

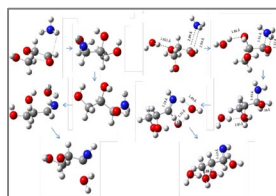
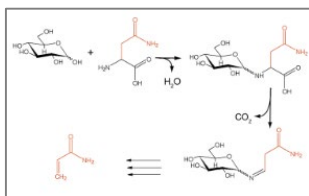
# BOOK OF ABSTRACTS

9<sup>th</sup> International conference  
**CHEMICAL REACTIONS IN  
FOODS IX  
(CRF 2023)**

September 13-15, 2023

Prague, Czech Republic

Jana Pulkrabová, Monika Tomaniová, Marco Arlorio, Vincenzo Fogliano,  
and Jana Hajšlová  
Editors



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9<sup>th</sup> International Conference on

# **CHEMICAL REACTIONS IN FOODS IX (CRF 2023)**

September 13-15, 2023  
Prague • Czech Republic

Organized by

**Department of Food Analysis and Nutrition,  
University of Chemistry and Technology, Prague (UCT Prague),  
Czech Republic**

**&**

**University of Piemonte Orientale, Italy (UPO)**

**&**

**Wageningen University & Research, The Netherlands (WUR)**



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and under auspices of the European Chemical Society (EuChemS), Division of Food Chemistry.*

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# **ORAL SESSIONS**

**L1****ON THE POSITIVE NEXUS BETWEEN FOOD (ULTRA)PROCESSING AND FOOD QUALITY****Vincenzo Fogliano**<sup>1\*</sup><sup>1</sup> Wageningen University, Wageningen, Netherlands

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Food processing is the means through which humankind, throughout its history, has treated raw materials to obtain more digestible and safer (e.g. to remove toxins) food products. Through the ages, humans have treated raw commodities they have hunted, gathered, grown or derived through domestication of animals to reduce spoilage and extend shelf life, improve their palatability and create safer food products. It is abundantly evident that human survival and evolution were unquestionably aided by the ability to process foods. The modern food industry also exploited two other obvious advantages of food processing: the mass production of long-shelf life products makes food affordable also to people with limited economic possibilities, and a striking reduction of perishable food waste was achieved. To further improving the sustainability of food processing is an ongoing challenge and the shift from a linear to a circular food production system is still at its infancy. In this presentation, some examples will be provided showing how playing with texture at macro-meso and micro-scale can be a smart way to use processing strategies to get foods having the quality characteristics desired by consumers.

**Keywords:** food processing, food quality, texture, consumer acceptance

**L2****111 YEARS OF THE MAILLARD REACTION - ANYTHING LEFT TO EXPLORE?****Thomas Henle**<sup>1\*</sup><sup>1</sup> Chair of Food Chemistry, Technische Universität Dresden, Dresden, Germany

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Chemical reactions occurring during heating or storage are of prime importance for food safety and quality. Among these, the Maillard reaction (also referred to as or nonenzymatic browning or "glycation") has been thoroughly investigated since the first paper published by Louis-Camille Maillard in 1912, primarily aiming to understand the impact of glycation on the flavour of processed foods. Furthermore, compounds such as acrylamide reached particular importance as "process contaminants". Despite extensive research, however, we still only partially understand the reaction process and related technological and physiological aspects. This applies in particular to the biological utilization of glycation products, whether in the human body or during biotechnological fermentation processes. In the presentation, the formation of individual glycation products during food and feed processing will be presented on the basis of individual case studies, a quantitative assessment of the daily uptake will be carried out and the relevance of related post-translational modifications for the functional properties of proteins will be explained. It will be shown that brewer's yeasts differ in their ability to detoxify dicarbonyl compounds via oxidative or reductive pathways, possibly as a result of selective domestication, and that the human intestinal microbiota can degrade Amadori products by deglycation processes and use them as a source of lysine. The overall picture is that even after decades of research, the Maillard reaction still raises numerous unresolved interdisciplinary questions, ranging from food quality to the unsolved mysteries of human evolution.

**Keywords:** Maillard reaction, biological utilization of glycation products, case studies



**L3**  
**CANCELLED**

**L4****INTERACTION OF FOOD CONTACT MATERIAL WITH PACKED FOOD: HOW TO EVALUATE THE PRODUCT SAFETY****Erich Leitner**<sup>1\*</sup><sup>1</sup> Graz University of Technology, Institute of Analytical Chemistry and Food Chemistry, Graz, Austria

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The global food packaging market size was valued at USD 362.9 billion in 2022 and is expected to expand at a compound annual growth rate of 5.7% from 2023 to 2030 [1]. There are different types of packaging materials on the market, one of the largest groups are various polymer materials ("plastic"). Approximately 40% of virgin polymers and 50% of produced paper are used for packaging applications. In the light of the ongoing discussions about global pollution there is a huge pressure on industry for the development of new packaging material. Also, in accordance with the European Strategy it is planned preventing packaging waste, boosting reuse and refill, and making all packaging recyclable by 2030 [2]. A new commission regulation released in September 2022 deals with the use of recycled polymers as food contact materials [3]. Paragraph 2 in this document clearly mentions that "a pre-requisite to any increase in recycled content in food packaging and other food contact materials remains the need to secure a high level of protection of human health." So, there is a high demand to develop and use different methods to proof the safety of these materials. In this presentation strategies for safety evaluation by various analytical techniques will be discussed.

[1] <https://www.grandviewresearch.com/industry-analysis/food-packaging-market>[2] [https://ec.europa.eu/commission/presscorner/detail/en/ip\\_22\\_7155](https://ec.europa.eu/commission/presscorner/detail/en/ip_22_7155)[3] <https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32022R1616>**Keywords:** food contact materials, food safety, analytical techniques

**L5****PHENOLIC COMPOUNDS ORIGINATED FROM EDIBLE PLANTS STRONGLY INHIBITING PANCREATIC LIPASE****Aristeidis Tsagkaris<sup>1\*</sup>, Anna Louckova<sup>2</sup>, Jana Hajslova<sup>3</sup>**

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According to the latest information by the World Health Organization (WHO, <https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight>, last accessed 30/6/2023), 39% of adults and 18% of children and adolescents are overweight or obese, which is a major risk factor for a metabolic syndrome development. This highlights the urgent need to investigate strategies to control obesity. Among the available approaches [1], utilizing natural bioactive compounds such as polyphenols to inhibit the activity of pancreatic lipase (PNLIP) (EC 3.1.1.3) has been promising [2]. PNLIP hydrolyses triacylglycerols to monoacylglycerols and free fatty acids in a high rate, up to 70%. Considering that dietary fats are mostly constituted of triacylglycerols and their contribution to energy intake, inhibiting PNLIP may be an effective strategy in reducing the synthesis of adipose tissue preventing from excessive fat deposition [3]. Importantly, PNLIP inhibitors have been identified in edible plants widely consumed around the globe indicating potential candidates for the production of "nutraceuticals". Nutraceuticals encompass a wide range of dietary sources including dietary supplements and processed foods with a high abundance in phenolic bioactive compounds with reported antioxidant, cardiovascular protective, anti-inflammatory properties and PNLIP inhibitory effects [4]. In this study, to identify edible sources with potential nutraceutical applications, 145 dietary plants were screened in terms of their in vitro PNLIP inhibitory effect. Both aqueous and non-polar extracts were tested, aiming to isolate different bioactive compounds in each occasion. After performing a high throughput screening of these crude extracts, a cut-off inhibitory level was set (70%) and only extracts exceeding this level were further analysed using an ultra-high-performance liquid chromatography hybrid quadrupole time-of-flight mass spectrometry (UHPLC-q-TOF-MS) system. A metabolomic workflow based on suspect screening was applied and bioactive compounds were tentatively identified highlighting their potential as PNLIP inhibitors.

