, Ratzenberger, Piltz, Hermansson and Varlamov) which are usually used for qualitatively estimation of the drying sensitivity of clays and consequently the prediction of the drying behavior. After a literature check it was found that only Bigot and Ratzenberg drying sensitivity index were compared. The second task was to estimate the clay drying behavior using all five methods. The results have confirmed that the most suitable conclusion about the drying behavior of the tested clay is obtained when results from Bigot, Piltz and Varlamov method are available.

Keywords: *Drying sensitivity, Clays, Bigot, Varlamov, Piltz, TG curve*

Acknowledgements: *This paper was realized under the project III 45008 which was financed by ministry of education and science of Serbia.*

ANTICORROSION ADDITIVES – SPRAY DRYER PRODUCTION AND INFLUENCE OF POST-PRODUCTION TREATMENT ON THE SIZE DISTRIBUTION

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Growing trends in the powder and liquid coating industries demand development of additional coating components with the specific functional targets, such as the anticorrosion properties for the metal coatings for example. The product development of such additives requires production optimization in order to obtain a component compatible with the basic powder and liquid coating formulations. This study has a focus on the particles size distribution. The size distribution measurements were performed with the LUMiSizer (LUM GmbH, Berlin), while the tested additive was produced by Niro EX Minor spray dryer. The examined substances were anticorrosion additives based on silica spheres and kaolin nano-tubes.

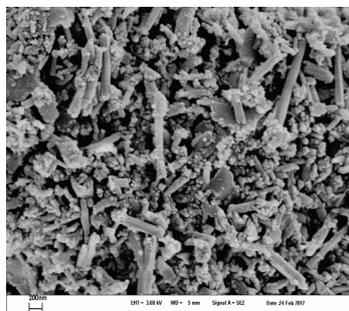


Fig. 1. Scanning electron microscopy (SEM) photo of kaolin nano-tubes.

The production governed with the spray dryer has some limitations, for instance the particles size of the loading material. In this work we show that, depending on the nozzle diameter, the particle size of the final product can be controlled, until some certain point. However, the final product properties are directly influenced by the loading parameters (temperature, pressure, pump speed etc.). Too big additive particles can produce some mechanical instabilities after mixing in the powder and liquid coatings. In order to decrease the particles size, two processes can be extra included. The first approach is the additional milling of the final additive and the second is the cleansing of the final additive in order to remove the remaining inhibitors crystals from the particles surfaces. Finally, production of the powder additives with the acceptable physical properties requires the optimisation of all production parameters at each production scale.

Keywords: *Coating industries, Anticorrosion additives, Particles size distribution*



FUNCTIONAL BIO-BASED MATERIALS IN WOUND DRESSING

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Wound healing is a dynamic and complicated process that requires a suitable environment to promote the healing process. The damaged skin needs to be immediately covered with a dressing capable of restoring tissue integrity and its function. Therefore, a medical dressing has to be sterile, non-toxic and non-allergic, protect against bacterial infection, enhance epidermal migration, allow gas exchange between wounded tissue and environment, should be non-adherent to the wound and easy to remove after healing. For this purpose, nano-structured highly skin-compatible non-woven textiles based on biopolyesters polyhydroxyalkanoates (PHAs) combine with natural antimicrobial agents (chitin-lignin nanocomplexes) using electrospinning processing have been developed. Thanks to the use of electrical forces, based on liquid atomization, electrospinning enables the production of short to continuous fibers or particles and the structures with variable density. The controlled release of active ingredients has been achieved through a porous structure of electrospun nanofiber meshes.

Keywords: *Biopolyesters, Electrospinning, Nanofibre meshes*

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BIOLOGICAL AND ELECTROCATALYTIC APPLICATIONS OF Ag, Au, AND Cu NANOPARTICLES, AND THEIR POLYANILINE BASED COMPOSITES

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As the field of nanotechnology is growing novel nanomaterials became significantly important for various application. Among them, metal nanoparticles, such as Ag, Au, and Cu have been widely investigated. These nanoparticles show very different physicochemical properties compared to the bulk counterparts which can be attributed to their high surface to volume ratio. The unique tunability of their plasmon resonance properties through variation of their size, shape, composition, and medium allows chemists to design nanostructures for specific applications. Due to these unique properties, they make an excellent candidate for biomedical and catalytic applications since the variety of biological/catalytic processes occur at nanometer scales. This work presents the Au, Ag and Cu nanoparticles of various sizes (5-40 nm) and shapes (spheres, rods, prisms) currently investigated in the field of microbiology, optics, and catalysis. It is focused on individual and particular polymer molecules functionalized nanoparticles which are prevented from agglomeration and oxidation, and also have improved physicochemical characteristics and functions. As biocompatible material, that also displays good conductivity and electrochemical activity, polyaniline polymer was a good choice to form functional nanocomposites with metal nanoparticles by in-situ polymerization processes. Relationships correlating their physicochemical properties (size, shape, chemical functionality, structures, and conductivity), cell models, and voltammetric response are discussed on the basis of data analysis. The antimicrobial applications of these systems - interaction with biological entities (*E. coli*, *S. aureus*, and *C. albicans*), and electrocatalytic applications - electrochemical oxygen reduction reaction are presented. Both activities, the electrochemical and antimicrobial of these materials, regardless of metal mass fraction, size and shape of nanoparticles, and polyaniline morphology are mostly related to the particles' surface effects, i.e., reactive (111) crystallographic planes present on their surface. These nanocomposites proved to be promising Pt-free oxygen reduction reaction catalysts and antimicrobial agents.

Keywords: Metal nanoparticles, Nanocomposites, Antimicrobial agents, Electrocatalysts

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DIELECTRIC CHARACTERIZATION OF ENGINE COOLANTS

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In an internal combustion engine, only one-third of the total energy produced works to propel the vehicle forward, one-third is removed as heat energy by the exhaust system and the remaining one-third of heat energy produced is taken away by the engine coolant. In addition coolant protects engine from freezing while defending components against corrosion. Nearly all engines use mix of ethylene glycol and water to provide freeze protection. In addition to the base fluid, there are a small amount of other ingredients including corrosion inhibitors, antifoams, dyes and other additives. While these other ingredients make up only a small fraction of the coolant, they are what differentiate one coolant from another. The electromagnetic specification of liquids provide useful information to improve design, processing, quality and control of product. The selection of dielectric measuring technique is basically associated with the tested material and depends onto several factors, e.g. frequency, temperature, required accuracy, sample size. Using the coaxial probe method, dielectric properties of different varieties of engine coolants have been examined at several temperatures between 20 and 50 degrees Celsius, and at various dilution ratio. The applied frequency range goes from 200 to 2400 MHz in steps of 100 MHz. Differences were also found in the dielectric properties of engine cooling fluids produced by different technologies.

Keywords: Engine coolant, Dielectric properties, Coaxial probe method

NON-INVASIVE STUDIES OF EARLY BRONZE AGE POTTERY FRAGMENTS EXAVATED AT THE SITE MEANISTE IN RANUTOVAC, SERBIA

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The examination of archaeological ceramic fragments is performed to reveal the possible source of the raw material and the technology of preparation. The Early Bronze Age ceramic fragments were excavated at the archaeological site Meaniste in the Ranutovac village near Vranje. The studies of fourteen samples with well-known archaeological context were analyzed using noninvasive analytical approach. The elemental composition of the ceramic fragments was determined by the pEDXRF spectrometric technique and compared with the elemental composition of the clay from the pit nearby the archaeological site. Based on the obtained results it can be assumed that the local raw material is most likely to be used for the preparation of the investigated samples. Additional examinations for double-fragmentation shreds are required. The rFTIR spectrometric technique was used to estimate the temperature of thermal treatment of clay during the production of the object. According to the results it can be assumed that most of the analyzed fragments were thermally treated at 800 °C, while some of the potsherds were additionally fired at a higher temperature (900 - 1000 °C).

Keywords: EDXRF, FTIR, Pottery samples

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MESOPOROUS SILICA – HYDROXYAPATITE COMPOSITES AS CARRIERS OF SOIL NUTRIENTS

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Mesoporous silica-based materials exhibit various favorable characteristics, such as thermal stability, biocompatibility, high porosity and large specific surface area, which makes them suitable for numerous industrial, environmental and biomedical applications. Their capability to load, retain and release cargo molecules in a controlled manner makes them promising carriers of different chemicals – drugs, soil and plant nutrients, pesticides, etc.

Potential for controlled release of nutrients and pesticides is a particularly attractive research area regarding the application of mesoporous silica nanoparticles (MSNs) in agriculture.

The aim of this work is to synthesize a series of composite materials based on mesoporous silica and hydroxyapatite (MSN-HA) for applications as biodegradable carriers of agriculture-important molecules. Novel materials are characterized by scanning electron microscopy, nitrogen sorption analyses, powder X-ray crystallography and infrared spectroscopy. Initial experimental results show that the MSN-HA composites are highly porous, have a large specific surface area, which therefore exhibit a large storage capacity for agricultural nutrients that can be gradually released into soil or plants.

Keywords: Composite material, Mesoporous silica, Hybrid fertilizer

Acknowledgements: The authors are grateful to the Ministry of Science and Technological Development of the Republic of Serbia for financial support (Grant III44006).

RE-SYNTHESIZING CATHODE MATERIAL FROM SPENT LI-ION BATTERIES AND ITS EXAMINATION IN AN AQUEOUS SOLUTION OF NaNO_3

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Li-ion batteries contains of: Al cathode with another cathode material (Li, Co, Mn, Ni, Fe) on the top of it, a Cu anode with graphite and Al crust above them, as well as an organic toxic electrolyte. The aim of this work was to find a re-synthesis method of the cathode material and test it in NaNO_3 aqueous solution, in order to investigate the possible foundations of more environmentally friendly batteries.

Five used and depleted batteries (Nokia BL-5C) were cut by a hacksaw. After removal of the crust, cathode material was obtained by dissolving of the Al in 10 M NaOH. Consequently, 6.262 g of obtained material were dissolved in 250 ml 2 M HNO_3 . The solution was analyzed by atomic absorption spectrometry, in accordance with erstwhile studies founded in the literature. Chemical formula of prepared material is $\text{Li}_{0.8}\text{Co}_{0.415}\text{Mn}_{0.435}\text{Ni}_{0.15}\text{O}_2$. A re-synthesis of the obtained cathode material was done through citrate-gel combustion. Volume of the solution was 50 ml. Firstly, pH was adjusted to 7 using 17.05 ml NH_3 . In the next step, 15.96 g of citric acid was added into solution of the cathode material to adjust molar ratio on $n(\text{NO}_3)/n(\text{CA})=3.6$. Finally, a further $\text{Li}:\sum\text{M}=1:1$, 18.6 ml of 0.1094 M LiNO_3 was added.

A layered structure of the re-synthesized material was confirmed by X-ray diffraction (XRD). The morphology of the material was investigated by SEM and showed presence of the nanosized particles which are favorable for a successful intercalation process. The capability of the layered structure to intercalate Na^+ ions in NaNO_3 was confirmed by Cyclic Voltammetry. The Pt electrode was counter as the saturated calomel electrode was a reference when the $\text{Li}(\text{Co-Mn-Ni})\text{O}_2$ was pasted to the working electrode. The capacities were found to be 36.9 (charge) and 27.9 mAh g^{-1} (discharge) which is satisfying capacity for application in an aqueous electrolytic medium.

Layered $\text{Li}(\text{Co-Mn-Ni})\text{O}_2$ structure was successfully re-synthesized by the citrate-combustion method. The resynthesized material was found to be capable of intercalating Na^+ ions in an aqueous solution of NaNO_3 , which makes this material suitable as cathode for sodium-based aqueous rechargeable batteries.

Keywords: Recycling, Li-ion batteries, Na-ion batteries, Aqueous electrolyte

Acknowledgements: This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia; Project III45014.

INFLUENCE OF BRICK CLAY CHARACTERISTICS TO THE QUALITY OF ADOBE CLAY BRICKS

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In this study, 139 heavy clay samples from Serbia are tested. The effects of macro-oxides content, granulometry analysis (contents of sand-, alevrolite-, and clay-sized particles), remains on the 0.063 mm sieve, and contents of total carbonates on the characteristics of raw and adobe clay bricks were mathematically analysed in this research. The observed parameters were characteristics of wet raw material (shaping moist, plasticity coefficient according to the method by Pfefferkorn, and drying sensitivity following Bigot) and adobe clay bricks (drying shrinkage and compressive strength). Correlations and principal component analysis were done to sum the important effects of inputs over the outputs. It was revealed that Al₂O₃ mostly influenced shaping moist, Na₂O and K₂O were influential to dry compressive strength and drying until the critical point. Increase in clay-sized particles induced higher shrinkage and compressive strength. A rise in the quantity of allevrolite-sized particles decreased drying sensitivity, drying shrinkage and compressive strength.

The analysis is done as the preliminary check of the data, before building Artificial neural networks.

Keywords: Adobe clay brick, Correlations, Plasticity, Drying sensitivity

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INVESTIGATION OF SOLVATOCHROMIC PROPERTIES OF SELECTED NEWLY SYNTHESIZED AZO-DYES DERIVATIVES

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In this paper, solvatochromic properties and intermolecular interactions of six newly synthesized structural isomers of arylazo pyridone dyes, 5-(substituted phenylazo)-3-cyano-6-hydroxy-4-methyl-2-pyridones, were obtained. These compounds belong to a group of dispersed and hydrophobic colors with good qualities, which show yellow or yellow-greenish shades. They are very intense colors with high molar extinction coefficient and they are stable during washing. Mostly, they are used for staining hydrophobic fibers and fabrics, for the production of reactive dyes and as stabilizers in the solar cell production. Further potential application of these compounds depends on the presence of intermolecular interactions. Due to that reason, these interactions are the subject of study in this paper. As a suitable method for this research, UV/VIS spectroscopy was used. Absorption spectra of all compounds were recorded in fifteen solvents of different properties in the range from 300 to 600 nm. All compounds show one intense absorption maximum in the range from 400 to 500 nm, attributed to the hydrazone form. Position of absorption maxima depends on the interactions of tested compound with the surrounding medium. Observing the changes in the position of the absorption maxima in different solvents and using correlation analysis by the method of multiple linear correlations of solvatochromic energies, we obtain quantitative information about influence of the existing interactions on the solvatochromic properties of investigated compounds.

Keywords: *Arylazo pyridone dyes, Solvatochromic properties, Intermolecular interactions, Absorption spectra*

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MINERAL BY-PRODUCTS AS ADDITIVES IN NEWLY SYNTHESIZED CEMENT

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On daily basis large quantities of mineral by-products are allocated from industrial plants in Serbia and disposed on landfills, with possible negative impacts on the environment including occupation of large areas, pollution of surface and ground water and of air and soil. These by-products could be reused as raw materials, especially in construction and production of building materials. This approach could provide useful and economically profitable products which is the purpose of this research. In order to examine the possibility of adding mineral by-products in cements as well as their impact on the general characteristics of the newly synthesized belite cements, it is necessary to perform in-depth characterization of materials. Through these investigations, the standard methods were used to analyze mineral, chemical and textural properties of the samples of ash from the Thermal Power Plant Kostolac B, metallurgical slag from the Smeltery in Bor and granulated blast-furnace slag from Železara in Smederevo (HBIS Group Iron & Steel). This research aims to evaluate solutions for mineral by-products and their reuse as secondary raw materials for environmental-friendly cements and adapt technological processes and thermal regimes traditionally used for the production of cements, decreasing production costs. The results of the research are presented in this paper.

Keywords: Characterization, Ash, Metallurgical slag, Granulated blast-furnace slag, Cement

Acknowledgements: Laboratory for materials in cultural heritage (Faculty of Technology Novi Sad), Laboratory of Crystallography (Faculty of Mining and Geology Belgrade), Laboratory for scanning electron microscopy with energy-dispersive spectrometer (Faculty of Mining and Geology Belgrade) and Laboratory for solid fuels (Mining institute).



FAR INFRARED RADIATION BIO-CERAMIC FIBERS A NEW GENERATION OF FUNCTIONAL TEXTILES

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Far-Infrared Radiation (FIR) textiles are part of the new category of functional and smart textiles for more comfort and user well-being and to improve blood flow in targeted areas of the body. FIR textiles are often made by addition of nano or micro sized ceramic powder to polymers prior spinning. Bio-ceramic powders zirconium, iron oxide, silicon carbide and germanium-based compounds can be incorporated into the structure of textiles. In this context, the concept of the Far-Infrared Radiation bioceramic-matrix textile was studied. This structure captures the positive contributions of the sun, far-infrared, which strengthen our muscular activity and our tone, to recover some of the loss of energy that escapes from our body in the form of heat and wavelengths during the effort and restores it. Far Infrared Radiation Fiber (RIL) is called ceramic fiber because the ceramic is built inside the fiber, which allows the fabric to absorb the body heat and then release a far infrared ray of 8 to 12 μ m wavelength. This is very similar with the release wavelength of the human body (9.36 μ m), the far infrared ray can thus penetrate deep into the skin to make the water molecule resonate and give heat that will allow blood vessels to dilate thermal stimulation of blood circulation. In this work the uses of ceramic fibers in textile structures are also discussed (the advantages of ceramic-textile matrix, the methods of preparation of these materials and the mechanism of Far Infrared radiation effect).

Keywords: Ceramic fiber, Comfort, Bioceramic, Functional textile

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PROTIC IONIC LIQUIDS IN CONSERVATION OF THE PAPER HERITAGE ARTEFACTS

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Protic ionic liquids (PILs) are one of the promising alternatives to replace conventional organic solvents. Due to their unique properties, namely low volatility, high thermal stability and conductivity, ionic liquids have been investigated for the purpose of greening various processes in order to reduce or completely eliminate the use of commercial solvents. In this work, new protic ionic liquids with 1-ammonium-2-propanol cation and six different anions: acetate (Ac), trifluoroacetate (TFA), trichloroacetate (TCIA), chloroacetate (CIA), 3-chloropropionate (3-CIP) and 4-chlorobutyrate (4-CIB) were prepared and fully characterized in order to study their antimicrobial activity and possible application for fungi and bacteria removal from deteriorated paper heritage artefacts. Densities of ionic liquids and their diluted aqueous solutions have been measured over the whole composition range at selected temperatures from 293.3 to 353.15 K for pure ionic liquids, and from 293.15 to 313.15 K for diluted aqueous solutions and at atmospheric pressure. Influence of specific functional groups in the anion structure on densities and volumetric properties as well as nature of interactions in aqueous solutions have been discussed in the terms of their antimicrobial activity and toxicity. The results indicated the possibility for molecular design of new ionic liquids with strong inhibition properties against the specific bacterial, mould and yeast strains. The significant antimicrobial properties observed in this research suggest that studied PILs may have potential applications in the paper art and artefact cleaning and conservation replacing thus, conventional solvents and organic substances that are toxic for humans and the environment.

Keywords: Protic ionic liquids, Green chemistry, Antimicrobial activity, Paper, Cultural heritage

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CORRELATION OF ED-XRF AND ICP-OES METHODS APPLIED IN CHEMICAL CHARACTERIZATION OF CEMENTITIOUS MATERIALS

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The aim of the investigation was to provide elaborate analysis of the accuracy of energy-dispersive X-ray fluorescence (ED-XRF) method for determination of the major and trace elements present in cement binders. In order to prove that ED-XRF, which is rapid and non-destructive procedure, can successfully substitute inductively coupled plasma - optical emission spectrometry (ICP-OES), the performances of both techniques were compared and results were correlated. The instrumental analyses were employed to assess variations in the chemical composition of construction materials designed on the basis of cement and mineral additives. Coal combustion ash was utilized as pozzolanic additive. The binders' mix-design was supplemented with sorptive clays to prevent migration of toxic elements from fly ash. Eleven binders containing cement CEM I 42.5, fly ash, zeolite and/or bentonite additions were prepared in accordance with chemometric experimental design rules. The chemical analyses included identification and quantification of 8 major (Si, Al, Fe, Ca, Mg, S, Na, K) and 6 trace elements (Cr, Zn, Cu, As, Ni, Pb). The validation procedure was conducted with respect to several statistical parameters including: selectivity, working range, repeatability, accuracy, reproducibility, linearity, limit of detection and limit of quantification. Certified reference materials and reference materials were employed in the validation procedure, primarily for establishing the working range. Limit of detection (LOD) and limit of quantification (LOQ) are important performance characteristics in validation. Calculation from the standard deviation of the blank was used for estimation of detection and quantification limit. Software data was used for LOD testing. LoQ was scrutinized by employing of C_{Cal}/C_{CRM} ratio, i.e. quotient of two concentrations of analyte - one for the calculated and the other for the certified or reference mass fraction. Nord test was applied in evaluation of measurement uncertainty. It was concluded that higher concentrations of elements induce improved repeatability and reproducibility. Also, it was highlighted that ED-XRF can be reasonably employed instead of ICP-OES for identification and quantification of major and trace elements in cement based construction materials.

Keywords: *Chemical analysis, Analytical modeling, Cement binders, Clay, Fly ash*

Acknowledgements: *This investigation was supported by Serbian Ministry of Education, Science and Technological Development and it was conducted under the project III 45008.*

POSSIBILITY OF CONSTRUCTION MATERIAL PROTECTION IN COASTAL CONDITIONS USING NEW ENVIRONMENTALLY FRIENDLY INHIBITORS

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Investigation of the corrosion process in the coastal regions is of great importance to human society since about half of the world's population lives in these regions and the industrialization of developing countries tends to concentrate production plants close to the sea. However, surprisingly little attention has been paid to the action of marine chlorides and the research was limited to just a small number of studies. Thus, the aim of this study was to test the inhibition properties of the selected ionic liquids for protecting construction materials against corrosion in coastal conditions as ecologically friendly compound. Newly synthesized ionic liquid was studied as a corrosion inhibitor of selected construction materials in coastal conditions, applying gravimetric and electrochemical methods. Investigation was performed in order to analyze the influence of different condition parameters and the applied ionic liquid concentration on its inhibition efficiency, and the mechanisms of material corrosion in marine atmospheres. The obtained results showed that the effect of the investigated ionic liquid in selected construction materials protection is manifest through the increase of the total resistance and to less damaged construction materials surfaces compared with the basic solution.

Keywords: Corrosion inhibition, Ionic liquids, Construction materials, Coastal conditions

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PHOTOCATALYTIC COATING BASED ON ILITE CLAY/TiO₂ COMPOSITE

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Natural clays have attracted great attention in regard to synthesis of clay-based photocatalysts, due to their layer structure, large specific surface area and remarkable adsorption capacity. The aim of this work was to obtain photocatalytic coating based on titanium-dioxide and ilite clay. TiO₂ (Degussa Company) was used as the active component while ilite clay acts as a support. In order to initiate the interaction between the active component and the support, TiO₂ in content of 3 and 10 wt% was impregnated by mechanical activation in attritor and planetary mill. The obtained ilite clay/TiO₂ suspensions were applied and investigated on three types of substrates: non-porous (glass), porous (clay roofing tile) and highly porous (clay-fly ash composite). The system ilite clay/10wt% TiO₂ impregnated in attritor mill showed better characteristics in the aspect of grain size, morphology, photocatalytic efficiency and durability of coatings. Photocatalytic efficiency of the synthesized coating was assessed by photodegradation of the model pollutant Rodamin B performed before and after durability tests. The investigation showed that photocatalytic activity of the ilite clay/TiO₂ coating generally depends on impregnation conditions and applied TiO₂ content.

Keywords: TiO₂, Clay, Photocatalysis, Photocatalytic coatings, Smart materials

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ADHESION OF BACTERIA ON PROTEIN-TERMINATING POLYELECTROLYTE MULTILAYERS

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Adhesion of bacteria on various material surfaces can be reduced by applying different types of coatings. In our study silica surface on which polyelectrolyte multilayers were formed, with various proteins being the terminating layer, was used as a material surface. We examined three different types of proteins: bovine serum albumin (BSA), lysozyme and glucanase. Adhesion experiments were performed with two bacterial strains: *Escherichia coli* and *Pseudomonas aeruginosa*. For the characterization of the surface prior bacterial adhesion the surface roughness and hydrophobicity were determined. Profilometry, AFM and tensiometry were used for that purpose. Additionally, the surface charge was estimated by the zeta potential measurements of silica particles which were covered with polyelectrolyte and protein layers. The extent of adhered bacteria was examined using scanning electron microscopy. It was shown that in the case of BSA and glucanase terminating layer specific protein-bacteria interactions are dominant. On the other hand, in the case where lysozyme was the outermost layer the lysis takes place. It was assessed that in the examined cases the adhesion strongly depends on the protein specificity while surface physical properties do not play the key role in the adhesion process.

Keywords: Bacterial adhesion, E. coli, P. aeruginosa, Polyelectrolyte multilayers, Proteins

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EVALUATION OF THE PROCESS PARAMETERS OF CERAMIC PRODUCED FROM CLAY AND C&DW

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Construction and demolition waste (C&DW) produced in Republic of North Macedonia presents a major economic and environmental problem, because all C&DW ends up in the local dumps or is disposed of in landfill. The aim of this research, was the determination of the physical and chemical properties of C&DW obtained at Macedonian demolition sites and further to investigate the possibility of waste bricks from this C&DW for the production of clay based ceramics by optimization of the main process parameters. Determination of the chemical composition of the waste demolition brick by means of X-ray fluorescence showed that it consisted of different oxides such as SiO₂, Al₂O₃, Fe₂O₃ and CaO and minor contents of alkaline and earth alkaline oxides. SEM analysis was applied for investigation of the morphology of the particles. Specific weight and particle size distribution were obtained and discussed. Production of the ceramics were conducted through consolidation of the clay and waste demolition brick with evaluation of the main process parameters such as: sintering temperature (ST), content of waste demolition brick (WDB) and isothermal period at the final temperature (IP). The porosity and bending strength of the clay based compacts were the response function. The optimization was performed through implementation of main effect plots, Pareto charts and 3D surface method using “Statgraphics Centurion” software package. The final model equations for porosity and bending strength dependence of the main process parameters have been obtained and are presented. Microstructure of the ceramics determined by SEM was also reported and discussed.

Key words: Construction and demolition waste, Clay, Process parameters, Bending strength, Porosity

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THERMAL STABILITY AND DEGRADATION KINETICS OF ALIPHATIC POLYURETHANE/NANOSILICA COMPOSITES

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In this work, a series of aliphatic polycarbonate-based polyurethane (PU) hybrid materials with different nanosilica content were synthesized by one-step preparation procedure different content of nanosilica during one-step preparation procedure. The study of filler effect on thermal stability and thermal degradation mechanism of obtained materials was carried out by simultaneous thermogravimetry–differential scanning calorimetry (TGA-DSC). The influence of different heating rates on thermal behavior of prepared elastomeric materials was investigated also. Observing the shape of DTG curves for prepared polyurethane nanocomposites, overlapping thermal degradation process in noticed. Homogeneity of prepared hybrid materials was confirmed with the absence of change in the degradation mechanism, regardless filler loading. The onset temperature was significantly shifted to higher temperatures by increasing filler content, indicating the presence of additional hydrogen bonding that caused enhancement of thermal stability of polycarbonate-based polyurethanes. The examination of the silica influence on the activation energy values, as an important parameter of degradation kinetics of synthesized elastomeric materials, was performed using different mathematical models (Flynn-Wall and Toop). The highest activation energy was found for the sample with the lowest silica content (0.15 wt%).

Keywords: Polyurethane hybrid materials, Nanosilica, Thermal stability, Thermal degradation, Mathematical models

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ANALYSIS OF GIBBSITE SAMPLES OF BAUXITE AND POSSIBILITY OF APPLICATION IN BAYER PROCESS

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Bayer process is currently the most used method for obtaining alumina worldwide. This process is one of the most energy – demanding technological processes, and the main raw material, bauxite, is exploited on a daily basis. Because of that, there are tendencies for improvement of the process from aspect of raw material usage, decreasing energy consumption etc. One of the ways for that is usage of gibbsitic (hydrargilite) bauxites, which can lead to decrease in energy consumption and consumption of other raw materials in Bayer process, due to its specific properties. In this paper, by reviewing literature and by visiting alumina plant "Alumina doo" basic differences between processing gibbsite and boehmite type bauxites have been shown (regarding unit operations, conditions of operations and material and energy flows). Also, chemical, mineralogical, and analysis of leaching of these bauxite types have been performed. All analysis has been performed in Research laboratory of plant "Alumina doo". Composition of bauxite has been analyzed by means of standard chemical methods in combination with UV-spectrophotometry (UV-1800), atomic absorption spectrophotometry (AA-7000 Shimadzu) and optical emission spectroscopy; leaching was performed in laboratory autoclave and furnace, and mineralogical composition was determined by diffractometer D8 Endeavor. On the basis of analysis, conclusions have been made about possibility of application of gibbsitic bauxites in the Bayer process.

Keywords: Alumina, Bayer process, Bauxite, Boehmite, Gibbsite

A CRITICAL REVIEW OF CERAMICS FRACTURE TOUGHNESS MEASUREMENT METHODS AND OBSERVATIONS ON SEVNB METHOD IN CASE OF TRADITIONAL CERAMICS

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By definition, fracture toughness is the measure of a material resistance to a brittle fracture when a crack is present. Its precise and reliable determination is of great importance for selection and application of ceramic materials, especially in case of newly developed ceramic materials. There are many methods currently used to measure the fracture toughness, like Surface Crack in Flexure (SCF), Chevron Notch (CN), Single-Edge-Pre-cracked Beam (SEPB) or Indentation Fracture (IF) method. However, these methods are often difficult to realize, unreliable or expensive. Recently, a new method was introduced to measure fracture toughness of advanced ceramic materials: Single-Edge-V-Notched Beam (SEVNB) method. In SEVNB method, a saw cut is tapered to a sharp V-notch using a razor blade sprinkled with diamond. This approach is user-friendly, easy, and last but not least, not expensive. This review, describes and analyze variances between SCF, CN, SEPB, IF and SEVNB method in detail, and present their application on several advance ceramics, from alumina 998 and 999, gas pressure sintered silicon nitride (GPSSN), yttria-stabilized tetragonal zirconia (Y-TZP), and traditional ceramics for roof tiles. It is showed that the repeatability and reproducibility of SEVNB method were very good. Further, the method proved to be forgiving and robust with respect to notch preparation for ceramics having a major microstructural feature greater than about 1 μm in size. During testing there were no mayor difficulties conducting the measurements. The SEVNB method is user-friendly, easy and cheap to conduct, reliable, and accurate.

Keywords: Fracture toughness, Monolithic ceramics, Traditional ceramics

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PREPARATION AND CHARACTERIZATION OF FILMS BASED ON MESOPOROUS SILICA AND ZEIN

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In recent years, much effort has been made to develop novel materials in packaging systems, including active packaging which can improve safety of food and extend shelf life of food products through controlled release systems. Biopolymer-mesoporous silica composites with incorporated antimicrobial agents exhibit a great potential for application in such food packaging systems, especially as antimicrobial films or coatings. Moreover, thanks to biocompatibility and nontoxicity of biopolymers and mesoporous silica, these kinds of packaging materials would be environmentally compatible. One of the biopolymers that is well known for its film forming and coating abilities is zein, a corn protein.

In this work, films casted from dispersions of zein nanoparticles and mesoporous silica nanoparticles (ZN-MSN films) were prepared and characterized for mechanical, optical and water barrier properties, and were benchmarked against films obtained only from zein nanoparticles dispersions (ZN films).

The initial experimental results showed that the ZN-MSN films have comparable, if not improved, characteristics to the pure zein films, which is encouraging toward constructing smart biocompatible composite materials for packaging and coating applications.

Keywords: Zein, Mesoporous silica, Biopolymer films

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FIFTEEN YEARS OF COLLABORATION OF THE FACULTY OF TECHNOLOGY AND THE GALLERY OF MATICA SRPSKA

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Cultural heritage field is characterized by the growing interest of the scientists, leading to the whole new area called conservation science. Conservators, coming mostly from humanistic area (art historians, restorers, archeologists) as well as architects, saw great benefit from this collaboration. The expertise of scientists (natural sciences and material science) is generally divided in two big areas: characterization of the original artistic materials on one side, as well as production and testing of the new materials suitable for the CH preservation on the other side.

The Faculty of technology from Novi Sad understood the emerging area and among first in Serbia started to build experience, gain knowledge and organize specialized analytical facilities. On the other side, cultural heritage institutions as the Gallery of Matica srpska were open for such type of activities, thanks to dedicated individuals working inside.

The history of fifteen years of collaboration is the history of enthusiasm, learning and the development of networks. If we take the Conference dedicated to the moisture (Gallery of Matica srpska, 2004) as the first milestone and the Lanterna conference (The Gallery of Matica srpska, 2014) as the second, we can see fast growing of knowledge in the conservation science in Serbia. In that period The Gallery of Matica srpska and the Faculty of technology collaborated on the pigments characterization on the religious paintings and objects, using mostly analysis of paint samples. Moreover, big efforts and a lot of energy were dedicated to the development of partnerships and learning opportunities.

The introduction of the non-invasive techniques and the creation of the Laboratory for Cultural heritage materials (2014, following Heromat project) brought to the Technological faculty international recognition in the field of conservation science. Besides the characterization of artistic materials using far broader set of analytical methods, the collaboration with the Gallery of Matica srpska was enlarged to the field of education (Summer school, master classes and so on).

The area of conservation science is characterized by the high level of uncertainty in the interpretation of the analytical results, due to the uniqueness of the artwork as the product of a creative process. Therefore a close collaboration between different fields of science and humanities is necessary throughout the whole process. The paper will give detailed insight in the history of the partnership, projects and results in collaboration with the Gallery of Matica srpska, contributing to the study and preservation of the national cultural heritage.

Keywords: Cultural heritage, Collaboration, Paint characterization, Gallery of Matica srpska

MODIFICATION OF STRUCTURE AND PROPERTIES OF BaTiO₃ THIN FILMS BY ADDITION OF Sr²⁺ AND Zr⁴⁺ IONS

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Barium titanate based thin films, due to the perovskite structure and ferroelectric behavior, were recognized as useful materials for production of capacitors, memories, sensors, etc. However, new fields for barium titanate application are recently opened. Perovskite structure provides change of divalent or tetravalent cations in crystal lattice by cations with similar ion radii and consequently modification of material properties. Partial substitution of Ba²⁺ or Ti⁴⁺ with Sr²⁺ or Zr⁴⁺ decreases the Curie temperature of BaTiO₃ and makes this material paraelectric near to the room temperature. Paraelectric phase of BaTiO₃ is recognized as good potential material for application in microwave tunable technologies.

BaTiO₃, Ba_{1-x}Sr_xTiO₃ (x=0.1, 0.2, 0.3, 0.4) and BaZr_xTi_{1-x}O₃ (x=0.1, 0.2) thin films were prepared by chemical solution deposition (CSD) method. In first step, metal salts were dissolved in acetic acid and 2-methoxy ethanol in order to obtain clear and stable solutions. Obtained solutions were deposited on commercial wafers by spin coating method and sintered at temperatures up to 1000 °C. Structural and functional characterization of prepared samples showed predominantly ferroelectric behavior of pure BaTiO₃. On the other hand, by increasing of Sr²⁺ or Zr⁴⁺ content, changes in structure were noticed and leads to creation of paraelectric phase.

Keywords: *Barium titanate, Thin films, Ferroelectrics*

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REUSE OF CEMENT BYPASS DUST FOR ENVIRONMENTAL FRIENDLY PRODUCTION OF BUILDING MATERIALS

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In the attempts to offer economically attractive products which are sustainable in regard to environment, cement industries these days are facing global challenges in waste management, which force them to continuously search for solutions and new sources of alternative fuels (AF). However, increased consumption of AF leads to new problems affecting stable process conditions and even causing production malfunctions. To prevent these, a part of the kiln gasses needs to be extracted and cooled together with kiln dust. By cooling, almost all volatiles are captured and removed from the kiln system as cement kiln bypass dust (CBD).

The chemical composition of CBD depends strongly on AF and raw materials used. If CBD contains low proportions of alkalies, chlorides and sulphur, it can be returned directly into the kiln as part of the raw mixture, or can be mixed as a minor component in cement and binder production. Yet, it is rarely the case and CBD composition could raise environmental footprint of the production process. Therefore, handling, transport and disposal of such dust have been of great concern worldwide, requesting new possibilities of environmental friendly CBD reuse. The main goal of our research is valorisation and evaluation of the CBD as raw material for production of environmental friendly building materials. In order to achieve presented goal, the research stages include: characterisation of CBD, evaluation and valorisation of CBD reuse potentials, design of building materials, characterisation of the developed building materials for the targeted usage in construction industry and lifecycle assessment. For the first phase of experimental work the following techniques were used: chemical characterization by XRF and FTIR, particle size distribution, mineralogy and microstructure by Optical microscopy and scanning electron microscopy (JEOL JSM 6460 LV), textural and mechanical properties. The obtained results are promising and demonstrate that CBD has a great reuse potential for design of novel materials in construction industry, which could remain fully in compliance with the relevant EU environmental regulations.

Keywords: *By-products, CBD, Alternative fuels, Environmental friendly products*

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VALORISATION OF FLY ASH AS SECONDARY RAW MATERIAL TOWARDS IMPLEMENTATION OF CIRCULAR ECONOMY

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Industrial waste like metallurgical slags, fly and bottom ashes, red mud, mine residues represent potential secondary raw materials which could be used in the production of a variety construction products such as cement, concrete, bricks, glass-ceramics etc. Currently, huge amounts of these residues have low recycling rate or are landfilled, especially in East & South-East Europe region. Based on the aims of circular economy these industrial wastes from one industry could be treated as raw material resources for other industry.

One of the ways of recycling these materials is synthesis of mineral binders with high Al-content which can be further used as eco-friendly binders. The high Al content presents the main goal for synthesis of innovative Al-rich binders, since it is mainly used from natural valuable resource - bauxite. The replacement of bauxite with Al-rich residues presents the challenge in the synthesis of innovative binders and this is the main challenge in RIS-ALiCE project in the frame of EIT Raw materials Programme.

Characterization of fly ash (as received and mechanically activated) from materials point (chemical and mineralogical composition, morphology of particles, granulometric composition) and radiological point (radionuclides Ra-226, Th-232, K-40 concentration) will be present in this study and its potentiality for synthesis innovative binders will be discussed.

Keywords: Fly ash, Secondary raw material, Circular economy, Mechanical activation

Acknowledgements: This activity has received funding from the European Institute of Innovation and Technology (EIT), a body of the European Union, under the Horizon 2020, the EU framework Programme for Research and Innovation.

MESOPOROUS SiO₂ PARTICLES WITH CHROMOPHORIC SYSTEM AND THEIR USE IN INDICATION OF RELATIVE HUMIDITY

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Mesoporous SiO₂ particles with defined and ordered porosity (pore size 2–50 nm), high surface area, allowing variations in pore volumes and morphology with adjustable pore diameter, presents an interesting material with applications in various fields such as drug delivery, sensing and catalysis. Desired properties can be tuned by adjusting the synthesis parameters, such as pH, the type of the structure directing agent (surfactants and polymers), and temperature of reaction mixtures, choice of silica precursor, addition of swelling agents, etc. We have synthesized various mesoporous SiO₂ materials with hexagonal arrangement of the pore channels, e.g., MCM-41 (Mobil Composition of Matter No. 41) and SBA-15 (Santa Barbara Amorphous) and with radial arrangement of pore channels. We further functionalized and treated the materials to incorporate chromophore-containing compounds into the mesoporous substrate obtaining a chromophoric system. The structural and morphological properties of the various mesoporous materials were characterized using electron microscopy (STEM), N₂ physisorption and FT-IR spectroscopy. The water vapour sorption capabilities were determined gravimetrically using IGA-100 gravimetric analyzer. The change of colour was evaluated via measurement of CIELab colour space, performed before and after exposure of the materials to a range of environments containing air with different relative water vapour pressures. The synthesized materials exhibited different amounts of adsorbed water at specific water vapour pressures depending on the material properties. The physical reaction of chromophoric system with water in vapour phase are mainly dependent on the additional functionalization of the external surface of mesoporous substrate rather than the pore channel diameter according to the evaluation using the change of the colour of the incorporated chromophore.

Keywords: *Mesoporous SiO₂, Humidity indicator, Capillary condensation, Sensors*

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Nutraceuticals and Pharmaceuticals





THE ROLE OF MICROBIOTA IN THE IMMUNONUTRITION FIELD

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Under healthy conditions, when the nutritional status is adequate, the immune system acts by protecting the host against any pathogen trying to invade the organism. However, when the nutritional status is inadequate, the immune system is depleted, and an immunodeficiency secondary to a malnutrition status is generated.

Nowadays, the study of microbiota is being a key factor since there is wide scientific evidence about the relationship between the immune system and the microbiota, since both of them mature simultaneously.

The interest for the study of microbiota has dramatically increased during the last two decades. While in 2000 there were 78 papers published in the scientific literature, the number in 2016 had raised up to more than 5000, and nowadays, the results found when searching pubmed in 2019 are near 50,000, ten more times in only 3 years!

Gut microbiota in humans is a very complex community made up of billions of microorganisms with the capability of producing benefits to the host throughout the extraction of nutrients and energy from all the polysaccharides consumed in the diet. A portion of these polysaccharides are not degraded by enzymes and others are fermented by the microbiota installed at the colon. The possibility to be able to characterize all bacteria at the gut is essential to understand all the mechanisms acting at this level and the impact they can have on the immune system, and hence, on the host health.

Although a standard microbiota has not been defined up to date, there are several factors related to lifestyle that seem to be involved in shaping the gut microbiota composition. The fact that the composition of microbiota can be diverse and balanced will depend on the following determinants: diet (solid foods and liquid beverages), food behaviour, physical activity, exercise, sleep quality and quantity, as well as different situations of stress.

Indeed, an unbalanced lifestyle will be related to dysbiosis, that means a very high risk to develop inflammatory diseases, such as obesity, type 2 diabetes, cardiovascular and neurodegenerative diseases, all of them related to most of the organs.

Keywords: immunonutrition, microbiota, obesity, type 2 diabetes



***IN SILICO* SIMULATIONS OF FREEZE DRYING OF PHARMACEUTICAL FORMULATIONS**

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Development of protein therapeutic formulations relies on the selection of excipients that stabilize protein and/or prevent aggregation. Experimental strategies to optimize formulations often use force degradation studies and involve the screening of many combinations of excipients and buffers. However, these effective methods do not readily provide information on intermolecular interactions responsible for the protective effects of excipients. *In silico* methods allow evaluation of interactions that may form between selected therapeutics during freeze drying pharmaceutical formulations. A combination of molecular docking methods has been successfully utilized to predict hotspots for protein excipient interfaces on a model protein, which were confirmed by molecular dynamics in presence of explicit solvent and buffer components. Furthermore, simulated annealing simulations of freezing and drying processes provide insights into protective effects of formulations on a molecular level.

This combination of approaches could provide information about the interactions of excipients in formulations and guide the computer-aided and rational development of future formulations.

Keywords: freeze drying, proteins, molecular docking



PHARMACEUTICALS IN THE ENVIRONMENT: A GROWING PROBLEM

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The World Health Organization essential medicines and health products programme works to increase access to essential, high-quality, safe, effective and affordable medical products. Pharmaceuticals are fundamental products for traditional and modern medicine in fighting diseases. However, environmental pollution caused by human and veterinary pharmaceuticals became an emerging problem among other in relation to antimicrobial resistance to human health. The excretion in the form of metabolites and the non-stability of some pharmaceuticals transformed into degradation products increase the complexity of the research of these emerging pollutants in the environment. The identification of the main sources has been one of the concerns to avoid the dispersion in the environment. Wastewater treatment plants (WWTPs) are one of the important focus since its technology was not planned to eliminate these compounds. Hospital effluents and intensive livestock production are other sources identified as problematic. Commission implementing decision (European Union) 2018/840 of 5 June 2018 establishes a watch list of substances for Union-wide monitoring in the field of water policy. The selection of the substances to be monitored is based on different criteria not neglecting the fact that analytical methods cannot entail excessive costs. The analytical methodology based on Solid Phase Extraction (SPE) and liquid chromatography with tandem mass spectrometry (HPLC-MS/MS) is the most appropriate choice for the determination of multi-class pharmaceuticals in waters.

Pharmaceuticals belonging to the nonsteroidal anti-inflammatory drug/analgesics, antibiotics, and psychiatric drugs were monitored in surface water and in influent and effluent of WWTPs located in Portugal. The levels of contamination of river waters are generally lower than those found in wastewater, and there is no standard that relates the composition of the influents and their respective effluents, as well as the composition of the effluents and the treatment processes implemented in the WWTPs. The environmental risk posed by pharmaceuticals detected in waters was assessed toward different trophic levels (algae, daphnids and fish).

Keywords: *Monitoring, Pharmaceuticals, Portugal, Waters, Environmental risk*

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DEVELOPMENT OF FUNCTIONAL PRODUCTS USING BIOLOGICALLY ACTIVE SUBSTANCES

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This article describes the problems of functional nutrition. The results of scientific research and practical developments on approaches to creation of functional food products are presented. Functional food products and their role in human nutrition, use of biologically active additives of natural origin in therapeutic and preventive food products are considered. The use of natural sources of biologically active substances in formulations of functional food products is justified.

The necessity of using such products in dietary therapy aimed at preventing gastrointestinal diseases and reducing cholesterol content in blood plasma were established. According to results of the research, functional bakery products with natural sources of biologically active substances were developed.

Keywords: Functional products, Food products, Biologically active substances, Prevention of diseases

RHEOLOGICAL PROPERTIES OF CHITOSAN-SODIUM LAURYL ETHER SULFATE COMPLEXES

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Chitosan is a cationic biopolymer, which attracts more and more attention in recent years, due to its exceptional physical and chemical properties, expressive biocompatibility and possibilities of obtaining from renewable sources. Due to the pronounced non-toxicity, biodegradability, antibacterial and antiviral activity, chitosan is widely used in products of the cosmetic, pharmaceutical and food industries. Polymer/surfactant interactions in aqueous systems significantly change the system's properties at the interfaces and in the solution, which can result in different effects, such as emulsifying, solubilization, increasing viscosity and colloidal stability. Formed polymer/surfactant complexes affect changes in the rheological properties as well and the final result is the formation of coacervates. The purpose of this study was to investigate the rheological properties of aqueous solutions of cationic polyelectrolyte, chitosan and sodium lauryl ether sulfate, an anionic surfactant, widely used in the cosmetics industry.

Using the Thermo Haake RS600 rheometer, changes in the rheological and elastic properties of chitosan and sodium lauryl ether sulfate (SLES) mixtures have been identified, gained as a result of the interaction of the components. In all examined samples coefficient of thixotropy was increasing with increase in concentration of SLES and achieves a maximum value at the mass ratio chitosan:SLES 1:2, after which it reduces. The oscillatory measurements in mixtures, performed by amplitude sweep method at low oscillating frequency 1 Hz, show that the linear viscoelastic region increases with increasing SLES concentration up to the same chitosan:SLES mass ratio. Such findings point to the structuration of the system caused by electrostatic and hydrophobic bonding of SLES molecules to the chitosan chains and formation stronger polymer-surfactant 3D network. By monitoring the changes in the rheological parameters of the mixtures over five days, it was observed that the viscosity, the coefficient of thixotropy and elasticity were increasing, indicating that changes in the system occur over a longer period of time. In that manner, obtained results indicate the possibility of using rheological methods for a more detailed description of the interaction in the chitosan/SLES mixtures, important for their application in cosmetics and pharmaceutical industries.

Keywords: Chitosan, Sodium lauryl ether sulfate, Rheological properties

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VARIATION IN THE MAIN NUTRIENTS, PHYTOCHEMICALS AND SENSORY PROPERTIES AMONG VARIOUS SWEET MAIZE GENOTYPES

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The present study evaluated the effect of genotypes on the composition of nutrients and phytochemicals in fresh sweet maize kernels and sensory properties of boiled maize samples. The main nutrients (starch, sugar, protein, oil and cellulose contents) and phytochemicals (free phenolic acids - galic, protocatechuic, vanilic, sinapic, p-coumaric, ferulic and cinnamic contents) of fresh kernels of twelve ZP sweet maize genotypes were determined. Tested samples showed significant differences in terms of the content of free phenolic acids and the nutrients as well as sensory properties. Conversely to common maize (phenolics present mainly in bound form), the phenolics determined in sweet maize kernels mainly existed in free form. Protocatechuic acid was the predominant phenolic acid (24.7-56.4 µg/g dry matter) in the all selected samples followed by p-coumaric (7.5-19.7 µg/g), ferulic (5.5-16.8 µg/g), cinnamic (5.6-16.0 µg/g), and galic acid (1.1-23.3 µg/g). Starch, sugar, protein, oil and cellulose content in the fresh materials ranged from 50.1-57.7%, 6.2-10.1%, 11.4-14.4%, 4.5-6.2% and 2.9-4.7%, respectively. The main nutrient composition showed significant effect on the sensory ratings of the boiled maize genotypes while the phytochemical composition did not show any influence in the ratings. Sweetness, starchy flavor, crispness and tenderness were the key sensory attributes strongly correlated with total sugars, sucrose, starch and cellulose. Differences in kernel chemical composition caused by genetic background can influence the sensory properties and consumer appreciation of sweet maize. The information from this study could be useful for the maize breeders to further improve the quality parameters of the sweet maize genotypes.

Keywords: *Sweet maize, Nutrients, Phytochemicals, Sensory properties*

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THE STUDY OF THE PROPERTIES OF THE SUBSTANCES FOR THE PHARMACEUTICAL INDUSTRY

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Biologically active substances (BAS) are the special group of organic compounds. With their help the processes of metabolism, growth and development of living organisms are carried out and regulated. At present the great interest is evoked by the BAS of medical, food and agricultural purposes (antibiotics, vaccines, vitamins, enzymes, polysaccharides, hormones, etc.) because one of the most important areas of medicine is the study of various natural and synthetic substances capable to regulate diverse processes in living cells. For the production of synthetic drugs it is necessary to study their properties, which can be used to create and improve existing methods for the production and purification of substances by means of the computer programs. Such programs allow to realize the computer synthesis based on the data on the structure of the compounds, the thermodynamic parameters of substances and the chemical reactions is selected as the main criteria for programming. Study of structural properties and polymorphism has great importance for the pharmaceutical industry because the polymorphic forms of a drug differ in physicochemical properties, such as solubility, dissolution rate, chemical and physical stability, flowability, hygroscopicity, tableting, and biological activity, bioavailability and toxicity. This goal can be achieved by studying of the physico-chemical properties and structural-functional relationships of biological objects by methods such as, vacuum adiabatic calorimetry, combustion calorimetry, x-ray diffraction, low-temperature x-ray diffraction, differential scanning calorimetry. Over the past few years, our team has obtained a significant amount of data on the calorimetric and structural properties of biologically active substances, namely: riboflavin, nicotinic acid, myo-inositol, folic acid dihydrate, L-carnitine, cyanocobalamin, prednisolone, hydrocortisone acetate etc. For the first time, their temperature dependence curves of the heat capacity were studied, and the enthalpies of combustion were determined using combustion calorimetry. From the obtained experimental data, the complete sets of thermodynamic functions of the compounds under study were calculated. In addition, the crystal structures of prednisolone, methylprednisolone, methylprednisolone aceponate, ethanol solvate of acetate cortisone and propanol solvate of acetate cortisone were resolved.

Keywords: *Biologically active substances, Proteins, Thermodynamic properties, Heat capacity, Vitamins*

POLYMORPHISM IN STEROID HORMONES

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Study of structural properties and polymorphism has great importance for the pharmaceutical industry because the polymorphic forms of a drug differ in physicochemical properties, such as solubility, dissolution rate, chemical and physical stability, flowability, hygroscopicity, tableting, and biological activity, bioavailability and toxicity. In addition the structure of molecules of biologically active substances is necessary to determine its reaction centers and selection of a preferred reaction center (regioselectivity). Investigations of polymorphism are interesting for the fact that a certain modification may be responsible for a specific property that may not display in any other form. For the compounds exhibiting polymorphism (more than 50% of biologically active substances) knowledge of the thermodynamic stability is needed for a better understanding of phase transitions and the conditions which are necessary for these transitions. Therefore the goals of this work include x-ray study of the modifications of the steroid hormones with the purpose of understanding of the possible reasons of polymorphism.

Table 1. Crystallographic parameters of methylprednisolone aceponate (form I, II), ethanol solvate of acetate cortisone and propanol solvate of acetate cortisone.

Formula	$C_{27}H_{36}O_7$	$C_{27}H_{36}O_7$	$C_{23}H_{30}O_6 \cdot C_2H_5OH$	$C_{23}H_{30}O_6 \cdot C_3H_7OH$
T, K	150(2)	90(2)	100 (2)	100 (2)
M	472.56		448.54	462.56
Symmetry,	Orthorhombic $P2_12_12_1$,	Orthorhombic $P2_12_12_1$,	Monoclinic $P2_1$,	Monoclinic $P2_1$,
Z	4	12	2	2
a, Å	6.57348(14)	14.8592(2)	9.6973 (4)	9.8941 (3)
b, Å	14.8295(3)	19.6844(5)	7.4950 (3)	7.3833 (2)
c, Å	26.2214(5)	26.1626(4)	16.1828 (7)	16.5816 (5)
β , °			93.559 (4)	90.110 (3)
V, Å ³	2556.11(9)	7652.4(2)	1173.92 (8)	1211.30 (6)
ρ , g·cm ⁻³	1.228	1.231	1.269	1.268

All single crystal X-ray diffraction experiments were carried out using XtaLAB Pro MM003 diffractometer (MoKa) with Cobra Oxford Cryosystems cooler. Data collection and reduction were carried out using CrysAlis Pro software.

Keywords: *Crystal structure, Polymorphism, Steroid hormones*



THE PHYTOCHEMICAL AND PHARMACOLOGICAL ACTIVITY OF CITRUS LIMETTA PEEL EXTRACTS

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Phytochemical screening of citrus peel extracts was performed for the presence of various phytochemicals. The phytochemical screening results indicated the presence of different phytochemical and bioactive compounds such as flavonoids, steroids, alkaloids, terpenoids, tannins, phenols carotenoids, minerals, and vitamins. Citrus plants are used for different purposes such in the food and beverage industries, cosmetic, and medicinal applications. The Citrus plants are not only providing nutritional supplements but also help for the reducing risk of several illnesses. Some research articles suggested that Citrus limetta is a potential source of antibacterial, antioxidant, anti-inflammatory and antitumor activities. We focused on Citrus limetta peels that are the important constituents of phenolic compounds and its free radical scavenging ability facilitated by their hydroxyl groups. The result indicates a good radical scavenging ability of Citrus limetta peel that will be useful in medicine, pharmaceutical, and cosmetic industries. It can be also used as a preservative in food industries due to its antioxidant properties. This study highlighted the industrial, pharmaceutical and medical potentials of the Citrus limetta peel.

Keywords: Citrus limetta peel, Phytochemicals, Phenolic compounds, Free radical scavenging ability

STUDIES ON PHYTOTHERAPEUTIC QUALITY OF *EQUISETUM ARVENSE* L. IN CLUJ, ROMANIA

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The sterile strains of horsetail are used in phytotherapy and homeopathy due to the content of polyphenols and silica. They proved to have good diuretic effects, being also natural silicon supplements. This plant from spontaneous flora is known as a excellent source of silica - beneficial for bones repair in case of fractures, while strengthening the tendons and ligaments. In ethnomedicine it is also used to improve the quality of hair and brittle nails.

The phytochemical profile of the hydroalcoholic extracts obtained from the horsetail (*Equisetum arvense* L.) sterile stem was evaluated using chromatographic and spectral methods. The evaluation was performed during 3 years, being identified and quantified the polyphenols as well as the antioxidant potential. Plants samples were harvested in the Cluj area (Romania); polyphenols and other biologically active compounds were identified using thin layer chromatography; separations were achieved on silica plates, using as eluent a mixture of ethyl acetate - formic acid - 98% acetic acid - water (67: 7.5: 7.5: 18, v / v) and HPLC. The DPPH and FRAP assay methods were used to evaluate the antioxidant effect.

Keywords: Horsetail, Phytochemical profile, Antioxidant effect

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EMULSIFYING PROPERTIES OF POTATO PROTEINS IN COMPARISON TO WHEY PROTEINS

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Food and pharmaceutical industries are constantly searching for novel sources of functional proteins. One of the particularly important properties of these proteins in food and drug systems is their emulsifying ability, which highly depends on the source of protein, its concentration, isolation technique, environmental conditions (pH, ionic strength), etc. The aim of this work was to investigate the feasibility of replacing common protein emulsifier, such as whey protein, with potato protein isolate in creating fish oil-in-water emulsions.

Oil-in-water emulsions (5, 15 and 30% oil concentration) stabilized by 4% (w/w) whey or potato protein were prepared using ultrasound assisted homogenization. These emulsions were characterized for their particle size distribution, rheological properties and susceptibility to creaming.

The results indicated that replacing whey protein with potato protein did not significantly affect creaming stability and emulsion rheology and had little effect on droplet size. While at lower oil concentrations (5 and 15%) emulsion stabilization with potato protein resulted in smaller mean droplet sizes in comparison to whey protein, the opposite effect was noticed at higher oil concentration (30%). In general, increasing oil-phase volume fraction resulted in increase in emulsion apparent viscosity, average droplet size and creaming stability.

According to obtained results it can be concluded that potato proteins, as alternative protein sources, would be suitable for the preparation of stable emulsions with adequate rheological properties and droplet size distribution, similar to those measured in whey protein emulsions.

Keywords: Protein, Emulsion, Droplet size, Rheology, Creaming stability

Acknowledgements: This work was financially supported by the Provincial Secretariat for Higher Education and Scientific Research, Republic of Serbia [grant number 142-451-2458/2018] as a part of the project entitled "Techno-functionality of proteins from alternative plant sources from Vojvodina region".

STUDY OF THE TEMPERATURE DEPENDENCE OF THE HEAT CAPACITY OF INULIN AND HEVEIN

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Hevein is a small protein (MW 4.7 kDa) with four disulphide bridges and one of major components (20%) of the bottom fraction of rubber tree (*Hevea brasiliensis*) latex. The protein has chitin binding property, which most likely responsible for its antifungal properties, and considered responsible for allergic reactions. Inulin (CAS: 9005-80-5) is a water soluble storage polysaccharide and belongs to a group of non-digestible carbohydrates called fructans. Both compounds are biologically active substances and have great potential for use in the food and pharmaceutical industry.

To measure the heat capacity C_p° of the tested substances in the range from 6 to 343 K a BKT-3.0 automatic precision adiabatic vacuum calorimeter with discrete heating was used.

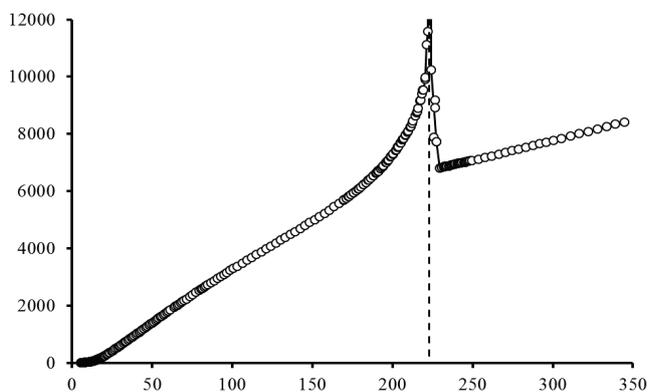


Fig. 1. Temperature dependence of heat capacity of hevein.

The experimental values of the molar heat capacity of hevein over the range from 5 K to 345 K and the averaging $C_p^\circ = f(T)$ plot are presented in Fig. 1. The heat capacity C_p° of this substance in intervals from 5 K to 220 K and from 230 K to 344 K gradually increases with rising temperature and does not show any peculiarities, but from 220 K to 230 K there is seen an anomalous heat capacity.

The transition temperature interval was determined graphically on the $C_p^\circ = f(T)$ curve. The transition temperature of the

sample under study $T_{tr}^\circ = (222.8 \pm 0.3)$ K was estimated as the temperature of maximal C_p° value within the temperature interval of the transition. The heat capacity C_p° of inulin substance gradually increases with rising temperature and does not show any peculiarities. The general aim of these investigations was to report the results of the thermodynamic study of the hevein and inulin.

Keywords: Hevein, Heat capacity, Phase transition, Inulin



WILD GROWING AND CULTIVATED POMEGRANATE: CHEMICAL PROFILING OF JUICES

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Pomegranate (*Punica granatum* L., Punicaceae) is one of the oldest edible fruits and is a native to many tropical and subtropical regions. Mainly, it is consumed as a fresh fruit, but nowadays there are a lot pomegranate products on the market, predominantly juices. Previous studies showed that pomegranate juice contains ellagic acid and its derivatives, ellagitannins such as punicalin and punicalagin, as well as proanthocyanidins, anthocyanins and flavonoids. Due to such chemical composition, a lot of biological activities were reported for pomegranate juice such as anti-inflammatory, antioxidant, antimicrobial, anticancer etc. For punicalin and punicalagin, significant *in vitro* antioxidant activities has been reported. Wild growing pomegranate fruits were collected at natural locality near village Daljam (PW), Montenegro in October 2017 and from the local grower in the same village. Also, two type of samples were obtained from the grower, wild growing transferred to plantation (PWT) and commercial variety Barski (PB). Ten fruits (approximately 2.5 kg) from four trees (2-3 fruits from each tree) were harvest at full maturity stage. The fruits were pressed using a press and the supernatant juice was decanted and frozen at -20°C until analysis. Juices were filtered through membrane filters and analyzed without any further sample pretreatment. HPLC analyses were carried out on Agilent 1200 RR HPLC instrument (Agilent, Waldbronn, Germany), with DAD detector, on a reverse phase Zorbax SB-C18 (Agilent) analytical column (150 mm \times 4.6 mm i.d.; 5 μm particle size). The amounts of the detected compounds were calculated using calibration curves made with standard compounds (ellagic and gallic acids, punicalin, punicalagin, cyanidin-3-glucoside and delphinidin-3-glucoside, cyanidin-3,5-diglucoside and delphinidin-3,5-diglucoside). The amounts of ellagic and gallic acids varied from 0.05 - 0.40 mg/L and 0.05-9.80 mg/L respectively. The amount of punicalin was from 0.04 – 172.40 mg/L, punicalagin from 9.20-176.70 mg/L while the content of total of 4 analysed anthocyanins varied from 180.60-744.50 mg/L of juice with the cyanidin-3-glucoside as the most abundant. For all tested compounds the trend was the same i.e. the highest content of the bioactive compounds was found in PWT followed by PW and PB.

Keywords: Pomegranate, Juice, Phenolics

Acknowledgements: This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia, project number III 46013.

INFLUENCE OF DRYING METHOD ON CHITOSAN/XANTHAN POLYELECTROLYTE COMPLEX CHARACTERISTICS

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Polyelectrolyte complexes are novel drug delivery systems obtained by establishing ion interactions between two oppositely charged polymers. Complexes between chitosan and xanthan could be used as biocompatible pharmaceutical excipients for controlled drug delivery. The aim of this study was to evaluate the influence of drying method on polyelectrolyte complex characteristics that could affect drug release kinetics if used as carriers for drug delivery. Prepared chitosan/xanthan hydrogels with polymers concentrations of 0.65% at pH 5.6, were dried under ambient conditions or freeze dried (Beta 2-8 LDplus, Martin Christ, Germany), and used for solid state characterization by X-ray diffraction (X) (D8 Advance, Bruker, Germany), infrared spectroscopy with *Fourier* transformation (FTIR) (Nicolet iS10, Thermo Fisher Scientific, USA) and scanning electron microscopy (SEM) (JSM-6390LV, JEOL, Japan). X-ray diffraction spectra indicated that complexes obtained by drying under ambient conditions had semi-crystalline structure with sharp peak at 9.48° and broad peak 20.04°, whether the freeze dried complex had amorphous structure with two broad peaks at 10.76° and 20.43°. Infrared spectra of dried polyelectrolyte complexes showed the absence of absorption bands characteristic for amines in the structure of chitosan (1652.23 cm⁻¹, 1374.98 cm⁻¹, and 1312.84 cm⁻¹) and shifting of absorption bands characteristic for carboxylic groups in the structure of xanthan (1604.26 cm⁻¹ and 1402.16 cm⁻¹) that could be the consequence of ion interaction establishment between the polymers. Scanning electron microscopy showed that freeze drying method resulted with significantly smaller particles of polyelectrolyte complex compared to those obtained by drying under ambient conditions. Both methods of drying resulted with non-porous particles. It can be concluded that drying method significantly influenced the characteristics of polyelectrolyte complex that can be reflected on drug release kinetics if these complexes were used as carriers for drug delivery.

Keywords: *Chitosan/xanthan polyelectrolyte complex, Freeze-drying, FTIR, SEM, XRD*

Acknowledgements: *This investigation was financially supported by research projects TR 34007 and III 46010 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia.*

***IN VITRO* CHARACTERIZATION OF REHYDRATION PROCESS AND DISSOLUTION OF IBUPROFEN FROM CHITOSAN/XANTHAN POLYELECTROLYTE COMPLEX BASED DRUG DELIVERY SYSTEMS**

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Ibuprofen is one of the most frequently used analgesic with good risk/benefit ratio. Due to its short half-life ($t_{1/2} \sim 2$ h), frequent administration of immediate release dosage forms is necessary. Chitosan/xanthan polyelectrolyte complexes can potentially influence the release kinetics of orally administered drugs due to their swelling capacity. The aim of this study was to investigate the influence of the used acid and pH of medium for chitosan/xanthan polyelectrolyte complex preparation on *in vitro* rehydration process and release kinetics of ibuprofen. Prepared polyelectrolyte complex hydrogels at pH 3.6, 4.6 and 5.6 using hydrochloric acid and acetic acid were dried under ambient conditions and redispersed using phosphate buffer pH 7.2 at concentration of 2% on LSB18 shaker (Grant, UK) during 24 h, at 37 ± 1 °C and 100 rpm. Controlled shear rate measurements were performed on rheometer Rheolab MC120 (Paar Physica, Germany) with Z3 DIN measuring device by increasing shear rate from 0 to 100 s⁻¹ and back to 0 s⁻¹, at 37 ± 0.1 °C. Physical mixtures of polyelectrolyte complexes and ibuprofen in mass ratio 1:2 were filled into capsules size 1 and *in vitro* release profiles in the paddle apparatus (50 rpm) (Erweka DT70, Germany) were obtained using 900 ml of phosphate buffer pH 7.2 at 37 ± 1 °C. All rheograms demonstrated flow with thixotropy. Rehydration capacity evaluation was based on apparent viscosities (minimal at 100 s⁻¹/maximal at 22.2 s⁻¹). The highest apparent viscosities had samples prepared with acetic acid at pH 3.6 (789/2960 mPa·s), and lowest with hydrochloric acid at pH 4.6 (409/1240 mPa·s). Immediate drug release was observed in samples with both acids at pH 3.6 where polyelectrolyte complex did not have any influence on drug release. All other samples showed extended release of ibuprofen with approximately 50% (for complex prepared at pH 5.6 with hydrochloric acid) to 70% (for complex prepared at pH 4.6 with hydrochloric acid) drug released after 10 h. It can be concluded that extended release of ibuprofen can be achieved by using chitosan/xanthan polyelectrolyte complexes with the highest dissolution rate observed in samples with lowest apparent viscosity after rehydration.

Keywords: Chitosan, Xanthan, Polyelectrolyte complex, Ibuprofen, In vitro dissolution test

Acknowledgements: This investigation was financially supported by research projects TR 34007 and III 46010 funded by the Ministry of Education, Science and Technological Development, Republic of Serbia.

THE EFFICIENCY OF ZINC SUPPLEMENTATION

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Minerals are important for human body to function normally. In dynamic conditions of life for a modern man it is almost impossible to fully satisfy needs for micronutrients from everyday nutrition. Therefore, in many cases it is important to additionally bring micronutrients in the form of dietary supplements. The aim of the study was to determine the role of health professionals in advising users on the application of zinc supplements, as well as the degree of user satisfaction after application of these preparations. In the research the original questionnaire used for surveyed 120 users of zinc supplements of both sexes in the pharmacies of the Farmanea Balkan institution in Belgrade. The obtained results were processed using descriptive statistics. The efficiency of each pharmaceutical preparation depends on whether it is used properly. Although the majority of 74.20% of respondents received advice on how to properly use dietary supplement from health professionals, it is worrying that 10.00% of respondents received advice from unqualified persons, while 15.80% of respondents did not receive any advice. The supplements that contain zinc in the chelated form are the most recommended, since this form of zinc is better absorbed in the body compared to the zinc in the form of oxides or free salts. The results of the survey show that the largest number of the sample subjects (30.90%) used a preparation in which zinc is chelated with gluconic acid in zinc gluconate form. Among research participants a high level of interest for multicomponent preparations was observed, especially for those in combination with vitamin C (28.20%). Zinc and vitamin C allow one another more efficient influence on the good general condition of the body because of the synergistic effect. According to the results of our research, a significant percentage of users (42.50%) took zinc preparations with a meal. Due to the fact that certain food components can affect the absorption of minerals from supplements, it is necessary to educate users about nutritional factors that inhibit or increase the digestibility of zinc in the body. As an indicator of the efficiency of zinc preparations the degree of user satisfaction on achieved results was analyzed. In our survey, zinc supplements proved to be very efficient, as the positive results were reported by 92.50% of respondents, while only 7.50% did not notice improvement. It is likely that satisfied users will continue to use the preparations, remaining loyal to specific producers. The results of our research confirm this, and so the vast majority of the sample subjects (90.80%) are planning to continue with zinc supplementation. The duty of all health care workers, and in particular pharmacists at the primary level of health care, is to provide complete information on dietary supplements in order to make the use of these preparations rational and efficient. Researches on the degree of user satisfaction of food supplements provide opportunities for producers to increase the overall level of user satisfaction, as well as to strengthen relationships with them, and thus not only maintain but also expand its overall user base.

Keywords: Zinc, Dietary supplements, User satisfaction

THERMODYNAMIC AND KINETIC PROPERTIES OF EQUIMOLAR AQUEOUS ANIONIC AND CATIONIC SURFACTANTS MIXTURES – CATANIONICS

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The new trends in industrial product development include the combination of several different surfactants in one formulation in order to improve the product properties. For example, in aqueous solutions the mixtures of anionic and cationic surfactants due to mutual electrostatic attraction form complexes or ion pairs, which show strong surface activity at very low bulk concentrations.

In this work we measured interfacial tensions of sodium alkyl sulphate and alkyl trimethylammonium bromide surfactants with shorter and longer chain lengths at the water/hexane interface. The experiments were performed with the Drop Profile Analysis Tensiometer PAT-1 (SINTERFACE Technologies, Germany). The experimental results were fitted with the Frumkin and Reorientation adsorption models.

When mixing positively (DoTAB) and negatively (SDS) charged surfactants in equimolar solutions, complexes of very high surface activity are formed. These SDS + DoTAB complexes adsorb at the solution/hexane interface at bulk concentrations three orders of magnitude lower than for the single surfactants (Fig. 1), showing a spectrum of new adsorption properties as surfactant ion pairs.

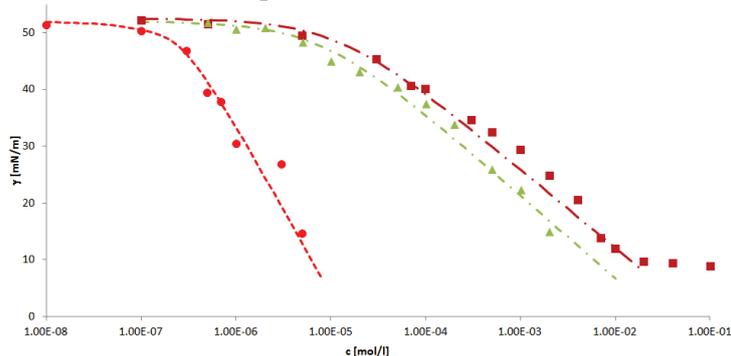


Fig. 1. Equilibrium interfacial tension vs. bulk concentration at the solution/hexane interface for SDS, and DoTAB and their mixture; (●) SDS + DoTAB (dimer complexes), (■) DoTAB, (▲) SDS; the solid line was calculated with the Frumkin adsorption model.

Keywords: *Surfactants mixture, Catanionics, Thermodynamic and kinetic properties*



IN SILICO STUDY OF THE BIOLOGICAL POTENTIAL OF THE SELECTED AMIDE DERIVATIVES

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In contemporary design of new molecule, in silico study of its properties precedes its synthesis. First step requires establishing the quantitative dependence between structure, physico-chemical properties and biological activity of the future molecule. Lipophilicity represents crucial molecular descriptor related to behaviour of the compound in biological medium. For selected amide derivatives, lipophilicity was determined by using reversed phase thin-layer chromatography (RPTLC18F_{254s}) in the presence of two organic modifiers, as well as applying adequate software packages. The experimentally obtained parameters of lipophilicity of the tested amide derivatives (R_M^0 , m and C_0) were correlated with the mathematically calculated values of the partition coefficient, $\log P$, the important pharmacokinetic predictors and the parameters of the toxicity by linear regression, respectively. Statistical quality of the obtained models was improved by performing leave-one-out (LOO) cross-validation. Values of basic and cross-validation statistical parameters confirmed high-quality of obtained mathematical models.

Keywords: Lipophilicity, Thin-layer chromatography, Pharmacokinetics, Toxicity

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REACTION OF INOCULATED CANCERS ON NITRIC OXIDE HYPERPRODUCTION, GLUCOSE ENERGY, FOLATE AND VITAMIN B₁₂ INHIBITION IN YOUNG HAMSTERS

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We investigated the effect of the nitric oxide hyperproduction, glucose energy, folate and vitamin B₁₂ inhibition caused by nitroglycerin and metformin on fibrosarcoma in young hamsters. The 24 Syrian golden hamsters of approximately 40 g, both sexes, were randomly allocated in 3 experimental and 1 control groups of 6 animals in each. 2 x 10⁶ BHK-21/C13 cells in 1 ml were injected subcutaneously on the back of animals in all groups. The first experimental group started daily peroral treatment with nitroglycerin 50 mg/kg, second with metformin 500mg/kg and third with a combination of nitroglycerin 50 mg/kg and metformin 500 mg/kg, via gastric probe 3 days before tumor inoculation. After 2 weeks, when the tumors were ~2cm in the control group, all animals were sacrificed, blood collected for glucose and other analyses, tumors excised, weighed, diameters measured, tumor samples pathohistologically (HE) and immunohistochemically (Ki-67, CD 31, COX IV, GLUT-1, iNOS) assessed and main organs toxicologically analyzed. Tumor volume was determined using the water displacement method and the formula $L \times S^2 / 2$, L - the longest, S - the shortest diameter. Ki-67-positive cells in tumor samples were quantified. Images were taken and processed by imaging software. Statistical significances were determined by the one way ANOVA. The combination of nitroglycerin and metformin significantly inhibited fibrosarcoma growth in hamsters without toxicity. Administration of nitroglycerin with metformin might be an effective and safe approach in novel nontoxic adjuvant pediatric anti-cancer treatment.

Keywords: *Fibrosarcoma, Young hamsters, Nitroglycerin, Metformin*

Acknowledgements: *To Autonomous Province of Vojvodina, Provincial Secretariat for High Education and Scientific Research [grant no. 142-451-2413/2018 (JP)] and Republic of Serbia, Ministry of Science [grant nos. 171039 (JS) and 172013 (DM)].*

FOLATE, VITAMIN B₁₂, GLUCOSE ENERGY DEFICIENCY AND NITRIC OXIDE OVERPRODUCTION INHIBITORY EFFECTS ON FIBROSARCOMA IN ADULT HAMSTERS

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We investigated the effect of folate, vitamin B₁₂, glucose energy deficiency and NO overproduction caused by metformin and nitroglycerin on fibrosarcoma in adult hamsters. The 24 Syrian golden hamsters of approximately 100 g, both sexes, were randomly allocated in 3 experimental and 1 control groups of 6 animals in each. 2 x 10⁶ BHK-21/C13 cells in 1 ml were injected subcutaneously on the back of all animals. The first experimental group started peroral treatment with metformin 500 mg/kg daily, second with nitroglycerin 50 mg/kg, and third with combination of metformin 500 mg/kg and nitroglycerin 50 mg/kg, via gastric probe 2 days before tumor inoculation. After 2 weeks, when the tumors were approximately 2 cm in control group, all animals were sacrificed, blood collected for glucose and other analyses, tumors excised, weighed, diameters measured, tumor samples pathohistologically (HE) and immunohistochemically (Ki-67, CD 31, COX IV, GLUT-1, iNOS) assessed and main organs toxicologically analyzed. Tumor volume was determined using the water displacement method and the formula $L \times S^2 / 2$, L - the longest, S - the shortest diameter. Ki-67-positive cells in tumor samples were quantified. Images were taken and processed by imaging software. Statistical significances were determined by the one way ANOVA.

The combination of metformin and nitroglycerin significantly inhibited fibrosarcoma growth in adult hamsters without toxicity.

Administration of metformin with nitroglycerin might be an effective and safe approach in novel nontoxic adjuvant anti-cancer treatment.

Keywords: *Fibrosarcoma, Hamsters, Metformin, Nitroglycerin*

Acknowledgements: *This study was supported by the Republic of Serbia, Autonomous Province of Vojvodina, Provincial Secretariat for High Education and Scientific Research [grant no. 142-451-2413/2018 (JP)] and Republic of Serbia, Ministry of Science [grant nos. 171039 (JS) and 172013 (DM)].*

POTENTIAL FOR USE OF VINE SHOOTS EXTRACT IN PHARMACEUTICAL INDUSTRY

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Certain types of agricultural waste may be considered as rich source of high-value phytochemicals, with commercial application in many fields of health industry. Annual pruning of vineyards generates large amounts of vine shoots waste, rich in phenolic compounds. One of the prominent compounds in this group is *trans*-resveratrol, proven to have significant biological and pharmacological properties. In this study we have optimized extraction of active principles from vine-shoots and tested cytotoxic effects of obtained extract on cells in the culture. Subcritical water extraction (SWE) of polyphenols from vine shoots was optimized by simultaneous maximization of the total phenolics and flavonoids yield and antioxidant activity. Box–Behnken experimental design (BBD) on three levels and three variables was used for optimization. Influence of temperature (120–200 °C), HCl (0 - 1,5 %), and extraction time (15–35 min) on each response was investigated at constant pressure of 30 bar. The impact of the extract was examined on three human cell lines: breast cancer (MCF-7), cervical cancer (HeLa), and fetal lung fibroblasts (MRC-5). Cells were incubated 24h and 48h with 20-125 µM of vine shoot extract and cytotoxic effect was evaluated using MTT assay. Pure *trans*-resveratrol was used in the same concentration range and incubation times as reference. The extract showed similar effect to resveratrol on malignant cell lines, MCF-7 and HeLa, while cytotoxic effect on normal lung fibroblasts was not pronounced. It is worth noting that selective cytotoxicity to tumor cells may be of significance for future use of vine shoots extract.

Keywords: Vine shoots, Phenolics, Subcritical water extraction, Antiproliferative effect

Acknowledgements: This research was supported by Provincial Secretariat for Higher Education and Scientific Research, Autonomous Province of Vojvodina, 142-451-2839/2018-01/01



CHEMICAL COMPOSITION AND CYTOTOXICITY OF *HELICHRYSUM ITALICUM* EXTRACT GROWN IN URBAN CONDITIONS

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Helichrysum italicum (Roth) G. Don fil. (Asteraceae), also called immortelle or sandy everlasting, is one of the most renowned species within genus *Helichrysum*. It is endemic Mediterranean species, preferably growing on sunny, rocky slopes and around sandy areas. Blossom buds are commonly collected for obtaining crude drug which have been traditionally used for wide variety of medicinal purposes. Aerial parts of immortelle are used predominantly for isolation of essential oils. Traditionally, immortelle is being used for wound healing and other skin conditions such as hematoma or scars.

The study, reports about the growth of *H. italicum* in urban environment, on vertical surface in the form of green wall. Supercritical CO₂ was used as a method of choice to obtain extract rich in phytochemicals and free of pollutants. Chemical characterization of extracts was performed by GC, while *in vitro* tests on cell culture provided insight in the cytotoxic effects of active principles on malignant and non-transformed cells in culture. The importance of reported approach to the sustainable development of the cities, gives additional value to the study.

Keywords: *H. italicum*, Cytotoxicity, Chemical composition, Supercritical CO₂ extraction

Acknowledgements: This research was supported by City Administration for Environmental Protection of Novi Sad, approved by the Decision of Mayor No. 501-2/2018-18/6-11 OA from 26.07.2018.

ANTIOXIDANT ACTIVITY OF *MELISSA OFFICINALIS* EXTRACTS

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Melissa officinalis L., also known as lemon balm, is species which belongs to genus *Melissa*, family Lamiaceae. Lemon balm is a perennial herb native to Southern Europe and Mediterranean region. It mostly grows at sand and loam fertile soils. *M. officinalis* is cultivated for its lemon-scented leaves which are currently being used in pharmaceutical, cosmetic and food industries. Also, lemon balm has been traditionally used for nervous complaints, lower abdominal disorders, nervous gastric complaints, hysteria and melancholia, chronic bronchial catarrh, nervous palpitations, vomiting, migraine, nervous debility, headache and high blood pressure. Furthermore, this plant is also known as available source of natural antioxidants and as potential food supplement. Previous research demonstrated that lemon balm leaves contain high amount of phenolic compounds, which are recognized as phytonutrients with significant beneficial biological activities based on their ability to prevent oxidative stress and minimize oxidative cell injury. Therefore, the purpose of this study was to estimate antioxidant activity of different ethanolic *M. officinalis* extract. The extraction was performed with different concentrations of ethanol (45%; 75% v/v) during two times of extraction (24h and 72h). In order to measure the antioxidant activity of plant-derived antioxidants, 1,1-diphenyl-2-picryl hydrazyl (DPPH) radical scavenging method was conducted spectrophotometrically. Results have demonstrated that analyzed extracts achieved IC₅₀ value in range from 5.56 to 7.61 µg/mL. Based on the obtained results, it could be concluded that increased period of extraction as well as concentration of ethanol, directly effects the quality of extract and antioxidant activity .

Keywords: *Melissa officinalis*, Antioxidant activity, DPPH



MICROENCAPSULATION OF JUNIPER BERRY ESSENTIAL OIL (*JUNIPERUS COMMUNIS* L.)

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The microencapsulation of essential oils provides their protection from oxidative decomposition and evaporation, flavour retention or masking unpleasant taste. The medical properties of some essential oils such as antioxidant, anti-inflammatory and antimicrobial activity makes them useful for the preparation of microcapsules, which structure can provide controlled release of active ingredients of encapsulated oil, allowing their usage in functional foods or pharmaceutical products.

The aim of this paper was to characterize microcapsules loaded with Juniper berry essential oil, which were prepared using different wall materials - maltodextrin (MD), gum arabic (GA) and their mixture. Microcapsules were prepared by spray-drying process. The emulsions for spray-drying were prepared by homogenisation as the first step, followed by ultrasonication to produce fine and stable emulsion. The moisture content, wettability and solubility of microcapsules, powder density, particle size, morphology of microcapsules and encapsulation efficiency (EE) were investigated.

Microcapsules with MD as a wall material had the biggest diameter ($D_{4,3} = 16.42 \mu\text{m}$), due to the biggest emulsion droplet diameter. Considering that GA has certain interfacial activity, which allows production of small droplet sizes emulsion without added emulsifier, microcapsules prepared with GA had the smallest average powder diameter ($D_{4,3} = 5.35 \mu\text{m}$). Further, these microcapsules have shown high oil retention of $83.5 \pm 2.5\%$, with encapsulated of $61.52 \pm 3.13\%$ of the oil, but somewhat higher moisture content ($6.21 \pm 0.73\%$) compared to microcapsules containing mixture of MD/GA as a wall material.

Keywords: Microencapsulation, Essential oil, Juniperus communis, Spray-drying

Acknowledgements: This work was supported by the Ministry of Education and Science of the Republic of Serbia, Project No. 46010.



PRODUCTION OF FUNCTIONAL PROTEIN ISOLATES FROM VEGETABLE BY-PRODUCTS

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Every year tons of vegetable by-products (stems and leaves) are generated and their stockpiling can cause environmental pollution. Taking into consideration the fact that they can be a good source of leaf proteins, by-products from cauliflower, broccoli, cabbage and beetroot were used to extract proteins and to investigate their functional properties. The production of functional proteins from vegetable by-products has recently attracted increasing attention.

The protein isolates were obtained with alkaline extraction of soluble proteins at pH 11, and their isoelectric precipitation at pH 4. Protein content of protein isolates was in the range of 53.5% - 72%. Minimum solubility was observed at pH 4, which confirms their point of isoelectric-precipitation. SDS-PAGE electrophoresis indicated similar composition of all proteins with the major subunits of about 45 kDa and 14 kDa. Their digestibility was studied using gastrointestinal proteases. The DPPH radical cation activities of all protein isolates was confirmed. All proteins show favourable emulsifying abilities, fat and water absorption capacities and foaming capacity and stability. These results indicate that obtained protein isolates, owing to their good functional properties, could be used as potential ingredients of health-promoting food and cosmetic products.

Keywords: Leaf protein, Vegetable by-products, Alkaline extraction, Functional properties, Digestion

CHEMICAL PROFILING OF WINES OBTAINED FROM AUTOCHTHONOUS GRAPE VARIETY PROKUPAC

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The moderate consumption of red wine is proved to have beneficial effects on cardiovascular health. The father of *French Paradox* suggested that low mortality caused by coronary heart disease in France, despite high intake of saturated fatty acids, can be partially due to regular wine consumption. The positive effects are attributed to chemical constituents, dominantly phenolic compounds such as anthocyanins. The anthocyanins are potent anti-oxidants and reactive oxygen species (ROS) scavengers that have been shown to inhibit oxidation of lipids and protect cultured endothelial cells from oxidant injury.

The aim of this study was to determine and compare phenolic profile of five commercial samples of red wine produced from Serbian autochthonous grape variety Prokupac (vintage 2015) with the special attention paid to anthocyanins. The total phenolics and proanthocyanidins were quantified using UV-VIS spectrophotometry. Analysis of anthocyanin compounds was performed using HPLC-DAD technique.

The content of total phenolics ranged from 160.3 to 320.7 mg GAE/100 mL, while the content of proanthocyanidins varied from 30.4-102.2 mg CE/100 mL. The anthocyanin profile was similar in all examined samples with nine different anthocyanin compounds identified, mainly monoglucosides of malvidin, delphinidin and peonidin. The most prominent compound was malvidin-3-*O*-glucoside (4.6-8.4 mg per 100 mL).

The results revealed quantitative differences regarding polyphenolic content which can be explained not only by different grown conditions (soil, climate), but also with different production technology.

Keywords: *Wine, Prokupac, Anthocyanins*

Acknowledgements: *This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia, project number III 46013.*

ULTRASOUND-ASSISTED EXTRACTION OF CANNABIDIOL AND Δ^9 -TETRAHYDROCANNABINOL FROM CANNABIS AREAL PARTS AND PROCESS MODELING

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Cannabis sativa L. contains more than 480 known compounds from 18 different chemical classes. The most important biologically active compounds of industrial hemp are cannabinoids: Δ^9 -tetrahydrocannabinol (THC), cannabidiol (CBD), cannabinol (CBN), cannabigerol (CBG), cannabichromene (CBC), and cannabinodiol (CBND). Besides this, cannabis also contains hydrocarbons, nitrogen-containing compounds, carbohydrates, flavonoids, noncannabinoid phenols, simple alcohols, aldehydes, ketones, acids, esters, and lactones.

The extraction of CBD and THC of industrial hemp was carried out by a ultrasound-assisted technique, an emerging environmentally-friendly technology. The effects of different extraction parameters (extraction temperature (40-80°C), extraction time (20-60 min), and ultrasonic power (24-60 W/L) on the extraction of CBD and THC were investigated using a response surface methodology. In obtained extracts content of CBD was in the range from 0.6158 to 0.8752 mg/mL, and content of THC was in the range from 0.0412 to 0.0458 mg/mL. Experimental results were described by the second order polynomial model. Model was estimated using analysis of variance (ANOVA). The optimization process carried out in order to obtain the most optimal content of desired types of cannabinoids.