[1] Müller TD, Blüher M, Tschöp MH, DiMarchi RD (2022) Anti-obesity drug discovery: advances and challenges. *Nat Rev Drug Discov* 21:201-223. <https://doi.org/10.1038/s41573-021-00337-8>

[2] Kumar A, Chauhan S (2021) Pancreatic lipase inhibitors: The road voyaged and successes. *Life Sci* 271:119115. <https://doi.org/https://doi.org/10.1016/j.lfs.2021.119115>

[3] Liu T-T, Liu X-T, Chen Q-X, Shi Y (2020) Lipase Inhibitors for Obesity: A Review. *Biomed Pharmacother* 128:110314. <https://doi.org/https://doi.org/10.1016/j.biopha.2020.110314>

[4] Ammendola S, Scotto d'Abusco A (2022) Nutraceuticals and the Network of Obesity Modulators. *Nutrients* 14:5099

**Keywords:** obesity, enzyme inhibition, bioactivity, phenolic compounds, high throughput screening

**Acknowledgement:** This work was supported by METROFOOD-CZ research infrastructure project (MEYS Grant No: LM2023064) including access to its facilities.

**L6****‘SMART FOODS’ DESIGNING: CHALLENGES FOR INTERDISCIPLINARY RESEARCH**

Jana Hajslova<sup>1\*</sup>, Jana Pulkrabova<sup>2</sup>, Monika Tomaniova<sup>3</sup>, Milena Stranska<sup>4</sup>

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The critical analysis of current research challenges aiming at further increase of the quality of future generations' life, clearly shows the importance of food quality, not only with regards to safety issues, but also taking into account their nutritional profile. In this context, the scientifically justified concept of the production of 'smart' foods is highly promising. The new, innovative strategy, is demonstrated on several concrete examples. Interdisciplinary research activities are based on: (i) utilization of the potential of modern sustainable agriculture for production of premium raw materials; (ii) application of modern, mild (bio)technologies to produce sensorially attractive 'smart' foods with the required profiles of limiting nutrients and rich in other bioactive substances; (iii) critical evaluation of the expected (positive) health effects as a result of consuming an appropriate daily dose of 'smart' foods through a clinical intervention study.

**Keywords:** smart food, premium food crops, mild food processing technologies, health benefits

**L7****PEPTIDE FORMATION IN FOOD: IMPLICATIONS FOR QUALITY, BIOACTIVITY AND AUTHENTICITY CONTROL****Monika Pischetsrieder**<sup>1\*</sup>

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During food production, a wide variety of peptides are formed by partial enzymatic hydrolysis of food proteins. Proteolytic enzymes are either endogenously present in the food or externally added during fermentation or hydrolysis processes. More than 250 peptides have been identified for example in raw milk, which are released from caseins by endogenous milk proteases, such as plasmin or cathepsins. Heating and storage further changes the peptide profile of milk either by residual proteolytic activity or by thermal protein cleavage. The number and variety of peptides strongly increase during the fermentation of milk, mainly by the activity of proteases secreted by starter cultures. In hypoallergenic milk products, milk proteins are completely converted into peptides by the addition of technological enzymes to whey products. Apart from milk and milk products, a high variety and quantity of peptides occur in many other animal and plant based food products. The advancement of various mass spectrometry methods, such as MALDI-TOF-MS and particularly high resolution LC-MS/MS allows the recording of comprehensive peptide profiles of various foods, which are composed of several hundreds to thousand components. Based on these peptide fingerprints, it is now possible to predict and describe their bioactivity and their impact on technological properties. Of particular importance is the antimicrobial activity of peptides. Depending on their primary structure, peptides can form amphiphilic helical conformations with a positive net charge, which can specifically interfere with bacterial cell membranes. Because of this known structure-activity relation, virtual screening selected Leg1 and Leg2 from enzymatic hydrolysates of chickpea storage proteins. Leg1 and Leg2 show strong antibacterial activity against several food spoilage bacteria and food pathogens including a multi resistant strain. Additionally, antifungal and antioxidative side activity of both chickpea peptides was observed. Since they are derived from a traditional food source, Leg1 and Leg2 can be considered as natural and safe food preservatives. Another important application of food peptide profiles is the control of food fraud. The peptide profile of a food sample is influenced by various internal and external parameters so that it is a fingerprint of the sample reflecting many aspects of authenticity. Thus, peptide markers have been identified, which can determine the species, heating conditions, or storage of milk or the identity of a fermented milk product. Finally, peptides influence, among others the emulsifying, foaming, and gelling properties as well as the taste of a food product and form complexes with other food ingredients, such as starch or polyphenols. Therefore, comprehensive peptide profiles are important to monitor the quality of the industrial production of hydrolyzed protein products.

**Keywords:** protein hydrolysis, peptides, fingerprints, markers, bioactivity

**L8**  
**CANCELLED**

## L9

# EFFECT OF FLUIDIZED BED ROASTING ON COCOA CHARACTERISTICS

Ruth Fabiola Peña-Correa<sup>1</sup>, Burçe Ataç Mogol<sup>2\*</sup>, Christos Frygas<sup>3</sup>, Martinus A.J.S. van Boekel<sup>4</sup>, Vincenzo Fogliano<sup>5</sup>

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Roasted cocoa nibs are the key raw material of chocolate manufacturing. Roasting benefits the proper processing of cocoa due to a decrease of water content. In addition, the Maillard reaction occurring during roasting, leads to the desired aroma formation, which contributes to the chocolate characteristics. Cocoa nibs are conventionally roasted (110-150°C, up to 2 hours) until the water content decreases to 2%. However, alternative roasting techniques with high convective heat transfer, such as fluidized bed roasting, could shorten the roasting time. The study evaluates the impact of two different roasting techniques, namely Slow Roasting (SR) in a conventional oven and Fast Roasting (FR) in a fluidized bed roaster, on cocoa's characteristics. For both roasting techniques, cocoa beans were roasted at 110, 120, 130, and 140°C for various times. Measurements of water content indicated that FR-140°C (fluidized bed roasting) was 16 times faster than SR-140°C (convective oven roasting). The color analysis of the cocoa nibs showed that the b\* value was higher in SR-nibs compared to FR-nibs. Fructose, glucose, and sucrose contents decreased by roasting, and the decrease in fructose was more prominent than others. The content of free amino acids was significantly decreased by both roasting techniques, showing that temperature had a greater effect than roasting technique. Among the volatile organic compounds analyzed, ketones, alcohols, esters, and sulfur compounds were found to be higher in SR-cocoa nibs. Certain aldehydes and pyrazines were found to be higher in FR-nibs, which contribute to the characteristic aroma of chocolate. In conclusion, this study showed that fluidized bed roasting could shorten the roasting time of cocoa nibs while favoring the formation of desired chocolate aroma compounds.

**Keywords:** cocoa, roasting, Maillard reaction

**Acknowledgement:** This work was supported by ICETEX (Instituto Colombiano de Crédito y Estudios Técnicos en el Exterior) under the program "Pasaporte a la Ciencia Doctorado", grant number 3674604.