Keywords: C. sativa L., Ultrasound-assisted extraction, THC, CBD, RSM

Acknowledgements: This study was carried out within the project 'The renaissance of industrial hemp in the light of current agronomic and medical challenges', N 114-451-2178/2016-03 financed by Provincial Secretariat for Science and Technological Development, Autonomous Province of Vojvodina, Republic of Serbia.

OPTIMIZATION OF EXTRACTION OF PHENOLICS FROM *GENTIANA LUTEA* ROOT

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The root of *G. lutea* is used for preparation of bitter beverages and pharmaceutical products. Benefits of *G. lutea* root are associated with the presence of various bioactive compounds, including secoiridoid, iridoid and phenolic compounds. In this regard, the aim was to optimize the extraction of phenolic compounds from *G. lutea* root by using maceration as a traditional extraction method. Optimization of the extraction of total phenolics from *G. lutea* root was performed in two stages. Statistical screening of the influence of seven factors (solid to solvent ratio, ethanol concentration, particle size, extraction time, extraction temperature, stirring, and microwave pretreatment) on the total phenolic content (TPC) was carried out by Plackett-Burman (PB) design. Four critical factors: extraction temperature (20-80°C), extraction time (8-180 minutes), solid to solvent ratio (1:10-1:50) and ethanol concentration (10-70% v/v) were optimized by using a response surface methodology (RSM) based on the central composite design. TPC of extracts was determined spectrophotometrically by Folin-Ciocalteu method and expressed in gallic acid equivalent (GAE) per g of dry weight (dw). According to the screening results, TPC of *G. lutea* root extracts was influenced predominantly by the solid to solvent ratio, ethanol concentration, extraction time and extraction temperature. Consequently, these four factors were further optimized by RSM and the optimal extraction conditions were selected: the highest temperature (65 °C), extraction time of 129.01 minutes, solid to solvent ratio of 1:20 and 51.96% ethanol concentration. Moreover, the second order polynomial models gave a satisfactory description of experimental data. It was shown that TPC was influenced significantly by linear effect of temperature and time, interaction of solid to solvent ratio and ethanol concentration and quadratic effect of time and ethanol concentration. The adequacy of the model obtained by RSM was demonstrated by the significant p value (< 0.0001), nonsignificant lack of fit (0.0975) and close agreement of experimentally obtained result (19.41 ± 1.03 mg GAE/g dw) for TPC with the predicted values (19.01 ± 1.10 mg GAE/g dw) under the optimal extraction conditions. In this research, an efficient maceration procedure for the extraction of phenolic compounds from the root of *G. lutea* was designed by using screening and optimization method.

Keywords: *Gentiana lutea*, Maceration, Total phenolic content, RSM

Acknowledgements: This research was supported by the Ministry of Education, Science and Technological Development of Republic of Serbia [grant numbers TR 34007 and 46013].

BIOACTIVITY POTENTIAL OF A MEDITERRANEAN BRACKISH DIATOM CULTIVATED IN PHOTOBIOREACTORS

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Benthic microalgae developed strategies through evolution to cope with a variety of abiotic conditions as well as grazing and competition for resources so to tolerate and also thrive in a wide range of aquatic ecosystems. We isolated a strain of the benthic, colonial diatom *Staurosirella pinnata* (Ehrenberg) D. M. Williams & Round from sediments of a Mediterranean dystrophic lagoon (Cabras, Sardinia, Italy). The stock culture was maintained in Diatom Medium (DM) and used as inoculum for mass cultivation in an indoor 30 L photobioreactor, at 25 °C, irradiance of 80 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ and 12:12h L/D cycle. The biomass was harvested at the stationary phase, by settling and centrifuging, and freeze-dried. A crude extract was obtained using a methanol aqueous solution (20% v/v) and separated into two different fractions, hydrophilic and lipophilic. Bioactivity of the hydrophilic fraction was then assessed by means of cytofluorimetric analysis on HaCaT, human immortalized keratinocytes, and CHL-1, human melanoma cell lines. The antiproliferative activity was tested in terms of cell death induction and cell cycle variations in a 24 h dose-response assay. Results showed a strong, dose-dependent cytotoxic effect on the CHL-1 cell line, up to 69.75% for the highest extract concentration tested (10 mg/ml). Conversely, extract administration to the HaCaT cells did not induce significant cell death levels. Further, we quantified the percentage of cells in each phase of the cell cycle. HaCaT showed a progressive and significant dose-dependent increase of cells in the S phase, at the lower doses. Whilst, administration to CHL-1 cells induced a completely different behaviour: 1) a progressive dose-dependent decrease in the percentage of cells in the G1 phase (from 48.87% of the control cells down to 25.20% at 3.2 mg/ml); 2) an increase of cells in the S phase (from 29.72% up to 42.10% at 3.2 mg/ml); 3) an increase of cells in G2/M phase (from 20.27% up to 30.02% at 3.2 mg/ml).

The compared analysis of cell death and cell cycle data suggests that the hydrophilic fraction of the crude extract of the diatom *S. pinnata* contains bioactive molecules that affect different targets or cellular pathways in malignant and normal cells. A more exhaustive and detailed analysis of the soluble compounds present in this fraction, as the future identification, isolation and characterization of new active metabolites will open novel perspectives for biomedical exploitation.

Keywords: *Diatom, Bioactivity, Cancer, Drug discovery*

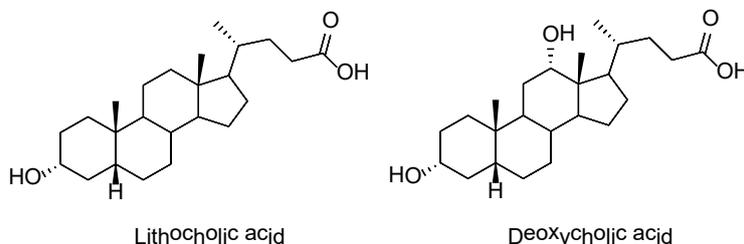
MICROWAVE-ASSISTED SYNTHESIS OF BILE ACID DERIVATIVES AS POTENTIAL LIGANDS OF GLUCOCORTICOID RECEPTOR AND ALDO-KETO REDUCTASE (AKR1C)

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Bile acid derivatives are widely used in modern medicine and pharmacy, where they can increase absorption of various drugs and also serve as therapeutic agents. Interest in microwave-assisted organic synthesis is rapidly increasing. This unconventional energy source drastically reduces reaction times and side product formation, while increasing final yields. From an ecological perspective, microwave-assisted synthesis reduces solvent requirements for reactions and during product isolation, in line with the principles of „green chemistry“. Here we describe microwave-assisted synthesis of 4-bromo-3-oxo derivatives of lithocholic and deoxycholic acids, as well as conjugated 3-oxo derivatives of lithocholic and deoxycholic acids, by dehydrohalogenation of the corresponding 4-bromo-3-oxo derivatives, which were obtained by oxidative halogenation using *N*-bromosuccinimide. Synthesis conditions will be presented in detail. Synthesized compounds were tested for affinity to glucocorticoid receptor *in vitro*, using a yeast-based fluorescence assay. Potential of the synthesized compounds as inhibitors of human aldo-keto reductase 1C (AKR1C) was tested using recombinant enzyme and an *in vitro* enzymatic assay. Several compounds displayed selective affinity for the glucocorticoid receptor or AKR1C suggesting their potential for design of compounds with anti-inflammatory or anti-cancer properties.



Keywords: *Microwave-assisted green chemistry, Bile acid derivatives, Anti-inflammatory, Glucocorticoid receptor, Aldo-keto reductase*

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SYNERGISM IN MIXED MONOLAYERS OF POLOXAMER 407 AND POLYSORBATES

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Polysorbates (PS) are surfactants commonly used as excipients in pharmaceutical formulations and in food preparation. Because of their ability to adsorb at interfaces polysorbates have important role in emulsions, suspensions and in therapeutic protein aqueous formulations. Since surfactant mixtures can have improved properties as compared to individual surfactants, mainly because of synergism, we have investigated nonideal behavior in mixed monolayers of poloxamer 407 (P407) and polysorbates (PS60, PS80 and PS85), at the air/water interface.

To investigate the nonideal behavior in mixed monolayers, we have monitored dependence of surface tension value on natural logarithm of concentration of individual surfactants and P407/PS surfactant mixtures. The surface tension measurements were carried out using du Noüy ring method on a Krüss Easy Dyne tensiometer.

Based on the experimentally obtained data, we have determined the concentrations of individual surfactants and surfactant mixtures (c_{12}) required to produce given surface tension of 45 mN/m. We have obtained the maximum surface excess concentration (Γ_{\max}) and the minimum surface area per molecule (A_{\min}) values of individual surfactants and surfactant mixtures. The corresponding ideal values and interaction parameter values were calculated, as well.

The experimentally obtained c_{12} and calculated A_{\min} values were lower than corresponding ideal values, for all investigated mixtures, while Γ_{\max} values of surfactant mixtures were higher as compared to the ideal values. This indicates existence of synergism in surface tension reduction effectiveness, packing of surfactants at the interface and adsorption effectiveness. The negative values were obtained for the interaction parameter, as well, confirming the synergism in the mixed monolayers. The observed synergism was highest in P407/PS85 mixtures, while lowest in P407/PS60 system. The nonideality can be attributed to dipole-induced dipole interactions in hydrophobic domain of surfactants, i.e., between polypropylene-oxide chains of P407 and fatty acid residues of polysorbate surfactants. Additional dipole-dipole interactions in P407/PS80 and P407/PS85 mixtures most probably exist because of the presence of the double bond in oleic residues of PS80 and PS85, what leads to stronger synergism in the mixed monolayer.

Keywords: *surfactants, polysorbates, poloxamer*

***IN VITRO* TECHNIQUES FOR *DROSERA ROTUNDIFOLIA* SPECIES FOR FUTURE USE IN THE PHARMACEUTICAL FIELD**

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Our aim was to study *Drosera rotundifolia* L. species, given the interest that biotechnologists have for *in vitro* cultures. This technique offers the possibility to rhythmically obtain definite biomass quantities, usable as prime material in the extraction of secondary metabolites for phytotherapy.

We've initiated a *Drosera* vitroculture that afterward we've subcultivated on media with different nutritional input, composition and concentrations of calusogenesis and organogenesis precursors. Particular attention was given to aspects of regenerative capacity of *Drosera rotundifolia* vitrocultures, cytological, histological and morpho-anatomical changes due by the fact that the phytoinoculums were cultivated on special media (with thidiazuron) to stimulate the proliferation processes of minirosettes or with other organic compounds to protect the vitrocultures from the severe regime imposed to reduce the subculturing interval, for 3-4 years. Investigations were performed by use of different cultures media, photonic and electronic microscopic observations of vegetative organs, and by chromatographic and spectral methods to evaluate the extracts obtained from these plantules.

Keywords: Drosera rotundifolia, Vitroculture, Polyphenols

THE EFFECT OF MAGNESIUM STEARATE AND SODIUM STARCH GLYCOLATE ON POWDER FLOWABILITY

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The powder flow plays an important role in the manufacture of dosage form such as direct compression tablets. The objective of this research was to investigate the individual and interaction effects of magnesium stearate (MgSt) (Merck Ltd., Germany) as a lubricant and sodium starch glycolate (SSG) (Primojel®) as a superdesintegrator on powder flowability. Powder formulations were prepared by mixing different concentration of MgSt (0.25% and 5%) and SSG (0.5% and 4%) with microcrystalline cellulose (MCC) (Vivapur®101) or spraydried lactose (LAC) (Super Tab 21AN) as fillers. Eight powder formulations were prepared using as filler LAC and MCC, as a superdesintegrator SSG and as a lubricant MgSt in ratios as follows: P1 (LAC:0.5%SSG:0.25%MgSt); P2 (LAC:4%SSG:5%MgSt); P3 (LAC:0.5%SSG:5%MgSt); P4 (LAC:4%SSG:5%MgSt); P5 (MCC:0.5%SSG:0.25%MgSt); P6 (MCC:4%SSG:5%MgSt); P7 (MCC:0.5%SSG:5%MgSt); P8 (MCC:4%SSG:0.25%MgSt). The powder formulations were evaluated using indirect methods of flowability evaluation according to 9th European Pharmacopoeia: measurement of tapped and bulk density (Ph.Eur.9, 2.9.34) and angle of repose (Ph.Eur. 2.9.36). Also, compressibility index, Hausner's ratio were calculated. A full factorial design with three factors at two levels and response surface methodology were applied to evaluate the influence of various concentrations of MgSt and SSG on powder flowability.

The results indicate that flow properties improved with decrease of SSG content and with increase of MgSt content. Magnesium stearate in powder formulations with microcrystalline cellulose has a greater influence on the improvement of the flow properties than powder formulations with spray-dried lactose. Keywords:

Keywords: Magnesium stearate, Sodium starch glycolate, Powder, Flowability, Factorial design

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EXPOSURE OF PATIENTS TO SODIUM FROM EFFERVESCENT DOSAGE FORMS

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Sodium in form of sodium salts is often used as an excipient in order to increase product solubility. Effervescent dosage forms contain sodium bicarbonate and sodium carbonate which with acidic agents in water produce carbon dioxide. Effervescent and soluble dosage forms contain high levels of sodium salts. Long-term increase of sodium intake can lead to increased blood pressure across all age ranges.

Data about sodium levels in effervescent dosage forms was obtained from Summary of Product Characteristics from website of Medicines and Medical Devices Agency of Serbia (MMDAS). Data about Recommended Dietary Allowance and Adequate Intake of sodium was obtained from official website of Food and Nutrition Board, Institute of Medicine, National Academies. Maximum daily doses recommended by the manufacturer and approved by MMDAS were observed. Average exposure to sodium through consumption of maximum daily doses for each age group was calculated as percentages of the adequate daily intake.

This study has shown that average exposure of adults to sodium through consumption of effervescent powders was 53.85%, effervescent granules 34.73% and effervescent tablets 94.11%. The highest exposure to sodium was found in effervescent tablets containing acetylsalicylic acid (400mg) and ascorbic acid (240mg) as active pharmaceutical ingredients: 217.20 % for people aged 18 to 50 years; 250.62 % for people aged 51 to 70 years; 271.50 % for people aged 70 and older.

Because of amount of sodium to which patients are exposed through consumption of effervescent dosage forms, health professionals should make effort to inform patients about potential risks. Also, pharmaceutical industry ought to develop effervescent forms with reduced sodium content.

Keywords: Sodium, Excipients, Effervescent dosage forms, Increased blood pressure

Acknowledgements: This study was supported by The Ministry of Education, Science and Technological Development, Republic of Serbia (project TP31071-2, 41012 and 172053)

OIL EXTRACTION FROM PLUM SEEDS (*PRUNI DOMESTICAE* L.)

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The plum seed oil is a rich source of essential fatty acids, E and B vitamins, beta-carotene, and beta-phytosterol, which makes it an excellent antioxidant. It is commonly added to the various products for skin care because it possesses the hydration properties and leads to skin regeneration. The aim of this study was to analyze the effect of solvent polarity on the yield and some physicochemical characteristics (density, refractive index, acid value, saponification value, and peroxide value) to assess the quality and functionality of plum seed oil. The Soxhlet extractions were performed using hexane, heptane, ethyl acetate, acetone, and chloroform:methanol (2:1, v/v) at the liquid-to-solid ratio of 10 cm³ g⁻¹ and boiling point temperature of the solvents for 6 h. The extracted plum seed oil with non-polar solvents (hexane and heptane) was pale yellow and limpid, but using polar solvents the color was little dark and turbid. The oils were pH neutral and odorless. The highest yield of plum seed oil of 30.5% was obtained with heptane and the lowest yield of 23.5% with ethyl acetate. The density was varied from 0.4 g cm⁻³ (hexane) to 1.1 g cm⁻³ (acetone). The oils have not exhibited significant variations in the refractive index, which ranged from 1.4695 to 1.4750. The low variation degree of refractive index values may indicate their relative purity. The higher acid value was obtained with ethyl acetate (2.8 mg KOH g⁻¹ of oil), while in other cases this value was lower than 2.2 mg KOH g⁻¹ of oil. The lower acidity of oil fractions shows a lower variation in the hydrolytic deterioration and they could have a long shelf life. The saponification values of the oil samples were found to be in the range of 180-198 mg KOH g⁻¹ of oil, whereby the lowest value was noticed for acetone oil fraction. The higher saponification value indicates the presence of low molecular weight fatty acids in a larger quantity. The peroxide values were in the range of 1.8 and 4.28 mmol O₂ kg⁻¹ of oil. The highest peroxide value was noticed for ethyl acetate fraction and the lowest for acetone fraction. Since the peroxide values were lower than 10 mmol O₂ kg⁻¹, the obtained oils can be considered as good quality and suitable for human consumption. The obtained results showed that the extraction of the plum seed oil is better to carry out using the more nonpolar solvents (hexane, heptane). The choice of solvent depends on the desired fraction to be extracted. The oil fractions with these physicochemical properties can have potential benefits in the cosmetics and pharmaceutical industries.

Keywords: Seed oil, Extraction, Characterization

Acknowledgements: This work was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia under the project TR-34012.

INVESTIGATION OF VOLUMETRIC AND VISCOMETRIC PROPERTIES OF ECSTASY IN AQUEOUS SOLUTIONS

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Ecstasy or 3,4-methylenedioxymethamphetamine (MDMA) is a member of the psychoactive substances called entactogen. Entactogens increase the sense of empathy and closeness, but regarding the similarity in the chemical structure with amphetamines they also belong to a large group of amphetamine-type drugs. It is one of the most popular and most represented drugs among young people. In tablets, MDMA in a form of a hydrochloride salt (MDMA-HCl) is mixed with adulterants such as caffeine, paracetamol, phenacetine and inert cutting agents like lactose, glucose, starch or microcrystalline cellulose. They are essential for shape and design, as well as for the taste of the pills. Besides the fact that it is a highly addictive psychoactive substance with a high potential for severe health damage, in a proper dosage and in a controlled environment under medical supervision, due to its stimulative and psychedelic properties it may have favorable pharmacological effect. Regarding this, MDMA-HCl is included in psychiatric therapeutic research for people with post-traumatic stress disorder and depression which do not respond to a classical treatment. Therefore, it is very important to study the interactions of MDMA-HCl in aqueous solutions and in (water + D-lactose) ternary mixtures, since the water is the most abundant solvent in human organism and lactose is one of the most common inert components in the pharmaceutical formulations. Examination of volumetric and viscosimetric features enables understanding of these interactions within the solution, as well as the influence of these interactions on transport properties and bioavailability of various molecules in human body. Experimental density and viscosity of MDMA-HCl in aqueous solutions and in (water + D-lactose) ternary mixtures have been investigated between (293.15 to 313.15) K and in molality range from (0.05 to 0.5) mol·kg⁻¹. From calculated apparent molar volumes, apparent molar volume at infinite dilution, Hepler's coefficient and viscosity *B*-coefficients, hydration number of MDMA-HCl of solute, it was found that MDMA-HCl acts as structure maker. The taste of MDMA-HCl was evaluated through calculated values of apparent specific molar volumes at infinite dilution. It was concluded that the taste of MDMA-HCl is basically bitter and becomes bitterer in the presence of lactose, which is explained by desolvation properties of the lactose solution.

Keywords: *Ecstasy, Volumetric properties, Viscosity, Taste, Psychoactive substances*

Acknowledgements: *This work was financially supported by the Ministry of Education, Science and Technological Development of Republic of Serbia under project contract ON172012.*

GREEN EXTRACTION OF POLYPHENOLS FROM NETTLE LEAVES (*URTICA DIONICA* L)

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Nettle (*Urtica dioica* L), as a wild-growing and widespread annual plant, possess a great economic potential due to its widespread application in food, pharmaceutical, animal feed and other industries. It has been used in a traditional medicine since ancient times due to the high content of various biologically active compounds, such as polyphenols which exert many pharmacological effects (i.e. antioxidative, hypoglycemic, antiviral, immunomodulatory etc.). Hence, nettle extracts are being prepared by conventional and/or novel extraction techniques (i.e. Pressurized Liquid Extraction, PLE). The advantages PLE are primarily related to shorter processing time, lower solvent consumption, higher efficiency and reduced energy costs, as well as the possibility to use "green solvents". This extraction process can compatible with the principles of green chemistry. Therefore, the aim of this study was to evaluate green extraction approach to optimize the valorization of dried nettle leaves in terms of total phenolic content (TPC) by PLE. PLE was conducted under different conditions by varying cycle numbers (1, 2, 3, 4), static times (5 and 10 min) and temperature (20 °C and 50 °C) with 96% ethanol as the generally recognized as safe and affordable extraction solvent (ASE 350, Dionex, Sunnyvale, CA, USA). The total phenolic content was estimated by Folin–Ciocalteu colorimetric method. The averages for TPC in the nettle extracts ranged from 106,44 ± 0.11 to 739,97 ± 0.XX mg Gallic Acid Equivalent per g of dry material. Increased static time and cycle number resulted in higher extraction yields with, the highest TPC in nettle extracts was obtained by 4 cycle process and 10 min of static times. With respect to temperature, results show a rise in TPC as extraction temperature increases (20 °C vs. 50 °C) probably due to more efficient degradation of cellular walls and easier release of bond phenolic compounds. In conclusion, PLE could be considered as an advanced and promising green approach that could overcome current limitations of conventional extraction and provide valuable nettle extracts that could find their application in industry and in emerging global markets.

Keywords: Nettle, Pressurized liquid extraction, Total phenolic content, Green recovery

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THE INFLUENCE OF ULTRASOUND ASSISTED EXTRACTION ON THE ISOLATION OF BIOACTIVE COMPOUNDS FROM NETTLE LEAVES

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Urtica dioica L., commonly known as nettle, is a perennial plant from the *Urticaceae* family. Nettle leaves are rich in bioactive compounds, such as the phenolic compounds, which are exceptionally important for the growth and reproduction of the plant. These molecules are potent antioxidants and are considered to have a significant effect on human health. Phenolic compounds are isolated using various extraction techniques, such as conventional methods that require extended extraction times and a higher amount of solvent. One of the most commonly used non-thermal eco-friendly techniques for isolation of bioactive compounds from plants is ultrasound assisted extraction (UAE), with shortened extraction time, reduced solvent consumption, lower extraction temperature and energy consumption. Hence, the aim of this study was to determine the optimal conditions for the extraction of phenolic compounds in dried nettle leaves (*Urtica dioica* L.) using ultrasound assisted extraction (UAE) and 96% ethanol as generally recognized as safe solvent. Parameters varied during the extractions conducted at ambient temperature were: the amplitude of ultrasound (25-100 %) and the extraction time (5-40 min). The total polyphenolic content (TPC) of nettle extracts was determined spectrophotometrically using the Folin-Ciocalteu method. The highest yield of the total phenolics extraction was 563,12 mg GAE/100g, achieved with the following extraction conditions: 20 minutes at an amplitude of 75 %.

Keywords: *Nettle, Ultrasound assisted extraction, Bioactive compounds, Total phenolic content*

Acknowledgements: *The authors would like to acknowledge the Croatian Science Foundation for financing our project titled “ Isolation and encapsulation of bioactive molecules of wild and cultivated nettle and fennel and effects on organism physiology“ (IP-01-2018-4924).”*

ANTIADHESIVE AND ANTIBIOFILM POTENTIAL OF SUBCRITICAL WATER EXTRACTS DERIVED FROM MASHROOM *INONOTUS OBLIQUUS*

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Mushroom *Inonotus obliquus* has been traditionally used in China, Korea, Russia and in most Baltic countries as health promoting remedy. The study was designed to investigate antimicrobial activity of subcritical water extracts derived from mushroom *I. obliquus* originating from Mongolia (IM) and from mountain Vlasina, Serbia (IS), including their antiadhesive and antibiofilm capability. Antimicrobial potential of subcritical water extracts was tested by microdilution method. The antiadhesive and antibiofilm abilities were monitored using Gram positive bacteria *Listeria monocytogenes*, *Enterococcus faecalis* and *Staphylococcus aureus*, as well as Gram negative *Salmonella enteritidis*, *Escherichia coli* and *Pseudomonas aeruginosa*. The most significant biofilm production was observed in *E. faecalis* (the third category) and *S. aureus* (the second category). It was noticed that IS and IM were able to provide up to 90% of *E. faecalis* and *S. aureus* adhesion and biofilm reduction, i.e. to decrease the ability of these two bacteria to adhere and form a biofilm (the first or almost zero category, after contact with extracts). In general, Gram positive bacteria appeared as more susceptible to the presence of both tested subcritical water extracts (MBC - 20 mg/mL in all experiments). According to the obtained MIC values, IM (5 mg/mL, 2.5 mg/mL, 2.5 mg/mL) exhibited a stronger effect than IS (10 mg/mL, 5 mg/mL, 5 mg/mL) on *L. monocytogenes*, *E. faecalis* and *S. aureus*, respectively.

This study revealed that tested subcritical *I. obliquus* water extracts are very effective in preventing the formation of biofilms, especially those of selected Gram-positive bacteria. This discovery could be very useful in combating biofilms, taking into account all the problems and dangers that they bring.

Key words: Inonotus obliquus, Antiadhesion ability, Antibiofilm ability

ANTIMICROBIAL ACTIVITY OF *MCENWTC'RQO KHGTC*"DRY EXTRACTS

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Plant produce a huge variety of secondary compounds as natural protection against microbial and insect attack. Many of these compounds have been isolated from whole plants or plant parts giving extracts with different biologically significant properties. *Maclura pomifera* (Rafin.) Schneider belongs to the *Moraceae* or the mulberry family. The whole plant or their parts have been widely used in traditional medicine with anti-microbial, anti-inflammatory, anti-malarial and anti-insect activities.

The objective of this work was to evaluate the antimicrobial properties of *Maclura pomifera* leaf extracts obtained with two different extraction solvents (30%, 50% and 70% EtOH and DCM). The preliminary screening of antimicrobial activity was performed by disk diffusion method. The obtained results have been shown that gram-negative bacteria, yeast and fungal strains were resistant to all tested extracts. Gram-positive bacteria tend to be more sensitive to all extracts, except DCM, which was found to have no antimicrobial effect against any of the examined microorganisms. The highest antimicrobial activity in the case of 70% EtOH extract was determined against *Enterococcus faecalis* ATCC 19433, while the lowest antimicrobial activity was assessed against *Bacillus cereus* ATCC 11778. In addition, it can be noticed that tested ethanol extracts have moderate antimicrobial activity against gram-positive bacteria. The minimal inhibitory concentration was determined for all tested gram-positive bacteria, which indicated that 70% EtOH extracts of *Maclura pomifera* has a great antimicrobial potential, so it can be considered as a food preservative or to be incorporated in pharmaceutical/cosmetic formulation with antimicrobial effect.

Keywords: *Antimicrobial activity, Maclura pomifera, Moraceae*

FREEZE DRIED BERRIES AS SOURCE OF NUTRITIONALLY VALUABLE COMPOUNDS

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Mulberry and elderberry are the oldest and popular medicinal plants, have been used in medicinal purposes since prehistoric times. Various scientific researches were found that bioactive components are present in all parts of plants (leaves, flowers, bark, fruits), which used for different examinations. The objective of this research was to show the phytochemical profile and different biological activity of berries extracts, which obtained from freeze-dried fruits. In this study for isolation of secondary metabolites were applied modern (microwave and ultrasound assisted extraction) and traditional (maceration) extraction techniques and used 50% EtOH and water as solvents. The spectrophotometrically analyses included determination of total phenolic, flavonoid and anthocyanins content. Phytochemical profile was performed using the HPLC method. Biological activity was investigated using different in vitro antioxidant and enzyme inhibitor assays. Antioxidant potential was analyzed using ABTS (2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid), DPPH (2,2-diphenyl-1-picrylhydrazyl), CUPRAC (cupric reducing antioxidant capacity) and phosphomolybdenum assays. Enzyme inhibitory effects were tested against cholinesterases, tyrosinase, α -amylase, and α -glucosidase. Based on the analysis, it has been established that all extracts are characterized by the high content of phenolic compounds (flavonoids and phenolic acids). Extracts of both berry fruits obtained by modern extraction techniques expressed better antioxidant potential in all applied assays than extracts obtained by maceration. The best enzyme inhibitory activity was showed mulberry extract obtained by microwave-assisted extraction and 50% EtOH solvent according to the enzyme acetylcholinesterase (7.56 ± 0.02 mg KAE/g extract), while elderberry extract obtained same extraction technique and some solvent was achieved the best inhibitory effect against tyrosinase (73.91 ± 0.27 mg KAE/g extract). Because of growing interest of consumers in healthier lifestyle and functional foods with enhanced nutritional and therapeutic values, results of this research, are very interesting and important, because it was established that fruits of mulberry and elderberry, which are not used in human nutrition, could be initial material for the production of new functional products in food industry and also this berries could be exceptional plant material for obtaining different cosmetics products.

Keywords: *Mulberry, Elderberry, Extraction techniques, Biological activity, New products*

Acknowledgments: *This research study was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (Project No. TR31013)*

COMPARISON OF THE EFFICIENCY OF DIFFERENT METHODS FOR HESPERIDIN AND NARIRUTIN EXTRACTION FROM ORANGE PEEL

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During industrial citrus processing, large quantities of waste material are produced mainly as citrus peel. These food by-products often contain significantly high amount of bioactive components, therefore representing a highly valuable raw material for production and development of new products. Orange peels contain diversity of bioactive compounds, among which hesperidin is the most abundant flavonoid present in citrus products. Hesperidin possesses different properties, such as positive effects on vascular or cardiovascular system, anti-inflammatory activity, anticancer activity, antimicrobial activity, antioxidant activity, while narirutin exhibits good antioxidant, anti-inflammatory, and produces antidepressant-like properties.

Systematic comparison between microwave-assisted (MAE) and ultrasound-assisted extraction (UAE) was performed. In both MAE and UAE methods, the influence of following extraction parameters was investigated in order to perform adequate comparison; temperature (30, 50, 70 °C), extraction time (5, 10, 15 min), and solvent type (water, 80 % aqueous ethanol solution, 50 % aqueous methanol solution), at investigated microwave power (300 - 800 W) in MAE, as well with frequency (37 Hz) and power (50 W) of ultrasound in UAE method. Extracts were characterized with different levels of hesperidin and narirutin (determined and quantified by HPLC) depending on the applied process parameters and extraction method. Both methods have demonstrated predominance of hesperidin in orange peel compared to narirutin. Extraction efficiency for both compounds was enhanced in aqueous ethanol and methanol solutions.

This study showed that MAE could obtain better extraction yields of hesperidin and narirutin in shorter extraction time, at lower heat exposure and with adequate solvent application compared to the UAE method.

Keywords: *Bioactive compounds, Extraction, Hesperidin, Narirutin, Orange peel*

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EFFECT OF ANTIHISTAMINE (LORATADINE) TO ACTIVITY OF CATALASE *IN VITRO*

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In this paper, the influence of H1-antihistamine from loratadine on the enzyme catalase was examined by spectrophotometric method. H1 antihistamine is used to alleviate the symptoms of allergic reactions, and antiallergic activity of loratadine is based on disabling the function of the secondary Ca^{2+} messenger in the release of histamine. Using the Lineweaver-Burk diagram, the values of the Michaelis-Menten constants (K_m) and the maximum velocity (V_{max}) were determined without the presence of different concentrations of loratadine. The values of the maximum velocity were constant, while the values of the Michaelis-Menten constants change, which indicated that in the presence of hydrogen peroxide as a substrate, loratadine proved to be a partial competitive inhibitor. In the case of partial inhibition of catalase, the decomposition of peroxide continues to be partially performed after loratadine inhibited the enzyme-substrate complex.

Keywords: *Loratadine, Catalase, Enzyme kinetics, Spectrophotometric method*

MULTI-RESPONSE OPTIMIZATION OF POLYPHENOLS RECOVERY FROM *THYMUS SERPYLLUM* BY ULTRASOUND-ASSISTED EXTRACTION

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The main goal of this study was to valorize *Thymus serpyllum*, the by-product of filter-tea production, for extraction of polyphenols with high antioxidant capacity by ultrasound-assisted extraction (UAE). In order to optimize the process, three levels, three variables, face-centered central composite experimental design in combination with response surface methodology (RSM) was applied. Temperature (50, 65 and 80°C), extraction time (40, 55 and 70 min) and ethanol concentration (45, 60 and 75%) were used as independent variables. Responses that gave us complete and useful information were total extraction yield (Y) and yields of total phenolics (TP) and flavonoids (TF) contents, as well as antioxidant activity parameters obtained by DPPH, ABTS and FRAP assays. Results were fitted to a second-order polynomial model and goodness of fit was determined by regression analysis and analysis of variance (ANOVA). The highest values of target responses experimentally obtained were 44,03 mg GAE/g DW, 60,92 mg CE/g DW, 52,49 mg TE/g DW and 118,94 mg TE/g DW for TP, TF, DPPH and ABTS, respectively. Good accordance between experimental and predicted values were achieved which suggested that obtained models could be used for multi-response optimization in order to maximize yields of target responses and improve antioxidant potential of *T. serpyllum* liquid extracts. It could be concluded that *T. serpyllum* herbal dust could be used as raw material for production of antioxidant-rich extracts.

Keywords: Thymus serpyllum, Ultrasound-assisted extraction, Polyphenols, Antioxidant activity

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CHEMICAL COMPOSITION OF FLOWER ESSENTIAL OIL *SANTOLINA CHAMAECYPARISSUS* L.

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The genus *Santolina* (Asteraceae family) is a taxonomically complex group of plant species widely distributed in the Mediterranean region. *Santolina chamaecyparissus* L., commonly referred to as cotton lavender, is a hardy aromatic evergreen shrub with a dense mound of attractive grayish-silver foliage. It has been widely used in traditional medicine since the ancient times due to its biological properties. The volatile components of the *S. chamaecyparissus* flowers from Medicinal plant collection garden Institute of Field and Vegetable Crops Novi Sad, were analyzed by GC and GC-MS. Among the 71 compounds presented in the essential oil, major components were artemisia ketone (36.1%) and vulgarone B (22.1%). Both of these compounds from *S. chamaecyparissus* essential oil possess strong antimicrobial activity. Apart from this, artemisia ketone has a herbaceous odour, and can be used as flavoring and as a natural food additive. It is used in pharmaceutical products for its functional properties, as well as in perfume industry for the fragrance.

Keywords: Cotton lavender, Volatile components, Artemisia ketone, Vulgarone B

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VALORISATION OF HOREHOUND EXTRACTS OBTAINED USING ULTRASOUND AND MICROWAVE ASSISTED EXTRACTION: ANTI-HYPERGLYCAEMIC ACTIVITY

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White horehound (*Marrubium vulgare* L.), is a grey-leaved perennial herb, belonging to Lamiaceae family, distributed in Eurasia and northern Africa zones. According to recent literature, horehound shows several *in vivo* and *in vitro* activities including antihypertensive, antioxidant, antiinflammatory, antidiabetic, effects on respiratory system, digestive stimulant, antiasthmatic, hypolipidemic, antibacterial and antifungal effects. In past, conventional extraction techniques have been most frequently employed in order to obtain bioactive compounds from horehound. Nowadays, modern extraction techniques offer a wide spectrum of possibilities to achieve higher yields for shorter time while saving solvent and energy consumption. Consequently, we decided to compare two modern techniques, ultrasonic-assisted extraction (UAE) and microwave assisted extraction (MAE) in reference to extraction yield, total phenol content, total flavonoid content, IC₅₀ value and EC₅₀ value of obtained extracts. Response surface methodology was used for optimization of process parameters in UAE and MAE. The optimal UAE parameters for maximized polyphenols and antioxidant activity were temperature of 73.6 °C, extraction time of 40 min and ultrasound power of 30.3 W/L, while in case of MAE optimal parameters were 63.8% ethanol, extraction time of 15 min and microwave power of 422 W. Many studies have already shown that intake of plant material rich in polyphenols can cause anti-hyperglycaemic effects in animals and in humans, possibly via α -glucosidase and/or α -amylase inhibition. In order to evaluate anti-hyperglycaemic activity of optimal UAE and MAE extracts, α -amylase and α -glucosidase inhibition tests were performed.

Keywords: Horehound, Ultrasound assisted extraction, Microwave assisted extraction, α -Amylase inhibition, α -Glucosidase inhibition

INFLUENCE OF SUPERCRITICAL FLUID EXTRACTION PARAMETERS ON GRAPE SEEDS OIL RECOVERY

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Winery processing by-products are rich source of valuable bioactive compounds which could be extracted by either conventional or novel extraction techniques. The main goal of this study was investigation of supercritical fluid extraction (SFE) parameters influence on recovery of grape seed oil. The effect of SFE parameters: pressure (250, 300 and 350 bar), temperature (40, 50 and 60°C), CO₂ flow rate (0.2, 0.3 and 0.4 kg/h) and particle size (315-800 and 800-2000 µm fraction) was evaluated using one-factor-at-a-time approach. Extraction curves for each run were obtained after 0, 15, 30, 45, 60, 90, 120, 180 and 240 min and total extraction yield (Y) was determined. Obtained grape seed oils were compared in terms of Y, fatty acid profile determined by GC-FID, functional quality indices, tocopherol content obtained by HPLC-DAD and *in vitro* antioxidant activity. Results suggested that the highest Y (12.23%) was obtained at following conditions: 350 bar, 60°C and 0.4 kg CO₂/h. Grape seed oils were characterized with particularly high content of α- and γ-tocopherols. Fatty acid profile consisted mostly of polyunsaturated fatty acids (>70%) with linoleic acid (C18:2n6c) being the most abundant. Due to appropriate chemical profile and high functional quality of samples, application of SFE was used for production of solvent-free oils of good quality. Therefore, grape seeds obtained as a by-product from winery industry were successfully valorized as a raw material for recovery of high quality vegetable oil which could be further used in cosmetic and food products.

Keywords: *Grape seeds, Supercritical fluid extraction, Polyunsaturated fatty acids, Tocopherols*

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PARTICLE ENGINEERING IN MODERN DRUG FORMULATION

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Modern pharmaceutical technology is concentrated on formulations which are targeted to the exact site at the appropriate time, with maximum efficiency and with reduced side-effects. The various methods used to decrease particle size into the micro- or the nanosize range can be divided into two main categories: as bottom-up and top-down techniques. These two categories are not separated sharply, because use of the top-down together with the bottom-up technique is important to control the particle size.

Our aims are to develop new technological protocols based on conventional and non-conventional procedures resulting micro and nanoparticles. The presentation will introduce novel technological methods (laser ablation, co-milling and co-spray-drying) to produce predispersions containing micro- or nanosized drugs, which are suitable for further drug formulations.

In this approach, poorly water-soluble compounds are formulated as nanometer-sized drug particles (nonsteroid anti-inflammatory agents, antibiotics and pharmaceuticals effect the central nervous system). We have applied as bottom-up methods the melt emulsification, solvent diffusion and solvent evaporation techniques using different drugs. We also apply technological procedures as co-milling, high pressure homogenization, high intensity ultrasound, solvent-antisolvent precipitation, nano-spray-drying, freeze-drying and coating. The micrometric and physicochemical properties, structure, compatibility, stability, *in vitro*, *ex vivo*, *in silico* and *in vivo* evaluations are used.

By using nanoparticles in nasal/pulmonar formulation (alternative drug administration), innovative and more efficient products can be achieved, which may lead to the improvement of different therapies.

Keywords: Nanocrystals, Microparticles, Alternative administration, Pharmaceutical formulation

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PROTEIN HYDROLISATES FROM FOOD BY-PRODUCTS AS NEW ANTI-DIABETIC COMPOUNDS

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In recent years there is an increasing interest for bioactive peptides because of their high therapeutic potential from the wide spectrum of action *in vivo*, including antioxidant, antibacterial, antihypertensive, immuno-modulatory, anti-diabetic activity, etc. Diabetes and obesity have become a major problem in global health care because of changes in lifestyle, food habits, and age-related metabolic disorders. Diabetes mellitus is one of the most common diseases, affecting millions of people worldwide. The most commonly used therapeutic approaches to low the concentration of glucose level is to decrease the rise of the blood glucose level by inhibiting the hydrolyzing enzymes, such as α -amylase and α -glucosidase. Inhibition of these hydrolyzing enzymes, which delay carbohydrate digestion, decreases the glucose absorption rates, therefore blunting the postprandial plasma glucose rise.

The aim of this study was to evaluate the α -amylase and α -glucosidase inhibitory activity of proteins, derived from food by-products, hydrolyzed under various enzymes. Protein isolate as source of bioactive peptides was extracted from defatted oil cakes (pumpkin and plum) by alkali solution along with isoelectric precipitation. Different proteases such as alcalasa, bromelain, pepsin and trypsin were used for obtained peptides with anti-diabetic properties. Therefore, proteins from by-products could be promising sources of bioactive peptides which are have potential as ingredients of functional foods or nutraceutical applications for the control of diabetes.

Keywords: Protein isolate, Enzymatic hydrolysis, Bioactive peptides, Anti-diabetic activity



***IN SILICO* PHARMACOKINETIC AND TOXICOLOGICAL ANALYSIS OF BENZOXAZOLES AS ANTIMICROBIAL AGENTS**

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Among all the heterocyclic compounds, benzoxazole scaffold is one of the most important heterocycles exhibiting numerous pharmacological activities thus providing the basis for the design of many relevant therapeutic agents. Pharmacokinetic and toxicological properties of 35 benzoxazoles and standard drugs isoniazid, ampicillin, gentamicin and ketoconazole were evaluated using computational techniques namely molinspiration, AdmetSAR and Data warrior. 33 heterocyclic derivatives and antimicrobial drugs except gentamicin met all criteria for satisfying oral bioavailability on the basis of Lipinski's rule. Bioactivity score of the compounds was predicted for drug targets including G protein-coupled receptor and nuclear receptor ligands, protease, kinase and other enzymes, and ion channel modulators. The majority of compounds expressed mainly moderate interactions with all drug targets. However, the most promising 24 compounds which are predicted to act by more than 4 proposal mechanisms are identified. Ketoconazole may have effects on all proposed targets, while ampicillin and gentamicin may have effects on 4 drug targets. Otherwise, the lack of activity is predicted for isoniazid. Absorption properties prediction of all proposed benzoxazoles, isoniazid and ketoconazole showed good blood-brain barrier penetration and human intestinal absorption. 11 benzoxazole is supposed to have positive colon adenocarcinoma permeability, as well as isoniazid and ketoconazole. None of the evaluated compounds and standard drugs was predicted as cytochrome P450 isoenzyme 2C9 (CYP2C9) and/or isoenzyme 2C9 (CYP2D6) substrate, while 27 and ketoconazole were predicted as CYP3A4 substrate. All of the studied benzoxazoles, as well as isoniazid, were predicted to have high CYP 450 inhibitory promiscuity. The larger part of tested heterocyclic derivatives is considered non-toxic. Compound 10 is predicted to have high mutagenic and tumorigenic risks, whereas compound 11 is predicted to have a high tumorigenic risk. For 13 compounds are likely to exhibit the irritant effect. Isoniazid is predicted to exhibit all four toxic effects, whereas the other standard antimicrobial drugs are considered safe to use. *In silico* analysis showed that some of the tested compounds are expected to have the most favourable toxicological and pharmacokinetic profiles, and can be selected as new promising bioactive compounds for further studies.

Keywords: Benzoxazoles, In silico study, Pharmacokinetic properties, Toxicological properties

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QUALITY CONTROL AND COUMARIN CONTENT DETERMINATION OF CINNAMON SAMPLES FROM THE SERBIAN MARKETS

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In vitro and *in vivo* researches of cinnamon indicate the high medical potential of this widely used spice. Significant effects are antimicrobial, antiinflammatory, the effect on lowering cholesterol levels and lowering blood glucose levels. It is also an ingredient in various dietary supplements intended for coronary disease and diabetes prevention. However, the problem arises because there are hundreds of species and subspecies of cinnamon plant, official source being *Cinnamomum verum* (*zeylanicum*) -Ceylon cinnamon, while *Cinnamomum cassia*-Chinese cinnamon and *Cinnamomum burmanii*-Indonesian cinnamon are commonly found on the market. They may differ greatly in quality and safety of use. The important difference is that cassia cinnamon may contain significantly higher content of coumarin, a compound that has hepatotoxic activity. The aim of this paper was to examine the quality of cinnamon that is available on the Serbian market according to the Rules on the quality of spices. Also the aim was to determine coumarin content in each sample and to analyze their main component in essential oils. The hydro-distilled volatile oils (according to Ph. Eur. IV) of three cinnamon types were analyzed using GC-MS. The content of coumarin was determined by HPLC method.

Results showed that only the powder of the organic Ceylon cinnamon meets the standard of the regulation for the content of essential oil. While all samples have satisfied the regulation for the maximum content of total ash and ash insoluble in HCl. The GC-MS analysis showed that cinnamaldehyde was the major component in both samples. But values of cinnamaldehyde mostly were below the lower limit prescribed by the 6th Ph. Eur. (55% to 75%). The coumarin vary between different cinnamon species. Since cinnamon is widely used as food additive with health benefits, all stages of its supply chain should undergo stricter criteria in order to avoid potential health risks.

Keywords: Cinnamon, Food safety, Coumarin, Essential oils, Food fraud

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ISOLATION OF ESSENTIAL OIL FROM ORGANIC *MENTHA PIPERITA* L. BY EMERGING EXTRACTION AND DISTILLATION TECHNIQUES

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Essential oils are gaining interest from the academic and industrial communities due to wide specter of biological activities and potential for application in pharmaceutical and food sector. The arrival of the „green” chemistry set its demands for reduction of toxic organic solvents consumption and improved valorization of natural resources. Emerging technologies development and application aims to ensure the intensification of processes and profitable production of quality extracts. The aim of this work is to investigate the possibility of using different extraction techniques for isolation of essential oil from organic peppermint (*Mentha piperita* L.). Soxhlet extraction and hydrodistillation were used as a conventional techniques, while microwave-assisted hydrodistillation (MWHD), ultrasound-assisted (UAE), microwave-assisted (MAE) and supercritical fluid extraction (SFE) were applied as novel techniques. Obtained extracts and essential oils were compared in terms of total extraction yield, chemical profile of terpenoids determined by GC-MS and HPTLC methods and *in vitro* antioxidant activity determined by DPPH assay. Results suggested that MWHD significantly improved kinetics and yield comparing to conventional techniques for recovery of pure essential oil. On the other hand, SFE was particularly suitable for production of antioxidant-rich and solvent-free extracts.

Keywords:* *Mentha piperita, Extraction, Hydrodistillation, Microwaves, Supercritical fluid extraction

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**EXTRACTION OF WILD THYME (*Thymus serpyllum* L.):
COMPARISON OF SUPERCRITICAL CO₂ EXTRACTION WITH CONVENTIONAL METHODS**

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Wild thyme (*Thymus serpyllum* L.) is a medicinal and aromatic plant native to Mediterranean region that contains valuable essential oils with endorsed therapeutic properties. In this work, wild thyme oil was extracted with supercritical CO₂ as green extraction method and compared with conventional extraction methods – Soxhlet extraction with methylene chloride and hydrodistillation. Extraction parameters were as follows: pressure 100, 200 and 300 bar, which is corresponding to supercritical CO₂ density of 628.7, 839.9 and 910.0 kg/m³, temperature 40°C, CO₂ mass flow rate 0.192 kg/h and extraction time 4 h. The results showed that supercritical CO₂ extraction parameters had a significant influence to extraction yields that ranged from 0.84 to 1.44 %. The extraction yield was improved with longer extraction time and with increase in pressure, i.e. increase in supercritical CO₂ density. During the first hour of extraction, fast extraction period was noted for all extraction conditions, but the difference was noted after the first hour of extraction. At the density of 628.7 kg/m³ and 910.0 kg/m³, a slow extraction period was noted for the next hour, followed by period of fast extraction for last 2 hours of extraction. The extraction yield of the Soxhlet extraction was 3.08±0.03 % which is 2.14 times higher compared to supercritical CO₂ extraction, while it was the lowest for hydrodistillation (0.08±0.03 %). In conclusion, the highest yield of supercritical extraction was obtained with 300 bar. Significantly higher yield was obtained with Soxhlet extraction, but considering the long extraction time, usage of organic and toxic solvents and inability to adjust selectivity of the Soxhlet extraction, it is not an optimal method for wild thyme extracts. Supercritical CO₂ extraction is considered a green alternative extraction method that is in terms of sustainable production, meaning that environmental and health safe product are obtained with high yield of lipophilic extracts.

Keywords: *Thymus serpyllum* L., Supercritical CO₂, Essential oil, Green extraction, Sustainable production

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PHENOLIC PROFILE AND ANTIOXIDANT PROPERTIES OF STRAWBERRY WINE

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Strawberries are rich source of many natural active principles. The most interesting among them are those one which exhibit antioxidant properties. As a derived product, strawberry fruit wine, is rich source of those natural active principles. Fruit wines were produced in microvinification which was conducted with two different pure selected yeast cultures and enzymatic preparation glycosidase. Determinations of total phenolic content (TPC) were conducted using Folin-Ciocalteu method. Antiradical activity estimated by DPPH method, while FRAP method was also applied. Selected phenolic compounds were quantified by UPLC TQ-MS/MS. The TPC for the strawberry wines were in interval 1372-1757 mg GAE/L while FRAP was 37.1- 55.7 mmol/L Fe²⁺. The IC₅₀ anti DPPH radical activity was from 4.7 to 7.7%. Also were quantified compounds which exhibit antioxidant properties which are hydroxybenzoic acid derivatives, such as vanillic, gallic, protocatehuic and *p*-hydroxybenzoic acid. The obtain results indicate that strawberry wine is good source of antioxidant compounds. Antioxidant properties and quantity of phenolic compounds depends from the technological process applied in the production of fruit wines. Strawberries and their derived products have beneficial health effect for overall health.

Keywords: Fruit wine, Strawberry, Phenolic profile, Antioxidants, Phenolic acids



IONIZATION BEHAVIOR OF URSODEOXYCHOLIC AND DEOXYCHOLIC ACID IN THE BINARY MIXED MICELLES WITH NONIONIC SURFACTANT TWEEN 60

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Bile acids, as biosurfactants, have been extensively studied for decades. Nevertheless, their application has been limited because they build micelles with low aggregation number. In order to overcome this problem, the mixed micelles systems with other types of surfactants, were developed. In this paper, the binary mixtures of ursodeoxycholic acid (UDHA) and deoxycholic acid (DHA) with Tween 60 (T60) in aqueous solution are examined: The pK_a values of UDHA and DHA are determined using the method of potentiometric titration, in the mixtures with different molar ratios of T60 and with different ionic strength of aqueous solution. The pK_a values of UDHA and DHA in the mixed micelles with Tween 60 (T60) were determined by potentiometric titration, using Radiometer Analytical TIM870 Titration Manager, with hydrochloric acid as a titrant. Micellar systems with different molar fractions of UDHA/DHA anions and T60 (0% T60, 10% T60, 20% T60, 30% T60, 40% T60 and 50% T60) were titrated. Ionic strength was adjusted by adding sodium-chloride in different concentrations (100 mmol/dm³, 300 mmol/dm³ i 500 mmol/dm³). Results show that pK_a values of UDHA and DHA in the mixed micelles were increasing as the molar fraction of T60 was raising. Series of solutions with the highest ionic strength showed have the highest pK_a values, for both UDHA and DHA in the binary mixtures with T60.

Based on the structure of mixed micelles with higher molar ratio of bile acids, whose exterior part is made of the negatively charged UDHA/DHA anions and interior part of T60, it would be expected that the increase of molar ratio of T60 would lead to the decrease of the pK_a values of UDHA/DHA. The obtained results could be explained by the change in the structure of the mixed micelle with higher molar ratio of T60, where the interior part of the micelle is made of T60, while the UDHA/DHA anions are tangentially positioned on the surface of the micelle. The protonated state of bile acids therefore would allow better incorporation into the hydrophilic pockets of the micelles, which are presented in the mixed micelles with higher content of T60. Increase in ionic strength of the solution additionally stabilize the micelle, due to neutralisation of carboxyl groups with sodium ions, which explains the increase of pK_a values of UDHA/DHA in the solutions with higher ionic strength. Increase in molar ratio of T60 in mixed micelles with UDHA/DHA leads to the changes in the structure of the micelles, which is reflected by higher pK_a values of the acids incorporated in micelles.

Keywords: Binary mixed micelles, Bile acid, Tween 60, pK_a



CHEMICAL COMPOSITION OF *CLINOPODIUM VULGARE* L. EXTRACTS AND THEIR ANTIOXIDANT AND CHOLINESTERASE INHIBITION POTENTIAL

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Clinopodium vulgare L. (Lamiaceae) is widely distributed in Europe, Northwest Asia, North Africa and America. In ethnopharmacology of Bosnia and Herzegovina, *C. vulgare* is used in the treatment of gastrointestinal disorders. Bioactive components of extracts have an important role in the prevention and treatment of chronic diseases related to oxidative stress. One of them is Alzheimer's disease, the most common form of dementia. In the treatment of Alzheimer's disease, inhibitors of cholinesterases are also used. Medicinal plants are potential source of cholinesterase inhibitors which is why they are subject of numerous research.

The aim of this work was to determine chemical composition as well as antioxidant and anticholinesterase potential of *C. vulgare* hot water and methanol extracts. Using HPLC/DAD technique, rosmarinic acid, elagic acid, and ferulic acid were identified as dominant components of the extracts. The results of analyzed extracts showed good antioxidant activity tested by DPPH and FRAP assay. Inhibition of acetylcholinesterase and butyrylcholinesterase was determined using Ellman's method. The obtained results showed low acetylcholinesterase inhibition potential and absence of butyrylcholinesterase inhibition.

Keywords: Clinopodium vulgare, Extracts, Anticholinesterase potential, Antioxidant potential

ENCAPSULATION OF B-CAROTENE FROM CARROT WASTE BY EXTRUSION TECHNIQUE

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After carrot industry processing, up to 50% of the raw material remains as pomace which is generally disposed as feed or manure. However, carrot pomace contains remarkable amounts of residual β -carotene which could be profitably utilized for the fortification and development of functional foods, pharmaceuticals, and medicines. β -Carotene serves as an important source of provitamin A and natural edible pigment, but the application is limited because of its instability toward environmental factors such as oxygen, temperature, and light. Encapsulation provides effective methods aimed at improving their stability, and the principal advantage of the extrusion method is the stability of the bioactives against oxidation. In the present study, carrot waste oil extract was encapsulated in Ca-alginate gel microbeads by electrostatic extrusion technique. Generally, alginate has been approved as a coating material by the Food and Drug Administration (US) and the European Food Safety Authority (Europe). After lyophilization of obtained microbeads, the high encapsulation efficiency (72.76%) was observed. Lyophilization significantly affected morphology and bead size, and the SEM micrographs showed that the extract was successfully incorporated into the particles. FTIR analysis doesn't suggest any strong interaction between alginate and oil/carotenoids mixture, which indicates that after the formation of Ca-alginate microbeads these two phases most remain separated. Due to three months storage period, dry microbeads showed the high retention (81.53%) and potential for preservation of β -carotene suitable for food or pharmaceutical industry.

Keywords: Carrot, Waste, β -carotene, Encapsulation, Storage stability

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PHYTOCHEMICALS CONTENT AND BIOACTIVITY OF PEACH POMACE

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Peach (*Prunus persica* L) is fruit which is nutritionally and economically important and one of the most popular fruit consumed worldwide. Nowadays, there is a growing interest in bioactive compounds of fruits and vegetables due to their known role in preventing diseases such as diabetes, cancer, stroke, arthritis, and aging. Waste products generated primarily during juice processing, are also promising sources of bioactive compounds which may be used because of their favorable technological or nutritional properties.

In this study, the freeze-dried peach pomace was extracted with mixture of acetone and methanol (36:64 v/v). Bioactive compounds (polyphenolics, flavonoids and carotenoids), antioxidant (AA) and anti-inflammatory (AIA) activity in obtained peach pomace extract (PPE) were determined spectrophotometrically. Some individual phenolic compounds were identified and quantified by HPLC. AA of PPE was examined using three different tests: DPPH[•], ABTS^{•+} and reducing power. Antidenaturation of egg albumin method was chosen to evaluate AIA of PPE. Diclofenac sodium (DS) was used as positive control. Total polyphenolic, flavonoid and carotenoid contents were: 642.77 mg gallic acid/100g, 1013.82 mg rutin/100g and 0.27 mg β -carotene/100g, respectively. The most dominant phenolic compound in PPE was chlorogenic acid. The obtained IC₅₀ values in antioxidant tests were: IC₅₀^{DPPH} = 19.43 mg/ml, IC₅₀^{ABTS} = 11.28 mg/ml and IC₅₀^{RP} = 9.30 mg/ml, respectively. The percent inhibition of AIA was 75% for PPE at the concentration of 75.76 mg/ml, while for DS the same percent inhibition was achieved at the concentration of 18.24 mg/ml.

These results suggest peach waste as a rich source of phytochemicals, and relatively strong antioxidant and anti-inflammatory activity, which could be used as functional food, food additive and in pharmaceutical industry.

Keywords: Peach, Pomace, Phytochemicals, Bioactivity

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COLD PRESSING VS. SUPERCRITICAL CO₂ EXTRACTION OF PLUM (*PRUNUS DOMESTICA* L.) KERNEL SEED

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Plum (*Prunus domestica* L.) is a fruit widely grown across Europe and its processing accounts for an important part of the total plum output. Consequently, after processing a considerable amount of plum, kernels are discharged as by-product. This creates a new opportunity to exploit plum kernels in order to provide an alternative to conventional edible oils.

The main aim of this study was to obtain high-quality oil from plum kernel seeds, without refining or other post-extraction treatments, using traditional cold pressing (CP) and supercritical carbon dioxide extraction (ScCO₂) as a modern technology. The oils were characterized based on the chemical composition of fatty acids and tocopherols. The ScCO₂ extraction was performed using the following parameters: pressure 300 bar, extraction time 5 h, and temperature 40 °C. Cold pressing was conducted using the following parameters: head presses temperature 60 °C, frequency 20 Hz and, a nozzle ID 6 mm.

In obtained oils, 12 fatty acids were identified. The oleic acid was the most dominant in both oils (68.66% in oil obtained by ScCO₂, 65.86% in oil obtained by CP), followed by linoleic acid (22.24-25.44%). Further, a significantly lower amount of saturated fatty acids like palmitic and stearic acids were determined. The majority of $\beta+\gamma$ -tocopherols were obtained after two hours of ScCO₂. After 1 h of ScCO₂, the amount of α -tocopherol is 2-fold higher than the amount obtained by CP. The similar ratio is noticed after 1 h of ScCO₂ in $\beta+\gamma$ -tocopherols and δ -tocopherol amounts compared to the ones obtained by the traditional method. It is evident that the highest solubility of tocopherols was in the first hour of the extraction. Solubility decreases with further extraction, and after 5 hours, a 25 times lower amount of α -tocopherol was extracted in comparison to the first hour. Total tocopherols content in oil obtained by ScCO₂ was 4 to 5.8-fold higher than CP. Obtained results justify the further processing of plum kernels as by-products of the fruit industry for the production of oil for potential food and pharmaceutical applications.

Keywords: *Prunus domestica, Supercritical carbon dioxide extraction, Cold pressing*

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SPRAY DRYING OF LIQUID EXTRACT OBTAINED FROM *ARCTOSTAPHYLOS UVA-URSI* HERBAL DUST

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During the production of filter tea from *Arctostaphylos uva-ursi*, from 10 to 20% of *A. uva-ursi* by-product, or "uva herbal dust" is produced, which is usually discarded as waste. *A. uva-ursi* is traditionally used to treat inflammations of the urinary tract, it has strong antioxidant, antibacterial, and diuretic effects. Due to the high content of pharmacologically active components in the native material, the assumption was that uva herbal dust could be used as a starting raw material for production of new herbal preparations.

Considering the advantages of dry extracts over liquid ones (simplified handling, transport and storage, longer shelf life...), the main goal of this study was to produce dry extract (powder) from the optimal liquid extract of *A.uva-ursi* obtained by subcritical water extraction. For this purpose spray drying was applied and the results were analyzed through the impact of process parameters (inlet and outlet temperature, concentration of the carrier – maltodextrin) on the characteristics of the obtained powders: hygroscopicity, bulk density, moisture content, water absorption and water solubility index and process efficiency.

In the spray drying process maltodextrin (MD) in different concentrations (0 to 40%) was used as a drying agent, while the inlet drying temperature was 120-140 °C. Efficiency of drying was higher than 60% in all cases. When inlet temperature was increased to 140 °C, the moisture content decreased (5.514%). It was noticed that with an increase of MD concentration hygroscopicity decreased. However, the results indicate that increased temperature and increased MD content in combination may affect conversely, that is to lead to the increase of hygroscopicity. Based on the results, by using spray drying technique, the liquid extract of *A. uva-ursi* herbal dust was successfully transformed into a dry extract in powder form, and as such it was used to produce a granulate that can be applied in the form of instant tea, which would be used for the normal functioning of the urinary tract, but also as a natural source of antioxidants.

Keywords: Spray drying, A. uva-ursi, Maltodextrin

EFFECT OF DIFFERENT SPRAY DRYING CONDITIONS ON MICROMETRIC AND STRUCTURAL CHARACTERISTICS OF BRINE OBTAINED FROM CABBAGE FERMENTATION

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Brine samples investigated in this research originated from the controlled cabbage fermentation which was set in industrial conditions. Cabbage was fermented at 18 °C, with 7% NaCl and with addition of starter culture. Obtained brine was then exposed to spray drying at different conditions of drying. Brine was dried at temperatures 120 and 140 °C with addition of 10, 30, and 50% maltodextrin as a carrier. The main goal of this research was to investigate influence of these different conditions of spray drying on the micrometric and structural characteristics of obtained dried brine powder. Thus, the main micrometric and structural analysis, such as particle size analysis, scanning electron microscopic analysis, bulk and tapped density, flowability and powder wettability were observed. The structure was investigated by thermoanalytical method and X-ray powder diffraction. The size of obtained micronized products was around 3-4 µm, except in the sample obtained at temperature 120 °C with 10% of maltodextrin, where it was around 8 µm. All particles had a spherical shape consisting of nanosized aggregates ranging between 200 and 500 nm in size. Hydrophilic properties and semi-crystalline structure of powders were detected.

Keywords: Brine fermentation, Spray drying, Micrometric and structural characteristics, Powder flow properties

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Petroleum Refining and Production



SYNTHESIS OF COLD FLOW ADDITIVES TO ECOLOGICAL PURE DIESEL FUEL BY METHODS OF CONTROLLED RADICAL POLYMERIZATION

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The most effective method of modifying the properties and characteristics of environmentally friendly diesel fuels of Euro-5 class is the introduction of additives into their composition. In particular, to improve the low-temperature properties of diesel fuels, additives based on homo- and copolymers of vinyl acetate, acrylonitrile, and methacrylic monomers are introduced into their composition. At the same time, the molecular weight characteristics of the synthesized polymers, as well as the composition and structure of the copolymers, have a key influence on the effectiveness of the synthesized additives. The most effective tool for the directed synthesis of polymers with desired molecular weight characteristics and properties is controlled radical polymerization or “living” radical polymerization.

In this work, the methods of controlled radical polymerization using the Reversible Addition-Fragmentation Chain Transfer (RAFT) and Atom Transfer Radical Polymerization (ATRP) methods were used to synthesize homo- and copolymers based on stearyl methacrylate, acrylonitrile, vinyl acetate and glycedyl methacrylate. The influence of the composition and structure of stearyl methacrylate copolymers (statistical or block copolymers) on the effectiveness of their use as depressor additives to Euro-5 diesel fuel has been evaluated. Catalytic systems based on copper and ruthenium complexes (ATRP), as well as thiobenzoates as regulating agents (RAFT), were used to initiate polymerization.

The optimal range of molecular weight of stearyl methacrylate homopolymers and copolymers has been established experimentally when used as depressant and depressant dispersant additives. It was shown that the introduction of polar comonomers, in particular, acrylonitrile and glycedyl methacrylate into the composition of the polymer molecule leads to an increase in the effectiveness of the action of additives. At the same time, statistical copolymers based on stearyl methacrylate and the above comonomers are more effective as additives than the corresponding block copolymers.

The effect of the synthesized homo- and copolymers of stearyl methacrylate on the oxidative stability of diesel fuel has been evaluated. It was shown that a number of synthesized additives have a complex effect: they simultaneously lower the pour point of diesel fuel and limiting (maximum) temperature of filterability and increase its oxidative stability.

Keywords: Polymers, Diesel fuel, Depressant additives

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PETROLEUM REFINING IN SERBIA: INVESTMENTS, DEVELOPMENT PROJECTS AND ACTIVITIES

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Most of the petroleum refineries in the region of south-eastern Europe were built in the 1960s and 1970s when the main petroleum product on the market was gasoline. The main petroleum refinery on the Serbian market is oil refinery Pančevo. This refinery was put into operation in 1968. To achieve Euro 5 standards of fuel quality (diesel and gasoline) and improve the energy efficiency of the downstream sector, the NIS a.d. made large investments in the modernization of petroleum refinery Pančevo. In 2019 NIS a.d. will finish the construction of deep conversion complex, or bottom-of-the barrel, as part of an ongoing modernization program to improve energy efficiency and boost production of improved quality petroleum products. In this paper, the transformation of the oil refinery in Pančevo is analyzed, as well as the impact of plans to execute several major investment projects. The most crucial projects of modernization of refining capacities are the construction of delayed coking unit and combined heat and power plant. It is expected that the start of delayed coking unit will increase processing efficiency of refinery Pančevo from a current 85.5% to 99.2%, as well as the production of high-quality, low-sulfur diesel and gasoline by more than 38%. Furthermore, it is expected increasing of jet fuel production by 11%, and LPG by 7%, while Nelson's complexity index will be increased from 8.5 to 9.5. The cross effects between existing and newly built units, as well as the impact of the change of crude assay on the output of petroleum products will be analyzed.

Keywords: Petroleum refining, Residue conversion, Delayed coking unit

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DEMULSIFICATION OF WATER-IN-CRUDE OIL EMULSIONS USING NEURAL NETWORKS

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During the crude oil exploitation, emulsions are formed due to the presence of natural surfactants in both the water and oil phase. The breaking of crude emulsions usually requires the presence of demulsifier in order to separate the crude oil from the water. This work investigates the impact of various water-soluble and oil-soluble demulsifiers on the separation efficiency of water from crude oil. Factors such as the demulsifier chemical structure and concentration, water content, and temperature were taken into account during testing. The procedure is developed to achieve reliable removal of water from crude oil and to optimize the dosage amount of the selected demulsifier. Reliable mathematical models for optimization of the demulsification are scarce due to the complex phenomena involved in the separation of water from crude oil. The objective of this work was to investigate the possibility of using artificial neural networks for the analysis and prediction of the separation efficiency. Full factorial design was used to investigate the separation efficiency of water from crude oil emulsion over a broad range of working conditions. Six commercially available demulsifiers were tested in order to separate water-in-crude-oil emulsions of crude oil from Serbia. The following crude oil properties were experimentally determined or estimated using standard methods: density, kinematic viscosity, paraffin content, total asphaltene content, mean relative molecular mass, pour point, and salt content. For each emulsion prepared, basic sediment and water content of crude oil were measured. Demulsification was performed using bottle test in the wide temperature range. Demulsifier concentration was varied from 20 to 50 ppm. The effect of demulsifier's type was followed using dependence of the separation efficiency on settling time. It was found that demulsification efficiency increases with increase in temperature and settling time for all applied demulsifiers. The tested neural networks enable reliable and accurate prediction of demulsification efficiency with the lowest values of raw residuals. The proposed statistical approach greatly simplifies the estimation of demulsification efficiency.

Keywords: Demulsification, Crude oil, Emulsion, Artificial neural network

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EFFECT OF EXTENDER OIL NATURE ON THE RUBBER CROSSLINK DENSITY

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Specific properties of natural rubber based products depend on the concentration and nature of reinforcing fillers, extender oil, as well as additives, stabilizers, antioxidants and vulcanization agents. Natural rubber as the main ingredient for rubber compounding is elastomeric material with a low degree of crosslinking. Crosslink density is characterized by the number of crosslinks per gram of rubber or the average molecular weight between crosslinks. Equilibrium swelling experiments in toluene were carried out on rubber samples produced by varying mineral extender oil concentration (0, 10, 20 and 30 phr). The experiment lasted for 72 hours at room temperature and the mass and volume of each sample were measured over time. The classical Phantom and Flory–Rehner approaches were used in this study. These models consider the volumetric fraction of rubber and the average molecular weight between crosslinks and were applied for data modeling. The results showed that the crosslink density depends on the paraffinic, naphthenic, and aromatic content of investigated mineral extender oils. The samples with the naphthenic oil showed the lowest crosslink density, while the paraffinic exhibited the highest value.

Keywords: Natural rubber, Swelling, Crosslink density, Extender oil

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AN OVERVIEW OF ARTIFICIAL LIFT SELECTION METHODS WITH CASE OF SELECTION USING MULTI-CRITERIA DECISION-MAKING METHODS

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The most commonly used methods of oil exploration in the world are Gas Lift (GL), Sucker Rod Pumps (SRP), Electric Submersible Pumps (ESP), Progressive Cavity Pumps (PCP), Hydraulic jet type Pump (HP). To achieve efficient production, an important role is the selection of most adequate artificial lift methods. The best selection of the method achieves economic production. In order to achieve the optimal selection of artificial lift methods use different approaches expert systems and multi-criteria analysis.

In this paper is presented overview of artificial lift selection methods and analysis of the multicriteria models application (VIKOR, TOPSIS, ELECTRE, AHP, and PROMETHEE)

Keywords: Artificial lift methods, Oil production, Decision making, Optimization, Multicriteria models



RENEWABLE JET FUEL PRODUCTION FROM VEGETABLE OIL: ASPEN SIMULATION AND TECHNO-ECONOMIC STUDY

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With the growing demand of air transport and concerns about its environmental impacts, alternative jet fuels derived from non-conventional sources have become an important strategy for achieving a sustainable and green aviation. The use of alternative jet fuels is being considered to reduce the dependency of the air transport sector on fossil derived fuel. Jet fuel produced from plant-derived and renewable sources has the potential to decrease the net greenhouse gas (GHG) emissions of the aviation industry. These renewable energy sources have high potential being well-matched with existing gas turbine powered aircraft engines and technology pathway has been already available on commercial stage. Waste cooking oil is a promising renewable feedstock for bio jet fuel production by hydroprocessing. The search for a commercially feasible and sustainable process that produces a long-lasting renewable jet fuel with low production costs and low greenhouse emissions are major objectives in this attractive and developing field.

Many technologies that convert biomass-based materials into jet fuel substitutes are available. Some are available at commercial or pre-commercial scale, and others are still in the research and development stage. Although a variety of feedstock and processes exist, only three processes have been approved for use in conventional aircraft. These include the gasification with Fischer-Tropsch synthesis (GFT) process, the HEFA process (process that hydroprocesses vegetable oil to fuel) and the direct fermentation of sugar to jet (DFSTJ) process. Industries are currently working on developing optimal processes that utilize sustainable feedstocks and can be produced economically. So far, bio-jet fuels from Fischer-Tropsch (F-T) synthesis and oil hydro-processing technologies have been approved by ASTM International Method D7566 for blending into conventional jet at levels up to 50%.

In this work the simulation and optimization of hydroprocessing technology for bio-jet fuel is performed by Aspen Plus model with techno-economic analysis. This research has shown, by sensitive analysis, that the overall cost of bio jet fuel production needs to be lower either by the reduction of raw material cost, by the decrease of energy consumption (process incorporated in refinery), or by the application of more efficient processing technology.

Keywords: *Bio-jet fuel, Aspen simulation, Techno-economy study*

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SIMULATION AND MODELLING OF AN INDUSTRIAL HYDROTREATING PROCESS USING VACUUM AND LIGHT GAS OIL - IMPACT OF CATALYST WETTING EFFICIENCY AND HYDRODEAROMATISATION REACTION KINETIC'S TYPE

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Over the years, hydrotreating process has greatly developed in order to reduce level of emissions from vehicles, through the setting of stringent limits of the concentrations of sulphur and aromatics contained in fuels. Improved understanding of hydrotreating has been acquired over the past decades through systematic study of different aspects of this complex process. Due to the complexity of hydrotreating it would be beneficial to develop further understanding of the process through mathematical models which are based on the industrial scale data and real feedstock. A reliable mathematical model can be crucial for process design, determination of operating parameters, and for the understanding of the entire process. Studying and improvement of hydrotreating operation should be considered through appropriate kinetics and process modelling. In this research work, different kinetic models for reaction of hydrodearomatisation and the influence of reaction rate on performances of the industrial trickle bed reactor for hydrotreating of gas oil and light cycle oil have been analyzed. Moreover, several models for wetting efficiency in TBR on industrial level are examined and influence on reaction rate and reactor performance is discussed.

Mathematical model for co-hydrotreating of straight run gas oil blended with fluid catalytic cracking naphtha and light cycle oil was developed previously using axial distribution of phase equilibrium and effective wetting in the catalytic reactor. Model assumes that hydrodesulphurization and hydrodearomatisation reactions occur on the catalyst surface which is in contact with vapour or liquid phase. Hougen-Watson type kinetic equations for different classes of sulphur compounds were applied in separate material balance equations for vapour and liquid phase. Mass transfer effects were taken into account using overall catalyst effectiveness factor for each of the reactions and overall energy balance equation was included in the model. Model results were validated using the industrial test run data. The change of temperature, catalyst wetting efficiency, and conversions of sulphur and aromatics have been determined. The results of the simulations have been compared with experimental data from the industrial test run and the best model for the observed process was analyzed comparing agreement between experimental point and model predictions of temperature, aromatic and sulphur conversions.

Keywords: *Hydrodesulphurization, Hydrodearomatisation, Modeling, Catalyst wetting efficiency*

Acknowledgements: *Financial support through project III-45019 of the Ministry of Education and Science of the Republic of Serbia is gratefully acknowledged.*

SYNTHESIS OF Re/Pd AND Co/Mo HETEROGENEOUS CATALYSTS ON MESOPOROUS SILICA USING SOL-GEL METHOD FOLLOWED BY SUPERCRITICAL DRYING WITH EXCESS SOLVENT

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Textural characteristics of catalysts and catalysts supports are of prime importance when it comes to their application in processing of high molecular weight compounds. Mesoporous silicas doped with Ti-ions is founding promising application as heterogeneous catalysts support obtained by sol-gel procedure. Drying procedure is a key factor in preservation of wet gel properties. In this paper, the effect of supercritical drying with excess solvent on textural characteristics of novel Pd/Re and conventional Co/Mo catalysts is addressed. PdRe on Ti doped mesoporous silica and Co/Mo silica/alumina (1:1) catalysts are prepared using sol-gel method followed with supercritical drying procedure. Specific surface area, mesoporosity and pore volume of the supports are analysed as the most important parameters for the catalyst application. The materials are characterized using FT-IR spectroscopy, N₂ physisorption and SEM micrographs.

Keywords: Catalyst, Mesoporous Silica, Ti-HMS, Sol-gel, Aerogel, Supercritical drying

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CRUDE OIL MONITORING CHALLENGES IN MODERN REFINERY

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Modern refineries are facing very volatile market, growing of raw material and energy prices and tightening of environmental requirements during last years. This resulted with increase of operation costs and need for operations optimization and flexibility. One of the tasks for achieving these goals is proper crude oil selection. Alternative crude oils, so called opportunity crudes, are getting more interesting because of their growing availability and lower prices. On the other side, they are challenging to process due to unknown composition and compatibility issues that may arise. Understanding of crude oil behavior is very important nowadays, not only in refining process but during distribution chain and storage as well. This work will present crude oil quality monitoring schedule in NIS Pančevo Refinery and laboratory analysis used in Research Department in order to provide appropriate crude oil quality control.

Keywords: Crude oil, Refinery, Monitoring



TESTING OF EXPLOITATION PROPERTIES OF HYPOID OIL

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Engine and transmission technologies have advanced rapidly in recent years. Consequently, the development and production of new transmission types, have gone into volume production contributing to the increase in hypoid oil consumption. Hypoid oil (gear oil) is a lubricating oil produced specifically for transmissions, transfer cases, and differentials in the automotive industry and other machinery. Usually, hypoid oil has a high kinematic viscosity and contains organosulfur compounds. The aim of this paper was to determine the degree of hypoid oil degradation as well as the optimal exploitation age of the hypoid oil and change in kinematic viscosity. The hypoid oil SAE 80W-90 produced in accordance with standard API GL-5 quality and commonly used as bus gear oil was tested. Therefore, this hypoid oil was tested in real conditions of exploitation. Exploitation testing involved the change in physical and chemical characteristics of hypoid oil, as well as qualitative and quantitative analysis of wear metallurgy, through standard laboratory methods. A sample of the hypoid oil was taken after the 70,000 km crossed, which is equivalent to a one-year period of using the hypoid oil in transmission system. The results were analyzed by analytical and statistical methods showing that the optimal replacement period of the hypoid oil is 100,000 km.

Keywords: hypoid oil, lubricating oil, gear oil, exploitation



NEW APPROACHES TO THE THIN-LAYER CRACKING OF ACID TARS

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Acid tars are close in their hydrocarbon composition to vacuum residue produced by distillation of petroleum: they mainly consist of high-boiling-point polyaromatic hydrocarbons, resins, oils, and asphaltenes. Liquid fuel, asphalt materials, and coke are industrially produced by the thermal cracking of petroleum vacuum residue. This line in hydrocarbon processing can also be applied to acid tars, which should be considered a promising source of various types of useful hydrocarbon-containing products. Up to now, attempts to obtain quality liquid fuel and asphalt materials by the thermal cracking of acid tars have been unsuccessful. The main reason for the failure is the presence in acid tars of considerable amounts of sulfonic acids and sulfuric acid, which fundamentally change the mechanism of thermal cracking. In acid tar, hydrolysis and desulfurization of sulfonic acids and redox reactions of sulfuric acid and hydrocarbons begin already at 120°C. Low-temperature cracking of sulfonic acids and decomposition of sulfuric acid in acid tars does not lead to a positive outcome, and only thermal degradation of high-boiling-point polyaromatic hydrocarbons, resins, and asphaltenes at 400–450°C or above 500°C allows liquid fuel and asphalt materials or liquid fuel and coke, respectively, to be obtained. If the cracking of sulfonic acids and high-molecular-mass hydrocarbons of acid tars be combined in one technological process, it will be possible to obtain asphalt materials and a wide-boiling-range hydrocarbon fraction. For this purpose, the method of thin-layer thermal cracking of acid tar has been developed at Lobachevsky University. Processes for the manufacture of liquid fuel, coke, and an asphalt binder from acid tars by thin-layer cracking method and the designs of thin-layer cracking devices for their implementation have been patented.

The chemical processes of thin-layer cracking of acid tar hydrocarbons obey the same laws as and the cracking of petroleum residue hydrocarbons. The cracking of higher petroleum hydrocarbons follows the first-order rate law at a small depth of thermal decomposition regardless of the component nature. At 200–350°C, the reactions of thermal degradation of sulfonic acids and desulfurization of asphaltenes and resins yielding a certain amount of liquid hydrocarbons mainly proceed in acid tars, whereas the processes of cracking of high-molecular-mass compounds to asphaltic products and volatile hydrocarbons occur above 400°C. To preclude the low-temperature hydrolysis of sulfonic acids and the catalytic action of sulfuric acid on the thermal degradation of the acid tars components, an acid tars sample washed of sulfuric acid and soluble sulfonic acids with water was used in the kinetic studies.

Keywords: *Acid tars, Cracking, Asphalt material, Thermal studies*



STRUCTURE-BASED MODELS FOR THE PREDICTION OF FLASH POINT OF MULTI-COMPONENT FUEL MIXTURES

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New models based on quantitative structure–property relationship (QSPR) have been developed to estimate the flash point of petroleum fractions. The fractions were analyzed as multi-component mixtures. For the model development, a large data set including experimental data on the flash point of different mixtures including numerous chemical families was applied. Enhanced replacement method was employed for reducing the independent variable dimensionality. The artificial neural network (ANN) was proposed in order to select and map input structure–property variables known as descriptors to the flash point output variables. Having in mind that ANN results, including the weight values depend on the initial assumptions of parameters necessary for ANN construction and fitting, the series of different ANN topologies were used. Moreover, the transfer functions and training algorithms received special attention in order to ANNs obtain better performances. The prediction capability of the developed models was evaluated using statistical criteria. In the end, all applied statistical methods confirmed that the proposed models are reliable and accurate.

Keywords: Petroleum fractions, Quantitative structure–property relationship (QSPR), Artificial neural network (ANN), Multi-component mixtures, Flash point



ADVANCED OIL AND GAS PRODUCTION AND PROCESSING TECHNOLOGIES FOR ARCTIC CONDITIONS

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The future trends of oil and gas industry are heading to the Arctic region characterized by long winters, low mean temperatures, permafrost and ice cover. Current projects of oil and gas production are operated at temperatures from -20°C to -60°C and all offshore structures must withstand at these extreme conditions. Commonly used construction materials like stainless steel and concrete can change their mechanical properties at low temperatures and become more brittle, which can lead to fracture generation and equipment failure. Overcoming the Arctic harsh environmental conditions requires innovative technologies in crude oil production and processing. All applied technologies in the Arctic region must be efficient, reliable and environmentally-friendly. In this paper oil and gas, production and processing aspects were analyzed with the emphasis on the Arctic conditions. The existing technologies are reviewed and discussed for application in the Arctic environment. Additionally, guidelines and improvements for future projects are discussed and analyzed.

Keywords: Arctic, Oil and gas, Production, Processing

ASPEN HYSYS SIMULATION OF NATURAL GAS DEHYDRATION PROCESS

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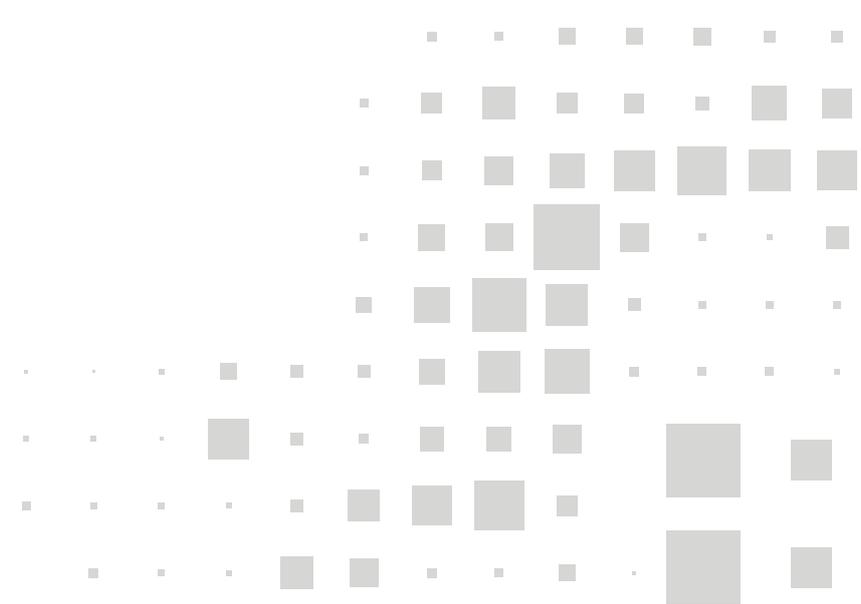
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Despite the trend of replacing fossil fuels with some renewable energy sources in the global energy production over the past ten years, natural gas will certainly remain a significant source of energy and a raw material for the chemical industry in the future. Namely, among the fossil fuels, natural gas has the lowest carbon dioxide (CO₂) emission factor per unit of energy released, but also the smallest emission of nitrogen (NO_x) and sulfur (SO_x) oxides, as well as particulate matter (PM). Thus, natural gas is considered to be an ecological fuel.

Natural gas obtained from different wells is usually collected at gas or oil-gas collection stations and subsequently treated within gas preparation facilities, prior to be delivered to customers, in order to fulfill the requirements defined by the regulations. Natural gas preparation processes are used to remove water and/or substances like CO₂ and hydrogen sulphide from the gas. Water in natural gas increases the risk of damage of the transportation facilities due to corrosion in contact with H₂S and CO₂, but also because of the formation of hydrates when transporting gas through pipes at low temperatures and high pressure.

This paper provides results of Aspen Hysys software package simulations of the natural gas dehydration process. A low and a medium pressure natural gas streams were gathered up in the gas collection station and the obtained wet natural gas stream was dried in an absorption unit, using triethyleneglycol (TEG) as the absorbent, prior to be delivered to the main gas pipeline. The production capacity of the dry natural gas was about 25000 kg per day. The influence of the change in the TEG flow rate on the moisture content of the outgoing gas was examined at different pressures. As it was expected, an increase in TEG flow decreased the moisture content of the output gas. The result shows that at an absorption pressure of 35·10⁵ Pa, moisture in the dehydrated gas is lowered to a value below 100 ppmw at a TEG flow of 0.04 m³/h; and at a pressure of 8·10⁵ Pa at TEG flows greater than 0.2 m³/h, which is about 5 times higher consumption of TEG. As the gas pressure increases, the moisture content of the outgoing gas decreases, and the effect of the pressure change is more significant at the smaller TEG flows. A drastic drop of moisture content in the gas was observed at manometric pressures of gas up to 14·10⁵ Pa, for all TEG flows.

Keywords: Natural gas, Dehydration, Aspen HYSYS, Triethyleneglycol, Absorption



**Sustainable Development, Chemical and
Environmental Engineering**





LAST ADVANCES ON THE LARGE SCALE PRODUCTION OF BIOACTIVE COMPOUNDS FOR AGRICULTURE AND AQUACULTURE FROM MICROALGAE

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In the last year relevant advances have been developed on the large-scale production of biostimulants, biopesticides and feed additives, in addition to biofertilizers and aquafeed, using microalgae. This research is part of activities related with SABANA project focus to the development of large-scale microalgae based biorefineries. The reliability of large-scale low-cost raceway reactors has been validated, the reactors being operated in full recirculation mode to save water and nutrients. Harvesting of the biomass has been optimized by combining a pre-concentration and dewatering step using dissolved air flotation and nozzle separator. Concerning cell disruption, this step is mandatory to adequately obtain end products. Although ultrasounds allow to achieve the larger breakage of the cells, the utilization of high-pressure homogenizers allows to obtain enough cell disruption for further extraction processes, but saving energy. Extracts obtained from the biomass sludge demonstrate relevant effects both as biostimulant and biopesticide. Concerning biostimulant effect both Auxin-like and Cytokinin-like activities were found, in some cases increases in the plant growth performance larger than 200% being measured. Concerning biopesticides, results demonstrate the inhibition of growth of five of the most relevant fungal pathogens, up to 60%. These effects have been demonstrated at small scale, in experiments performed at laboratory and culture chambers, it being now validated in real field trials. Concerning aquaculture, although the biochemical composition of the microalgae biomass is highly valuable, the digestibility of the biomass is largely dependent of the strain. To improve the digestibility of the biomass it is recommendable to perform a previous cell disruption, both mechanically or enzymatically. Fish trials performed incorporating biomass of different microalgae strains and percentages to Aquafeed demonstrate that the major advantage of incorporating microalgae biomass is not related with the improvement of growth but with the enhancement of health of fishes as probiotic. First end products are being evaluated, prior to market evaluation by companies. Thus, SABANA is expected to conclude with real commercial processes demonstrated at commercial level.