## L10

# EVALUATING THE IMPACT OF REFRIGERATION AND CELLULOSE MOLECULAR FILTRATION ON EXTRA VIRGIN OLIVE OIL COMPOSITION DURING STORAGE: A COMPREHENSIVE 2-YEAR STUDY

Konstantina Lazarou<sup>1</sup>, Aristeidis Tsagkaris<sup>2</sup>, Sofia Drakopoulou<sup>3</sup>, Antonis Kiriakopoulos<sup>4</sup>, Charalampos Proestos<sup>5</sup>, Nikolaos Thomaidis<sup>6</sup>, Marilena Dasenaki<sup>7\*</sup>

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Extra virgin olive oil (EVOO) features unique nutritional value due to its composition, resulting in a product of high market share. However, EVOO is susceptible to compositional changes during storage leading to significant decrease of its quality [1]. It is therefore an imperative need to identify the most effective storage conditions that preserve its composition along with its distinctive sensory features. In this study, a thorough investigation was performed on the stability of "Kolovi" EVOO, a variety originating from Lesvos Greek island, under different storage conditions over a 24-month period. The impact of a patented molecular filtration technique on EVOO stability was also evaluated. Physicochemical characteristics, sensory profile, bioactive content, pigments and squalene, as well as fatty acid profile and diglycerides were monitored, providing a comprehensive overview of EVOO compositional alterations during storage. The positive impact of refrigeration was observed and most of the measured parameters were preserved stable for a longer period when EVOO samples were stored at 4 °C. Importantly, even better results were acquired by combining refrigeration and molecular filtration of EVOOs, accounting for certain characteristics improvement of stability, e.g., acidity value or lutein content. On the downside, neither the applied storage conditions nor molecular filtration were able to induce the stability of 4 monitored parameters, namely the K232 index, the monounsaturated fatty acids to polyunsaturated fatty acids ratio (MUFAs/PUFAs) and the tocopherol content. Overall, the presented study provides a temporal compositional profile of Kolovi EVOO and highlights the effect of different storage conditions. The great effect of refrigeration and molecular filtration in EVOO characteristics could provide a valuable insight to the establishment of a proper control storage which is crucial to maintain for longer EVOO quality and to enhance its commercialization.

[1] Jimenez-Lopez C, Carpena M, Lourenço-Lopes C, Gallardo-Gomez M, Lorenzo JM, Barba FJ, Prieto MA, and Simal-Gandara J. *Foods* MDPI; 9:1014 (2020).

**Keywords:** olive oil stability, molecular filtration, refrigeration, quality parameters, bioactive content



**L11****ADEQUATE POSTHARVEST-TREATMENT OF CLIMACTERIC FRUITS IS ESSENTIAL FOR THE FLAVOUR FORMATION OF THE FRUITS****Barbara Siegmund<sup>1\*</sup>, Thomas Lehner<sup>2</sup>, Niklas Pontesegger<sup>3</sup>**<sup>1 2 3</sup> Graz University of Technology, Graz, Austria

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Many fruit and vegetable varieties that can be found on the supermarket shelves throughout the whole year belong to the group of climacteric fruits and vegetables. Apples, bananas and mangoes are prominent representatives of this group. In contrast to non-climacteric fruits, climacteric fruits show postharvest-ripening. This behaviour allows to harvest these fruits in a mature stage before the onset of the so-called climacteric rise where the respiration rate significantly increases and induces the ripening processes. Postharvest-storage under appropriate conditions allows to significantly decelerate the ripening processes and to induce the postharvest ripening processes at a later stage. This behaviour enables us to bring fruits of high quality onto the market even months after the harvest. Even though these techniques have been applied and continuously refined for many years, it is of great importance to understand the specific requirements of different cultivars. During on-tree ripening, postharvest- or poststorage-ripening, the formation of fruit flavour is induced by the formation and/or activation of enzyme systems such as LOX, ADH and ACC for apples. Furthermore, for fruits such as mangoes - whose flavour is dominated by terpenes - the expression of terpene synthases plays an additional important role. In this contribution we demonstrate that a deep understanding of the biochemical reactions in the fruits are required to be able to bring fruits of high quality on the market. At the example of Crimson Crisp apples - which is a new and scab-resistant apple variety - we show that postharvest storage under different conditions (i.e., storage under regular air with temperature control, CA storage as well as CA storage with additional MCP application) has significant impact on the postharvest ripening behaviour of Crimson Crisp apples as the applied conditions significantly impact the biochemical reactions in the apples [1]. The results also show clearly that experiences from other apple varieties may not be directly transferred to new apple varieties. At the example of mangoes, we demonstrate that the arrangement of the fruits in the postharvest ripening chambers may impact the ventilation and, as a consequence, the respiration of the fruits. Dense fruit arrangement - followed by insufficient oxygen supply to the fruits - leads to a degradation of terpenes, a reduced formation of reaction products from the lipoxygenase pathway and less pronounced fruitiness and mango flavour [2].

[1] N. Pontesegger, T. Rühmer & B. Siegmund, *foods*, 2023, <https://doi.org/10.3390/foods12091876>[2] T. Lehner & B. Siegmund, *Food Chemistry*, 2002, <https://doi.org/10.1016/j.foodchem.2020.126608>**Keywords:** climacteric fruits, flavour formation, postharvest storage, postharvest ripening, apples & mangoes**Acknowledgement:** The authors would like to thank Agnello GmbH, Graz and the Research Centre for Fruit Growing & Viticulture Haidegg, Graz for the good cooperation and for providing the samples and storage facilities

## L12

### HYPERSPECTRAL IMAGING AND MACHINE LEARNING FOR ASSESSING LETTUCE QUALITY

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Food composition is typically analysed with chemical measurements, to ensure quality of the food and safety of the customers. However, these measurements are time-consuming and costly. We study the feasibility of building non-invasive and cost-efficient methods for quality assessment by using hyperspectral imaging. Spectral imaging has previously been successfully used with targeted chemical measurements to estimate the freshness and chemical composition of the food we eat. The broad scope of our project is to combine hyperspectral imaging, machine learning, analytical chemistry, plant biology and food science to assess the quality of fresh-cut vegetables. One of the aims of the project is to develop well-working non-invasive methods in analyzing the chemical composition of fresh-cut lettuce and extend their shelf life. To this end we are developing hyperspectral imaging platform and machine learning methodology to accurately combine the biological and chemical measurements and the hyperspectral data. The general idea of hyperspectral imaging is to combine spectroscopy to computer vision: the idea is to measure the reflected or transmitted light spectrum of each pixel in a photograph. This allows for greater accuracy in determining the color changes of the object. The hyperspectral camera used in this study, Specim IQ, also measures short wave infrared, which allows us to see phenomena not visible with the naked eye. The hyperspectral imaging platform we have developed is based on Specim IQ hyperspectral pushbroom camera, Effilux hyperspectral led lights, and optical board. The platform is relatively low-cost, mobile and takes approximately 0.17 m<sup>2</sup> of table space when functional. By using a handsewn light cover, we can take the hyperspectral images in relatively light environment, which makes our setup more mobile. By adding a conveyor belt, the setup can be fully automated, though better cameras for such setup exist, e.g., Specim FX10. In machine learning approaches, we are looking at semi- or self-supervised machine learning and deep learning methods and representation learning, most notably transformer and self-attention networks, to estimate the relevant parameters of the imaged lettuce sample. By using a proxy task, such as predicting the control group the sample belongs or the time point the image is taken, we learn a representation of the lettuce, which can be transferred to estimate e.g., chlorophyll concentration, sugar concentration or amount of browning or pinking. The presentation will cover the imaging platform, the machine learning methodology and empirical results on ice lettuce.

**Keywords:** hyperspectral imaging, food quality control, machine learning, lettuce

**L13****INFLUENCE OF CHIA SEEDS ON THE FORMATION OF ACRYLAMIDE IN BISCUITS**

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Acrylamide is formed as a process contaminant during the Maillard reaction when the free amino acid asparagine reacts with reducing sugars at high temperatures and low water activity [1]. The popularity of bakery products with added chia seeds (*Salvia hispanica*) has steadily increased in the recent years due to their attributed nutritional properties [2]. Chia seeds are classified as novel food and their use in foodstuffs is limited in quantity. According to reg (EU) 2020/24, e.g. a maximum of 10% whole chia seeds may be used in baked products. This is based a.o. on evidence that the addition of chia seeds can cause increased acrylamide levels in foods [3]. In a study by Mesias et al. partial replacement of wheat flour with chia flour resulted in increased formation of acrylamide in biscuits compared to the control sample [4]. In contrast, Galluzzo et al. did not find any significant difference in the acrylamide content of breads with and without different proportions of chia seeds [5]. Hence the aim of our studies was to investigate the influence of chia seeds in different processing stages (whole seeds, ground, roasted) and in interaction with other ingredients such as baking agents or different sugars as well as acrylamide influencing factors like temperature or time. Furthermore, the content of free asparagine in various chia seed batches was determined. It ranged between 400 and 550 mg/kg [6]. In our baking experiments, the processing state of the chia seeds was shown to be decisive for acrylamide formation. Whereas whole chia seeds seem to have no influence on the formation of acrylamide or rather reduce it, the addition of ground chia seeds led to a significant increase in the acrylamide content. In biscuits containing 15% ground chia seeds we found about three times as much acrylamide as in the control samples. Other ingredients such as ammonium bicarbonate showed the well-known effect of increased acrylamide formation also in combination with chia seeds. In contrast, citric acid significantly reduced the formation of acrylamide in the presence of chia seeds, too [6]. The analysis of bread and biscuit samples containing chia seeds from different retail stores showed that only one out of 26 samples exceeded the respective acrylamide benchmark level. Flaxseed flour is increasingly used as a food ingredient, too, e.g. as a substitute for egg yolk. As flaxseeds have a very similar nutrient composition to chia seeds, we now investigate their influence on acrylamide formation. Initial studies show that replacing conventional flour with flaxseed flour promotes the formation of acrylamide in a similar way as chia seeds do.

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**Keywords:** process contaminants, acrylamide, chia seeds, chia flour, flaxseed flour

**L14****TISSUE DISRUPTION AND ACID ADDITION AFFECT THE ENZYMATIC GLUCOSINOLATE HYDROLYSIS IN RED CABBAGE****Franziska Hanschen**<sup>1\*</sup><sup>1</sup>Leibniz Institute of Vegetable and Ornamental Crops (IGZ), Großbeeren, Germany

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Glucosinolates are sulfur rich plant secondary metabolites in *Brassica* vegetables such as cabbage or broccoli. They are precursors of bioactive and health-promoting compounds such as isothiocyanates (ITCs). Especially ITC are valued for their cancer preventing properties that are based on chemopreventive mechanisms [1]. Upon tissue disruption the enzyme myrosinase initiates enzymatic hydrolysis of glucosinolates and ITCs can be formed. However, in presence of specifier proteins often mainly nitriles and epithionitriles are formed [2]. Further, ITC can be quickly converted to amines by an enzyme-like mechanism in cabbage [3]. So, it was the aim of this study to evaluate how tissue disruption, incubation time and addition of domestic acids such as vinegar or lemon juice affect the fate of glucosinolates to identify conditions best to yield high ITC levels. Acidification of cabbage during salad/smoothie preparation was found to be ideal to obtain high levels of ITC, while amine formation was enhanced by homogenization. Taken together glucosinolate hydrolysis is very complex and many factors influence which compounds are up taken, which affects glucosinolate-related effects on humans.

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**Keywords:** glucosinolate hydrolysis, amines, isothiocyanates, cabbage

**L15****ERYTHRITOL AND FOOD SAFETY: CHEMICAL REACTIVITY IN FOODS AND NUTRITIONAL INSIGHTS**

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Erythritol ((2R,3S)-butan-1,2,3,4-tetraol) is a sweetener polyol present in nature, produced to date by biotechnology, gaining favour within consumers and food industry. It is widely used as food additive in calorie-reduced foods, depending on its “zero calories” impact (ranging from 0 to 0.2 calories/gram) [1]. Erythritol allows it to use it to reduce glycaemic index of foods, also giving a benefit for oral health. This additive (permitted in many Countries since 1990) was approved by EC in the list of food additives in 2006 (Dir. 2006/52/EC).

An antioxidant activity has been reported for erythritol, suggesting new functionality at technological level [2]. Recent studies highlighted health concerns related to the consume of erythritol, particularly referring to the cardiovascular diseases-correlated risk [3,4].

No reports are available to date on the possible interaction of erythritol with food components or on its fate during food processing operations. Even if quite unreactive in ambient conditions, thermal treatment of polyols can start dehydration and other reactions, leading to breakdown molecules able to react in different pathways in food systems. Erythritol is reported to undergo thermal degradation over its melting point, more pronounced in air than in inert atmosphere [5]. A link with the glyoxal formation under thermal processing was previously suggested [6]. Considering that  $\alpha$ -dicarbonyl compounds (e.g. glyoxal, methylglyoxal) can originate by thermally processed lipids, some interplays with the parallel antioxidant activity of erythritol could be suggested.

This communication aims to clarify the reactivity of erythritol, regarding i) its influence on acrylamide formation/reduction in bakery products (HPLC-MS studies), ii) its degradation under thermal processing at high temperature (NMR and MS studies), iii) its effective antioxidant capacity (in vitro, lipid model study, monitoring by HS-GC-IMS) as well as iv) its impact at nutritional level by in vivo trials in humans, coupled with metabolomic studies.

The outcomes confirm the technological usefulness for erythritol, albeit highlighting criticisms correlated to its reactivity, at physiological level. These results suggest a new scenario in the risk evaluation of erythritol, requiring more studies to assess its effective safety.

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**L16**  
**CANCELLED**

**L17****SAVOURY BISCUITS PREPARED WITH ADDITION OF CRICKET FLOUR: A COMPREHENSIVE QUALITY ASSESSMENT**

**Beverly Hradecká<sup>1\*</sup>, Michal Stupák<sup>2</sup>, Hana Pírová<sup>3</sup>, Lucie Stará<sup>4</sup>, Maria Filatová<sup>5</sup>, Ivan Švec<sup>6</sup>, Marcela Sluková<sup>7</sup>, Pavel Skřivan<sup>8</sup>, Jana Hajšlová<sup>9</sup>**

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Savoury biscuits are popular traditional ready-to-eat snacks prepared with basic ingredients such as wheat flour, fat, water, and salt. However, their poor nutritional value and high consumption contribute to the development of different civilization diseases. Therefore, enhancing the nutritional value by adding, e.g., fibre, proteins or legumes is challenging. Recently, a perspective source of proteins presents edible insect powders known for their beneficial composition, including essential amino acids, bioactive compounds (vitamins, minerals), unsaturated fatty acids, fibre, and a low carbohydrate content contributing to the reduction of the glycaemic index. However, when protein-rich insect flours are used for biscuit production, free amino acids and reducing sugars serve as important reactants in the Maillard reaction. In addition to the development of desirable flavour and colour, a heat-induced contaminant such as well-known acrylamide (a probable human carcinogen, group 2A) can arise. Additionally, the fat used for the biscuit's preparation can also be a source of processing contaminants 3-monochloropropane-1,2-diol fatty acid esters (3-MCPDEs) and glycidyl fatty acid esters. They are degraded in vivo by gastrointestinal lipases to form 3-MCPD and glycidol, classified as a probable human carcinogen (group 2B) and a potential human carcinogen (group 2A), respectively. In the recent years, several insect species (e.g., house crickets, yellow mealworms, and grasshoppers) have been authorized in the EU as novel foods, finding various uses. In this study, we aimed to investigate the impact of incorporation of cricket flour (*Acheta domesticus*) into the biscuit dough to assess the impact not only on the sensorial properties of the final product, but also on its nutritional value and chemical safety. The basic recipe (standard biscuit) contained wheat flour or whole-grain wheat flour, rapeseed oil, and salt. In the modified recipe cricket flour was added in different ratios (5, 10 and 15 %). The biscuits were baked at 180 °C for 25, 35 and 45 min and then subjected to analysis of acrylamide, MCPD and glycidyl esters. Finally, the aromatic profile was measured and a sensory evaluation was conducted.