Keywords: microalgae, biostimulants, biopesticides, large scale, aquaculture, biofertilizers



BIOLOGICAL CONVERSION OF GREENHOUSE GASES INTO ADDED VALUE BIO-PRODUCTS: MOVING TOWARDS GHG BIOREFINERIES

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CH₄ and N₂O emissions represent 20 % of the anthropogenic greenhouse gas (GHG) emissions, and their share is expected to increase in this 21st century based on the steady increase in human population. The European Union has committed to reducing its GHG emissions by 40% by 2030 and 60% by 2040 (compared to 1990 levels). Apart from the current actions oriented to reduce CO₂ emissions from fossil fuel combustion, additional measurements such as an active CH₄ and N₂O abatement must be considered in order to achieve these target emission cuts. Despite the increasing environmental relevance of CH₄ and N₂O, minimum attention has been paid to date to these greenhouse gas emissions from a legislation and abatement viewpoint worldwide. This has limited the development of cost-efficient and environmentally friendly end-of-pipe treatment technologies. Physical/chemical treatment methods for CH₄ abatement such as incineration are either inefficient or costly at the low concentrations (1-5%) typically found in emissions from waste treatment activities, coal mining or animal farming. On the other hand, conventional NO_x treatment technologies such as selective catalytic/non-catalytic reduction, adsorption or scrubbing present prohibitive operating costs when treating large volumes of air containing low concentrations of NO_x (as a result of their intensive use of energy and chemical). In this context, biotechnologies could become, if properly tailored, a low-cost and environmentally friendly treatment alternative to physical/chemical methods for the abatement of CH₄ and N₂O emissions. Biotechnologies, which are based on the enzymatic conversion of these GHGs into less harmful products such as CO₂, N₂ or H₂O, have been consistently proven as robust and efficient methods for the treatment of industrial volatile organic compounds and malodours, exhibiting lower operating costs and environmental impacts than their physical/chemical counterparts. Unfortunately, conventional bioreactors such as biofilters, biotrickling filters or bioscrubbers are limited by the mass transfer of these GHGs from the gas to the liquid phase as a result of their high Henry law constant. Under optimal operating conditions, GHGs can be also used as feedstock to synthesize high added-value products such as biopolymers, exopolysaccharides, vitamins or ectoine. The valorization of GHGs through their bioconversion into commodities with a high market value will turn their abatement into a sustainable and profitable process. This keynote will critically review the recent advances in biotechnologies devoted to CH₄ and N₂O removal, and the potential bioconversion of these GHGs into added-value products.

Keywords: bioconversion, climate change mitigation, greenhouse gases, methane, nitrous oxide

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TINY MICROALGAE FOR HUGE APPLICATIONS. BIOREFINERY APPROACH

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Current technical and economical limitations of conventional wastewater treatment have triggered the search for sustainable and cost-effective alternatives. The integration of microalgae into wastewater treatment, not only offers environmental benefits, but also helps support microalgae production costs.

Symbiotic microalgae-bacteria systems for treating wastewaters rely on the microalgal potential to supply oxygen, through photosynthesis, to the degradative bacteria. This process allows the recovery of nutrients into a valuable biomass which can be used as a biofertilizer, bioplastics, animal feed and for biofuel production. Moreover, the microalgal high-value compounds can be recovered by subcritical water extraction to obtain functional products.

This work showed a biorefinery of microalga *Scenedesmus obliquus* cultivated in brewery effluents and the obtained products from the biomass. The achieved removal efficiencies were around 30% for phosphorus, 61% for COD, and above 90% for nitrogen. The subcritical water extraction of the biomass allowed the recovery of high-value compounds such as: phenols (0.249-1.016 mg Gallic acid equivalent/mL) and flavonoids (0.05-0.167 mg Catechin equivalent/mL). In addition, the microalgal biomass was converted into biohydrogen through dark fermentation, achieving a yield of 67 mL H₂/g (Volatile Solids). A thermochemical route was also followed, with yields of bio-oil higher than 60% in pyrolysis experiments (with 30 % of bio-char); for gasification experiments, the yields were around 60 % of syngas. Finally, the microalga was tested as a biofertilizer/biostimulant and showed an enhancing effect on seed germination and plant growth (number, length and leaves).

Keywords: *Scenedesmus obliquus*, Wastewater treatment, Subcritical water extraction, Thermochemical conversion, Biofertilizers

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THERMODYNAMICS STUDIES OF COPPER AND CHROMIUM(VI) IONS ONTO SUGAR BEET SHREDS AS ADSORBENTS

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Adsorption is a process significantly affected by temperature and thermodynamic studies are frequently investigated when new potential adsorbents are evaluated. Thermodynamic parameters provide insights into adsorption mechanisms which allow further modification and optimization of the process.

The changes in Gibbs free energy (ΔG°), enthalpy (ΔH°), and entropy (ΔS°) for the adsorption of copper and chromium(VI) ions onto sugar beet shreds (SBS) were determined by performing batch adsorption experiments at room temperature (23°C), 35°C and 45°C. Adsorption isotherms were obtained by adding 5 g/l of SBS in aqueous solutions of copper (initial pH 4) and chromium(VI) (initial pH 2) of different ion concentrations. Initial and equilibrium concentrations of ions were determined by complexometric (for copper ions) and oxidation-reduction titration (for chromium(VI) ions).

Results showed that the adsorption capacity of copper and chromium(VI) ions increased with temperature but the increase was considerably greater for copper ions. Estimation of thermodynamic parameters showed that adsorption of both copper and chromium(VI) ions gave a positive value of the enthalpy change which indicated that the processes were endothermic. The negative change of Gibbs free energy implied spontaneity of the processes and high affinity of these ions for adsorbent. Decrease of ΔG° with increase of the temperature indicated that processes took place easier at higher temperatures. The positive change of entropy for adsorption of both types of ions showed the increased randomness at the solid/solution interface during the adsorption process. Dubinin-Radushkevich model was used to determine if the mechanism of copper and chromium(VI) ions binding is chemical, ion-exchange or physical sorption. The results suggested that these processes onto SBS predominantly took place as ion exchange.

Keywords: Thermodynamics, Adsorption, Heavy metals, Sugar beet shreds

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SUPERCRITICAL CO₂ EXTRACTION OF TOBACCO WASTE

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Production and processing of tobacco is a constantly growing industry, which is developing new products and try to ensure high production efficiency. Nevertheless, a large amount of tobacco waste is produced, which is hazardous due to the high content of nicotine. Treatment of wastes such as recycling and reusing are an imperative today due to rigorous environmental protection legislation. Tobacco waste is consisted of leaf parts named tobacco scrap, very small particle named dust and midrib parts. Since they are derived from tobacco leaves, they contain all compound as leaves but in lower concentrations. Supercritical CO₂ extraction of bioactive components, including nicotine, is a new possibility of utilization such type of waste.

Tobacco waste (type: scrap) was obtained from “Fabrika Duhana Sarajevo” from Bosnia and Herzegovina. A series of operational parameters of supercritical CO₂ extraction of tobacco waste (pressure: 100–300 bar, temperature: 40–80°C), were investigated in a laboratory scale apparatus. CO₂ mass flow rate was kept constant during the process. The results show that the extraction yields were significantly affected by applied operational extraction parameters. The increase in pressure and temperature improved the extraction yield. Nicotine content in obtained extracts was analyzed using GC/MS.

Supercritical CO₂ extraction showed to be effective at two levels. Firstly, it can be used for production of selected high-valuable bioactive compounds, which can be implemented into new products. Secondly, after extraction of nicotine, tobacco waste becomes more suitable for some other application or disposal.

Keywords: *Tobacco waste, scrap, supercritical fluid extraction, extraction yield*

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THERMOCHEMISTRY ASPECTS OF MECHANOCHEMISTRY ACTIVATION OF THE FLOTATION PROCESSES

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The energy that holds together atoms in molecules is a significant value that is measured in tens and hundreds of thousands of joules per mole substance. These molecules, which built most of the world available to us, held together other, much weaker forces. Their energy is measured only in tens of joules per mole, values that we are sometimes prone to neglect by comparing them with the aforementioned. Cohesive pressure is the phenomena in condensed phases and is associated with intermolecular forces. Driving forces in the process of mass transfer of substance in multiphase disperse systems, driven by molecular forces, which resultant is formation of concentration and temperature gradient. Flotation process is carried out in the pulp, apropos, dispersed system which is in dynamic equilibrium with the energy of mixing. Selective forming of surface alloys on minerals in the pulp has a diffusion character is service of mass transfer. Separation processes, liquid-liquid, are actually processes of treating the mineral raw material by flotation contributing to a greater or lesser extent, distribution of useful minerals. The paper presents the process ore flotation Veliki Krivelj and statistically analyzed the mechanism of action the collectors xanthate and aeropromoter.

Keywords: Flotation process, Intermolecular forces, Mass transfer mechanisms

HYDRODYNAMICS OF ROTATING DISC CONTACTOR EXTRACTION COLUMNS

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A rotating disc contactor (RDC extraction column) is a type of process column widely used in the chemical industry for liquid-liquid extraction. It is used primarily for filtering lubricating oil from furfural, in processing of deasphalting oil with propane, in removal of the mercaptan from a gas tank, wherein the separation of metals, in the food industry, as well as methods for retention of basic organic synthesis. The apparatus consists of a vertical cylindrical vessel, divided length wise into a number of equally spaced compartments by a series of rings. A rotor disc supported by a shaft is centered in each compartment. The process of mixing and separating the two liquids is reinforced by the rotation of the rotor discs. Varying the column diameter, column height, compartment height, flow speed, and rotation speed allows for adjustment of the droplet size in the dispersed phase.

This paper presents the comparison of hydrodynamic characteristics of the different RDC columns such as drop size, dispersed-phase holdup, flooding characteristics, and column efficiency. On the other hand, the investigation was performed for various values of the rotor speed, total throughput and solvents feed ratio. In the experimental works the different liquid-liquid systems were presented, such as: water-acetic acid-methyl isobutyl ketone, toluene-water-acetone, and n-butanol-succinic acid-water. Sauter mean drop sizes (d_{32}) generated from a hole distributor in liquid-liquid extraction in the RDC columns were studied under various conditions. The two correlations are the first of their type to consider the distributor hole inlet diameter in RDC column. Dispersed-phase holdup and flooding behavior were studied in a pilot plant scale perforated rotating disc contactor (PRDC) using three different liquid-liquid systems. The influence of various operational parameters including rotor speed, continuous and dispersed phases flow rates on flooding velocities and holdup have been studied.

Keywords: *Rotating disc contactor, Solvent extraction, Drop size, Holdup, Column efficiency*

OXIDATIVE DEGRADATION OF CYAN FLEXO DYE WITH HETEROGENEOUS FENTON REAGENT - $\text{Fe}_2(\text{MoO}_4)_3$ PARTICLE

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Heterogeneous Fenton catalyst $\text{Fe}_2(\text{MoO}_4)_3$ was prepared and the process efficiency was evaluated for oxidation of water-based Cyan flexo dye in synthetic aqueous solution and printing wastewater. The removal process of printing dye was analyzed by measuring the absorbance of the aqueous solutions before and after the treatment with UV/VIS spectrophotometer, while dye mineralization was evaluated by the determination of total organic carbon content and chemical oxygen demand (COD). Four determinants of the heterogeneous Fenton system, including initial dye concentration, iron concentration, pH and hydrogen peroxide concentration were investigated. Statistical method, definitive screening design was applied to generate optimal operational conditions of the four variables, which maximizes the process of dye removal. The initial dye concentration of 20 mg/L, catalyst dosage of 0.75 mg/L, pH of 2 and H_2O_2 concentration of 11 mM were chosen as the best operational conditions, contributing to 82% of the process efficiency. The heterogeneous Fenton process efficiency of 79% was achieved within the treatment of printing wastewater under optimal conditions for a 90 minute reaction time. The maximum COD removal efficiency was 61.1%, while 67% mineralization was achieved. The obtained results confirmed synergistic effect of Fe^{3+} and MoO_4^{2-} which contributed to high catalytic activity and high efficiency of heterogeneous Fenton catalyst.

Keywords: Printing dye, Heterogeneous Fenton process, $\text{Fe}_2(\text{MoO}_4)_3$, Wastewater treatment, Definitive screening design

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PRELIMINARY STUDY OF CHROMIUM(VI) IONS ADSORPTION ON BOEHMITE

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For the past few decades a search for new adsorbents that could possibly replace activated charcoal in water and wastewater treatment has been intensive. A variety of different materials have been investigated but a perfect adsorbent for any specific purpose has not been found yet. Hence, there are still wide range of materials to be investigated. In this study possibility of chromium(VI) ions adsorption by microporous boehmite was studied.

Nanostructured boehmite was synthesized by hydrolysis of diluted sodium aluminate solution (Bayer liquor) with sulfuric acid. Aluminate solution was modified by adding glucose in order to obtain boehmite at room temperature. XRD confirmed single phase nanopowder. Powder's specific surface area was almost 300 m²/g with pore sizes about 3 nm.

Adsorption of chromium(IV) ions from aqueous solution by boehmite was investigated as function of time. Batch experiments were performed by mixing 0.5 g/l of adsorbent and chromium ions solution (initial concentration of 50 mg/l and pH 2) for 2 min to 24 h. Kinetic data obtained were fitted to the pseudo-first order and pseudo-second order model and Elovich model.

The results showed that more than 75% adsorbed amount of ions were bound in first 180min and the maximal adsorption capacity achieved after 24 h was 28.65 mg/g. Although all kinetic models provided high coefficient of determination the pseudo-second order fitted the experimental data the best ($R^2= 0,9971$).

Keywords: Boehmite, Adsorption, Chromium, Kinetics

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REMOVAL AND RECOVERY OF AMMONIUM FROM MILKING PARLOUR WASTEWATER USING POMEGRANATE PEEL

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Nitrogen compounds are widely used in agriculture as fertilizers and essential elements for the growth of living organisms. However, excessive ammonium nitrogen (NH_4^+) discharged in water sources can cause eutrophication leading to depletion of dissolved oxygen and toxicity to fish and other aquatic organisms. In our previous work, pomegranate peel (PgP) proves high ability to adsorb ammonium (NH_4^+) from the simulated wastewater under various experimental conditions, varying parameters such as pH, contact time, adsorbent dose, initial ammonium concentration, and stirring speed. Moreover, in addition to the abundant availability, low-cost and eco-friendly advantages, this biosorbent offers the possibility to recycle ammonium back for agricultural purposes. However, a study of the effect of interfering ions on the adsorption mechanism still missing and presents an important issue. In this work, the efficiency of pomegranate peel as biosorbent was investigated in order to remove ammonium from wastewater of a milking parlour. It consists of water, complex carbohydrates and nutrients, i.e. nitrogen, phosphorus and potassium. The initial concentration of total nitrogen (TN) and ammonium (NH_4^+) in examined sample were 66 mg/L and 56 mg/L respectively. In addition, other compounds found in this type of wastewater mainly include Sodium (Na), Calcium (Ca), Magnesium (Mg), Chlorides (Cl), Sulfates (SO_4) and Carbonates (CO_3) which could affect the final adsorption process efficiency. After the adsorption, microfiltration process is planned to separate adsorbent from the wastewater and test its efficiency as fertilizer.

Keywords: *Agriculture, Milking parlour wastewater, Ammonium removal*

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USING ERA5 DATA FOR ESTIMATION OF THE WIND ENERGY POTENTIAL IN SERBIA

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The wind data from the meteorological reanalysis are commonly used in estimation of wind energy potential over some region. The climate reanalysis produced by The European Centre for Medium-Range Weather Forecasts, ERA5 were recently realized. The spatial resolution of about 31 km, hourly time resolution and the wind speed at 100m height, the height relevant for modern wind turbines make those reanalysis suitable for the estimation of the wind energy potential in Serbia. The annual and seasonal wind energy potential at 100m height was estimated. The region with the highest wind energy potential is the Vojvodina region and the Eastern parts of Serbia. The wind energy potential is highest in the winter season and lowest in the summer season. The estimated wind energy potential is similar to those obtained in previous studies, but gives a more detailed time and spatial resolution of the available wind energy.

Keywords: wind energy, reanalysis, ERA5

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OPTIMIZATION OF STARCH SUSPENSION MICROFILTRATION PROCESS – ANN APPROACH

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Process intensification, in chemical engineering manner, demands higher productivity of the process, reduced waste production, and lower energy consumption. Intensification of the microfiltration process can be achieved using different techniques and one of them is the implementation of turbulence promoter in the tubular membrane channel. The efficiency of different turbulence promoters strongly depends on cross-flow microfiltration operation conditions such as feed concentration, cross-flow velocity, transmembrane pressure, etc. The aim of this study was the modeling of Kenics static mixer-assisted crossflow starch suspensions microfiltration process, using artificial neural networks (ANNs). The influence of transmembrane pressure, suspension flow rate and suspension concentration on the permeate flux in a system with and without Kenics static mixer, and on the reduction of the specific energy consumption was examined, in order to investigate the efficiency of turbulence promoter from the flux improvement and economic aspects. The second goal was multi-objective optimization in order to achieve the optimal solutions, from a cost-effectiveness point of view. Obtained results showed that the permeate flux in a system with and without static mixer is mostly influenced by suspension concentration, while the suspension flow rate had the biggest impact on the reduction of the specific energy consumption. High values of coefficient of determination (R^2 was in the range from 0.9860 to 0.9971, for three mentioned responses) indicate very good agreement between ANN predictions and experimental data. Results of multi-objective optimization indicate that starch suspension microfiltration process should be carried out at maximum transmembrane pressure, minimum suspension flow rate and minimum concentration. The increase of the flow rate in the system with static mixer led to a significant increase in the specific energy consumption, limiting the application of Kenics static mixer, from an economic point of view.

Keywords: *Artificial neural network, Microfiltration, Starch suspension, Kenics static mixer*

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VISCOSITY AND pH DETERMINATION OF CHOLINE CHLORIDE BASED DEEP EUTECTIC SOLVENTS CONTAINING AMIDES

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Given the use of large amounts of volatile and flammable organic solvents in industry, rapid development of green and sustainable technologies, as well as environmentally friendly solvents is required. Deep eutectic solvents (DESs) are a promising alternative to traditional solvents due to their suitable characteristics such as non-toxicity, thermal stability, easy preparation, biodegradability and price. DESs are generally based on mixtures of cheap and available components, nontoxic quaternary ammonium salts (usually choline chloride) with natural products (amides, sugars, alcohols, sugar alcohols, polyols and organic acids) as HBD. Since there is a possibility of tailoring their physicochemical properties making them appropriate for use in different processes, it is important to characterize the solvents well, especially physicochemical properties.

For the measurement of physicochemical properties (viscosity and pH), five choline chloride-based solvents with different urea derivatives and acetamide were prepared and characterized. The effects of water addition and temperature on these physicochemical properties were also evaluated. The measured physicochemical properties were provided in temperature range from 30 to 80 °C with different water contents in range 10-50 % (v/v).

The high viscosities of DESs can present a problem, especially in extraction and energy consumption needed for stirring and pumping, but luckily, the viscosity can be easily adjusted by obtaining higher temperatures or adding appropriate amount of water. For the DESs used in this study, the viscosity decreased in the following order: choline chloride:1,3 dimethylurea (1:2)>choline chloride:thiourea (1:2)>choline chloride:acetamide (1:2)>choline chloride:urea (1:2)>choline chloride: *N*-methylurea (1:3).

Increase of temperature and water content of DESs is followed by pH decreasing in all DESs except solvent with thiourea. pH values were in the range 9.30 – 7.20, 9.13 – 6.87, 7.59 – 5.08, 4.47 – 5.81 and 8.23 – 6.70 for *N*-methylurea, urea, acetamide, thiourea and 1,3-dimethylurea, respectively.

Keywords: *Deep eutectic solvents, Choline chloride, Viscosity, pH*

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AFB1 AND FUMONISIN B1 ADSORPTION AND DESORPTION USING ZEOLITES

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Zeolites are crystalline, hydrated tecto-alumosilicates of alkaline and earth alkaline cations, which have an infinite three-dimensional crystalline structure. Because of the porous, cation exchange capacity and layered structures, zeolites can adsorb mycotoxins and other molecules. In this study, *in vitro* tests were performed to determine the ability of zeolite (MINAZEL®) to bind aflatoxin B₁ (AFB₁) and fumonisin B₁ (FUM). Adsorption/desorption assays consisted of suspending zeolites (100 mg) in 10 mL of buffer solution (pH 3.0) spiked with 2 µg/mL of AFB₁ and FB₁. The samples were incubated at 37°C for 60 minutes at 180 rpm and then centrifuged. The supernatant was analysed by LC-MS/MS for adsorption of AFB₁ and FB₁. The remaining supernatant was removed and the pellet was resuspended in pH 6.5 buffer, incubated at 37°C for 30 minutes, centrifuged, and the supernatant was analysed by Agilent 6460 LC-MS/MS. Zeolite (MINAZEL®) had the greatest ability to bind 99 % AFB₁ and 72 % of FB₁ at pH 3. There was only minimal desorption of Aflatoxin (0.2 %), and FB₁ (4.5%) at pH 6.5. Therefore, Zeolite (MINAZEL®) may prove practical for mycotoxins remediation in animals.

Keywords: *zeolite, MINAZEL®, AFB₁, FB₁, LC-MS/MS*

DANUBE SEDIMENT CONTAMINATION WITH POLYCHLORINATED BIPHENYLS: NEW INTERPRETATION OF SEDIMENT QUALITY ASSESSMENT

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Apart from the contaminants of greatest concern, it is still possible to detect "old" classics such as polychlorinated biphenyls (PCBs) in aquatic systems. Since PCBs are detected in all environmental matrices and have been identified as harmful substances due to their toxicity, persistence and bioaccumulation in humans and wildlife, they are still one of the important groups of persistent organic pollutants (POPs). For this reason, this original approach studies the toxicological influence of PCBs, quantified in sediment samples collected at ten sites along the river Danube, by an application of advanced classification and clustering methods such as hierarchical cluster analysis (HCA) and Kohonen's self-organising maps (SOMs). Selected multivariate techniques were applied to the monitoring dataset in order to obtain visual images of the components distributed at each sampling site when all components are included in the classification and data projection procedure. After analyzing the data set using both techniques, groups that exhibit similar behavior were isolated. In the hexagon and dendrogram of variables three main clusters were distinguished. Regarding identification of pollutant spatial patterns, the SOM did not isolate a clear phenomenon probably due to the absence of local pollution sources contributing to the elevated concentrations of these compounds. The presented assumptions indicated that the supplemental application of SOM and HCA offers advantageous features over the usually rough interpretation of PCBs pattern and over the single use of the methods.

Keywords: PCBs, SOM, HCA, sediment, Danube

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THE USE OF GREEN SOLVENTS IN A PRE-TREATMENT OF LIPIDS EXTRACTION FROM GREEN MICROALGAE *CHLORELLA* SP.

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The use of green solvents and the process of green chemistry attract much attention of modern research, due to increasing environmental pollution and disturbing general health by using conventional solvents. Deep eutectic solvents (DES) are considered a special class of ionic liquids. The aim of this work was to examine the possibility of use of some DES for the pretreatment of biomass of green microalgae *Chlorella* sp. in order to obtain a higher yield of the extracted lipids. The pretreatments were carried out with eutectic mixtures of the quaternary ammonium salt choline chloride (ChCl) with: urea (1:2) (ChCl:U) and acetic acid (1:2) (ChCl:Aa). The biomass of algae was obtained by culturing in controlled laboratory conditions, under constant illumination and temperature of 27 °C. Bligh-Dyer method was applied for the extractions of lipids from the biomass treated with DES and the aqueous phase obtained after pretreatment of the microalgae biomass (aDES). Results of the lipid yields of biomass treated with ChCl:U and ChCl:Aa were 40.2 % and 37.9 %, (calculated for dry biomass), respectively. The lipid yields obtained from aDES were only 0.8 % and 0.1 % for ChCl:U and ChCl:Aa, respectively. Using a DES, satisfactory lipid yields have been obtained, which gives the opportunity for commercial use of these inexpensive, non-toxic and easily available solvents.

Keywords: microalgae, deep eutectic solvents, lipid extraction

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ECONOMIC FEASIBILITY OF AIR SOURCE HEAT PUMPS FOR RESIDENTIAL BUILDINGS IN VOJVODINA

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Households sector in province of Vojvodina, Serbia, offers great opportunities for energy savings and introduction of renewable energy sources in the future. An air source heat pump (ASHP) is a system which transfers heat from outside to inside a building, or vice versa. Under the principles of vapor compression, an ASHP uses a refrigerant system involving a compressor and a condenser to absorb heat at one place and release it at another. They can be used as a space heater or cooler and represent long-lasting system with low maintenance requirements. The aim of the present work is to assess economic feasibility of air heat pump systems for heating and cooling purposes in province residential sector.

Although from an environmental point of view the application of heat pumps air/water do not represent much progress at this moment, due to electricity production in Serbia, the economic indicators of the project realization are very satisfactory. Analysis of cumulative cash flow and return on investment shows a great economic justification for the application of air source heat pump. The funds invested in the implementation of this project will be returned within 3.4 years, while the investment that includes the installation of the under floor heating system will be returned in the period of 7.9 years. By analysing the risks involved in the implementation of this project, it is determined that the greatest impact on project viability has energy price in the base case object heating, i.e. natural gas. The least impact on the investment return period have initial costs with a relative impact of around 0.35; while the impact of the electricity price is 0.45. The influence of the natural gas price that is dominant has a negative impact on the return of the investment, that is, the decrease in its price leads to the prolongation of the investment return period, which is extremely unfavourable for the realization of the project. With a maximum natural gas price decrease of 50%, the project becomes unprofitable.

Keywords: Thermal-economic modeling, Air source heat pump, Residential building

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DETERMINATION OF THE TOTAL ANTIOXIDANT CAPACITY OF BLACK MUSTARD SEEDS (*BRASSICA NIGRA*) EXTRACTS OBTAINED BY DIFFERENT DEEP EUTECTIC SOLVENTS

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Brassica nigra, popularly known as black mustard, has been cultivated for thousands of years in different parts of the world for its medicinal and nutritional value. The present paper deals with the antioxidant activities of the black mustard seed extracts obtained using ethanol and different deep eutectic solvents such as triethanolamine : glycerol (TEOA : G), TEOA : G (with 25% of water), TEOA : G (with 25% of ethanol), triethanolamine : propylene glycol (TEOA : PEG), TEOA : PEG (with 25% of water), TEOA : PEG (with 25% of ethanol), choline chloride : urea (ChCl : U), ChCl : U (with 25% of water) and ChCl : U (with 25% of ethanol). The extraction was carried out at the seed-to-solvent ratio of 1:10 w/v and 65°C for 3 h. The IC₅₀ value of the extracts obtained by ethanol (0.15±0.04 mg/mL) and TEOA : PEG (0.15±0.03 mg/mL) was the same. The extract obtained with ChCl : U showed better antioxidant activity (IC₅₀ value = 0.10±0.03 mg/mL), while extract obtained with TEOA : G had a lower IC₅₀ value of 0.16±0.05 mg/mL, compared to ethanol. By adding 25% of ethanol to the TEOA : G and TEOA : PEG, the antioxidant activities of the obtained extracts increased compared to the antioxidant activities of extracts obtained using the sole DESs, so the IC₅₀ values were 0.11±0.09 mg/mL and 0.14±0.03 mg/mL, respectively. Addition of 25% of ethanol to ChCl : U extract did not influence antioxidant activity. By addition of 25% of water to the TEOA : G, TEOA : PEG and ChCl : U, the antioxidant activities of the obtained extracts reduced (IC₅₀ value were 0.17±0.03 mg/mL, 0.18±0.05 mg/mL and 0.14±0.04 mg/mL, respectively) compared to the antioxidant activities of the extracts obtained by using the sole DESs. The results showed that the DESs could improve the antioxidant activity of black mustard seeds extracts.

Keywords: extraction, deep eutectic solvents, black mustard seed, antioxidant activity

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KINETICS OF THE EPOXIDATION OF LINSEED OIL WITH IN SITU FORMED PERACETIC ACID

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Epoxidized vegetable oils have been commercially applied as plasticizer and stabilizer for PVC for a long time. More recently, they are also used as platform chemicals for the production of different polymer precursors. In industry, vegetable oils are epoxidized with organic peracid formed *in situ* from corresponding organic acid and hydrogen peroxide. Biphasic model for the epoxidation of vegetable oils with peracetic acid formed *in situ* in the presence of an ion exchange resin as a catalyst is proposed. The main reactions of the process, i.e. peroxidation and epoxidation, as well as the side reaction of the epoxy group opening with acetic acid, are considered. The influence of the fatty acid composition of the vegetable oil, as a significant property of this renewable raw material, on the kinetics of the epoxidation and epoxy group opening reactions is mathematically described. The proposed model was applied to the experimental data obtained for the epoxidation of linseed oil. The epoxidation was performed with peracetic acid at temperatures ranged from 60 to 85°C. The molar ratio of linseed oil unsaturation:acetic acid was 1:0.5, whereas the molar ratio of linseed oil unsaturation:hydrogen peroxide varied from 1:1.1 to 1:1.5. The loading of the catalyst Amberlite IR 120-H varied from 10 to 20% with respect to the total weight of acetic acid and 30% hydrogen peroxide aqueous solution. The proposed model agrees well with the obtained experimental data with a standard deviation of 0.02. At the average temperature of the runs of 346 K, the double bond in the linolenic acid chain which is firstly epoxidized was approximately eight times more reactive than the other double bonds in the triglycerides' fatty acid chains. Also, the corresponding epoxy group is approximately seventy times more reactive towards the opening reaction with acetic acid than the other epoxy groups. Therefore, its stability is significantly lower than the stability of other epoxy groups during the epoxidation process.

Keywords: *Kinetics, Model, Epoxidation, Peracetic acid, Ion exchange resin*

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MITIGATION OF MEMBRANE FOULING BY APPLICATION OF TURBULENCE PROMOTERS

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In membrane filtration, the most challenging is how to control and mitigate membrane fouling. The consequence of fouling is a decrease in flux so in overall process efficiency. The way of the minimisation of fouling thus alleviation of flux is an increase in shear rate near the membrane surface by the insertion of static turbulence promoters (TP) in a membrane channel. A static turbulence promoter uses only the energy of moving fluid to increase turbulence thus disrupting a boundary layer near the membrane surface and minimising fouling. Organic feeds like feeds containing proteins, oil, natural organic matters (humic acid), cause severe fouling with pronounced concentration polarization which can be combined with in-pore fouling. In these cases, conventional operation of microfiltration and ultrafiltration has low efficiency due to operation under low fluxes and high energy consumption under high cross-flow velocities. In this work, mitigation of membrane fouling by the application of static turbulence promoters has been investigated. Particularly, attention was given to the investigation of the influence of the geometry of turbulence promoter, on the flux alleviation in microfiltration and ultrafiltration.

Experiments were performed using a ceramic tubular membrane with inserted motionless turbulence promoter. Various geometries of turbulence promoters such as helical and blade geometry in different dimensions have been tested. Performance of metal and polymeric 3D printed turbulence promoters has been compared. Various organic or organic-inorganic feeds were used to show wide possibilities of application. Flux improvement and specific energy consumption have been chosen as indicators of efficiency.

Turbulence promoters proved to be very efficient in fouling minimisation providing very high fluxes two to six times higher compared to the conventional operation without a promoter. Helical promoters are more efficient than blade type promoters providing higher flux improvement with lower energy consumption. Differences in the efficiency of promoters of distinctive geometry rely on the differences in the flow field. The energy consumption was lower for helical promoters due to lower pressure drop compared to the blade type mixer. Helical promoters proved to be more efficient in fouling minimisation caused by proteins than that caused by oil droplets.

Keywords: turbulence promoter, fouling minimization, microfiltration, ultrafiltration

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ECOLOGICAL RISK MANAGEMENT WITH INTEGRATED MULTI-TROPIC AQUACULTURE APPROACH FOR MARINE ENERGY PROJECTS IN INDIA

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Economic development of India is associated with various environmental and social problems and increasing demand of energy sources. In India, there are limited marine energy resources but those may responsible for few environmental issues. To get clearance from environmental issues is critical challenge for any marine projects due to their potential of environmental harm. With this there are few plans of cultivation of seaweed nearby coastal areas of Gujarat state of India. Seaweed or macroalgae biorefinery, the sequential production of fuel, food, and chemicals from biomass, can serve as an alternative and attractive source of energy. The environmental impact of large scale seaweed cultivation will be depletion of nutrient level from the site area. To mitigate the possible impacts of offshore wind farm and seaweed cultivation, it is necessary to establish the large scale cultivation in such a way that will be beneficial for the better ecological performance. The ecological engineering (EE) aims to maintain the sustainability not only in all dimensions but also with special focus on the ecosystem management. The offshore aquaculture of seaweed and combined fish project is referred as integrated multi-trophic aquaculture (IMTA), can be successful example of EE such promising way in which seaweeds and bivalves can act as inorganic and organic extractive organisms respectively. In few parts of world, such IMTA projects are already in practice but in case of India, still there is no such project available in offshore environment. Here we are overviewing the possibility of ecological engineering with large scale seaweed cultivation in western coastal area of India where India's first offshore wind energy plant is expected to be established in coming years. We determined the components of possible integrated project on the basic visits to local market of fishes to get the idea of demand of seafood, the species which have economic importance, popularity. There are few major challenges for the implementation of IMTA in Indian seawater, we pointed out few major among those.

Keywords: Integrated multi-trophic aquaculture (IMTA); ecological engineering (EE); large scale seaweed farming; integrated project; Indian seawater

COMPARISON OF EXTRACTION AGENTS FOR METAL DETERMINATION IN SEDIMENTS FROM ARTIFICIAL LAKES AND RIVERS IN SERBIA

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The purpose of this paper was to investigate the effectiveness of three different extraction agents for extraction of 25 elements from sediment samples collected from 4 artificial lakes and 12 rivers in Serbia (33 samples in total). The extraction efficiency of the agents was evaluated by its ability to extract the highest quantity of the elements. For that purpose, three acids (1M HCl, 2M HNO₃ and 0.43M CH₃COOH) have been used. Inductively coupled plasma atomic emission spectrometry (ICP-OES) was used for quantitative determination of following elements: Al, As, B, Ba, Be, Bi, Ca, Cd, Cr, Co, Cu, Fe, Hg, K, Li, Mg, Mn, Ni, Na, Pb, Sb, Se, Sr, V and Zn. The extraction with 1M HCl has showed the best results for the majority of investigated elements (especially Sr, Mn and Ca). Antimony (Sb) was detected only after an extraction with 0.43M CH₃COOOH, while selenium (Se) could not be detected when 2M HNO₃ was applied as an extraction agent. The present study could be very useful for choosing suitable method for determination of specific elements and also can be helpful in evaluation of the contaminants in freshwater sediments in Serbia. This might contribute to environmental risk assessment of the present elements.

Keywords: Extraction agents, Metals, River and lake sediments

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PRODUCTION OF L (+) LACTIC ACID FROM CORN HUSK HYDROLYZATE

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Lactic acid has a broad variety of applications in food, pharmaceutical, chemical and polymer industries. In recent years, a growing interest in biodegradable plastics, such as poly-lactic acid, boosts the demand for lactic acid on nonedible feedstocks. Utilization of different lignocellulosic agro-industrial residues and wastes could enable cost-effective lactic acid production, improve food security and decrease environmental pollution. The pretreatment and hydrolysis of lignocellulosic biomass is considered as first and key step in its processing for lactic acid production. The main role of pretreatment and hydrolysis is to effectively provide fermentable sugars from structural carbohydrates, cellulose and hemicellulose, which can be converted into lactic acid by lactic acid producing microorganisms.

The main goal of the research described in this paper is to evaluate the process for L (+) lactic acid production from low-cost lignocellulosic biomass. Ground corn husk (PKB Corporation Belgrade, Padinska Skela, Serbia) was subjected to microwave-assisted alkali pretreatment and subsequent enzymatic hydrolysis using cellulase complex Cellic[®] CTec2. The obtained hydrolyzate was further used as a substrate in lactic acid fermentation by *Lactobacillus paracasei* NRRL B-4564. The saccharification and fermentation efficiency of pretreated corn husk was assessed and compared with untreated sample.

According to the obtained results, corn husk hydrolyzate was a good substrate for lactic acid fermentation by *L. paracasei*. Microwave-assisted alkali pretreatment led to an increase in the glucose yield during enzymatic hydrolysis and consequently, improved lactic acid concentration was achieved in fermentation by *L. paracasei*, compared to the control sample. Due to lower energy consumption and shorter processing time, microwave-assisted pretreatment of corn husk enables development of the process with reduced environmental impact, compared to the conventional chemical pretreatments conducted under high temperature or high pressure conditions, providing effective processing of agro-industrial residues into high-valuable product, such as lactic acid.

Keywords: corn husk, pretreatment, enzymatic hydrolysis, lactic acid fermentation, Lactobacillus paracasei NRRL B-4564

Acknowledgements: Research presented in this paper was funded by Ministry of Education, Science and Technological Development, Republic of Serbia, project number TR 31017 and Project #I-1 Scientific and Technological Collaboration of Republic of Serbia and PR China.



ASSESSMENT OF THE IMPACT OF WASTE WATER DISPOSAL ON THE BEGEJ

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The consequences of discharging wastewater into river flows increase the need for a thorough assessment of the impact of pollution in order to make appropriate decisions. When assessing the impact on the body of water, it is necessary to determine the status sensitivity of surface water bodies on the identified significant pressures and to carry out an assessment of the probability that surface water bodies will not meet the environmental objectives by using all previous information and other relevant data. Analysis of pressures and impacts finally implies the assessment of the risk of failure in achieving environmental goals. The investigated area is the river Begej, which springs in Romania and flows through Serbia, where it flows into Tisa. For the purpose of characterization of concentrated sources of pollution and assessment of their impact, monitoring of wastewater was carried out, as well as monitoring of the water of Begej itself. Monitoring was initiated several times in the period 2010-2018. and included data on the quality and quantity of waste and surface waters. It covered the period of different water flows in Begej, as well as the different operating modes of the industry. On this basis, simulations of all possible water flows were performed. The maximum load that the water can receive, with ought affecting its quality, is established. To assess the impact of wastewater discharge, risk assessment code was calculated for each recipient, the total maximum daily pollution input, taking into account the cumulative effects of the pressure and the characteristic (sensitivity) of the recipient. The results showed that a large number of concentrated sources of pollution represent a significant pressure on the quality of Begej. Begej is a river of male flowing power and a small capacity for self-purification, its unsatisfactory quality is already on entering our country. Water of Begej is burdened with organic materials, the effect of which is the reduction in the content of dissolved oxygen in water, the increase in BPK₅ and the frequent occurrence of anaerobic conditions, which entails not only the disturbance of the chemical regime, but also the destruction of the aquatic life of the watercourse. At different flows and even at the maximum flow of water in the watercourse, it has been established that the load from concentrated sources of pollution is much greater than the accepting power of the recipient when it comes to organic matter.

Keywords: *monitoring, pressure, impact assessment, wastewater*

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FUEL POTENTIAL AND PROPERTIES OF GRAPE POMACE HYDROCHAR

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Increasing fossil fuel depletion that leads to air pollution and global warming have become serious environmental problem. For this reason, a numerous of alternative biofuels have been developed and investigated as potential energy sources to substitute them. One of promising and highly effective technology for conversion of wet and waste biomass into multi-functional materials is a hydrothermal carbonization. In this study grape pomace was hydrothermally carbonized at different temperatures (180, 200 and 220 °C). Produced hydrochars were characterized in order to investigate its potential application as alternative and energy-efficient renewable fuels. The carbon, fixed carbon, sulfur and volatile matter contents were determined in all hydrochar samples. Obtained results showed that temperatures play significant role on the structural characteristics of produced materials. As expected, the carbon content and fixed carbon content in hydrochars were increased with temperature increasing (Table 1). Higher yields of carbon and fixed carbon observed in hydrochars indicated that intensive carbonization of biomass occurred. On the contrary, sulfur and volatile matter content were decreased. Observed reduction may be a result of dehydration and decarboxylation of grape pomace during hydrothermal treatment. These reductions are highly beneficial and improve the efficiency of solids' direct combustion. Decreased volatile matter content can potentially reduce the release of inorganic vapors and pollutant emission during combustion, while decreased sulfur content preventing generation and emission of harmful sulfur oxides, SO_x, compared to the parent biomass. The present study showed that hydrothermal carbonization improved fuel qualities and potential of grape pomace hydrochars among different reaction temperatures.

Table 1. Fuel characteristics of grape pomace and obtained hydrochars

Sample	Cfix (%)	C (%)	S (%)	VM (%)
Grape pomace	17,29	48,87	0,34	75,49
HC-180	22,16	56,01	0,27	68,27
HC-200	25,65	56,95	0,24	64,85
HC-220	27,27	58,38	0,22	62,79

Keywords: *Grape pomace, Hydrochar, Hydrothermal carbonization, Fuel*

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GREEN ENERGY PRODUCTION AT THE WASTEWATER TREATMENT PLANT IN SUBOTICA

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Anaerobic digestion is among the oldest methods used for solids and biosolids stabilization. The process consists of organic matter decomposition in an oxygen-free environment. A variety of microorganisms derive energy by metabolising organic matter under anaerobic conditions. The end products of digestion are biogas and stabilized sludge.

The wastewater treatment plant in Subotica utilizes anaerobic digestion for wastewater sludge stabilization. Biogas obtained through this process is used in combined heat and power plants. The energy gained this way is used on the wastewater treatment plant itself, for digester sludge heating as well as energy savings.

The aim of this paper is to present the energy production during one year. In the first quarter of the specific year, the production was very low, because of severe maintenance issues on the plant. After the repairs were done, the energy production returned to its normal state.

Keywords: biogas, wastewater treatment, energy production

INFLUENCE OF KENIX STATIC MIXER ON THE FLUX IMPROVEMENT IN THE MICROFILTRATION OF INDUSTRIAL WASTEWATER

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In the last decades, the large quantity of industrial wastewater discharged from chemical and food industries is becoming the key global environmental issue, the reason is the rapid development of industrialization. Membrane separation is extensively applied in the treatment of industrial wastewater stream but fouling is the major limitation for successful membrane application. The usage of turbulence promoters such as Kenix static mixer can reduce flux decline. The aim of this study was to investigate the influence of Kenix static mixer on the flux improvement in the microfiltration of industrial wastewater from soybean processing factory “Sojaprotein”, “Victoria group”, Serbia.

Cross-flow microfiltration on the ceramic membrane with a pore size of 200 nm was performed. The investigation was carried out at feed flows of 60 and 180 L/h, while the temperature (30°C) and pressure (1 bar) were constant during all the experiments. The two investigated filtration systems were the conventional system without any fouling mitigation method and system with Kenix static mixer.

The experiments indicated that higher feed flow had a positive effect on the reduction of flux decline in both investigated filtration systems. Comparison of the conventional system and system with Kenix static mixer showed that system with Kenix static mixer alleviated membrane fouling. The flux improvement was about 115% on both flow rates. Also, these preliminary results showed turbidity removal efficiency of 99%.

Keywords: Membrane separations, Microfiltration, Wastewater, Turbulence promoters

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A NEW APPROACH FOR MODELLING AND OPTIMIZATION OF RUBBER CURING PROCESS

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The rubber industry is considered as a huge energy consumer. In order to make the rubber production more energy efficient, it is important to determine the optimum curing procedure e.g. vulcanization time and temperature. The aim of this work was the development of an advanced kinetic model for simulation and optimization of the rubber curing process. Vulcanization kinetics of commercially available rubber gum for tire production is investigated using oscillating disc rheometer at six temperatures in the range from 130 to 180°C. Curing curves were fitted with a new kinetic model, that includes both vulcanization and degradation of rubber. The proposed model was transformed into the differential equations, coupled and solved together with the heat transfer equation. Numerical solutions were obtained for the rubber sphere, 5 cm in diameter. The value of rubber thermal diffusivity ($\alpha = 1.207 \cdot 10^{-7} \text{ m}^2/\text{s}$), necessary for the heat transfer equation, was experimentally determined for the rubber sphere. Specially developed optimization approach was used in order to calculate vulcanization temperature range and curing time. The temperature range was estimated from 136 to 161°C with corresponding vulcanization time from 83.3 to 31.6 min. The optimal condition for rubber sphere vulcanization was determined as the mean value of the temperature range (148.5°C and 45 min). The minimal degree of cure for the optimal condition was 0.961. The optimal solution was confirmed using the finite element method (FEM). FEM simulations were performed using COMSOL Multiphysics software. In this way, a new, fast and reliable procedure was developed in order to optimize the rubber curing process, and thus reduce energy consumption.