**Keywords:** acrylamide, MCPD esters, glycidyl esters, protein enriched biscuits, cricket flour

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## L18

# CHOLESTEROL OXIDATION PRODUCTS AS MARKERS OF NUTRITIONAL QUALITY OF MILK AND MILK PRODUCTS

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Cholesterol undergoes enzymatic and non-enzymatic oxidation, resulting in various cholesterol oxidation products (COPs), including 7 $\alpha$ OHC, 7 $\beta$ OHC, 7KC,  $\alpha$ -epoxy,  $\beta$ -epoxy, TRIOL, and 25OHC. Non-enzymatic COPs mainly stem from food degradation caused by heat, light, radiation, and oxygen, raising concerns about their health risks. GC-MS enables accurate measurement of oxysterols in raw and processed food. We examined oxysterols in whole milk powder (WMP) and chocolates with increasing shelf-lives, monitoring changes in industrially stored WMP with different packaging. Non-enzymatic COPs increased with WMP shelf-life, reflecting nutritional quality indicators. This trend was observed in chocolates too. Processing had a lesser impact on oxysterol generation compared to WMP autoxidation, making shelf-life the primary determinant. Proper packaging mitigated oxysterol formation in processed food, preserving characteristics. Although more data is needed, reducing consumption is advised due to potential adverse effects at high concentrations. Measuring enzymatic and non-enzymatic oxysterols can enhance commercial and nutritional value of milk ingredients and products, highlighting quality and freshness. These findings emphasize the need to redefine food quality parameters.

**Keywords:** COPs, oxysterols, food quality, nutritional markers



## L19

# MANIPULATING THE MAILLARD REACTION TO REDUCE ACRYLAMIDE AND MAINTAIN FLAVOUR

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The relationship between the formation of flavour and acrylamide is complex. Both are formed via the Maillard reaction, and as a rule, changes in starting materials and process conditions which reduce acrylamide also reduce the formation of colour and flavour. However, this is not always the case. By identifying exactly which aroma compounds are providing the characteristic aroma, and understanding the detail of the formation pathways and limiting precursors, it is possible to steer the Maillard reaction to produce more aroma compounds and less acrylamide. The talk will focus on pyrazines which are important in baked, roasted and toasted foods, particularly the trisubstituted pyrazines, which are much more odour-active than their disubstituted analogues. The formation pathways are complex and the same pyrazine may be formed from multiple precursors depending on the conditions and matrix. Their carbon skeleton can contain fragments of both the parent sugar and amino acids. This talk will cover several examples where we have manipulated the volatile profile. In one example we show that it is possible to enhance baked flavour in low acrylamide crackers (~80 ng/g acrylamide), by adding a specifically targeted combination of amino acids and key intermediates, without increasing acrylamide concentration and demonstrate the chemical synergy involved in generating the target aroma compounds.

**Keywords:** acrylamide, amino acids, baked potato cracker, flavour, pyrazines, Strecker aldehydes

**L20****ACRYLAMIDE REDUCTION STRATEGY IN COMBINATION WITH DEOXYNIVALENOL MITIGATION IN INDUSTRIAL BISCUITS PRODUCTION****Michele Suman<sup>1\*</sup>, Silvia Generotti<sup>2</sup>, Martina Cirlini<sup>3</sup>, Chiara Dall'Asta<sup>4</sup>**<sup>1</sup> Department for Sustainable Food Process, Catholic University Sacred Heart, Piacenza, Italy<sup>2</sup> <sup>3</sup> <sup>4</sup> Department of Food & Drug, University of Parma, Parco Area delle Scienze 95/A, 43124, Parma, Italy

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Acrylamide is formed during baking in some frequently consumed food products. It is proven to be carcinogenic in rodents and a probable human carcinogen. Thus, the food industry is working to find solutions to minimize its formation during processing. To better understand the sources of its formation, the present study is aimed at investigating how acrylamide concentration may be influenced by bakery-making parameters within a parallel strategy of mycotoxin mitigation (focusing specifically on deoxynivalenol–DON) related to wholegrain and cocoa biscuit production. Among Fusarium toxins, DON is considered the most important contaminant in wheat and related bakery products, such as biscuits, due to its widespread occurrence. Exploiting the power of a Design of Experiments (DoE), several conditions were varied as mycotoxin contamination levels of the raw materials, recipe formulation, pH value of dough, and baking time/temperature; each selected treatment was varied within a defined range according to the technological requirements to obtain an appreciable product for consumers. The developed predictive model suggested how some parameters can concretely contribute to limit acrylamide formation in the final product, highlighting a significant role of pH value (correlated also to sodium bicarbonate raising agent), followed by baking time/temperature parameters. The study represents a concrete example of how the control and optimization of selected operative parameters may lead to multiple mitigation of specific natural/process contaminants in the final food products, though still remaining in the sensorial satisfactory range.

Suman, M.; Generotti, S.; Cirlini, M.; Dall'Asta C.: "Acrylamide Reduction Strategy in Combination with Deoxynivalenol Mitigation in Biscuits Production", *Toxins* 2019, 11, pp. 499-511

**Keywords:** acrylamide, deoxynivalenol, multiple mitigation strategies, design of experiments, bakery food processing, biscuits

**L21****THE ROLE OF 5-HYDROXYMETHYLFURFURAL ACCUMULATION VIA SUCROSE DEGRADATION ON ACRYLAMIDE FORMATION IN LOW MOISTURE FOOD SYSTEMS****Dilara Şen<sup>1</sup>, Vural Gökmen<sup>2\*</sup>**

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Multiresponse kinetic modelling of reactions can be used as a powerful tool to understand complex reaction mechanisms and obtain a better insight into the whole mechanism. In this way, rate-determining steps can be determined and the link between reactants and products can be expressed quantitatively. This study aims at investigating the mechanism of acrylamide formation in nuts and seeds during roasting in order to propose a generic multiresponse kinetic model explaining the roles of sucrose degradation and 5-hydroxymethyl furfural formation for actual food systems low in moisture content and lacking in reducing sugars. Sunflower (*Helianthus annuus* L.), flaxseed (*Linum usitatissimum* L.), peanut (*Arachis hypogaea* L.), and almond (*Prunus dulcis*) were selected to represent low moisture actual food systems. The samples were roasted at 160, 180, and 200 °C for 5 to 60 minutes and analyzed to determine changes in the concentrations of reactants (free amino acids, sugars) and products (alpha-dicarbonyl compounds, 5-hydroxymethyl furfural, acrylamide). The proposed multiresponse kinetic model suggests that sucrose first degrades to glucose and fructofuranosyl cation forming 5-hydroxymethylfurfural mainly through the 3-deoxyglucosone pathway in all samples roasted at 160 and 180 °C. The results revealed that the reaction of asparagine with 5-hydroxymethylfurfural is the predominant pathway for acrylamide formation.