Keywords: Rubber, Vulcanization kinetics, Mathematical modelling, Optimization

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STATISTICAL OPTIMIZATION OF THE CORN GERM OIL METHANOLYSIS CATALYZED BY POTASSIUM HYDROXIDE

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Biodiesel is an alternative fuel obtained usually by the methanolysis of vegetable oils and animal fats. As the price of biodiesel is mostly affected by the oily feedstock cost, recent researches have been focused on using waste and cheap raw materials as feedstocks for biodiesel production. This work dealt with methanolysis of the oil obtained from corn germs, a by-product from corn-based starch production. The overall oil content in the corn germs, determined by Soxhlet extraction, was $45.2 \pm 0.2\%$. The oil was converted to fatty acid methyl esters (FAME) through a two-step process including the H_2SO_4 -catalyzed esterification of free fatty acids (FFA) and the KOH catalyzed methanolysis of the esterified oil. Both reactions were performed in a three-necked round-bottom flask, equipped with a reflux condenser and a magnetic stirrer. The FFA esterification was carried out at the H_2SO_4 concentration of 2 % (based on the oil mass), the methanol-to-oil molar ratio of 8.5:1 and 45 °C under atmospheric pressure. The methanolysis of the esterified oil was conducted at a methanol-to-oil molar ratio of 6:1 and different catalyst amounts and reaction temperatures according to a 3^3 factorial design with 3 central points. The FAME experimental data were fitted by the second-order polynomial equations, which validity was proven by a high coefficient of determination (0.985) and a low mean relative percentage deviation ($\pm 1.1\%$) between the calculated and experimental FAME contents, as well as a high F_{model} -value (144.9) and a low p -value (< 0.0001), meaning that the developed model fitted well. The optimal reaction conditions for achieving the highest FAME content determined by solving the model equation were 51 °C, 1.2 % KOH and 27.4 min, under which the experimental FAME contents agreed well with the predicted values (both being 99.1%).

Keywords: *Biodiesel, Corn germ oil, Methanolysis, Optimization*

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KINETIC MODELING OF CORN GERM OIL METHANOLYSIS USING CORN COBS ASH AS A CATALYST

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The kinetics of the corn germ oil (CGO) methanolysis catalyzed by corn cobs ash was modeled. The oil was recovered from corn germs by the maceration using *n*-hexane as a solvent while the ash was obtained by combusting corn cobs in a solid fuel furnace. The methanolysis of CGO was carried out in a batch reactor, equipped with a reflux condenser and a magnetic stirrer (900 rpm), at the methanol:oil molar ratio of 9:1, the catalyst loading of 20% (based on the oil weight) and the reaction temperatures of 40, 50 and 60°C under atmospheric pressure. All experiments were performed in duplicate. The kinetics of the CGO methanolysis was described by the simplified model that combined the changing mechanism and the first-order rate law with respect to triacylglycerols. The reaction rate constant depended on the reaction temperature according to the Arrhenius equation. The activation energy and the pre-exponential factor were calculated to be 51.5 kJ mol⁻¹ and 1.9·10⁷ min⁻¹, respectively. The used model described reliably the progress of methanolysis reaction supported by a relatively mean relative percentage deviation between the calculated and experimental triacylglycerol conversion degree (±7.6%, based on 53 data).

Keywords: *biodiesel, corn cobs ash, corn germ oil, kinetic modeling*

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FATTY ACID METHYL ESTER SYNTHESIS FROM OIL BLENDS

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The development of fatty acid methyl esters (FAMES) synthesis based on different types of oil blends is beneficial for reducing the financial risk because of fluctuation in the oil feedstocks price and uncertainty in their quality. In this study, the synthesis of FAMES from the oil blends of camelina, castor and used cooking sunflower oils over different type of catalysts was investigated. Blending of camelina and used cooking sunflower oils with castor oil having a high content of ricinoleic acid would lower its viscosity. The fatty acid composition of oil blend prepared by mixing 30 wt% of camelina oil, 30 wt% castor oil and 40 wt% of used cooking sunflower oil was determined by gas chromatography. The methanolysis of this oil blend was performed in a batch reactor at 60 °C and under atmospheric pressure. Four catalysts (5 % based on oil weight) were tested for FAME synthesis: calcium oxide, calcium oxide modified by choline chloride:glycerol deep eutectic solvent, the ash obtained by combusting rosehip seed cake at 800°C and potassium hydroxide. The reaction rate increased in the following order: ash < calcium oxide < modified calcium oxide < potassium hydroxide. The mass transfer resistance was noticed only in the reaction catalyzed by the ash. The lowest catalytical activity of the ash was ascribed to its low calcium oxide content (57.39%) indicating a lower concentration of catalytically active sites in the reaction mixture and the poor mixing efficiency because of the high amount of solid catalyst.

Keywords: Calcium oxide, Camelina oil, Castor oil, Methanolysis, Used cooking oil

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COMBINATION OF ALKALINE/ACIDIC AND MICROWAVE TREATMENT FOR ENHANCED ANAEROBIC DIGESTION OF SLUDGE

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Advantageous effects of microwave irradiation on sludge disintegration and anaerobic digestion are verified by many scientific papers. Acid or alkali dosage with thermal sludge processing has also positive effect on organic matter solubility and biogas yield. Combination of conventionally used chemical methods with microwave irradiation could be a promising pretreatment process with shortened process time required and enhanced biodegradability.

In this work the applicability and efficiency of continuous flow microwave irradiation of food industry sludge was investigated. The change of biodegradability was characterized by the specific biogas yield produced in mesophilic anaerobic digestion.

The results show that the exposure of the sludge to a standalone microwave treatment with an irradiated energy of 200 kJ L⁻¹ and microwave power of 600 W resulted in a biogas yield of 390 ± 23 mL g TS⁻¹. At lower power level (450 W) increasing of NaOH dosage or irradiated energy (kJ L⁻¹) led to increased biogas yield. Biogas yield of microwave-acidic pretreated sludge was lower than that obtained from microwave-alkaline pretreated samples. At the highest power level (750 W) there was no further significant increase in biogas yield when energy intensity increased from 150 to 250 kJ L⁻¹ or the NaOH dosage increased from 0.25 to 0.55 g NaOH/g TS. In microwave assisted alkaline sludge treatment the increased dosage of alkali enables reduction of the energy intensity of microwave irradiation, which enhances the overall energy efficiency of the process.

Keywords: *microwave, sludge, anaerobic digestion, biodegradability*

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ASEPTIC APPLE (*MALUS DOMESTICA* BORKH.) AND SOUR CHERRY (*PRUNUS CERASUS* N.) CHANGE IN THE CONTENT OF THE INGREDIENTS DURING STORAGE

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One of the possible ways of eliminating seasonal and spatial variations in the fruit-processing sector is aseptic preservation. This helps to increase the shelf life of natural, non-added aseptic fruit puree products without significant loss of quality.

Production of aseptic fruit-growing semi-finished products involves the production of ascorbic acid for color fixing. However, the latest customer demands the abandonment of ascorbic acid to provide an additive-free product.

During this research, changes in the content of the constituents of semi-finished products of preserved apple (*Malus domestica Borkh*) and sour cherries was observed (*Prunus cerasus L.*), during aseptic storage and during storage in the abdomen conditions. Samples of 5 kg of both fruits were prepared with the addition of ascorbic acid and acerola concentrate. Samples produced without control material were considered as control samples. Storage time was 8 months.

For both fruits, changes in water soluble solids, pH, and color characteristics (L^* , a^* , b^*) were monitored. In order to characterize changes in the content of the ingredients, the reducing capacity of the samples by the FRAP (Ferric Reducing Ability of Plasma) method and the total polyphenol content (TPC, Total Polyphenol Content) were examined.

In the case of sour cherry, the change in coloring was monitored by measuring the anthocyanin content. The acerola-treated type retained its color much better than the control. Fruit puree containing acerola shows that it has a positive effect on the amount of antioxidant molecules. Measurements for polyphenol compounds confirm that the additive has a higher polyphenol content, as the storage time extended, compared to apple.

Our results so far are considered to be encouraging and additional ingredients will be tested, in order to produce aseptic fruit puree with high quality, without the addition of ascorbic acid.

Keywords: aseptic preservation, apple, sour cherry, antioxidant ingredients

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MAJOR ENVIRONMENTAL LAW ASPECTS OF SUSTAINABLE DEVELOPMENT

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The object and subject of our text is to present some of the elements of the Major scientific project 47011, funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia, necessary for the protection of environmental rights and values from the negative impacts of certain industries, at the first place: mining, chemical, metal industry and energetic and to introduce with them the elements of the state apparatus and the economy. Researching is based on the full application of the normative methods of legal sciences: law normative, logic normative, formally normative, and materially normative, methods needed for the consideration of the law and sub-law elements of importance for the protection of: 1. Working environment, which protection at the same time is the protection of health and life of employees, 2. by human work produced environmental values, and 3. All the live and non alive natural environmental values, not only the eco mediums (waters, soil and air), protection that has to be established not only formally but in reality and, primarily, to cover the most important elements of the protection from numerous hazards and consequences, at first place within the mining, chemical and metal industry and energetic. Our conclusion is that this protection have to be treated as continuously evolving phenomenon.

Keywords: Sustainable development, Economic challenges, Environmental needs, Environmental law approach



ACTUAL AND REQUIRED WATER QUALITY IN ACCORDANCE WITH THE EU WFD ON THE DANUBE RIVER IN SERBIA

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The Water Framework Directive (WFD) established an innovative approach to water management based on river basins, natural geographic and hydrological units, and sets deadlines for Member States to protect aquatic ecosystems. The Water Framework Directive aimed to achieve the "good ecological and chemical status"- Class II of all European surface and ground waters by 2015.

Harmonized legislation in the area of waters of the Republic of Serbia recommended by the WFD has been implemented since 2012. The key objectives of the Directive are to protect the status of aquatic ecosystems, to implement the principles of sustainable water management at the level of the basin and to preserve the quality of the environment. It should be noted that in Serbia the adopted Rulebook on determination of water bodies of surface water and groundwater (Official Gazette of the RS, No. 96/2010), Rulebook on parameters of ecological and chemical status of surface waters, and parameters of chemical and quantitative status of ground waters (O.G.RS, No 74/11), as well Regulation on emission limit values for pollutants in surface and ground waters and sediments and the deadlines for their reaching (Official Gazette of the RS, No 50/12) have realized the conditions that monitoring of the Republic Hydrometeorological Service (RHSS) is carried out in accordance with the WFD.

The research area for monitoring the water quality of the Danube River in this work included ten measuring stations: Bezdan, Bogojevo, Novi Sad, Slankamen, Zemun, Smederevo, Banatska Palanka, Tekija, Brza Palanka and Radujevac along the course through Serbia in 2015 and the methodology defined by the Regulation on emission limit values for pollutants was applied in the surface and ground waters, as well as in sediments and the deadlines for their reaching (Official Gazette of the RS No 50/12). On the basis of the conducted survey of 35 parameters altogether at ten measuring stations of the Danube River, it was determined that 22 water quality parameters correspond to Class II, 12 parameters correspond to Class III and 1 parameter corresponds to class IV Class ecological status of surface water quality.

Keywords: *WFD, water quality, Danube, Serbia*

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ENHANCEMENT AND COMPARISON OF SUBMERGED AND SOLID-STATE AMYLASE PRODUCTION FROM WHEAT CHAFF BY STATISTICAL OPTIMIZATION

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By-products of the agroindustry sector represent a rising problem in terms of their enormous quantities being generated and disposal problems. Utilizing wheat processing waste (wheat chaff) for producing hydrolytic enzymes could solve not only the waste disposal costs but also amylase production costs. Choosing the right technique of cultivation for enzymes production under conditions which provide maximal results, in terms of enzyme yield and activity, represents a key segment in transferring this technology from the laboratory into practice.

The main objective of this work was to optimize process parameters (cultivation temperature, pH value and cultivation time) of cultivating *Trichoderma reesei* on media based on wheat chaff by submerged and solid-state technique, in order to enhance and compare two types of amylase production. Based on the results for amylase activity in filtrates after cultivating fungi on wheat chaff under different process conditions determined by the Box-Behnken Design experimental plan, the optimization of this bioprocess was carried out.

Results have shown that the optimal conditions for the submerged fermentation are 29.86°C, pH 5.08 and 6.43 days of cultivation, while for the solid-state technique the optimal numbers are 35°C, 6.00 and 6.34 days, respectively. The amyolytic activities of the obtained cultivation filtrates under optimal conditions are 0.2307 U/mL and 0.1845 U/mL for submerged and solid-state technique, respectively. Compared to the results obtained before optimization, this is a 29.71 % and 79.51 % enhancement of enzyme activity for submerged and solid-state production of amylases from wheat chaff, respectively. Further studies should be directed towards kinetic modelling of the production process in order to have a deeper insight about how to monitor and control amylase production from wheat chaff.

Keywords: Wheat chaff, Amylase, Cultivation, Trichoderma reesei, Solid-state

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ADSORPTION OF Cu(II) IONS FROM SYNTHETIC SOLUTION BY SUNFLOWER SEED HUSKS

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Sunflower seed husks are abundant by-product from the vegetable oil industry. Annual production of the sunflower seed in Serbia is between 650,000-720,000 tons. Husks are usually incinerated by vegetable oil producers in order to obtain energy. In this paper adsorption of Cu(II) ions from synthetic solution by unmodified sunflower seed husks was examined. Influence of the initial concentration of Cu(II) ions and adsorbent dosage on the adsorption capacity were investigated. The experimental data were fitted by Langmuir and Freundlich isotherm models. The maximal adsorption capacity for Cu(II) ions calculated from Langmuir adsorption isotherm is 34.89 mg/g. This result is suggesting that sunflower seed husks have a potential to be applied as an effective adsorbents of copper ions from contaminated waters.

Keywords: sunflower seed husks, copper, adsorption

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INVESTIGATION OF PHOTOCATALYTIC PROPERTIES OF ZnFe-MIXED OXIDES

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The exponential increase of environmental pollution with no systematic solution has become an overwhelming concern in the scientific community. Considering that one of the major organic pollutants are residual dyes from different industries, effective dye removal methods present a challenging and a motivating task. Due to the high demands in this field, photocatalysis has been increasingly investigated with the aim to design promising photocatalysts that would be efficient in dye removal using the solar light. The motivation for this research was to develop a simple and inexpensive synthesis method for ZnFe-mixed metal oxides with the desired properties in order to enhance their photocatalytic performances in the photodegradation of organic dyes.

Mixed metal oxides (ZnFe-S) were successfully synthesized and thermally activated (ZnFe-T). The photocatalytic tests were conducted in an open cylindrical thermostated Pyrex reaction vessel using ULTRA VITALUX 300 W lamp, which emission spectrum simulates solar light. The photocatalytic activity was estimated by Rhodamine B (RhB) photodegradation, monitoring the decrease of RhB concentration (starting concentration: 10 μM RhB solution) in time using UV-VIS spectrophotometer. It was concluded that both samples exhibited satisfactory photocatalytic activity. The decrease of RhB concentration was detected (5.5 μM RhB for ZnFe-S and 1 μM RhB for ZnFe-T after 300min) indicating good photocatalytic properties of both samples. More pronounced photocatalytic activity of the ZnFe-T sample could be attributed to the additional phase formation after thermal activation (ZnFe₂O₄ spinel phase). This research opens a new route to the design and synthesis of mixed metal oxides with promising photocatalytic properties that could further be investigated for their application in wastewater purification.

Keywords: *Photocatalysis, Wastewater purification, Mixed metal oxides*

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REDUCTION OF FOULING IN DAIRY WASTEWATER ULTRAFILTRATION PROCESS

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The treatment of wastewater from food industry is of crucial importance for environment point of view. Compared to conventional treatments, membrane filtration technology has many advantages over traditional methods such as less required space, energy and can be easily combined with other methods. However fouling of the membranes is still a limiting factor, it can be reduced via enhanced shear force on the membrane surface by the application of vibration, or using coagulation and flocculation, as pre-treatment methods.

In our work, different ultrafiltration (*UF*) membranes were tested and compared using classical and vibration devices. Sedimentation, centrifugation and different chemical pre-treatments with and without pH adjustment, as well as vibratory shear enhanced processing *UF* experiments were carried out with dairy wastewater samples. Ultrafiltration fluxes, membrane rejections of organic matter content (*COD*), electric conductivity (*EC*), total dissolved solids (*TDS*) and turbidity were measured and compared. Operational parameters, transmembrane pressure and module vibration amplitude, were gradually modified during the tests to investigate how they affect the separation intensity. Furthermore, membrane resistances were calculated and the contact angle values of the membranes were measured. In order to know the efficiency in more detail the effects of module vibration and transmembrane pressure on specific energy demand were examined. Moreover, concentration experiments were also implemented to compare laboratorial with industrial conditions.

Keywords: *ultrafiltration, dairy wastewater, fouling mitigation*

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EFFECTS OF COMMERCIAL ANTIOXIDANT FORMULATIONS ON OXIDATIVE STABILITY OF FATTY ACID METHYL ESTERS FROM DIFFERENT RAW MATERIALS

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Fatty acid methyl esters (FAMES) are main constituents of biodiesel, considered as one of the most promising petroleum diesel fuel substitutes. Important characteristics of biodiesel is an oxidation stability, which describes its relative susceptibility to oxidative degradation of the composition and consequently of the fuel characteristics. In fact, poor oxidation stability is a main disadvantage of biodiesel if compared to conventional diesel. Addition of antioxidant is the most efficient methods for improving the stability and delaying the oxidation degradation of biodiesel composition. Although effects of various antioxidants on FAMES stability have been often investigated, majority of the reported results refer to the addition of synthetic, single compounds to biodiesel, whereas only few studies dealt with commercial formulation. Thus, the aim of the study was to test an effectiveness of commercial antioxidant formulations in improving the stability of FAMES produced from two different raw materials (a soybean-sunflower oil blend 50:50 and waste cooking oil). Five of the tested additives were based on synthetic phenolic compounds, while one was amine type. Induction period (IP) of FAMES was measured by the Rancimat method before and after the addition of various dosages of commercial formulations.

The lowest additions of 0.05 mL and 0.10 mL were insufficient to improve the starting IPs of both FAMES. Amine-based additive influenced the oxidation stability of the treated FAMES in different manner in comparison to the phenolic additives. The results proved previous findings on TBHQ effectiveness over the other active compounds of the tested formulations. Significant improvement of IPs, well above 8 h, was achieved with rather high dosages of the additives, being in the range 0.5-4% depending on the additive itself, which was expected taking into account the comparison with available literature data on the same active compounds of the used additives. Further study should prove that the high dosages of additives does not introduce the unfavorable effect on overall quality of the studied FAMES.

Keywords: *Biodiesel, Phenolic antioxidants, Amine-based antioxidant, Induction period*

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HEMP BIODIESEL PRODUCTION: ANALYSIS OF MATERIAL AND ENERGY CONSUMPTION ON INDUSTRIAL SCALE

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Herbaceous crop, such as hemp (*Cannabis Sativa* L.), is increasingly important as a feedstock for biodiesel production. Therefore, it is a need for production models that include explicit material and energy calculations. The absence of an exhaustive thermodynamic database for both hemp seed oil and methyl esters and the absence of flowsheet are significant barriers to develop a reliable model of hemp biodiesel production.

The model presented in this study estimated the necessary data and placed it into a heuristic model using a commercial simulator Aspen Plus v.8.6. The hemp oil (triacylglyceride or TAG) was presented by the composite TAG5, which referred to a weight-average composite of the five triglycerides used. Similarly, hemp biodiesel (five methyl esters) were incorporated into the model as composite FAME5. Thermodynamic properties of TAG5 and FAME5, particularly enthalpy, free energy, molar volume, heat capacity, and vapor pressure were identified, too.

The hemp biodiesel model with a production capacity of 8,271 tones/year, included a conventional alkali-catalyzed continuous process. Hemp provided a competitively high biodiesel yield (0.99 g/g of hemp oil). Waste solid, salts as well as surpluses of glycerol in the market were side-products of hemp biodiesel production. Simulation analyses showed that the current energy bottlenecks for the large-scale production of hemp biodiesel are transesterification and solvent separation operations. It was found that the reactor and flash distillation column were the largest contributors to the energy cost.

If legal and perception challenges of this “niche” crop could be overcome, wide-scale hemp biodiesel production seemed to be possible.

Keywords: biodiesel, hemp seed oil, simulation, thermodynamics

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PURIFICATION OF DAIRY WASTEWATER COMBINING MEMBRANE FILTRATION WITH OZONE AND FENTON PRETREATMENTS

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There are several investigations aiming reuse of dairy wastewaters, and membrane processes are promising methods to achieve this goal. However, membrane fouling is a limiting factor of these processes. Beside proteins of the dairy wastewater, which were found to be severe foulants for the existing membrane materials, various matters may affect fouling; in this work, fouling propensities of typical dairy effluents were investigated and discussed. Advanced oxidation processes (AOPs), like ozone and Fenton-pretreatment, are widely used in the fields of water and wastewater treatments and may be appropriate pre-treatment methods before membrane filtration, due to their (micro)floculation effect, and oxidation efficiency.

In the present study the effect of the ozone and Fenton-reaction as a pre-treatment of real dairy waste waters were investigated. Fluxes, filtration resistances and pollutant retention were determined and compared. As according to earlier studies ozone can improve biogas production from food industrial waste, the biogas production from the concentration of dairy wastewater also was investigated.

Keywords: membrane filtration, ozone, Fenton

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CORROSION BEHAVIOUR OF STAINLESS STEEL EN 1.4301 IN 0.5 M HCl IN PRESENCE OF COMMERCIAL INHIBITOR

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In everyday application it is necessary to protect construction materials from impact of corrosion processes, not only because of material damage, but also for the protection of people and the environment. One of the most effective and often used methods is metal protection by corrosion inhibitors. This paper presents the results of the corrosion behaviour of stainless steel EN 1.4301 in 0.5 mol dm⁻³ hydrochloric acid, as well as the efficiency testing of commercial industrial corrosion inhibitor. For experimental research it was used DC-technique (Tafel extrapolation, linear polarization method, potentiodynamic polarization). The obtained results show that the inhibitor is effective in the protection of stainless steel from corrosion in 0.5 mol dm⁻³ HCl and the protection efficiency depends primarily on the concentration of the inhibitor. The highest efficacy was achieved at a concentration of inhibitor of 0.15 mL/L ($E_p = 55.7\%$). According to the corrosion mechanism the inhibitor is behaved as a mixed inhibitor.

Keywords: *Corrosion rate, protection efficiency, DC-technique*

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CLOUD POINT EXTRACTION PROCEDURE FOR PRECONCENTRATION AND DETERMINATION OF LEAD(II) IONS USING TRITON X-100 AT ROOM TEMPERATURE

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A Cloud point extraction (CPE) procedure was presented for preconcentration of lead (II) ions, after complexation by 18-crown-6 (18C6) and extraction with Triton X-100 at proposed experimental conditions. After separation of surfactant - rich phase, content of Pb(II) ions in remaining solution was measured by Flame Atomic Absorption Spectrometry (FAAS). The experimental conditions such as pH, temperature, concentration of Triton X-100, concentration of 18C6, time of incubation, type and concentration of added electrolyte, were evaluated. Results showed that among investigated electrolytes (NaCl, Na₂SO₄ and Na₂CO₃) the amount of 0.9 mol/L Na₂CO₃ lowers cloud point temperature of Triton X-100 to 22°C (room temperature during the experiment), thus simplifying the extraction procedure. After incubation time of 5 minutes and using the concentration of $1.2 \cdot 10^{-3}$ mol/L Triton X-100 and $1.5 \cdot 10^{-4}$ mol/L 18C6 (1:1 stoichiometric ratio for 18C6:Pb), 60% of lead (II) ions were efficiently removed from investigated solution.

Keywords: lead(II), Triton X-100, 18-crown-6, Cloud Point Extraction, FAAS

THE EFFECT OF POROSITY AND SURFACE AREA OF CLINOPTILOLITE AND ZEOLITE ON THE ADSORPTION OF THE PRODUCTS OF THERMO-OXIDATIVE DEGRADATION OF EDIBLE SUNFLOWER OIL

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Surface area, pore volume and porosity of solid materials have an effect on their adsorption behavior. These properties of zeolite and clinoptilolite are analyzed as parameters which determine the adsorption of the products of thermo-oxidative degradation of edible sunflower oil. When edible oils are heated above 150°C, they undergo thermo-oxidative changes depending on a variety of factors, such as temperature, heating time, type of oil, etc. These changes cause a series of chemical reactions (hydrolysis, oxidation, polymerization, etc.) resulting in the formation of free fatty acids, an increase in the peroxide number and the acid number. This study investigates the changes of the content of free fatty acids, the acid number, and the peroxide number in edible sunflower oil when heating at the temperature range from 110°C to 190°C for 30 minutes, as well as after the adsorption by a natural and a synthetic adsorbent (clinoptilolite and zeolite 4A). The results of the research show that these adsorbents are effective in removing the products of chemical reactions occurring in thermal degradation, which is particularly evident after the adsorption of oil heated at 190°C. Somewhat better results were achieved after the adsorption using clinoptilolite, with a 72% reduction in the content of free fatty acids, 83% reduction in the acid number, and 43% reduction in the peroxide number. After the adsorption by zeolite 4A, the content of free fatty acids was reduced by 65%, the acid number by 76%, and the peroxide number by 39%. From the values of pore volume, BET surface area, micropore surface area and adsorption cumulative surface area of pores, we would expect that clinoptilolite has much better adsorption properties. However, the results obtained indicate that the presence of cations in the pores of this porous material impedes the process of adsorption. Conversely, zeolite exhibits greater adsorption properties due to the lower content of cations as a result of the fact that it is obtained in controlled conditions. Further research should focus on the acid modification of the surface of clinoptilolite in order to increase its adsorption properties.

Keywords: Edible Oil, Porosity, Surface Area, Clinoptilolite, Zeolite

INTEGRATION OF FIRST AND SECOND GENERATION BIOETHANOL PRODUCTION FROM BY-PRODUCTS OF SUGAR BEET PROCESSING

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In recent years there has been an increasing need for sustainable, renewable and cheap alternative sources of energy that are economically viable, and one of the most promising alternatives to fossil fuels is bioethanol. Second generation bioethanol (lignocellulosic biomass) does not compromise food security, such as the conventional, first generation bioethanol (starchy grain and sugar-rich crops) and has the most potential to become the biofuel that will replace fossil fuels. However, second generation bioethanol production is still being developed and a major challenge for its commercialization is the low ethanol titer and low fermentation efficiency, while first generation bioethanol has already seen large scale commercialization. The efficiency of second generation bioethanol production can be improved by integrating the first and second generation bioethanol production, in order to increase the feasibility of cellulosic bioethanol and promote its industrial implementation.

This study aimed to examine the possibility of integrating the first and second generation bioethanol production process from by-products of the sugar beet processing industry, specifically molasses and sugar beet pulp. The first part of the study covered acid and thermal pretreatment of the lignocellulosic raw material, followed by enzymatic hydrolysis by *Trichoderma reesei*. The integration of the bioethanol production process was carried out by mixing the hydrolysates obtained from sugar beet pulp pretreatment and enzymatic hydrolysis, as well as molasses, and using the obtained mixture for bioethanol production by *Saccharomyces cerevisiae*. The obtained results show that the quantity of the produced bioethanol is affected by the efficiency of sugar beet pulp pretreatment and enzymatic hydrolysis, as well as the amount of molasses. In order to increase efficiency of this bioprocess further research should incorporate the optimization of each bioprocess segment. The obtained results have significant potential and represent a basis for the development of an integrated bioethanol production process.

Keywords: Bioethanol, Bioprocess, Fermentation, Sugar beet processing by-products

Acknowledgements: This study is part of the project TR-31002, which is supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia.



HOW RISK MANAGEMENT STANDARDS CAN CONTRIBUTE TO ACHIEVING SUSTAINABLE DEVELOPMENT GOALS

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The paper presents the most recent developments of risk management standards from the ISO 31000 series. This family of standards includes vocabulary (ISO Guide 73), guidelines (ISO 31000), risk assessment techniques (IEC 31010), and guidance for their application (ISO/TR 31004). This research discusses the correlation between these international standards and the latest Sustainable Development Goals of the United Nations. The conclusions and recommendations direct organizations on how to apply risk management principles and concepts for enhancing their performance and to contribute to greater ideals than their individual success.

Keywords: Risk Management, Risk assessment, ISO 31000 series, international standards, UN Sustainable Development Goals

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DEVELOPMENT OF WILD CYANOBACTERIAL BIOMASS (*APHANIZOMENON FLOS-AQUAE*) BIOREFINING SCHEMES

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Cyanobacteria are prokaryotic photosynthetic microorganisms that can have significant effects on the water quality as well as the functioning of aquatic ecosystems. In heavily eutrophicated systems, mass proliferation of cyanobacteria can lead to bloom formation with strong financial and environmental impacts. Recent reports indicate that removal of wild cyanobacterial blooms from the Curonian lagoon as a management measure should be prioritized. Herein we envisioned the utilization of wild cyanobacterial biomass as a feedstock within a biorefinery concept. Towards this end, fractionation of mentioned biomass was achieved with conventional and/ or high-pressure extraction techniques. Recovery of lipophilic products using supercritical CO₂ extraction was optimized by central composite design (CCD) and response surface methodology (RSM). Under optimal conditions (42.5 MPa, 55°C and 120 min of extraction), SFE-CO₂ yielded 4.43 g/100 g DW of non-polar extract with high contents of α -linoleic acid and α -tocopherol. In the next step, isolation of phycobiliprotein rich aqueous extracts was based on ultrasound assisted extraction optimized with CCD and RSM. The combination of homogenization, as a primary technique, followed by application of 8.75 min of ultrasounds at 84% amplitude, resulted in the highest phycobiliprotein yield. The residual biomass was further subjected to pressurized liquid extraction with increasing polarity solvents (acetone, ethanol, and water). The obtained extracts were evaluated for their functional properties with various *in vitro* radical scavenging and enzyme inhibition assays. Phytochemical composition by UPLC-ESI-TOF-MS revealed the presence of several natural pigments as well as auxins. Conclusively, this underutilized biomass could be considered as a feedstock for the recovery of various products with potential biotechnological, food, agrochemical, and pharmaceutical applications.

Keywords: cyanobacteria, biorefining, phycobiliproteins, α -linoleic acid

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INVESTIGATION OF HUMIC ACIDS EVOLUTION DURING EX SITU BIOREMEDIATION OF PETROLEUM CONTAMINATED SOIL

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Petroleum and its derivatives are among the major pollutants of soil. Bioremediation, a method that uses the ability of microorganisms to decompose toxic waste, is a prevailing trend in the decontamination of polluted soil contaminated by petroleum and its derivatives. Bioremediation procedures are in harmony with the principles of sustainable development, are categorized as “environmentally friendly” technologies, do not form waste and the treated soil can recover its natural biological activity.

The aim of this paper is to study the use of an active consortium of zymogenous microorganisms for *ex situ* bioremediation of a petroleum contaminant in the soil, along with simultaneous monitoring of the humification process.

In the pilot experiment, which lasted for 6 months, 150 m³ of soil contaminated by various petroleum derivatives was treated by *ex situ* bioremediation. The total petroleum hydrocarbons content was reduced from the initial 30 to 2.2 g/kg, while the content of humic acids increased by 47%. The analysis of humic acids from the studied samples showed that the structure of humic acids changed during bioremediation, i.e. the content of aliphatic carbon was reduced, while the content of aromatic and carboxylic carbon was increased. The degree of aromaticity of molecules of humic acids increased from 24.9% to 28.9% during bioremediation.

The results of our research based on bioremediation of oil-contaminated sites have shown pollutant removal efficiency of up to 92% with a simultaneous production of humic acids, which contributes to complete soil restoration.

Keywords: *Bioremediation, Zymogenous microorganisms, Petroleum, Humic acids*

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WHEAT SEEDLINGS UNDER THE SALINITY STRESS

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Salinity is one of the major problem in agriculture resulting in loss of yield, and in fields it often occurs in the combination with drought. In plants, salinity causes both osmotic and ionic stress. Aside it causes the perturbations at morphological, physiological and biochemical level, accumulation of NaCl in plants can be toxic for the plant. In this study winter wheat genotypes (BC Bernarda and BC Tena) were used to test their resistance to salinity. The seeds were sown in pots and exposed to 0, 90, 160 and 230 mM NaCl solution for five days. Seedlings were grown under 12 h light/12 h dark photoperiod and artificial illumination at 23±2 °C. The germination was tracked from 2nd day, while shoots and roots length was recorded last day of the experiment. The highest NaCl concentration (230 mM) reduced shoots and roots length compared to concentrations of 0, 90 and 160 mM in both genotypes, indicating that this salt level impeded growth. Germination percentage in samples with 230 mM NaCl in both genotypes after 2nd day of the experiment decreased for 307.7% compared to control sample, while at 160 mM NaCl inhibition of germination was from 47.1% in BC Bernarda to 56% in BC Tena. These results indicate that NaCl had greater negative impact in early stages of germination. Comparing variations among genotypes from 3rd to 5th day of the experiment, BC Tena showed higher germination percentage at all NaCl concentrations than BC Bernarda indicating that it poses greater salt tolerance. Present study reveals different genetic background of tested genotypes related with salinity resistance.

Keywords: wheat, shoot and root length, germination, salinity



HOW LCA DAMPENED THE ENTHUSIASM OF FORMER BIOFUEL ADVOCATES?

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Only a decade ago biofuels were seen as an ideal, eco-friendly replacement for the liquid fossil fuels that power the world's transport systems. Environmental advocacy organizations, policy makers and researchers alike were extolling the benefits of ethanol and biodiesel as a carbon-neutral bridge to a sustainable energy future. Concerns quickly followed that these first-generation biofuels, made from food crops, were often as harmful to the environment as fossil fuels when all factors associated with their life cycle were considered. Initially, Life Cycle Assessment (LCA) results supported the claim of biofuels' superiority. However, with further development of assessment techniques, and the expansion of the system boundaries to include new aspects of the product chain, LCA results of biofuel's carbon balance gradually shifted from favorable to unfavorable. Beleaguered by criticism and denounced by politicians and environmentalists, proponents of biofuels have been forced on to the defensive. But are there indeed reasons to doubt the reliability of LCA results? Probably yes, since the estimation of the environmental impact of biofuels is very complex, given the large variety of available methods and techniques used to address specific life cycle modelling issues and the general lack of consensus on appropriate LCA approaches. By reviewing scientific papers published in the last two decades on the environmental impact of biofuels, it will be demonstrated how the choice of the assessment methodology influences the LCA results and the understanding of its sustainability. Furthermore, the report summarizes the methodological challenges that must be resolved to enable LCA to effectively evaluate the environmental impact of different biofuels. These challenges may be relevant to many LCA efforts; however, the focus here is its implication on applying LCA to crop-based biofuels.

Keywords: *Biofuels, Life cycle assessment, Reliability*

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USE OF SEQUENTIAL EXTRACTION TO ASSESS METAL PARTITIONING IN FLOODED AGRICULTURAL SOIL

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Soil contamination with potentially toxic elements (PTEs) represents a worldwide problem mainly because these elements can be transferred and accumulated into the crops, thereby posing a hazard to human health. Different physico-chemical properties of soil influence mobility or sorption of PTEs, i.e. pH, conductivity, soil texture, organic matter content, etc., as well as the distribution of PTEs among the various soil fractions: soluble (*f1*), extractable (carbonate-associated PTEs) (*f2*), reducible (bound to Fe/Mn hydroxides) (*f3*), oxidable (bound to soil organic matter and sulfides) (*f4*), residual fraction (bound to mineral structures) (*f5*). The association of the PTEs with the specific soil fraction determines how strongly they are retained in the soil and how easy they may be mobilized to the hydrosphere and biosphere. Thus, the main objectives of this study were: (i) to determine the concentrations of seven PTEs (Pb, Cu, Ni, Cr, As, Co, and Cd) in 20 composite samples of previously flooded arable soil from the Jamena village located in the Northern Serbian Province (the area of intensive agricultural production), (ii) to investigate their distribution throughout the application of chemical fractionation which included the association of elements to specific soil fraction, and (iii) to evaluate the mobility of PTEs, which is in correlation with their bioavailability. The average value of soil pH and percent of organic matter were 7.38 ± 0.44 and 7.41 ± 2.18 , respectively. Particle size distribution revealed that analyzed soils consist of 65% of sand, 34% of slit and 1% of clay. The pseudo-total concentrations of Pb, Cu, Ni, Cr, As, Co, and Cd were 23.1, 34.9, 58.6, 28.0, 8.73, 7.04, and 0.444 mg/kg, respectively. The target value set by the Serbian regulation was only exceeded for Ni. The soil fractionation revealed three different partitioning patterns for PTEs (i) Pb, Cu, Ni, Cr, and As were mainly associated with the *f5* fraction, (ii) Cd was equally distributed in *f2*, *f3*, and *f5* fractions, while (iii) Co was mainly associated with the *f3* fraction. On the basis of the mobility of PTEs in soil assessed using a “Mobility Factor” index, the following order was observed $\text{Cd (39\%)} \gg \text{As (5.6\%)} > \text{Ni (3.5\%)} > \text{Cu (3.1\%)} \gg \text{Co (0.77\%)} > \text{Cr (0.37\%)} > \text{Pb (0.34\%)}$. As a consequence, Cd, As, Ni, and Cu are more available to be up-taken by plants and soil organisms.

Keywords: *Sequential extraction, Heavy elements pollution, Agricultural soil, Mobility*

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REMOVAL OF NATURAL ORGANIC MATTER AND EMERGING CONTAMINANTS FROM GROUNDWATER USING OZONATION AND GAC FILTRATION

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This work presents results from a pilot scale drinking water treatment plant used to investigate the performance of ozone oxidation and granulated activated carbon (GAC) adsorption in removing natural organic matter (NOM) and specific organic micropollutants from groundwater. The investigated groundwater has a relatively low NOM content (1.83 ± 1.01 mg C/L total organic carbon, TOC). Using gas chromatography/mass spectrometry (GC/MS) screening analysis with Deconvolution Reporting Software, a variety of different organic compounds were identified, including benzophenone, 2-phenoxyethanol, butylated hydroxytoluene and benzoic acid, all contaminants of emerging concern (CEC) identified by NORMAN. CEC have been detected in the environment, but are not yet included in routine monitoring programs at the EU level. Their fate, behavior and (eco)toxicological effects are not well understood. Application of the ozonation process resulted in a 4-20% NOM reduction, based on the TOC values. Estimated removal of CECs by ozone increased with increasing ozone dose (up to $1.0 \text{ g O}_3/\text{m}^3$) and was in the range 24-70%. Adsorption on GAC further improves total NOM and CECs removal compared to the ozonation alone. Combined use of ozone and GAC provides up to 16-33% TOC reduction as well as 70-82% CECs removal. UV absorbance values at 254 and 278 nm can serve as indicators of aromatic carbon content in water, and were significantly reduced after ozonation and GAC filtration (by up to 50%). Among the CECs investigated, benzophenone was the most prone to oxidation/adsorption treatment. In addition to the naturally present organic matter, CECs detected by GC/MS analysis can serve as indicators of anthropogenic pollution which may alter drinking water quality. Tracking their behavior during treatment allows assessment of the efficiency of the technological line and optimization of the oxidation process in the case of groundwater pollution by infiltration.

Keywords: *ozonation, GAC filtration, NOM, emerging substances*

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BIOCATALYTIC BIODIESEL SYNTHESIS WITH *BURKHOLDERIA CEPACIA* AND *PSEUDOMONAS FLUORESCENS* LIPASE

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Biodiesel is a mixture of fatty acid methyl esters, usually produced by a transesterification reactions in the presence of sodium hydroxide as catalyst. However, this most often used industrial process for biodiesel synthesis has certain shortcomings. Thus, the use of new catalysts, and among them biocatalyst such as lipases are preferred. Lipases (EC 3.1.1.3.), triacylglycerol acylhydrolases, are enzymes of plant, animal or microbial origin, which are capable to perform efficient transesterification reactions in the presence of organic solvents. Although the use of lipases in biodiesel synthesis is well documented, there are few questions which still need to be resolved, and among it optimal conditions for biodiesel biocatalytic production. Namely, various temperatures and pH values were reported for biodiesel production, which are in some instances different from optimal temperature and pH of lipases used in transesterification reaction. Moreover, determination of pH and temperature optimum for lipase activity has been performed by various substrates differing in their chemical structure and composition, which additionally complicates the selection of optimal conditions. Therefore, the aim of this study was to determine pH and temperature optimum of *Burkholderia cepacia* and *Pseudomonas fluorescens* lipases using two chemically and structurally different substrates: olive oil and *p*-nitrophenyl palmitate, and to examine the efficiency of biodiesel biocatalytic synthesis in batch reactor at determined optimal conditions for lipase activity. Results have shown that optimal conditions (pH and temperature) for lipase activity differed regarding substrate used, and that those determined by olive oil are well in accordance with the data obtained from manufacturer. Furthermore, during batch reactor biodiesel synthesis, higher amount of fatty acid methyl esters (FAME) were produced at optimal conditions determined with olive oil as lipase substrate, than with *p*-nitrophenyl palmitate. Data on the percentage of FAME in crude biodiesel has shown that *Burkholderia cepacia* lipase is more suitable for biodiesel synthesis, where more than 96.5% FAME were obtained after 24 hours of synthesis, in comparison to *Pseudomonas fluorescens* lipase with maximally 71.9% of FAME produced at optimal conditions.

The obtained data indicates that greater amount of biodiesel can be produced if optimal conditions for lipase activity (pH and temperature optimum) are applied during batch reactor synthesis. However, optimal conditions for lipase activity should be determined with olive oil test as more reliable.

Keywords: lipase, biodiesel, Burkholderia cepacia, Pseudomonas fluorescens

SOLAR-LIGHT PHOTOCATALYTIC ACTIVITY OF ZnFe_2O_4 NANOPARTICLES

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In the past decades, much attention was paid to the photocatalytic degradation of organic pollutants with different semiconductor compounds. Among them, solar-light-induced photocatalysts are of great interest because of their high efficiency in utilizing solar energy. The advantages of using the spinel ZnFe_2O_4 as photocatalyst are displayed by its chemical and thermal stability, low cost, magnetic properties and the narrow band gap of around 1.9 eV. The present paper is focused on the research in the photocatalytic activity of zinc ferrites nanoparticles synthesized via different synthetic routes: hydrothermal and coprecipitation method. In both synthesis, aqueous solutions of nitrate sols were used as a source of metal cations and NaOH was used as a precipitating agent. The X-ray diffraction patterns of the prepared nanoparticles indicate the formation of single-phase spinel structure, with crystallite sizes of around 5 nm. The photocatalytic activity of the synthesized samples was evaluated by Rhodamine B (RhB) photodegradation in aqueous solution with the starting concentration of 10 μM RhB under solar-light irradiation. The RhB photodegradation was monitored at the defined time of irradiation using UV-VIS spectrophotometer. Photocatalytic activity of both samples was observed. The photocatalytic efficiency after 360 min of solar-light irradiation reached 44% over hydrothermally prepared ZnFe_2O_4 , while it was slightly enhanced to 47% when ZnFe_2O_4 nanoparticles prepared by coprecipitation were used. These results have shown that zinc ferrite nanoparticles are one of the promising photocatalysts in the field of industrial photodegradation of organic pollutants.

Keywords: *zinc ferrites, nanoparticles, photocatalyst*

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MODELING OF ULTRAFILTRATION WITH DIFFERENT FOULING MECHANISMS

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Membrane filtration process is getting more attention and focus in food industry due to them advantages compared with other conventional separation methods. Unavoidably, membrane fouling always hinders the membrane performance. The characterization of membrane fouling mechanisms is highly important especially during the ultrafiltration process. In this work, Hermia's and Makardij's models were used to investigate the fouling and flux decline mechanisms. Ultrafiltration experiments were performed at different plant originated beverage (soy and oat) and operating parameters (pressure, stirring rate). The fouling mechanism and constant for flux change can be identified using the models. Polyether sulfone membrane was employed throughout this work with 10 kDa MWCO values. Ultrafiltration experiments were performed at a different plant originated beverage (soy and oat), transmembrane pressures (0.005 and 0.1 MPa) and stirring rate (100, 200, 300 and 400 rpm). In this study we used two mathematical models to describe permeate flux decline and identify the fouling mechanism.