**Keywords:** Maillard reaction, acrylamide, 5-hydroxymethylfurfural, low moisture foods, nuts and seeds, multiresponse kinetic modelling

**L22****FURAN AND ALKYLfurans IN BREAKFAST CEREALS - INFLUENCE OF INGREDIENTS AND PRODUCTION STEPS**

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Furan and alkylfurans are volatile process contaminants in foods that are formed during thermal processing. Of concern are production processes with high temperatures such as roasting, baking, extrusion cooking or pasteurization. Furans are therefore found in foods such as coffee and cocoa, canned foods, baby foods, as well as baked goods and breakfast cereals. Among these, breakfast cereals are a particularly relevant source of exposure as they are consumed by all age groups but especially by children. Furan is classified as a possible carcinogen (Group 2B) by the International Agency for Research on Cancer (IARC). Different formation pathways are described in literature and sugars, polyunsaturated fatty acids, amino acids, ascorbic acid as well as carotenoids can act as precursors for the formation of furan and alkylfurans. During the project, a validated headspace gas chromatography-mass spectrometry method with solid-phase microextraction (SPME) was used to screen for the presence of furan and alkylfurans in different types of breakfast cereals. In addition to furan, 2-methylfuran, 3-methylfuran, 2,5-dimethylfuran, 2-ethylfuran and 2-pentylfuran were also analyzed. Concentrations up to 230 µg/kg furan and 600 µg/kg total furan were detected in the samples, with puffed cereals showing the highest levels, followed by flakes and extruded products. In addition to furan and 2-methylfuran, 2-pentylfuran was also detected at comparably high levels. The results were evaluated with regard to production procedures and ingredients, like grain types or added sugar. The implementation of model experiments such as baking and extrusion cooking using systematically varied recipes and process parameters as well as the analysis of samples from various steps of the industrial processes enabled the identification of relevant factors for furan formation and indicates possible approaches towards minimization of the formation of furan and alkylfurans. This IGF project of the FEI (IGF 21305N) was supported via AiF within the programme for promoting the Industrial Collective Research (IGF) of the German Ministry of Economics and Climate Action (BMWK), based on a resolution of the German Parliament.

**Keywords:** breakfast cereals, GC-MS, furan, process contaminants, SPME

## L23

### IS FERMENTATION A GOOD OPTION FOR MITIGATING ACRYLAMIDE IN LEGUMES?

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Legumes as a high source of proteins and other nutritionally valuable compounds are in spotlights of consumers focused on healthy nutrition. The current popular trend is the consumption of legume-based products with a long shelf life as extruded or puffed ready-to-eat snacks. Products, not excluding legumes, undergoing heat treatment as puffing at high temperature and high pressure for a very short time, usually have a noticeable content of undesirable probably carcinogenic acrylamide, which depends, besides others aspects, on the amino acid asparagine content in the raw material. For improving digestibility of legumes and enhancing them with additional worthy components, the fermentation with fungi *Actinomucor elegans* was applied. In this study, the potential of fermented and unfermented legumes (peas, beans, and lentils) to form acrylamide in snacks was evaluated and verified.

**Keywords:** acrylamide, legumes, fermentation, mitigation strategies

**Acknowledgement:** This work was supported by the Ministry of Agriculture and Rural Development of the Slovak Republic under the contract No 1092/2022/MPRVSR-930 and it contributed to the activities of the COST Action 20128 Promoting Innovations in Fermented Foods (PIMENTO). Legume samples were complimentary provided by Celpo, Ltd. Očová, Slovakia.

## PROMOTING INNOVATION OF FERMENTED FOODS (PIMENTO) - COST ACTION CA20128

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Present in all European diets, fermented foods (FF) hold a strategic place due to the benefits they offer in terms of nutrition, sustainability, innovation, cultural heritage and consumer interest. The potential of FF for improving human health but also driving food innovation and local production in the next decades has become highly relevant. The PIMENTO project, a COST Action CA20128 (Promoting Innovation of ferMENTed fOods; <https://fermentedfoods.eu/>), which started in November 2021, is supported by COST (European Cooperation in Science and Technology; [www.cost.eu](http://www.cost.eu)). The challenge of PIMENTO is to federate the scientific community and other key stakeholders working on FF. The long-term goal of PIMENTO is to place Europe at the spearhead of innovation on microbial foods, promoting health, regional diversity, and local production at different scales, contributing to economic and societal development as well as food sovereignty in order to promote multi-modal innovation and respond to the expectations of European communities. The wide variety of stakeholders engaged will enable CA PIMENTO: i) to tightly connect and clarify scientific knowledge on health aspects of FF; ii) to tackle technical, societal and legislative bottlenecks behind FF-based innovations; iii) to contribute to the establishment of long-term scientific collaborations on FF; iv) to disseminate widely defined scientific knowledge on FF; v) to outline a strategic roadmap for future joint research. PIMENTO will contribute to the European Green Deal and the "Farm to Fork" strategy by enhancing research and innovation into fermentation-based solutions for food products and processes, improving nutritional, sensory and functional properties. This collaborative network of researchers that includes food scientists, innovators, entrepreneurs, microbiologists, biochemists, and nutritionists has a very broad geographical coverage with 396 partners from 283 institutions of 50 countries. This regional diversity will play an important role through considering a differentiated panel of FF in diets.

**Keywords:** fermented foods, innovation

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**L24****HIGH PRESSURE THERMAL STERILIZATION (HPTS) AND ITS EFFECT ON FOOD PROCESSING CONTAMINANTS AND QUALITY-RELATED PROPERTIES IN FOOD IN COMPARISON TO THERMAL-ONLY PROCESSING****Robert Sevenich**<sup>1\*</sup>

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High pressure in combination with high temperatures is an innovative and emerging technology to efficiently sterilize low acid food products over a long shelf-life. In this regard it is crucial to inactivate all pathogenic vegetative microorganism and in particular spore former and endospores, respectively. Compared to commercial sterilization techniques, with long dwell times and subsequent extensive heat impact, the additional pressure application enables faster heating and cooling rates and instantaneous heating throughout the whole product. Therefore, a less harsh impact on nutritional and sensorial qualities is generally attributed to the so-called High Pressure Thermal Sterilization (HPTS) by researchers, ideally resulting in a better overall quality of the food product. This work focuses on the comparison of thermal-only sterilization and high- pressure thermal treatment on theoretical basis, particularly affecting microbial stability, selected vitamins and bioactive compounds, colouring pigments, and food processing contaminants. Findings indicate that the additional pressure application can beneficially improve selected quality-related attributes, e.g. reduced formation of food processing contaminants like furan. Consequently, a higher quality product for consumer is attributed. Nevertheless, the review of articles and research work also revealed that it is not always possible to retain quality attributes in equal measure, regardless of the promising advantages of HPTS. Fine-tuning of the process parameters pressure, temperature and time is therefore mandatory to reach both goals, microbial stability and high quality.