According to the Hermia's models, there are four main fouling mechanisms: complete blocking, standard blocking, intermediate blocking and cake layer formation. The other model is the Makardij model where the flux change depends on two coefficients: k_1 is the rate constant for flux decline and k_2 is the rate constant for deposit removal from the membrane. Hermia model analysis showed that the cake layer formation model is the most typical fouling mechanism.

In the case of Makardij's model for transmembrane pressure of 0.005 MPa, the rate constant for deposit removal (k_2) and for transmembrane pressure of 0.1 MPa the value of k_1 is prevailing. The values of two different models give an exact and well-understood connected result of the mechanism of fouling. At lower pressures the rate constant for deposit removal is prevailing and in this case non cake layer formation model was observed in the Herma models.

Keywords: *fouling; Hermia's model; Makardij's model; ultrafiltration*

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INNOVATIVE FOOD SCIENCE AND BIOPROCESSES	1
CHARACTERISATION OF PROTEINS AND SURFACTANTS AT FLUID INTERFACES RELEVANT FOR FOAMS AND EMULSIONS IN FOOD	
T. Kairaliyeva, N. Mucic, J. Katona, E.V. Aksenenko, V.I. Kovalchuk, A.V. Makievski, V.B. Fainerman, S.B. Aidarova, <u>R. Miller</u>	2
BIOPOLYMERS AS VEHICLES FOR DELIVERING INNOVATIVE FUNCTIONALITIES IN FOOD SYSTEMS	
Costas G. Biliaderis	4
PROCESS SIMULATION IS AN ESSENTIAL TOOL IN PRODUCT DEVELOPMENT	
<u>Zivko Nikolov</u> , Laura Soto-Sierra.....	5
INNOVATING WITH BIOACTIVES FROM BRASSICA BYPRODUCTS - THE SPIN-OFF EXPERIENCE	
<u>Diego A. Moreno</u> , Raúl Domínguez-Perles, Paula García-Ibañez, Ángel Abellán-Victorio, Micaela Carvajal, Cristina García-Viguera.....	6
FROM FOOD TO SAFE FOOD: PROBLEMS AND CHALLENGES IN NEW PRODUCT DEVELOPMENT	
<u>Peter Raspor</u>	8
APPLICATION OF SUPERCRITICAL CARBON DIOXIDE FOR DRYING OF FRUITS AND VEGETABLES – SENSORY AND FOOD SAFETY PERSPECTIVES	
Nikola Tomić, Sara Spilimbergo, Gerard Hofland, Andreja Rajković	9
CHALLENGES IN SUGAR PRODUCTION - FOOD SAFETY MANAGEMENT	
<u>Biljana Bogdanović</u> , Branislav Bogdanović.....	12
GINGER ESSENTIAL OIL IMPROVES QUALITY OF COOKED PORK	
<u>Branislav V. Šojić</u> , Vladimir M. Tomović, Branimir M. Pavlić, Predrag M. Ikonić, Marija R. Jokanović, Maja Đ. Ivić, Snežana B. Škaljac	13
EFFECT OF WILD OREGANO ESSENTIAL OIL ON LIPID OXIDATION IN MARINATED PORK CHOPS	
<u>Maja Đ. Ivić</u> , Vladimir M. Tomović, Marija R. Jokanović, Snežana B. Škaljac, Branislav V. Šojić.....	14
THE EFFECT OF GENOTYPE (G), GROWING ENVIRONMENT (E), AND THEIR INTERACTION (G x E) ON THE CHEMICAL COMPOSITION OF WHEAT BRAN	
<u>Ankica Kondić-Špika</u> , Sanja Mikic, Dragana Trkulja, Novica Mladenov, Nada Grahovac, Zvonimir Sakač, Saša Krstović, Nikola Hristov	15



EXPERIMENTAL PRODUCTION OF HEAT-ACID COAGULATING CHEESE WITH ADDITION OF PLUM	
<u>Emina Kadrić</u> , Zlatan Sarić	17
LEAD CONTENT IN THE MEAT OF SAANEN GOAT MALE KIDS FROM VOJVODINA (NORTHERN SERBIA)	
<u>Vladimir M. Tomović</u> , Marija R. Jokanović, Branislav V. Šojić, Snežana B. Škaljac, Milana M. Lazović, Ivana M. Vasiljević, Igor B. Tomašević, Ivana R. Nikolić, Dragan D. Vujadinović, Mila S. Tomović	18
ANTIOXIDANT POTENTIAL AND SENSORY QUALITY OF NEW GREEN WALNUT LIQUEUR	
<u>Sonja P. Veljović</u> , Marija P. Petrović, Stanislava Ž. Gorjanović, Maja M. Natić, Predrag V. Vukosavljević	19
ORGANOCHLORINE PESTICIDES IN RAW MEAT ON THE SERBIAN MARKET	
<u>Brankica Kartalović</u> , Nikolina Novakov, Željko Mihaljev, Jelena Petrović, Jelena Babić, Krešimir Mastanjević	20
DEVELOPMENT OF NEW IN VITRO COLORIMETRIC MTT ASSAY FOR HIGH-THROUGHPUT SCREENING OF ANTI-YEAST ACTIVITY OF PLANT-DERIVED PRESERVATIVE CANDIDATES IN AN ORANGE JUICE FOOD MODEL	
<u>Jan Stas</u> , Marketa Houdkova, Jan Banout, Ladislav Kokoska	22
DISCRIMINANT ANALYSIS OF CHEMICAL AND ANTIOXIDANT PROPERTIES OF <i>Capsicum annuum</i> CULTIVATED UNDER DIFFERENT CONDITIONS	
Milica Ž. Karadžić Banjac, Strahinja Z. Kovačević, Žarko M. Ilin, Boris D. Adamović, Aleksandra N. Tepić Horecki, Zdravko M. Šumić, Anita S. Vakula, Lidija R. Jevrić, Sanja O. Podunavac-Kuzmanović	23
EFFECT OF STARTER CULTURE (<i>STAPHYLOCOCCUS XYLOSUS</i>) ADDITION ON COLOUR CHARACTERISTICS OF DRY FERMENTED SAUSAGE	
<u>Snežana B. Škaljac</u> , Marija R. Jokanović, Vladimir M. Tomović, Maja Đ. Ivić, Branislav V. Šojić, Tatjana A. Peulić, Predrag M. Ikonić, Natalija R. Džinić, Ljiljana S. Petrović	24
THE EFFECT OF FREEZING AND FROZEN STORAGE ON THE CALORIMETRIC PROPERTIES OF LIQUID EGG PRODUCTS	
<u>Karina I. Hidas</u> , Csaba Németh, Anna Visy, Adrienn Tóth, László Friedrich, Ildikó Cs. NyulasZeke.....	25
EFFECT OF HYDROCOLLOIDS ON THE RHEOLOGICAL BEHAVIOR OF YEAST RAISED DOUGH	
Zhour Sabri, Anikó Kovács, <u>Katalin Badakné Kerti</u>	27



EFFECT OF SUGAR REDUCTION AND SUBSTITUTION ON POUND CAKE	
<u>Anikó Kovács</u> , Katalin Badakné Kerti, László Somogyi	28
THE ANTIOXIDANT ACTIVITY AND COMPOSITION OF THE PHENOLICS FROM NON-GROUND NETTLE (<i>Urtica dioica</i> L.) SEEDS EXTRACTED BY DISTILLED AND TAP WATER	
<u>Jelena S. Mitrović</u> , Nada Č. Nikolić, Ivana T. Karabegović, Miodrag M. Lazić, Ljubiša B. Nikolić	29
APPLICATION OF ARTIFICIAL NEURAL NETWORK FOR OPTIMIZATION OF MICROWAVE-ASSISTED EXTRACTION (MAE) OF DEFATTED WHEAT GERM PHENOLIC ANTIOXIDANTS	
Nemanja Bojanić, Nemanja Teslić, Dušan Rakić, Aleksandar Takači, Zoran Zeković, Aleksandar Fišteš, Marija Bodroža-Solarov, Branimir Pavlić	30
RHEOLOGICAL, TEXTURAL AND SENSORY PROPERTIES OF HUMMUS SPREAD DEPENDING ON THE TECHNOLOGICAL PARAMETERS	
<u>Ivana Nikolić</u> , Milena Subotić, Ljubica Dokić, Aleksandar Takači, Zita Šereš, Dragana ŠoronjaSimović, Nikola Maravić	32
SUSCEPTIBILITY OF REFERENCE MICROBIAL STRAINS TO CHOKEBERRY POMACE EXTRACT	
<u>Aleksandra, S. Ranitović</u> , Dragoljub, D. Cvetković, Siniša, L. Markov, Vesna T. Tumbas Šaponjac, Jelena J. Vulić	33
COMPUTER VISION SYSTEM AS RAPID TOOL FOR VOLUME INSPECTION OF POGÁCSA	
<u>Hanieh Amani</u> , László Baranyai, Katalin Badak-Kerti	34
EVALUATION OF IN VITRO GROWTH-INHIBITORY EFFECT OF SPICE ESSENTIAL OILS AND SUPERCRITICAL CARBON DIOXIDE EXTRACTS ON FOOD PATHOGENIC BACTERIA IN LIQUID AND VAPOR PHASE USING BROTH MICRODILUTION VOLATILIZATION METHOD	
<u>Kateřina Vihanová</u> , Markéta Houdková, Trinop Promgool, Ondřej Horák, Somdej Kanokmedhakul, Ladislav Kokoška.....	36
THE POSSIBILITY OF USING EXTRUDED SUGAR BEET PULP FOR THE PRODUCTION OF COOKIES, FROM THE ASPECT OF MICROBIOLOGICAL PROFILE	
Jovana S. Petrović, Sunčica D. Kocić-Tanackov, Biljana S. Pajin, Ivana S. Lončarević, Aleksandar Z. Fišteš, Nemanja Đ. Bojanić, Jelena D. Pejin	37



QUALITY OF FERMENTED MILK PRODUCTS OBTAINED BY CONCENTRATED KOMBUCHA INOCULUM

Mirela D. Ilicic, Spasenija D. Milanovic, Marijana Đ. Carić, Katarina G. Kanurić, Vladimir R. Vukić, Dajana V. Vukić 38

STABILITY OF CAROTENOIDS IN PUMPKIN DURING THE DIFFERENT HEAT TREATMENTS

Dragana M. Paunović, Jovana M. Marković, Evica R. Ivanović, Biljana B. Rabrenović, Saša M. Despotović, Nebojša R. Banjac 39

TECHNOLOGY AND QUALITY OF FRESH CHEESE PRODUCED BY KOMBUCHA INOCULUM

Mirela D. Iličić, Katarina G. Kanurić, Vladimir R. Vukić, Dajana V. Vukić, Maja R. Bjekić..... 41

THE QUALITY OF SUNFLOWER OIL AND PALM OLEIN DURING THE PRODUCTION OF FRENCH FRIES

Dragana M. Paunović, Mirjana A. Demin, Tanja S. Petrović, Jovana M. Marković, Vesna B. Vujasinović, Biljana B. Rabrenović 42

IMMOBILISATION OF LACTOBACILLUS RHAMNOSUS IN COMBINED POLYVINYL ALCOHOL/CALCIUM ALGINATE MATRIX FOR L-(+)LACTIC ACID FERMENTATION

Miloš S. Radosavljević, Steva M. Lević, Jelena D. Pejin, Miona M. Belović, Milana Pribić, Sunčica D. Kocić-Tanackov, Aleksandra P. Djukić-Vuković, Ljiljana V. Mojović, Viktor A. Nedović 43

TRITICALE AS POTENTIAL BREWING RAW MATERIAL

Milana M. Pribić, Jelena D. Pejin, Sunčica D. Kocić-Tanackov, Miloš S. Radosavljević, Aleksandra P. Đukić-Vuković, Ljiljana V. Mojović..... 44

VERY HIGH GRAVITY FERMENTATION OF SUGAR BEET THICK JUICE BY *SACCHAROMYCES CEREVISIAE* IMMOBILIZED IN COMBINED ALGINATE-MAIZE STEM TISSUE BEADS

Vesna M. Vučurović, Vladimir S. Puškaš, Uroš D. Miljić, Jelena S. Filipović..... 46

TEXTURE CHARACTERISTICS OF DRY FERMENTED SAUSAGE AFFECTED BY DRYING PROCESS CONDITIONS

Marija R. Jokanović, Vladimir M. Tomović, Snežana B. Škaljac, Branislav V. Šojić, Predrag M. Ikonić, Tatjana A. Peulić, Maja Đ. Ivić, Nedim M. Čučević 47

POYPHENOL PROFILE OF HONEYDEW AND POLYFLORAL HONEY FROM NORTHER PART OF MONTENEGRO	
Milica M. Nešović, Uroš M. Gašić, <u>Živoslav Lj. Tešić</u>	48
ELECTROCHEMICAL STUDY OF VITAMIN B1 BY MEANS OF ADSORPTIVE STRIPPING CHRONOPOTENTIOMETRY	
<u>Tanja Ž. Brezo-Borjan</u> ¹ , Zvonimir J. Suturović, Snežana Ž. Kravić, Zorica S. Stojanović, Ana D. Đurović, Jovana J. Kos	50
INSTANT COFFEE ENRICHED WITH DIETARY FIBRE	
<u>Blaž Ferjančič</u> , Nina Petrovčič, Mojca Korošec, Nada Vahčić, Jasna Bertonecelj	51
EFFECT OF NITROGEN SOURCES ON BIOBACTERICIDE PRODUCTION BY <i>BACILLUS SUBTILIS</i>	
<u>Ida E. Zahović</u> , Zorana Z. Rončević, Jovana J. Đuran, Ivana Ž. Mitrović, Jovana A. Grahovac, Siniša N. Dodić	52
ANTILISTERIAL EFFECTS OF AUTOCHTHONOUS LACTOCOCCI IN CHEESES FROM ULTRAFILTERED MILK	
Marina P. Ivanović, <u>Zorica T. Radulović</u> , Milica M. Mirković, Ana V. Radulović, Jelena B. Miočinović, Dušanka D. Paunović, Nemanja L. Mirković	53
PRODUCTION OF TEXTURIZED ROUND-BEEF SNACK USING FREEZE AND MICROWAVE PRE-DRYING METHODS	
Anıl Bodruk, Feyza Elmas, Mehmet Koç, Fatma Meltem Serdaroğlu, Nurcan Koca, Figen Kaymak Ertekin.....	55
<i>IN VITRO</i> BIOLOGICAL ACTIVITY OF OILSEED MEAL PROTEIN HYDROLYSATES	
<u>Kristina Radošević</u> , Marijan Logarušić, Ivana Radojčić Redovniković, Marina Cvjetko Bubalo, Igor Slivac, Višnja Gaurina Srček	56
ANTIOXIDATIVE AND ANTICANCER EFFECT OF FLAXSEED PROTEIN HYDROLYSATES	
<u>Marina Cvjetko Bubalo</u> , Marjan Logarušić, Kristina Radošević, Igor Slivac, Višnja Gaurina Srček.....	57
GREEN BIOREFINERY FOR THE ENHANCEMENT OF BIOACTIVITY OF PECTIN FROM SUGAR BEET EXTRACTION WASTE	
Maja M. Milošević, Milica N. Perović, Tatjana R. Đorđević, <u>Mirjana G. Antov</u>	58

PHYSICAL AND NUTRITIONAL PROPERTIES OF EXTRUDED SNACKS WITH BREWER'S PROCESSING BY-PRODUCT ADDITION	
<u>Jovana D. Delić</u> , Predrag M. Ikonić, Marija R. Jokanović, Tatjana A. Peulić, Vojislav V. Banjac, Radmilo R. Čolović, Strahinja Ž. Vidosavljević.....	60
USE OF SAGE EXTRACT FOR GROWTH CONTROL OF <i>E. coli</i> IN MINCED PORK	
Bojana R. Danilović, <u>Natalija G. Đorđević</u> , Aleksandar D. Veličković, Dragiša S. Savić.....	61
EFFECTS OF DIFFERENT MARINATION PROCESSES ON DRYING OF TURKEY BREAST MEAT	
<u>Feyza Elmas</u> , Anıl Bodruk, <u>Mehmet Koç</u> , Fatma Meltem Serdaroğlu, Nurcan Koca, Figen Kaymak Ertekin.....	62
EFFECTS OF MICROWAVE DRYING PRIOR TO PUFF DRYING ON THE PHYSICAL QUALITY CHARACTERISTICS OF DRIED REDUCED- FAT WHITE CHEESE	
<u>Özgün Köprüalan</u> ¹ , Şeyma Arıkaya, Mehmet Koç, Nurcan Koca, Figen Kaymak-Ertekin	63
FACTORS THAT INFLUENCE DETECTION OF HUMAN NOROVIRUS IN RASPBERRIES	
Dragoslava D. Radin	65
PREPARATION OF ZEIN NANOPARTICLES AND SELF-STANDING FILM	
<u>Ljiljana M. Spasojević</u> ¹ , Jaroslav M. Katona, Sandra Đ. Bučko, Nejra F. Omerović, Slavica M. Savić, Jadranka L. Fraj, Jelena R. Milinković Budinčić, Lidija B. Petrović .	66
THE RELATION OF SHAPE AND MOISTURE CONTENT OF KASHAR CHEESE PIECES WITH PRE-DRYING METHODS FOR THE PRODUCTION OF DRIED CHEESE	
Şeyma Arıkaya, Özgün Köprüalan, <u>Nurcan Koca</u> ¹ , Mehmet Koç, Figen Kaymak-Ertekin	67
EFFECTS OF EXPLOSIVE PUFF DRYING AND FREEZE DRYING ON QUALITY CHARACTERISTICS OF HIGH NUTRITIVE PUMPKIN CHIPS	
Özgül Altay, Özgün Köprüalan, Anıl Bodruk, <u>Figen Kaymak Ertekin</u>	68
ENCAPSULATION EFFICIENCY OF CARVACROL IN ZEIN/ROSIN NANOPARTICLES	
<u>Danijela Z. Rajić</u> ¹ , Ljiljana M. Spasojević, Jaroslav M. Katona, Sandra Đ. Bučko, Jadranka L. Fraj, Jelena R. Milinković Budinčić, Lidija B. Petrović, Ljiljana Popović, Marko M. Ivanović, Altnay Sharipova, Saule Aidarova.....	70

THE INFLUENCE OF THE FREEZING RATE ON THE PHYSICO- CHEMICAL PROPERTIES OF PORK MEAT (<i>M. Longissimus dorsi</i>)	
<u>Danica M. Savanović¹</u> , Radoslav D. Grujić, Jovo M. Savanović.....	71
RHEOLOGICAL PROPERTIES OF VEGETEABLE CREAMS INCLUDING DIFFERENT TEXTURIZING AGENTS	
Bence Halasi, Diána Furulyás, Mónika Stéger-Máté, <u>Beatrix Szabó-Nótin</u>	73
CREATION OF INNOVATIVE MEAT PRODUCTS BY RE-USE OF SPENT BARLEY	
Korzeniowska M.	75
OIL AND PROTEIN CONTENTS IN RAPESEED (<i>BRASSICA NAPUS</i> L.) AS A FUNCTION OF ENVIRONMENTAL CONDITIONS DURING SEED FILLING PERIOD	
<u>Ana Marjanović Jeromela</u> , Nada Grahovac, Vladimir Šarac, Vladimir Aćin, Milan Mirosavljević, Ankica Kondić Špika	76
PRODUCTION AND EXTRACTION OF ANTIMICROBIAL COMPOUNDS EFFECTIVE AGAINST PHYTOPATHOGENIC <i>XANTHOMONAS</i> SPP.	
Ivana S. Pajčin ¹ , Jovana A. Grahovac, Vanja R. Vlajkov, Zorana Z. Rončević, Mila S. Grahovac, Aleksandar I. Jokić, Siniša N. Dodić	77
SENSORY AND SOME CHEMICAL CHARACTERISTICS OF OLIVE OILS PRODUCED IN LYBIA	
<u>Vesna B. Vujasinović</u> , Biljana B. Rabrenović, Seddiq M. A. Esalami, Sanja B. Dimić, Dragan V. Tešanović, Maja V. Banjac.....	79
SIMULTANEOUS ULTRASONICATION AND ENZYMATIC SACCHARIFICATION FOR FERMENTABLE SUGAR PRODUCTION USING <i>CHLORELLA VULGARIS</i>	
<u>Sibel Uzuner</u> , Sebnem Kurhan, Gulsun Akdemir Evrendilek	80
SEED SURFACE DISINFECTION METHODS: CURRENT APPLICATIONS AND NEW DIRECTIONS	
Bahar Atmaca, Nurullah Bulut, Sibel Uzuner, <u>Gülsün Akdemir Evrendilek</u>	81
BACTERIAL ADHESION ON KITCHEN SURFACES	
Davor Kovačević, Anamarija Zore, Mojca Jevšnik, Anže Abram, Valentina Runko, Irena Slišković, Katja Bezek, Peter Raspor, <u>Klemen Bohinc</u>	82
MICROWAVE-ASSISTED DILUTE ACID PRETREATMENT FOR IMPROVING PRESSURIZED FERMENTABLE SUGAR EXTRACTION FROM HAZELNUT SHELLS	
Sibel Uzuner	84



FATTY ACIDS PROFILE OF RED RASPBERRY (*RUBUS IDAEUS* L.) SEED OIL:
OPTIMIZATION OF SUPERCRITICAL FLUID EXTRACTION

Boško Marić, Biljana Abramović, Branimir Pavlić, Lidija Peić-Tukuljac, Nebojša Ilić,
Dušica Čolović, Marija Bodroža-Solarov, Zoran Zeković, Nemanja Teslić 85

COMBINATIONS OF HIGH PRESSURE PROCESSING AND HEAT TREATMENT:
SAFETY AND PROTEIN STRUCTURE OF LIQUID EGG WHITE

Adrienn Tóth, Csaba Németh, Réka Juhász, József Surányi, Tamás Csurka, Klára
Pásztor-Huszár, László Friedrich 86

MOLECULAR IDENTIFICATION OF AFLATOXIN BIOSYNTHESIS GENES IN
ASPERGILLUS SPECIES

Nataša Ž. Čurčić, Jelena A. Krulj, Aleksandra S. Bočarov Stančić, Jelena N. Perović,
Marija I. Bodroža Solarov 87

XENOBIOTICS IN *LACTUCA SATIVA* L.

Edward Muntean, Marcel Duda, Nicoleta Muntean 89

INFLUENCE OF pH VALUE OF CEMENT-BASED SUBSTRATES ON VIABILITY OF
BIOCALCIFYING BACTERIA *SPOROSARCINA PASTEURII* DSM 33

Olja Lj. Šovljanski, Ana M. Vidaković, Snežana B. Vučetić, Bojan B. Miljević, Jonjaua
G. Ranogajec, Siniša L. Markov 90

STUDY THE EFFECT OF DEEP FRYING IN OIL CONTENT CHANGES OF FATTY
ACIDS FOR SUNFLOWER OIL AND COTTON

Mahmoud Dahhan 91

SUPERCRITICAL CARBON DIOXIDE EXTRACTION OF CAROTENOIDS FROM
PHAEODACTYLUM TRICORNUTUM

Nadiia Khakimova, Michail Syrpas, Petras Rimantas Venskutonis 92

EFFECT OF FAT CONTENT ON AROMA RELEASE OF FLAVOURED PUDDINGS

Ceyda Dadalı, Yeşim Elmacı 94

USE OF LEGUMES IN GLUTEN-FREE PASTA PRODUCTION

Ceyda, Dadalı, Yeşim, Elmacı 95

THE INFLUENCE OF ROASTING TEMPERATURE ON THE PHYSICAL
PROPERTIES OF ARABICA AND ROBUSTA COFFEE

Božana Odžaković, Natalija Džinić, Marija Jokanović, Slavica Grujić 96



THE TECHNOLOGICAL QUALITY OF SUGAR BEET IN VOJVODINA DURING 2016-2018

Lidija E. Peić Tukuljac, Rada C. Jevtić-Mučibabić, Jovana S. Kojić, Zita I. Šereš, Jelena A. Krulj, Nikola R. Maravić, Marija I. Bodroža Solarov 98

PROPERTIES OF BIOPOLYMER FILM WITH ESSENTIAL OILS

Sandra N. Bulut, Senka Z. Popović, Nevena M. Hromiš, Danijela Z. Šuput, Vera L. Lazić 99

MECHANICAL PROPERTIES OF PUMPKIN OIL CAKE BASED COMPOSITE BIOPOLYMER FILMS

Nevena M. Hromiš, Senka Z. Popović, Sandra N. Bulut, Danijela Z. Šuput, Vera L. Lazić 100

QUALITY CHARACTERISTICS OF STRAWBERRY COATED WITH QUINCE SEED MUCILAGE AS EDIBLE COATING

Ali Kozlu, Yeşim Elmacı..... 102

IMPACT OF AFLATOXIN B₁ ON THE QUALITY OF STORED SPELT WHEAT

Jelena A. Krulj, Jovana S. Kojić, Nataša Ž. Čurčić, Lidija E. Peić Tukuljac, Jelena N. Perović, Marija I. Bodroža-Solarov 103

BETAINE CONTENT IN BUCKWHEAT ENRICHED WHOLEGRAIN WHEAT PASTA

Jelena N. Perović, Jovana S. Kojić, Dubravka J. Škrobot, Jelena A. Krulj, Lidija E. Peić Tukuljac, Nebojša M. Ilić, Marija I. Bodroža Solarov 104

OPTIMIZATION OF MICROWAVE ASSISTED ENZYMATIC EXTRACTION OF STEVIOL GLYCOSIDES AND PHENOLIC COMPOUNDS FROM STEVIA LEAF

Ahmet Görgüç, Esra Gençdağ, Fatih Mehmet Yılmaz..... 106

HYPOGLYCAEMIC AND ANTIHYPERGLYCAEMIC ACTIVITY OF NEW DRUG FORMULATION OF BASIL EXTRACTS

Branislava D. Teofilović, Aleksandar L. Rašković, Nebojša P. Stilinović, Svetlana S. Goločorbin Kon, Momir M. Mikov..... 107

PHENOLICS CONTENT AND ANTIOXIDANT CAPACITY OF BERRY NECTARS

Tijana D. Ilić, Anja N. Vujčić, Nevena M. Dabetić, Vanja M. Todorović, Bojana B. Vidović 108



APPLICATION OF CORRELATIVE ANALYTICAL TECHNIQUES FOR THE DETECTION OF HEAT TREATMENT OF HONEY <u>Zsanett Bodor</u> , Csilla Benedek, Tímea Kaszab, John-Lewis Zinia Zaukuu, Zoltan Kovacs	109
SUGAR BEET MOLASSES QUALITY ENHANCEMENT THROUGH TREATMENT WITH MODIFIED SUGAR BEET PULP Miljana Z. Djordjević, <u>Zita I. Šereš</u> , Nikola R. Maravić, Marijana Z. Djordjević, Dragana M. Šoronja-Simović, Cecilia Hodur, Naoufal Bellahsen	112
PASTING PROPERTIES OF SNACK PRODUCTS FROM SPELT WHOLEGRAIN FLOUR WITH ADDED BETAINE <u>Jovana S. Kojić</u> , Miroslav Hadnađev, Bojana Kokić, Jelena Krulj, Nebojša Ilić, Jelena Perović, Marija Bodroža Solarov	113
PREFERENCES TOWARDS SWEETS AND SALTY SNACK CONSUMPTION AMONG YOUNG POPULATION IN RELATION TO BMI <u>Dragana M. Šoronja-Simović</u> , Zita I. Šereš, Ferenc E. Kiš, Nikola R. Maravić, Biljana S. Pajin, Miljana Z. Djordjević, Žana Šaranović	114
SPECTROSCOPY AS A RAPID METHOD TO DETECT PAPRIKA POWDER ADULTERATION WITH CORN FLOUR <u>John-Lewis Z. Zaukuu</u> , Zsanett Bodor, Zoltan Gillay, Flora Vitalis, Viktoria Zsom-Muha, Zoltan Kovacs	115
THE USE OF ADVANCED CONTROL SYSTEMS IN FOOD PROCESSING OPERATIONS Safiye Nur Dirim	116
THE USE OF SPRAY DRYING AND AGGLOMERATION METHODS TO OBTAIN SPINACH JUICE POWDER AND AGGLOMERATES <u>Hira Yüksel</u> , Safiye Nur Dirim	118
PRODUCTION OF INSTANT LIQUORICE (<i>GLYCYRRHIZA GLABRA</i>) ROOT SHERBET <u>Bülent Başığit</u> , İbrahim Hayoğlu, Sara Bulut, Gülbahar Akyar	119
PHENOLIC FRACTIONS, ANTIOXIDANT AND ANTIDIABETIC POTENTIALS OF <i>QUERCUS INFECTORIA</i> GALL Bülent Başığit, Şehriban Yükksekaya, Ümran Cansu, <u>Hidayet Sağlam</u> , Mehmet Karaaslan	120



OXIDATIVE STABILIZATION OF COLD-PRESSED SUNFLOWER OIL BY CARAWAY (*CARUM CARVI* L.) ESSENTIAL OIL

Aleksandra S. Stojićević, Biljana B. Rabrenović, Vladislav Rac, Vladimir Šarac, Tatjana M. Šolević Knudsen, Mališa P. Antić 121

ASSESSMENT OF ANTIMICROBIAL ACTIVITY OF POMEGRANATE PEEL AND DETERMINATION OF ITS INDIVIDUAL PHENOLIC COMPOUNDS BY USING LC-ESI-MS/MS

Seba Al-Sataf, Bülent Başığit, Ümran Cansu, Hidayet Sağlam, Asliye Karaaslan, Mehmet Karaaslan..... 123

THE EFFICACY OF CONTINUOUS SYSTEM ULTRAVIOLET LIGHT TREATMENT ON THE INHIBITION OF MOLDS INOCULATED TO SURFACES OF YOGHURT WITH SURFACE CREAM

Gülten Tiryaki Gündüz, Ayça Korkmaz, Duygu Kışla, Nurcan Koca, Müge Urgan Öztürk, Sevcan Ünlütürk 124

Keywords: ultraviolet light, surface decontamination, mould inhibition, yoghurt 125

GUAR-XANTHAN EFFECT ON STARCH BIOPOLYMER FILMS PROPERTIES

Danijela Z. Šuput, Senka Z. Popović, Sandra N. Bulut, Nevena M. Hromiš, Vera L. Lazić 125

INNOVATIVE PROTOTYPE OF VACUUM DRYER FOR FRUIT DRYING

Anita S. Vakula, Zdravko M. Šumić, Branimir M. Pavlić, Marija R. Jokanović, Aleksandra N. Tepić Horecki 126

PHYSICO-CHEMICAL PROPERTIES OF VACUUM DRIED APRICOT: INFLUENCE OF DIFFERENT PACKAGING MATERIALS

Aleksandra N. Tepić Horecki, Vera L. Lazić, Senka Z. Popović, Nevena M. Hromiš, Anita S. Vakula, Danijela Z. Šuput, Sandra N. Bulut, Tatjana N. Daničić, Branimir M. Pavlić, Zdravko M. Šumić..... 128

SENSORY CHARACTERISTICS OF FERMENTED CABBAGE OBTAINED ON DIFFERENT CONDITIONS OF FERMENTATION

Zdravko M. Šumić, Mirna V. Drašković Berger, Anita S. Vakula, Marija R. Jokanović, Biljana R. Cvetković, Branimir M. Pavlić, Aleksandra N. Tepić Horecki 129

PREPARATION AND CHARACTERIZATION OF FIBROIN BASED pH SENSITIVE FILMS

Andrea Rac, Nejra Omerović, Ivana Podunavac, Branimir Bajac, Jaroslav Katona..... 130



THE EFFECT OF REPLACING BEEF FAT WITH OLIVE OIL AND PRODUCTION METHODS ON OXIDATIVE CHANGES IN FERMENTED SAUSAGE (SUCUK)

Aslı Zungur Bastioğlu, Berker Nacak, Meltem Serdaroğlu 131

COLOR INVESTIGATION OF COLD PRESSED OILS OF THE LATEST CONFECTIONARY SUNFLOWER HYBRIDS

Ranko S. Romanić, Tanja Z. Lužaić, Nada L. Grahovac, Nada T. Hladni, Snežana Ž. Kravić, Zorica S. Stojanović 132

INHIBITION OF SALMONELLA ENTERITIDIS GROWTH AND STORAGE STABILITY IN CHICKEN MEAT TREATED WITH BASIL AND ROSEMARY ESSENTIAL OILS ALONE OR IN COMBINATION

Zorica Z. Stojanović-Radić, Milica Pejčić, Marija Jokanović, Maja Ivić, Branislav Šojić, Snežana Škaljac 134

TOTAL QUALITY INDEX APPROACH IN THE CULTIVATION OF OYSTER MUSHROOM (*PLEUROTUS OSTREATUS*) GROWN IN CELLULOSE PLANT WASTE

Ana G. Doroški, Ilija V. Đekić, Jovana Đ. Vunduk, Miomir P. Nikšić, Anita S. Klaus 135

PERCEPTION OF SWEETNES INTENSITY AMONG STUDENTS FROM TWO COUNTRIES

Mojca Korošec, Jasna Bertonec, Elena Molac, Dragana Šoronja-Simović, Biljana Pajin, Zita Šereš 136

THE INFLUENCE OF FUNCTIONAL INGREDIENTS ON THE TECHNOLOGICAL AND QUALITY PARAMETERS OF WHEAT BREAD

Ana Griz, Jana Zahorec, Meta Sterniša, Alenka Levart, Zita Šereš, Dragana Šoronja-Simović, Sonja Smole Možina 137

HORSERADISH LYOPHILISATE AS BREAD INGREDIENT - EFFECT ON RHEOLOGICAL AND FERMENTATION PROPERTIES

Meta Sterniša, Matea Radešić, Jana Zahorec, Franz Bucar, Zita Šereš, Dragana Šoronja-Simović, Sonja Smole Možina 138

PHYSICAL AND MECHANICAL PROPERTIES OF DIFFERENT PROTEINBASED EDIBLE FILMS

Miroslav Hadnađev, Milan Marčeta, Nataša Jovanović-Lješević, Veljko Krstonošić, Tamara Dapčević-Hadnađev 140

pH VALUE, TEMPERATURE AND COLOR PARAMETERS L*, a*, b* EVALUATION IN RAW PORK DURING 72 HOURS POST-MORTEM

Goran Vučić, Radoslav Grujić, Ladislav Vasilišin 141



EXTRACTION OF BIOPOLYSACCHARIDES FROM MARINE BIOMASS USING ALTERNATIVE SOLVENTS

Naiara Fernández Hernández, Maha Abdallah, Ingeborg Heuschkel, Pavel Gurikov, Maria R. Bronze, Ana A. Matias..... 142

EXTRACTION OF POLYPHENOLS FROM OILSEED CAKES BY SUBCRITICAL WATER

Nataša M. Nastić, Biljana S. Pajin, Giorgia Spigno, Jaroslava V. Švarc-Gajić, Ivana S. Lončarević..... 143

SUBCRITICAL WATER EXTRACTION OF PHENOLIC COMPOUNDS FROM COCOA BEAN HULLS

Nataša M. Nastić, Biljana S. Pajin, Joachim Venus, Jaroslava V. Švarc-Gajić, Ivana S. Lončarević..... 145

MICROBIOLOGICAL STABILITY OF COOKED SAUSAGES AS FUNCTION OF REPLACEMENT INORGANIC SALTS WITH NATURAL ADDITIVES

Dragan P. Vujadinović, Milan S. Vukić, Vladimir M. Tomović, Ardea Ž. Milidrag, Marko M. Ivanović..... 146

INFLUENCE OF DIFFERENT FOOD BY-PRODUCTS ON RHEOLOGICAL BEHAVIOR OF WHOLEGRAIN WHEAT DOUGH

Aleksandra M. Torbica, Jelena M. Tomić, Elizabet P. Janić Hajnal..... 147

THE EFFECT OF DRYING TREATMENT ON THE RETENTION OF ANTIOXIDATIVE PROPERTIES OF STRAWBERRY

Snežana M. Stevanović, Tanja S. Petrović, Mirjana B. Pešić 148

CHARACTERISATION OF FRUIT WINE PRODUCED FROM QUINCE (*CYDONIA OBLONGA*) CONCENTRATED JUICE

Vladimir S. Puškaš, Uroš D. Miljić, Jovana J. Đuran, Vesna M. Vučurović..... 150

DETERMINATION OF BISPHENOL A IN BABY BOTTLES AND DRINKING CONTAINERS BY HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY

Gorica Č. Vuković, Marina R. Đukić, Vojislava P. Bursić, Tijana N. Stojanović, Aleksandra P. Petrović, Dušan M. Marinković, Bojan B. Konstantinović 151

NOVEL RAPID SENSORY PROFILING METHOD AS A TOOL FOR DIFFERENTIATION BETWEEN TRADITIONAL AND COMMERCIAL DRY FERMENTED SAUSAGES

Dubravka J. Škrobot, Jelena M. Tomić, Predrag M. Ikonić, Tamara R. Dapčević Hadnađev, Alena M. Tomšik, Miroslav S. Hadnađev, Jovana D. Delić..... 152



TESTING 3D PRINTED STATIC MIXER WITH IMPROVED GEOMETRY	
<u>Igor L. Gáspár</u> , Svetlana S. Popović.....	154
PHYSICOCHEMICAL PROPERTIES OF OLIVE OIL OLEOGELS STRUCTURED WITH MONOGLYCERIDES	
Alexandros Soniadis, <u>Costas G. Biliaderis</u> , Konstantina Zampouni, Thomas Moschakis, Athina Lazaridou, Eugenios Katsanidis	155
IMPROVEMENT OF THE NUTRITIONAL PROFILE OF GREEK SAUSAGES BY ANIMAL FAT AND NaCl SUBSTITUTION	
Konstantina Zampouni, <u>Costas G. Biliaderis</u> , Alexandros Soniadis, Athina Lazaridou, Thomas Moschakis, Eugenios Katsanidis	156
SPOILAGE BACTERIA <i>PSEUDOMONAS</i> – PRODUCTION OF HYDROLYTIC ENZYMES AND ABILITY TO GROW AT 5 °C	
Meta Sterniša, Mihael Čargo, <u>Sonja Smole Možina</u>	157
MATERIALS DESIGN AND APPLICATIONS	159
BIOLOGICAL OILS AS A PLATFORM FOR SUSTAINABLE DEVELOPMENT OF POLYMERIC MATERIALS	
Zoran S. Petrović	160
POLYCARBONATE-BASED POLYURETHANES AND POLYURETHANE NANOCOMPOSITES	
<u>Milena Špírková</u> , Jelena Pavličević, Jiří Hodan, Sabina Krejčíková, Luďka Machová, Jaroslava Budinski-Simendić	161
FOAMING OF ALKALI ACTIVATED MATERIALS	
<u>Vilma Ducman</u> , Katja Traven, Mark Češnovar, Lidija Korat, Barbara Horvat	162
RAMAN SPECTROSCOPY FOR CHARACTERISATION OF BRONZE OBJECTS IN DIFFERENT ENVIRONMENTS AND ITS PROTECTION SYSTEMS	
Polonca Ropret	163
NANO-MODIFIED BIO-FIBRES FOR ADVANCED CEMENT MORTARS	
Anna-Maria Sigouni, Fotini Kesikidou, Parthena Koltso, <u>Maria Stefanidou</u>	165
ZnO/TiO ₂ AND Ag/ZnO/TiO ₂ CATALYSTS FOR DECOLORIZATION OF MIXTURE OF ORGANIC DYES AND COMPARISON OF THEIR EFFICIENCY WITH TiO ₂ , ZnO, TiO ₂ +ZnO, Au/ZnO AND MnO ₂	
Zsigmond J. Papp	166



CRACK SELF-HEALING ABILITY OF BIO-MORTAR

Bojan Miljević, Francesco Lo Monte, Snežana Vučetić, Olja Šovljanski, John Milan van der Bergh, Ivan Ristić, Siniša Markov, Liberato Ferrara, Jonjaua Ranogajec 167

NON-INVASIVE INVESTIGATION OF PAINTING TECHNOLOGY OF PROMINENT 20TH CENTURY PAINTER MIĆA POPOVIĆ

Snežana Vučetić, John Milan van der Bergh, Bojan Miljević, Jonjaua Ranogajec, Bojan Nikolić, Marko Božović, Milica Konstantinović, Ognjen Kovačević, Veljko Džikić... 168

SELF-CLEANING OF HISTORICAL AND MODERN MINERAL MATERIALS – LABORATORY AND IN SITU APPROACHES

Rajko Travica, Radmila Mandić, Marija Tzoutzouli Malešević, Snežana Vučetić, Bojan Miljević, John Milan van der Bergh, Jonjaua Ranogajec..... 170

SYNTHESIS OF EPOXY BASED COATINGS

Ivan Ristić, Radmila Radičević, Milena Marinović-Cincović, Danica Piper, Maja Valčić, Aleksandra Miletić, Vladan Mičić 171

INTERDISCIPLINARY ASPECT OF EDUCATION AND MATERIALS SCIENCE

Nevena Vukić, Natalija Budinski, Zsolt Lavicza, Mirjana Jovičić, Vesna Teofilović, Tamara Erceg, Mlađan Popović 172

THE INFLUENCE OF THE TYPE OF ACTIVATION OF THE MONTMORILLONITE ON THE HYDROLYTIC STABILITY OF UREA-FORMALDEHYDE NANOCOMPOSITE

Milena Čanačević, Marija Kostić, Suzana Samaržija-Jovanović, Vojislav Jovanović, Branka Petković, Gordana Marković, Jaroslava Budinski-Simendić 173

THERMAL PROPERTIES OF AMINE CURED EPOXY HYBRID MATERIALS WITH DIFFERENT CONTENT OF MONTMORILLONITE

Vesna B. Teofilović, Mirjana C. Jovičić, Jelena M. Pavličević, Katalin Mészáros-Szécsényi, Radmila Ž. Radičević, Milena Špírkova, Ayşe Z. Aroğuz..... 174

STRUCTURING OF ENVIRONMENTALLY FRIENDLY TYRES USING SBR ELASTOMER AND COMBINED ACTIVE NANOFILLERS

Jelena Pavličević, Nada Lazić, Milenko Plavšić, Pero Dugić, Milanka Plavšić, Dejan Kojić, Jaroslava Budinski-Simendić 175

THE USE OF MATHEMATICAL MODELING FOR DETERMINATION OF OPTIMAL PMMA/NANO-OXIDE MATERIAL DESIGN

Mirjana C. Jovičić, Jelena M. Pavličević, Bojana B. Ikonić, Predrag S. Kojić, Aleksandar I. Jokić, Dragan D. Govedarica, Katalin Mészáros Szécsényi 176



MEASURING THE DRYING SENSITIVITY OF CLAYS: A REVIEW	
<u>Miloš Vasić, Zagorka Radojević</u>	177
ANTICORROSION ADDITIVES – SPRAY DRYER PRODUCTION AND INFLUENCE OF POST-PRODUCTION TREATMENT ON THE SIZE DISTRIBUTION	
M. Papovic, J. Skrbic, R. Rogovits, T. Sobisch, J. Katona, <u>N. Mucic</u>	178
FUNCTIONAL BIO-BASED MATERIALS IN WOUND DRESSING	
Aleksandra P. Miletić, Ivan S. Ristić, <u>Branka M. Pilić</u>	180
BIOLOGICAL AND ELECTROCATALYTIC APPLICATIONS OF Ag, Au, AND Cu NANOPARTICLES, AND THEIR POLYANILINE BASED COMPOSITES	
<u>Vesna Vodnik, Una Stamenović</u>	181
DIELECTRIC CHARACTERIZATION OF ENGINE COOLANTS	
Róbertné V. P. Kovács	182
NON-INVASIVE STUDIES OF EARLY BRONZE AGE POTTERY FRAGMENTS EXAVATED AT THE SITE MEANISTE IN RANUTOVAC, SERBIA	
<u>Maja D. Gajić-Kvašček, Velibor Dj. Andrić, Marija M. Vuksanović, Aleksandar Bulatović</u>	183
MESOPOROUS SILICA – HYDROXYAPATITE COMPOSITES AS CARRIERS OF SOIL NUTRIENTS	
<u>Kristina Živojević, Vladimir Ćirić, Branislav Jović, Nikola Knežević</u>	185
RE-SYNTHESIZING CATHODE MATERIAL FROM SPENT LI-ION BATTERIES AND ITS EXAMINATION IN AN AQUEOUS SOLUTION OF NaNO_3	
<u>Jelena. V. Senčanski, Ivana B. Stojković Simatović, Stevan N. Blagojević, MilicaM.Nešović, Milica J. Vujković</u>	186
INFLUENCE OF BRICK CLAY CHARACTERISTICS TO THE QUALITY OF ADOBE CLAY BRICKS	
<u>Milica V. Vasić, Lato L. Pezo, Zagorka M. Radojević</u>	187
INVESTIGATION OF SOLVATOCHROMIC PROPERTIES OF SELECTED NEWLY SYNTHESIZED AZO-DYES DERIVATIVES	
<u>Gorana S. Mrđan, Borko M. Matijević, Jelena Lađarević, Dušan Ž. Mijin, Đendi Đ. Vaštag, Suzana Lj. Apostolov</u>	189
MINERAL BY-PRODUCTS AS ADDITIVES IN NEWLY SYNTHESIZED CEMENT	
<u>Jasmina Nešković, Jonjaua Ranogajec, Milan Trumić, Snežana Vučetić</u>	190



FAR INFRARED RADIATION BIOCERAMIC FIBERS A NEW GENERATION OF FUNCTIONAL TEXTILES

Amine Haj Taieb 191

PROTIC IONIC LIQUIDS IN CONSERVATION OF THE PAPER HERITAGE ARTEFACTS

Slobodan B. Gadžurić, Nataša N. Dimitrić, Sanja D. Belić, Aleksandar S. Tot, Jovana J. Panić, Maja A. Karaman, Daniela Korolija-Crkvenjakov, Milan B. Vraneš 192

CORRELATION OF ED-XRF AND ICP-OES METHODS APPLIED IN CHEMICAL CHARACTERIZATION OF CEMENTITOUS MATERIALS

Nevenka Mijatović, Anja Terzić, Ljiljana Miličić, Dragana Živojinović 194

POSSIBILITY OF CONSTRUCTION MATERIAL PROTECTION IN COASTAL CONDITIONS USING NEW ENVIRONMENTALLY FRIENDLY INHIBITORS

Đendi Đ. Vaštag, Špiro N. Ivošević, Slobodan B. Gadžurić, Milan B. Vraneš, Tatjana M. Trtić Petrović, Senka Šekularac-Ivošević, Suzana Lj. Apostolov 195

PHOTOCATALYTIC COATING BASED ON ILITE CLAY/TiO₂ COMPOSITE

Vojo Jovanov, Vladimir Zečević, Jonjaua Ranogajec, Emilija Fidanchevska 196

ADHESION OF BACTERIA ON PROTEIN-TERMINATING POLYELECTROLYTE MULTILAYERS

Klemen Bohinc, Jerca Bajuk, Jasmina Jukić, Anže Abram, Martina Oder, Karmen Godič Torkar, Peter Raspor, Davor Kovačević 198

EVALUATION OF THE PROCESS PARAMETERS OF CERAMIC PRODUCED FROM CLAY AND C&DW

Biljana Angjusheva, Vilma Ducman, Emilija Fidancevska, Vojo Jovanov 199

THERMAL STABILITY AND DEGRADATION KINETICS OF ALIPHATIC POLYURETHANE/NANOSILICA COMPOSITES

Berta Hollo, Milena Špírková, Jelena Pavličević, Oskar Bera, Mirjana Jovičić, Bojana Ikonić, Katalin Mészáros Szécsényi 200

ANALYSIS OF GIBBSITE SAMPLES OF BAUXITE AND POSSIBILITY OF APPLICATION IN BAYER PROCESS

Dario Balaban, Duško Kostić, Mitar Perušić, Zoran Obrenović, Radislav Filipović 201

A CRITICAL REVIEW OF CERAMICS FRACTURE TOUGHNESS MEASUREMENT METHODS AND OBSERVATIONS ON SEVNB METHOD IN CASE OF TRADITIONAL CERAMICS

Dragan Rajnovic, Lepasava Sidjanin, Snezana Vucetic, Jonjaua Ranogajec 202



PREPARATION AND CHARACTERIZATION OF FILMS BASED ON MESOPOROUS SILICA AND ZEIN	
<u>Nejra Omerović</u> , Minja Mladenović, Nikola Knežević, Ljiljana Spasojević, Jaroslav Katona	204
FIFTEEN YEARS OF COLLABORATION OF THE FACULTY OF TECHNOLOGY AND THE GALLERY OF MATICA SRPSKA	
Daniela D. Korolija Crkvenjakov	205
MODIFICATION OF STRUCTURE AND PROPERTIES OF BaTiO ₃ THIN FILMS BY ADDITION OF Sr ²⁺ AND Zr ⁴⁺ IONS	
<u>Jelena Vukmirović</u> , Andrea Nesterović, Ivan Stijepović, Marija Milanović, Zeljka Cvejić, Jelena Bobić, Vladimir V. Srdić	206
REUSE OF CEMENT BYPASS DUST FOR ENVIRONMENTAL FRIENDLY PRODUCTION OF BUILDING MATERIALS	
<u>Damir Čjepa</u> , Željko Ivošević, Snežana Vučetić, Bojan Miljević, John Milan van der Bergh, Jonjaua Ranogajec	208
VALORISATION OF FLY ASH AS SECONDARY RAW MATERIAL TOWARDS IMPLEMENTATION OF CIRCULAR ECONOMY	
<u>Biljana Angjusheva</u> , Vojo Jovanov, Katarin Šter, Alexandra Momirov, Pece Murtanovski, Emilija Fidanchevski, Sabina Kramar	209
MESOPOROUS SiO ₂ PARTICLES WITH CHROMOPHORIC SYSTEM AND THEIR USE IN INDICATION OF RELATIVE HUMIDITY	
E. Švara Fabjan, <u>A. Sever Škapin</u> , P. Nadrah, A. Ajdovec, N. Zabukovec Logar, M. Mazaj, G. Dražić	210
NUTRACEUTICALS AND PHARMACEUTICALS.....	212
THE ROLE OF MICROBIOTA IN THE IMMUNONUTRITION FIELD	
Ascensión Marcos	213
<i>IN SILICO</i> SIMULATIONS OF FREEZE DRYING OF PHARMACEUTICAL FORMULATIONS	
Teresa Barata, Paul Dalby, Steve Brocchini, <u>Mire Zloh</u>	214
PHARMACEUTICALS IN THE ENVIRONMENT: A GROWING PROBLEM	
<u>Cristina Delerue-Matos</u> , Paula Paíga, Luísa Correia-Sá, Manuela Correia	215
DEVELOPMENT OF FUNCTIONAL PRODUCTS USING BIOLOGICALLY ACTIVE SUBSTANCES	
Evgeniy A. Shamin.....	216



RHEOLOGICAL PROPERTIES OF CHITOSAN-SODIUM LAURYL ETHER SULFATE COMPLEXES	
<u>Jelena R. Milinković Budinčić</u> , Lidija B. Petrović, Jadranka L. Fraj, Jaroslav M. Katona, Sandra Đ. Bučko, Ljiljana M. Spasojević	217
VARIATION IN THE MAIN NUTRIENTS, PHYTOCHEMICALS AND SENSORY PROPERTIES AMONG VARIOUS SWEET MAIZE GENOTYPES	
Marija S. Milašinović-Šeremešić, <u>Milica M. Radosavljević</u> , Jelena Ž. Srdić, Jelena Z. Mesarović	219
THE STUDY OF THE PROPERTIES OF THE SUBSTANCES FOR THE PHARMACEUTICAL INDUSTRY	
<u>Alexander V. Knyazev</u> , Anastasia S. Shipilova, Svetlana S. Knyazeva, Ekaterina V. Gusarova, Vladimir N. Emel'yanenko, Alexey A. Amosov	220
POLYMORPHISM IN STEROID HORMONES	
<u>Svetlana S. Knyazeva</u> , Alexander V. Knyazev, Anastasia S. Shipilova, Ekaterina V. Gusarova, Alexey A. Amosov	221
THE PHYTOCHEMICAL AND PHARMACOLOGICAL ACTIVITY OF CITRUS LIMETTA PEEL EXTRACTS	
Shyam Jee	222
STUDIES ON PHYTOTHERAPEUTIC QUALITY OF <i>EQUISETUM ARVENSE</i> L. IN CLUJ, ROMANIA	
Neli-Kinga Olah, Edward Muntean, <u>Marcel M. Duda</u>	224
EMULSIFYING PROPERTIES OF POTATO PROTEINS IN COMPARISON TO WHEY PROTEINS	
<u>Marina D. Kalic</u> , Veljko S. Krstonošić, Miroslav S. Hadnađev, Tamara R. Dapčević Hadnađev	225
STUDY OF THE TEMPERATURE DEPENDENCE OF THE HEAT CAPACITY OF INULIN AND HEVEIN	
<u>Anastasia S. Shipilova</u> , Alexander V. Knyazev, Svetlana S. Knyazeva, Ekaterina V. Gusarova, Alexey A. Amosov	226
WILD GROWING AND CULTIVATED POMEGRANATE: CHEMICAL PROFILING OF JUICES	
<u>Šavikin P. Katarina</u> , Zdunić M. Gordana, Teodora R. Janković, Jelena Č. Živković, Dubravka J. Bigović, Nebojša R. Menković	227



INFLUENCE OF DRYING METHOD ON CHITOSAN/XANTHAN POLYELECTROLYTE COMPLEX CHARACTERISTICS	
Ana R. Ćirić, Miodrag N. Mitrić, Vladimir D. Dobričić, <u>Ljiljana M. Đekić</u>	228
IN VITRO CHARACTERIZATION OF REHYDRATION PROCESS AND DISSOLUTION OF IBUPROFEN FROM CHITOSAN/XANTHAN POLYELECTROLYTE COMPLEX BASED DRUG DELIVERY SYSTEMS	
<u>Ana R. Ćirić</u> , Ljiljana M. Đekić	229
THE EFFICIENCY OF ZINC SUPPLEMENTATION	
<u>Jelena D. Nakomčić</u> , Tamara S. Zadrija	231
THERMODYNAMIC AND KINETIC PROPERTIES OF EQUIMOLAR AQUEOUS ANIONIC AND CATIONIC SURFACTANTS MIXTURES – CATANIONICS	
J. Skrbić, T. Kairaliyeva, L. Petrović, J. Katona, V.B. Fainerman, <u>N. Mucić</u> , R. Miller	233
IN SILICO STUDY OF THE BIOLOGICAL POTENTIAL OF THE SELECTED AMIDE DERIVATIVES	
<u>Suzana Lj. Apostolov</u> , Đendi Đ. Vaštag, Borko M. Matijević, Gorana S. Mrđan	234
REACTION OF INOCULATED CANCERS ON NITRIC OXIDE HYPERPRODUCTION, GLUCOSE ENERGY, FOLATE AND VITAMIN B ₁₂ INHIBITION IN YOUNG HAMSTERS	
Dušica J. Popović, Kosta J. Popović, Dušan Lalošević, <u>Jovan K. Popović</u> , Ivan Čapo, Dejan Miljković	235
FOLATE, VITAMIN B ₁₂ , GLUCOSE ENERGY DEFICIENCY AND NITRIC OXIDE OVERPRODUCTION INHIBITORY EFFECTS ON FIBROSARCOMA IN ADULT HAMSTERS	
Kosta J. Popović, Dušica, J. Popović, Dušan Lalošević, <u>Jovan K. Popović</u> , Ivan Čapo, Dejan Miljković	236
POTENTIAL FOR USE OF VINE SHOOTS EXTRACT IN PHARMACEUTICAL INDUSTRY	
<u>Nikola B. Jojić</u> , Aleksandra I. Jovanović Galović, Nataša M. Jovanović Lješковиć, Slobodan B. Gigov, Milan D. Ilić, Senka S. Vidović, Jelena Z. Vladić, Vesna V. Kojić, Dimitar S. Jakimov	237
CHEMICAL COMPOSITION AND CYTOTOXICITY OF <i>HELICHRYSUM ITALICUM</i> EXTRACT GROWN IN URBAN CONDITIONS	
<u>Zorica O. Mrkonjić</u> , Aleksandra I. Jovanović Galović, Nataša M. Jovanović Lješковиć, Mire F. Zloh, Senka S. Vidović, Jelena Z. Vladić, Milan D. Ilić, Manda I. Dizdar, Vesna V. Kojić, Dimitar S. Jakimov	238



ANTIOXIDANT ACTIVITY OF *MELISSA OFFICINALIS* EXTRACTS

Zorica O. Mrkonjić, Marija D. Jolić, Marina D. Kalić, Manda I. Dizdar, Aleksandra I. Jovanović Galović 239

MICROENCAPSULATION OF JUNIPER BERRY ESSENTIAL OIL (*JUNIPERUS COMMUNIS* L.)

Jelena D. Bajac, Branislava G. Nikolovski, Ivana S. Lončarević, Jovana S. Petrović, Lidija B. Petrović, Branimir M. Bajac 240

PRODUCTION OF FUNCTIONAL PROTEIN ISOLATES FROM VEGETABLE BY-PRODUCTS

Tea Sedlar, Jelena Čakarević, Ljiljana Popović 241

CHEMICAL PROFILING OF WINES OBTAINED FROM AUTOCHTHONOUS GRAPE VARIETY PROKUPAC

Milan D. Ilić, Jelena Č. Živković, Katarina P. Šavikin, Zorica O. Mrkonjić, Nikola B. Jojić, Nataša M. Lješković, Slobodan B. Gigov, Nebojša R. Menković 242

ULTRASOUND-ASSISTED EXTRACTION OF CANNABIDIOL AND $\Delta\Delta$ ⁹⁹ TETRAHYDROCANNABINOL FROM CANNABIS AREAL PARTS AND PROCESS MODELING

Zorica Drinić, Jelena Vladić, Senka Vidović, Anamarija Koren, Biljana Kiprovski, Nadežda Stojanov, Tijana Zeremski 243

OPTIMIZATION OF EXTRACTION OF PHENOLICS FROM *GENTIANA LUTEA* ROOT

Jelena R. Mudrić, Teodora R. Janković, Katarina P. Šavikin, Svetlana R. Ibrić, Jelena D. Đuriš 244

BIOACTIVITY POTENTIAL OF A MEDITERRANEAN BRACKISH DIATOM CULTIVATED IN PHOTOBIOREACTORS

C. Rodolfo, S. Savio, R. Congestri 245

MICROWAVE-ASSISTED SYNTHESIS OF BILE ACID DERIVATIVES AS POTENTIAL LIGANDS OF GLUCOCORTICOID RECEPTOR AND ALDO-KETO REDUCTASE (AKR1C)

Ljubica M. Grbović, Ksenija J. Pavlović, Bojana R. Vasiljević, Sofija S. Bekić, Maja A. Marinović, Edward T. Petri, Anđelka S. Čelić 246

SYNERGISM IN MIXED MONOLAYERS OF POLOXAMER 407 AND POLYSORBATES

Dejan M. Ćirin, Veljko S. Krstonošić, Maja Lj. Milanović 247



IN VITRO TECHNIQUES FOR *DROSER* *ROTUNDIFOLIA* SPECIES FOR FUTURE USE IN THE PHARMACEUTICAL FIELD

Violeta Turcuş, Viviane B. Bota, Elisabeta Chiş, Endre Mathe, Neli Olah 248

THE EFFECT OF MAGNESIUM STEARATE AND SODIUM STARCH GLYCOLATE ON POWDER FLOWABILITY

Gordana Švonja Parezanović, Mladena Lalić-Popović, Svetlana Goločorbin-Kon, Nemanja Todorović, Nebojša Pavlović, Jelena Jovičić-Bata..... 249

EXPOSURE OF PATIENTS TO SODIUM FROM EFFERVESCENT DOSAGE FORMS

Jelena Čanji, Nemanja Todorović, Katarina Jeremić, Nebojša Pavlović, Svetlana Goločorbin-Kon, Jelena Jovičić Bata, Mladena Lalić-Popović 250

OIL EXTRACTION FROM PLUM SEEDS (*PRUNI DOMESTICAE* L.)

Ivan M. Savić, Ivana M. Savić-Gajić 251

INVESTIGATION OF VOLUMETRIC AND VISCOMETRIC PROPERTIES OF ECSTASY IN AQUEOUS SOLUTIONS

Sanja D. Belić, Marija B. Petrin Miličević, Nataša Radosavljević-Stevanović, Milan B. Vraneš, Aleksandar S. Tot, Snežana M. Papović, Slobodan B. Gadžurić 252

GREEN EXTRACTION OF POLYPHENOLS FROM NETTLE LEAVES (*URTICA DIOICA* L)

Marta Maras, Valentina Kruk, Sanja Radman, Verica Dragović-Uzelac, Danijela Bursać Kovačević..... 253

THE INFLUENCE OF ULTRASOUND ASSISTED EXTRACTION ON THE ISOLATION OF BIOACTIVE COMPOUNDS FROM NETTLE LEAVES

Valentina Kruk, Marijana Jurković, Maja Repajić, Ivanka Žutić, Verica Dragović-Uzelac, Zoran Zorić..... 255

ANTIADHESIVE AND ANTIBIOFILM POTENTIAL OF SUBCRITICAL WATER EXTRACTS DERIVED FROM MASHROOM *INONOTUS OBLIQUUS*

Vesna V. Lazić, Jovana Đ. Vunduk, Jelena Z. Vladić, Senka S. Vidović, Anita S. Klaus..... 256

ANTIMICROBIAL ACTIVITY OF *MACLURA POMIFERA* DRY EXTRACTS

Snežana Đ. Filip, Ana M. Vidaković, Aleksandra S. Ranitović, Saša D. Đurović, Stevan N. Blagojević, Zoran P. Zeković..... 257



FREEZE DRIED BERRIES AS SOURCE OF NUTRITIONALLY VALUABLE COMPOUNDS	
Marija Radojković , Milena Vujanović , Gökhan Zengin, Tatjana Majkić, Ivana Beara, Saša Đurović, Zoran Zeković	258
COMPARISON OF THE EFFICIENCY OF DIFFERENT METHODS FOR HESPERIDIN AND NARIRUTIN EXTRACTION FROM ORANGE PEEL	
<u>Silvija Šafranko</u> , Martina Jakovljević, Senka Vidović, Stela Jokić	260
EFFECT OF ANTIHISTAMINE (LORATADINE) TO ACTIVITY OF CATALASE <i>IN VITRO</i>	
Edhem Hasković, <u>Safija Herenda</u> , Zehra Halilović, Denis Hasković, Ena Deljkić	261
MULTI-RESPONSE OPTIMIZATION OF POLYPHENOLS RECOVERY FROM <i>THYMUS SERPYLLUM</i> BY ULTRASOUND-ASSISTED EXTRACTION	
<u>Živan Mrkonjić</u> , Nemanja Teslić, Zoran Zeković, Branimir Pavlić.....	262
CHEMICAL COMPOSITION OF <i>SANTOLINA CHAMAECYPARISSUS</i> L. FLOWER ESSENTIAL OIL	
Biljana Kiprovski, Vladimir Sikora.....	263
VALORISATION OF HOREHOUND EXTRACTS OBTAINED USING ULTRASOUND AND MICROWAVE ASSISTED EXTRACTION: ANTIHYPERGLYCAEMIC ACTIVITY	
<u>Aleksandra Gavarić</u> , Senka Vidović, Jelena Vladić, Jelena Čakarević, Ljiljana Popović.....	264
INFLUENCE OF SUPERCRITICAL FLUID EXTRACTION PARAMETERS ON GRAPE SEEDS OIL RECOVERY	
<u>Ivana Dimić</u> , Živan Mrkonjić, Nemanja Teslić, Dušica Čolović, Zoran Zeković, Branimir Pavlić.....	265
PARTICLE ENGINEERING IN MODERN DRUG FORMULATION	
Rita Ambrus.....	266
PROTEIN HYDROLISATES FROM FOOD BY-PRODUCTS AS NEW ANTIDIABETIC COMPOUNDS	
<u>Jelena Čakarević</u> , Tea Sedlar, Ljiljana Popović	267
<i>IN SILICO</i> PHARMACOKINETIC AND TOXICOLOGICAL ANALYSIS OF BENZOXAZOLES AS ANTIMICROBIAL AGENTS	
<u>Jelena V. Živković</u> , Slavica M. Sunarić, Marko S. Denić, Vesna Lj. Savić, Milica I. Stanković, Katarina B. Bačević.....	268



QUALITY CONTROL AND COUMARIN CONTENT DETERMINATION OF CINNAMON SAMPLES FROM THE SERBIAN MARKETS

Katarina D. Jeremić, Neda S. Gavarić, Nebojša V. Kladar, Milica G. Aćimović, Nemanja B. Todorović, Maja D. Hitl, Biljana N. Božin 269

ISOLATION OF ESSENTIAL OIL FROM ORGANIC *MENTHA PIPERITA* L. BY EMERGING EXTRACTION AND DISTILLATION TECHNIQUES

Branimir Pavlić, Aleksandar Radivojac, Saša Đurović, Živan Mrkonjić, Zoran Zeković 270

EXTRACTION OF WILD THYME (*THYMUS SERPYLLUM* L.): SUPERCRITICAL CO₂ VS. CONVENTIONAL EXTRACTION METHODS

Marinela Nutrizio, Jelena Vladić, Aleksandra Gavrić, Anet Režek Jambrak, Senka Vidović 271

PHENOLIC PROFILE AND ANTIOXIDANT PROPERTIES OF STRAWBERRY WINE

Uroš Čakar, Aleksandar Petrović, Boris Pejin, Vlatka Vajs, Brižita Đorđević 272

IONIZATION BEHAVIOR OF URSODEOXYCHOLIC AND DEOXYCHOLIC ACID IN THE BINARY MIXED MICELLES WITH NONIONIC SURFACTANT TWEEN 60

Gorana G. Puača, Jelena V. Srbinov, Vesna B. Tepavčević, Zita J. Farkas-Agatić, Mihalj M. Poša 273

CHEMICAL COMPOSITION OF *CLINOPODIUM VULGARE* L. EXTRACTS AND THEIR ANTIOXIDANT AND CHOLINESTERASE INHIBITION POTENTIAL

Mejra Bektašević, Olivera Politeo, Ivana Carev 274

ENCAPSULATION OF B-CAROTENE FROM CARROT WASTE BY EXTRUSION TECHNIQUE

Vanja Šregelj, Steva Lević, Jelena Vulić, Ana Kalušević, Jasna Čanadanović-Brunet, Gordana Četković, Viktor Nedović, Vesna Tumbas Šaponjac 275

PHYTOCHEMICALS CONTENT AND BIOACTIVITY OF PEACH POMACE

Ksenija Bibovski, Vanja Šregelj, Jelena Vulić, Gordana Četković, Jasna Čanadanović-Brunet, Vesna Tumbas Šaponjac, Slađana Stajčić 276

COLD PRESSING VS. SUPERCRITICAL CO₂ EXTRACTION OF PLUM (*PRUNUS DOMESTICA* L.) KERNEL SEED

Jelena Vladić, Aleksandra Gavarić, Stela Jokić, Ljiljana Popović, Ana Matias, Naiara Fernández Hernández, Alexandre Agostinho, Senka Vidović 277



SPRAY DRYING OF LIQUID EXTRACT OBTAINED FROM *ARCTOSTAPHYLOS UVA-URSI* HERBAL DUST

Abdulahakim B. Naffati, Senka Vidović, Jelena Vladić..... 278

EFFECT OF DIFFERENT SPRAY DRYING CONDITIONS ON MICROMETRIC AND STRUCTURAL CHARACTERISTICS OF BRINE OBTAINED FROM CABBAGE FERMENTATION

Senka Vidović, Anita Vakula, Rita Ambrus, Jelena Vladić, Gabor Katona, Nataša Nastić 279

PETROLEUM REFINING AND PRODUCTION 281

SYNTHESIS OF COLD FLOW ADDITIVES TO ECOLOGICAL PURE DIESEL FUEL BY METHODS OF CONTROLLED RADICAL POLYMERIZATION

Dmitry F. Grishin, Elena V. Kolyakina, Marina V. Pavlovskaya 282

PETROLEUM REFINING IN SERBIA: INVESTMENTS, DEVELOPMENT PROJECTS AND ACTIVITIES

Dragan Govedarica 283

DEMULSIFICATION OF WATER-IN-CRUDE OIL EMULSIONS USING NEURAL NETWORKS

Hassan Al-Homigany, Mirjana Bulić, Anatoly Zolotukhin, Slavko Nešić, Vlada Streletskaya, Predrag Kojić, Dragan Govedarica 284

EFFECT OF EXTENDER OIL NATURE ON THE RUBBER CROSSLINK DENSITY

Julijana Blagojević, Predrag Kojić, Jelena Pavličević, Olga Govedarica, Mirjana Jovičić, Oskar Bera, Dragan Govedarica..... 285

AN OVERVIEW OF ARTIFICIAL LIFT SELECTION METHODS WITH CASE OF SELECTION USING MULTI-CRITERIA DECISION-MAKING METHODS

Miroslav P. Crnogorac, Dušan Š. Danilović, Vesna D. Karović Maričić, Lola D. Tomić..... 286

RENEWABLE JET FUEL PRODUCTION FROM VEGETABLE OIL: ASPEN SIMULATION AND TECHNO-ECONOMIC STUDY

Tijana Čarapić, Sandra B. Glišić 287

SIMULATION AND MODELLING OF AN INDUSTRIAL HYDROTREATING PROCESS USING VACUUM AND LIGHT GAS OIL - IMPACT OF CATALYST WETTING EFFICIENCY AND HYDRODEAROMATISATION REACTION KINETIC'S TYPE

Sandra B. Glišić, Aleksandar M. Orlović..... 288



SYNTHESIS OF Re/Pd AND Co/Mo HETEROGENEOUS CATALYSTS ON MESOPOROUS SILICA USING SOL-GEL METHOD FOLLOWED BY SUPERCRITICAL DRYING WITH EXCESS SOLVENT	
Dragana Prokić-Vidojević, <u>Sandra B. Glišić</u> , Aleksandar M. Orlović.....	289
CRUDE OIL MONITORING CHALLENGES IN MODERN REFINERY	
<u>Milka Jelić</u> , Marija Stepanović.....	290
TESTING OF EXPLOITATION PROPERTIES OF HYPOID OIL	
<u>Aleksandar Kekić</u> , Milana Đuričić, Milana Berber, Predrag Kojić, Julijana Blagojević, Dragan Govedarica.....	291
NEW APPROACHES TO THE THIN-LAYER CRACKING OF ACID TARS	
<u>Alexander V. Knyazev</u> , Valentina F. Zanozina, Anastasia S. Shipilova, Svetlana S. Knyazeva.....	292
STRUCTURE-BASED MODELS FOR THE PREDICTION OF FLASH POINT OF MULTI-COMPONENT FUEL MIXTURES	
Ali Younes Nagi Mosbah, <u>Predrag Kojić</u> , Oskar Bera, Olga Govedarica, Radovan Omorjan, Dragan Govedarica.....	293
ADVANCED OIL AND GAS PRODUCTION AND PROCESSING TECHNOLOGIES FOR ARCTIC CONDITIONS	
<u>Slavko Nešić</u> , Dragan Govedarica, Konstantin Pivovarov, Anatoly Zolotukhin, Vlada Streletskaya	294
ASPEN HYSYS SIMULATION OF NATURAL GAS DEHYDRATION PROCESS	
<u>Branislava G. Nikolovski</u> , Svetlana R. Kuzminac, Nevena B. Blagojević, Jelena D. Bajac, Biljana Miljković, Spasojević Đ. Momčilo, Milan N. Sovilj.....	295
SUSTAINABLE DEVELOPMENT, CHEMICAL AND ENVIRONMENTAL ENGINEERING.....	297
LAST ADVANCES ON THE LARGE SCALE PRODUCTION OF BIOACTIVE COMPOUNDS FOR AGRICULTURE AND AQUACULTURE FROM MICROALGAE	
<u>Fransisco G. Acién Fernández</u> , Cintia Gómez Serrano, Jose M. Fernández Sevilla, Emilio Molina Grima	298
BIOLOGICAL CONVERSION OF GREENHOUSE GASES INTO ADDED VALUE BIO-PRODUCTS: MOVING TOWARDS GHG BIOREFINERIES	
Raúl Muñoz	299



TINY MICROALGAE FOR HUGE APPLICATIONS. BIOREFINERY APPROACH	
Alice Ferreira, Sofia Carapinha, Jelena Vladic, Senka Vidovic, Dragoljub Cvetkovic, Lusine Melkonyan, Gayane Avetisova, <u>Luisa Gouveia</u>	300
THERMODYNAMICS STUDIES OF COPPER AND CHROMIUM(VI) IONS ONTO SUGAR BEET SHREDS AS ADSORBENTS	
<u>Dragana V. Kukić</u> , Marina B. Šćiban, Vesna V. Vasić, Jelena M. Prodanović	302
SUPERCRITICAL CO ₂ EXTRACTION OF TOBACCO WASTE	
<u>Marija Banožić</u> , Tanja Gagić, Željko Knez, Mojca Škerget, Stela Jokić	303
THERMOCHEMISTRY ASPECTS OF MECHANOCHEMISTRY ACTIVATION OF THE FLOTATION PROCESSES	
Milan M. Petrov.....	304
HYDRODYNAMICS OF ROTATING DISC CONTACTOR EXTRACTION COLUMNS	
<u>Milan N. Sovilj</u> , Momčilo Đ. Spasojević, Branislava G. Nikolovski	305
OXIDATIVE DEGRADATION OF CYAN FLEXY DYE WITH HETEROGENEOUS FENTON REAGENT - Fe ₂ (MoO ₄) ₃ PARTICLE	
<u>Vesna S. Gvoić</u> , Miljana Đ. Prica, Đurđa V. Kerkez, Milena Đ. Bečelić-Tomin, Aleksandra Z. Kulić, Anita S. Leovac Maćerak, Božo D. Dalmacija.....	307
PRELIMINARY STUDY OF CHROMIUM(VI) IONS ADSORPTION ON BOEHMITE	
<u>Dragana V. Kukić</u> , Vesna V. Vasić, Marija M. Milanović, Ivan Lj. Stijepović, Marina B. Šćiban, Jelena M. Prodanović	308
REMOVAL AND RECOVERY OF AMMONIUM FROM MILKING PARLOUR WASTEWATER USING POMEGRANATE PEEL	
<u>Naoufal Bellahsen</u> , Virág Nagypál, Edit Mikó, Zita Šereš, Szabolcs Kertész, Cecilia Hodúr.....	309
USING ERA5 DATA FOR ESTIMATION OF THE WIND ENERGY POTENTIAL IN SERBIA	
Zorica M. Podrascanin.....	311
OPTIMIZATION OF STARCH SUSPENSION MICROFILTRATION PROCESS – ANN APPROACH	
<u>Bojana B. Ikonić</u> , Jelena M. Pavličević, Oskar J. Bera, Aleksandar I. Jokić, Predrag S. Kojić, Arpad I. Kiralj, Nataša Lj. Lukić.....	312



VISCOSITY AND pH DETERMINATION OF CHOLINE CHLORIDE BASED DEEP EUTECTIC SOLVENTS CONTAINING AMIDES	
<u>Martina Jakovljević</u> , Melita Lončarić, Mario Komar, Maja Molnar.....	313
AFB1 AND FUMONISIN B1 ADSORPTION AND DESORPTION USING ZEOLITES	
Hunor Farkaš, <u>Jog Raj</u> , Robert Čepela, Zdenka Jakovčević, Jasna Bošnjak-Neumüller, Marko Vasiljević	314
DANUBE SEDIMENT CONTAMINATION WITH POLYCHLORINATED BIPHENYLS: NEW INTERPRETATION OF SEDIMENT QUALITY ASSESSMENT	
<u>Maja Brborić</u> , Borivoj Stepanov, Jelena Radonić, Maja Turk Sekulić	316
THE USE OF GREEN SOLVENTS IN A PRE-TREATMENT OF LIPIDS EXTRACTION FROM GREEN MICROALGAE <i>CHLORELLA</i> SP.	
Natalija G. Đorđević, Bojana R. Danilović, Vlada B. Veljković, <u>Dragiša S. Savić</u>	317
ECONOMIC FEASIBILITY OF AIR SOURCE HEAT PUMPS FOR RESIDENTIAL BUILDINGS IN VOJVODINA	
<u>Arpad I. Kiralj</u> , Aleksandar I. Jokić, Nataša Lj. Lukić, Jovana A. Grahovac, Bojana B. Ikonić, Jelena M. Pavličević, Svetlana S. Popović	318
DETERMINATION OF THE TOTAL ANTIOXIDANT CAPACITY OF BLACK MUSTARD SEEDS (<i>BRASSICA NIGRA</i>) EXTRACTS OBTAINED BY DIFFERENT DEEP EUTECTIC SOLVENTS	
<u>Biljana S. Đorđević</u> , Zoran B. Todorović, Ivica G. Đalović, Dragan Z. Troter, Petar M. Mitrović, Vlada B. Veljković	319
KINETICS OF THE EPOXIDATION OF LINSEED OIL WITH IN SITU FORMED PERACETIC ACID	
<u>Olga M. Govedarica</u> , Milovan R. Janković, Snežana V. Sinadinović-Fišer, Nevena R. Vukić, Ljiljana M. Tanasić, Vesna B. Teofilović, Ljiljana S. Korugic-Karasz	321
MITIGATION OF MEMBRANE FOULING BY APPLICATION OF TURBULENCE PROMOTERS	
Svetlana S. Popović	322
ECOLOGICAL RISK MANAGEMENT WITH INTEGRATED MULTITROPIC AQUACULTURE APPROACH FOR MARINE ENERGY PROJECTS IN INDIA	
Kapilkumar Nivrutti Ingle	323



COMPARISON OF EXTRACTION AGENTS FOR METAL DETERMINATION IN SEDIMENTS FROM ARTIFICIAL LAKES AND RIVERS IN SERBIA	
<u>Katarina Pantović Spajić</u> , Dragana Đorđević, Sanja Sakan, Tatjana Šošćarić, Zorica Lopićić, Aleksandra Janićijević, Ksenija Stojanović	325
PRODUCTION OF L (+) LACTIC ACID FROM CORN HUSK HYDROLYZATE	
<u>Dragana Mladenović</u> , Aleksandra Djukić-Vuković, Jelena Pejcin, Sunćica Kocić-Tanackov, Ljiljana Mojović	326
ASSESSMENT OF THE IMPACT OF WASTE WATER DISPOSAL ON THE BEGEJ	
<u>Vesna Ź. Pešić</u> , Milena R. Bećelić-Tomin, Đurđa V. Kerkez, Dejan M. Krćmar, Božo D. Dalmacija, Snežana P. Maletić	327
FUEL POTENTIAL AND PROPERTIES OF GRAPE POMACE HYDROCHAR	
<u>Jelena T. Petrović</u> , Marija L. Mihajlović, Marija S. Petrović, Marija M. Kojić, Marija R. Koprivica, Tatjana Šošćarić, Leposava Filipović-Petrović.....	328
GREEN ENERGY PRODUCTION AT THE WASTEWATER TREATMENT PLANT IN SUBOTICA	
<u>Biljana Isić</u>	330
INFLUENCE OF KENIX STATIC MIXER ON THE FLUX IMPROVEMENT IN THE MICROFILTRATION OF INDUSTRIAL WASTEWATER	
<u>Maja I. Jovović</u> , Aleksandar I. Jokić, Svetlana S. Popović, Zita I. Šereš, Biljana S. Pajin, Bojana B. Ikonić, Nataša Lj. Lukić	331
A NEW APPROACH FOR MODELLING AND OPTIMIZATION OF RUBBER CURING PROCESS	
<u>Jelena D. Lubura</u> , Predrag S. Kojić, Dragan D. Govedarica, Jelena M. Pavlićević, Bojana B. Ikonić, Mirjana C. Jovićić, Oskar J. Bera.....	332
STATISTICAL OPTIMIZATION OF THE CORN GERM OIL METHANOLYSIS CATALYZED BY POTASSIUM HYDROXIDE	
<u>Olivera S. Stamenković</u> , Petar M. Mitrović, Ivica G. Đalović, Milan D. Kostić, Vlada B. Veljković	333
KINETIC MODELING OF CORN GERM OIL METHANOLYSIS USING CORN COBS ASH AS A CATALYST	
<u>Milan D. Kostić</u> , Petar M. Mitrović, Ivica G. Đalović, Olivera S. Stamenković, Vlada B. Veljković	335



FATTY ACID METHYL ESTER SYNTHESIS FROM OIL BLENDS

Marija R. Miladinović, Ivana B. Banković-Ilić, Olivera S. Stamenković, Ivica G. Djalović, Petar M. Mitrović, Vlada B. Veljković 336

COMBINATION OF ALKALINE/ACIDIC AND MICROWAVE TREATMENT FOR ENHANCED ANAEROBIC DIGESTION OF SLUDGE

Zoltán Jákó, Sándor Beszédes, Balázs Lemmer, Andrea Vágvölgyi, Cecilia Hodúr.... 337

ASEPTIC APPLE (*MALUS DOMESTICA* BORKH.) AND SOUR CHERRY (*PRUNUS CERASUS* L.) CHANGE IN THE CONTENT OF THE INGREDIENTS DURING STORAGE

Ribárszki Ákos, Furulyás Diána, Rentsendavaa Chagnaadorj, Stéger-Máté Mónika 339

MAJOR ENVIRONMENTAL LAW ASPECTS OF SUSTAINABLE DEVELOPMENT

Vladan Joldžić 340

ACTUAL AND REQUIRED WATER QUALITY IN ACCORDANCE WITH THE EU WFD ON THE DANUBE RIVER IN SERBIA

Ljiljana M. Takić, Ivana I. Mladenović Ranisavljević, Dejan M. Vasović, Nenad V. Živković..... 341

ENHANCEMENT AND COMPARISON OF SUBMERGED AND SOLIDSTATE AMYLASE PRODUCTION FROM WHEAT CHAFF BY STATISTICAL OPTIMIZATION

Damjan G. Vučurović, Mirjana I. Jovanović, Bojana Ž Bajić, Siniša N. Dodić..... 342

ADSORPTION OF Cu(II) IONS FROM SYNTHETIC SOLUTION BY SUNFLOWER SEED HUSKS

Srđan Stanković, Tatjana Šoštarić, Mladen Bugarčić, Aleksandra Janićijević, Katarina Pantović-Spajić, Zorica Lopičić 344

INVESTIGATION OF PHOTOCATALYTIC PROPERTIES OF ZnFe-MIXED OXIDES

Milica S. Hadnadjev-Kostić, Tatjana J. Vulić, Jasmina M. Dostanić 345

REDUCTION OF FOULING IN DAIRY WASTEWATER ULTRAFILTRATION PROCESS

Szabolcs Kertész, Nikola Maravić, Szabolcs Gyula Szerencsés, Gábor Veréb, Sándor Beszédes, Zsuzsanna László, Cecilia Hodúr 346

EFFECTS OF COMMERCIAL ANTIOXIDANT FORMULATIONS ON OXIDATIVE STABILITY OF FATTY ACID METHYL ESTERS FROM DIFFERENT RAW MATERIALS

Milica Rankov Šicar, Radoslav Mičić, Milan Tomić, Nataša Đurišić-Mladenović..... 348



HEMP BIODIESEL PRODUCTION: ANALYSIS OF MATERIAL AND ENERGY CONSUMPTION ON INDUSTRIAL SCALE

Marija B. Tasic, Milan D. Kostic, Olivera S. Stamenkovic, Vlada B. Veljkovic 349

PURIFICATION OF DAIRY WASTEWATER COMBINING MEMBRANE FILTRATION WITH OZONE AND FENTON PRETREATMENTS

Mihály Zakar, Zita Šereš, Erika Hanczné-Lakatos, Gábor Keszthelyi-Szabó, Zsuzsanna László 350

Keywords: membrane filtration, ozone, Fenton 351

CORROSION BEHAVIOUR OF STAINLESS STEEL EN 1.4301 IN 0.5 M HCl IN PRESENCE OF COMMERCIAL INHIBITOR

Borislav Malinović, Tijana Djuričić, Duško Zorić 351

CLOUD POINT EXTRACTION PROCEDURE FOR PRECONCENTRATION AND DETERMINATION OF LEAD(II) IONS USING TRITON X-100 AT ROOM TEMPERATURE

Mersiha Suljkanović, Jasmin Suljagić, Nusreta Hasić, Selmina Hodžić 353

THE EFFECT OF POROSITY AND SURFACE AREA OF CLINOPTILOLITE AND ZEOLITE ON THE ADSORPTION OF THE PRODUCTS OF THERMO-OXIDATIVE DEGRADATION OF EDIBLE SUNFLOWER OIL

Sanja M. Dobrnjac, Ljubica C. Vasiljević, Stevan N. Blagojević, Miladin J. Gligorić, Zoran B. Obrenović, Vesna P. Cvijetinović, Dragan V. Tošković, Željko J. Ostojić 354

INTEGRATION OF FIRST AND SECOND GENERATION BIOETHANOL PRODUCTION FROM BY-PRODUCTS OF SUGAR BEET PROCESSING

Jovana D. Gucunski, Bojana Ž. Bajić, Damjan G. Vučurović, Siniša N. Dodić 355

HOW RISK MANAGEMENT STANDARDS CAN CONTRIBUTE TO ACHIEVING SUSTAINABLE DEVELOPMENT GOALS

Tzvetelin K. Gueorguiev 357

DEVELOPMENT OF WILD CYANOBACTERIAL BIOMASS (*APHANIZOMENON FLOS-AQUAE*) BIOREFINING SCHEMES

Michail Syrpas, Jolita Bukauskaitė, Loreta Bašinskienė, Petras Rimantas Venskutonis 358

INVESTIGATION OF HUMIC ACIDS EVOLUTION DURING EX SITU BIOREMEDIATION OF PETROLEUM CONTAMINATED SOIL

Jelena Avdalović, Mila Ilić, Srđan Miletić, Vladimir Beškoski, Nikoleta Lugonja, Jelena Milić, Miroslav M. Vrvic 359

WHEAT SEEDLINGS UNDER THE SALINITY STRESS	
<u>Maja Ižaković</u> , Katica Teskera, Matea Begić, Tihana Marček	360
HOW LCA DAMPENED THE ENTHUSIASM OF FORMER BIOFUEL ADVOCATES?	
Ferenc E. Kiss.....	361
USE OF SEQUENTIAL EXTRACTION TO ASSESS METAL PARTITIONING IN FLOODED AGRICULTURAL SOIL	
<u>Biljana D. Škrbić</u> , Igor S. Antić, Maja B. Buljovčić, Jelena R. Živančev.....	363
REMOVAL OF NATURAL ORGANIC MATTER AND EMERGING CONTAMINANTS FROM GROUNDWATER USING OZONATION AND GAC FILTRATION	
J. Molnar Jazić, M. Kragulj Isakovski, A. Tubić, T. Apostolović, M. Watson, S. Maletić, <u>J. Agbaba</u>	364
BIOCATALYTIC BIODIESEL SYNTHESIS WITH <i>BURKHOLDERIA CEPACIA</i> AND <i>PSEUDOMONAS FLUORESCENS</i> LIPASE	
<u>Sandra Budžaki</u> , Marta Ostojčić, Ivica Strelec.....	365
SOLAR-LIGHT PHOTOCATALYTIC ACTIVITY OF ZnFe ₂ O ₄ NANOPARTICLES	
<u>Marija Milanović</u> , Milica Hadnađev-Kostić.....	367
MODELING OF ULTRAFILTRATION WITH DIFFERENT FOULING MECHANISMS	
Nikolett Gulyás, Zoltán Jákói, Virág Nagypál, <u>Cecilia Hodúr</u>	368

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