**Keywords:** High Pressure Thermal Sterilization, processing parameters, quality related food composition, microbial stability, processing contaminants

**L25****UNTARGETED <sup>13</sup>C-LABELLING LC-HRMS BASED APPROACH TO STUDY THE FATE OF MYCOTOXINS DURING FOOD PROCESSING**

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Thermal food processing can be favorable for reduction of mycotoxins which are commonly present in raw material. Although, most of the mycotoxins are in general stable compounds, combination of high temperature, pressure and presence of complex matrix may lead to their decline in final products. However, reduction of mycotoxin level does not have to result in a mitigation of toxicological effects. The formed degradation products can be even more toxic than the parent toxins. To study the fate of mycotoxins during food processing is a very challenging task for several reasons: 1) Characterization and structural elucidation of degradation products in complex matrix is difficult, 2) matrix composition varies over the production process, 3) availability of analytical standards is limited, 4) preparation of "in-house" analytical standards in larger amounts needed for structural confirmation and for toxicological assessment is costly and time-consuming. Therefore, most of the published studies were mainly focused on the targeted detection of the parent mycotoxins and calculation of the rate of decline over the production process. The untargeted stable isotope labelling (SIL)-assisted metabolomic approach developed for metabolization of deoxynivalenol in wheat [1] was applied to study of the fate of mycotoxins during food processing. The work flow of SIL is as follows: raw material used for food production is treated with a mixture of non-labelled and <sup>13</sup>C-labelled standard of mycotoxin, intermediate products and final product are then analyzed by liquid chromatography-high resolution mass spectrometry (LC-HRMS). Detected pair of signals originated from non-labelled and isotopically labelled compounds are extracted from the full-scan chromatogram by the software algorithm MetExtract II. [2] The application of the SIL-LC-HRMS will be presented on the case studies focused on the fate of deoxynivalenol in bread, biscuits and crackers [3]. Moreover, preliminary results about fates of relevant mycotoxins in production of gluten free pasta (such as aflatoxins, fumonisins and zearalenone) will be presented.

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**Keywords:** mycotoxins, food processing, degradation products, untargeted stable isotope labelling (SIL)-assisted metabolomic approach



**L26****FORMATION AND DEGRADATION OF GLYCATION COMPOUNDS DURING BEER PRODUCTION**

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During thermal treatment of food, glycation products are formed from amino components and reducing sugars through the Maillard reaction, which can act as precursors for aroma and color compounds. In beer production, malt contributes a baseline level of glycation products [1], and such compounds have been identified in beer [2] with some acting as precursors for aging aroma compounds [3]. However, little is known about the kinetics of glycation products during the malt production and brewing processes. In our study, we produced malt and beer at a laboratory scale and conducted a screening of free glycation products, including Amadori compounds, dicarbonyl compounds, and advanced glycation end products (AGEs), throughout the entire process in order to evaluate the impact of individual process steps on glycation reactions. The methods used in the study include LC-QQQ-MS stable isotope dilution assay for Amadori compounds and AGEs, and HPLC-UV after derivatization with ortho-phenylene diamine for dicarbonyl compounds. Our findings showed that alpha-N-Amadori compounds behaved similarly while dicarbonyl compounds and AGEs showed different effects. Amadori compounds stay constant during mashing and decrease by wort boiling. 3-Deoxyglucosone and the late-stage product MG-H1 increases in the brewhouse while glucosone, 3-deoxypentosone and pyrrolidine don't show any effects. In addition, wort boiling and fermentation are the steps with the highest influence in concentration changes. Furthermore, we evaluated the impact of different malt types, mash temperature profiles, and yeast strains on the formation of these compounds. Our results showed that the choice of malt had a significant impact on the levels of glycation compounds, and the use of different yeast strains also had an effect on the concentration of some substances in the final beer. The mash temperature profile had the lowest impact.

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[2] Hellwig et al. (2016) Free and Protein-Bound Maillard Reaction Products in Beer: Method Development and a Survey of Different Beer Types, 38: 7234-7243.

[3] Nobis & Kwasnicki et al. (2021) The Influence of De Novo Formation of Aging Aldehydes 10: 2668

**Keywords:** Maillard, glycation, beer, malt, brewing

**L27****ROASTING IMPACT ON COCOA AND COCOA BEAN SHELLS: AN OVERVIEW ON BIOACTIVE COMPOUNDS AND NEW UP-CYCLED INGREDIENTS**

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Cocoa beans (*T. cacao* L.) are subjected to the roasting process, a strong thermal impact which leads to significant modification in their chemical composition, allowing the microbial and enzymatic stabilization as well as the formation of the typical aroma [1]. Cocoa bean shells (CBS) are a by-product generated during the roasting phase, when beans are de-hulled and shells discarded [2]. Cocoa and CBS contains bioactive compounds such as methylxanthines, polyphenols and prebiotic fibers [3], characterized by physiological implications, but significantly impacted by roasting. The chemical characterization of CBS confirmed their use as basic raw material for "up-cycled" ingredients production for pharmaceutical, nutraceutical and cosmetic purposes.

CBS are characterized by a significant content of dietary fiber (DF), up to 59% of dry matter [4], with a ratio of insoluble dietary fiber (IDF) to soluble dietary fiber (SDF) reaching 3:1. IDF, and particularly SDF are often prebiotic ingredients able to boost the production of Short Chain Fatty Acids (SCFAs) by the colonic microorganisms at gut level, thus conferring a health benefit to the host [5].

This communication aims to clarify the impact of roasting on polyphenols of cocoa beans from different geographical origins, showing the strategies to isolate new functional ingredients eliminating contaminants. Isolation of pectin fraction [6], clovamide [7] and antioxidant polyphenols and pigments [8], and, more recently, the enzyme-driven production of prebiotic oligosaccharides is reported and critically discussed, confirming the usefulness of cocoa-related by-products/wastes for a new smart nutrition. More particularly, the bio-based process to produce new fiber and polyphenols-rich ingredients from CBS will be described, also reporting a preliminary evaluation of the thermal processing (model study) on volatilome of new ingredients, adding conventional ingredients like sucrose, lipids (sunflower oil) and wheat flour, by HS-GC-IMS.

All these outcomes are a promising opportunity to open a new scenario in the bio-valorisation of CBS, according with the concepts of the bio-based industry and the circular economy.

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**L28****PEPTIDE PROFILING OF PASTEURIZED MILK FERMENTED WITH YOGURT STARTER CULTURES *S. THERMOPHILUS* AND *LB. BULGARICUS* BY MICROLC-IM-QTOF-MS/MS**

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Yogurt is traditionally produced by fermentation of milk with mixed cultures of *Streptococcus thermophilus* and *Lactobacillus bulgaricus*. These lactic acid bacteria are multiauxotrophic and therefore not only digest the milk carbohydrates but also the proteins. Extracellular bacterial proteases hydrolyse intact milk proteins to form peptides, which subsequently get further hydrolysed by intracellular peptidases to form smaller peptides and free amino acids. This causes the release of bioactive peptides which contribute to the health benefits of yogurt, including peptides with antihypertensive, antimicrobial and antioxidative properties. Peptide profiling is an effective tool to better understand the proteolytic processes during yogurt fermentation which lead to the formation of bioactive peptides. However, the influence of the starter cultures on the peptide profile has not yet been adequately researched. Thus, pasteurized cow's milk was fermented with single cultures and a mixed culture of *S. thermophilus* (DSM 20259) and *Lb. bulgaricus* (DSM 20080) for 24 h and 48 h, respectively. The degree of hydrolysis of the fermentation products was determined using an OPA assay adapted to the matrix. Peptides were extracted by solid-phase microextraction and analysed by microLC-IM-QTOF-MS/MS in the data dependent acquisition mode (DDA). The peptides were identified using the software PEAKS<sup>®</sup> Online, which uses a combination of de novo sequencing and a database search. Data was further evaluated in RStudio. Based on the peptide profile, fermented samples could be distinguished from unfermented control samples. The identified peptides mostly derived from the four caseins. With up to 2000 peptides, the number of peptides identified in samples fermented with only *Lb. bulgaricus* or a mixed culture of both investigated organisms was up to 50% higher than in unfermented samples. In particular, *Lb. bulgaricus* significantly increased the number of peptides from  $\beta$ - and  $\kappa$ -casein. These results can be attributed to the high proteolytic activity of *Lb. bulgaricus* which can also be seen in the results of the OPA assay. In addition to this, the number of bioactive peptides already described in literature was noticeably higher in samples containing *Lb. bulgaricus*. *S. thermophilus* on the other hand hardly influences the peptide profile in the mixed culture due to its low proteolytic activity. In the samples fermented with a *S. thermophilus* single culture, fewer peptides with a higher average peptide length than in the other samples were identified. In combination with the results of the OPA-assay, this indicates that *S. thermophilus* digests mainly short-chain peptides, generating shorter peptides and free amino acids which were not covered by the peptide profiling. In conclusion, *Lb. bulgaricus* has a higher influence on the changes of the peptide profile during yogurt fermentation than *S. thermophilus* and is responsible for the release of bioactive peptides.

**Keywords:** milk proteins, fermentation, peptide profiling, untargeted proteomics

**L29****REACTIVE  $\alpha$ -DICARBONYL COMPOUNDS IN MAILLARD CHEMISTRY OF MONO- AND OLIGOSACCHARIDES****Karel Cejpek<sup>1\*</sup>, Anna Šírová<sup>2</sup>, Anna Průšová<sup>3</sup>, Vít Tobiška<sup>4</sup>, Zuzana Procházková<sup>5</sup>**<sup>1 2 3 4 5</sup> University of Chemistry and Technology Prague, Department of Food Analysis and Nutrition, Prague, Czech Republic

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During caramelization, Maillard reaction, and other non-enzymatic browning reactions, reducing sugars easily undergo a series of enolization and dehydration reactions, and alternatively also oxidation, fragmentation and other reactions to form corresponding  $\alpha$ -dicarbonyl intermediates and products, such as deoxyglycosuloses and methylglyoxal. They are key intermediates of non-enzymatic browning reactions and can cause the decrease of nutritional value and sensory, redox, toxicological and technologically-functional changes in food. Foodborne  $\alpha$ -dicarbonyl compounds ( $\alpha$ -DCs) can react with many constituents of human gastrointestinal tract and probably also act in plasma. The formation of  $\alpha$ -DCs from monosaccharides is quite clearly described but oligosaccharides can react in more complex pathways. Structural differences between reducing and non-reducing sugars, oligosaccharides and monosaccharides, and oligosaccharides with different glycosidic linkages lead to both different reactivity and different pools of  $\alpha$ -DC intermediates and products in food. In addition to the effects of many extrinsic factors, the reactivity of reducing saccharides depends on the relative content of their acyclic and cyclic forms, structural conformations, and electron effects of vicinal substituents affecting reactions with nucleophiles, e.g., in the Maillard reaction. Our experiments were focused principally on  $\alpha$ -DCs arising from oligosaccharides with (1 $\rightarrow$ 4)-glycosidic linkage such as lactose and their isomers. HPLC-PDA and LC-MS methods were used for analyses of  $\alpha$ -dicarbonyl compounds in the form of quinoxaline derivatives. Kinetics of  $\alpha$ -DCs was evaluated in complementary sets of reaction models. The model reactions systems varied in several parameters such as reactants; caramelization or Maillard reaction conditions; aqueous or dry systems; and *ex-post* or *in-statu-nascenti* derivatization of  $\alpha$ -DCs. The levels of  $\alpha$ -DCs were determined in selected foods with significant content of reducing oligosaccharides (e.g., dairy products). The scavenging effect of some phenolic compounds towards  $\alpha$ -DCs in selected samples was also shown. The results acquired both from model experiments and food samples may contribute to better understanding and controlling the Maillard reaction in dairy and starch containing products. In addition to  $\alpha$ -DCs with a glycosidically bonded sugar unit(s), the typical aldose-derived  $\alpha$ -DC is 1,4-dideoxyhexosulose while the determining products for ketosides are 4-deoxyhexosulose, 3-deoxypentosulose and 3,4-dideoxypentosulose. In acidic food such as carbonated soft drinks sweetened with sucrose and syrups, the development of  $\alpha$ -DCs is strongly affected by the sugar used. Besides the findings on oligosaccharide-related  $\alpha$ -DCs and mechanisms of their formation, the results revealed that ketoses and ketosides are superior to aldoses and aldosesides in the production of  $\alpha$ -dicarbonyl compounds.

**Keywords:** Maillard reaction, caramelization,  $\alpha$ -dicarbonyl compounds, oligosaccharides, dairy products

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**L30****FORMATION OF PROTEIN-BOUND MAILLARD REACTION PRODUCTS DURING THE STORAGE OF MANUKA HONEY****Marcus Thierig<sup>1\*</sup>, Eva Siegel<sup>2</sup>, Thomas Henle<sup>3</sup>**<sup>1 2 3</sup> TU Dresden, Dresden, Germany

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Manuka honey is exclusively produced in New Zealand and in South Australia and provides a pronounced non-peroxide antibacterial activity mainly caused by methylglyoxal (MGO) besides other synergistically active compounds [1]. While non-manuka honeys just contain small amounts of MGO up to 5 mg/kg, in manuka honeys MGO contents up to 1540 mg/kg were quantitated [2,3]. MGO is a reactive dicarbonyl compound that is able to glycate lysine- and arginine residues of honey protein. Previous work indicated that the contents of the protein-bound Maillard reaction products (MRPs) N- $\epsilon$ -carboxyethyllysine (CEL) and "methylglyoxal-derived hydroimidazolone 1" (MG-H1) in isolated honey protein correlate with the MGO content in the honeys. [4]. Therefore, these MRPs are potential marker substances for the authentication of manuka honey. To evaluate the formation of CEL, MG-H1 and the MGO-independent MRPs N- $\epsilon$ -fructosyllysine (FL), N- $\epsilon$ -carboxymethyllysine und pyrroline, as well as their contribution to the glycation pattern, an accelerated storage of manuka honey spiked with 100-700 mg/kg MGO was performed for 10 weeks at 37°C. After isolation of the honey protein and enzymatic hydrolysis, protein-bound MRPs were quantified via LC-MS/MS. For quantification, the content of protein-bound leucine was used as quasi-internal standard. During the incubation protein-bound CEL (3-42  $\mu$ mol/mmol Leu) and MG-H1 (0.4-6.5  $\mu$ mol/mmol Leu) were formed continuously. Thereby, higher MGO contents in the honey lead to an increased formation of these compounds. At the same time FL formation decreased at high MGO levels (46-24  $\mu$ mol/mmol Leu). Thus, competing reactions of MGO and glucose for lysine side chains can be assumed. Additionally, it has been shown that 27 to 33% of the available lysine residues are glycated during 10 weeks storage. However, the degree of lysine modification is independent from the MGO content. The honey proteins differ solely in their glycation pattern. FL is the most abundant MRP in low-MGO honeys, while higher MGO contents lead to a shift towards CEL as the predominant MRP. Furthermore, it has been shown that both the contents of protein-bound MRP as well as the glycation patterns in the model honeys are comparable to authentic manuka samples with similar MGO contents. Therefore, our results contribute to the authentication of manuka honey.

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**L31****CORRELATION OF MAILLARD REACTIONS COMPOUNDS WITH INDUSTRIAL "DOCE DE LEITE" COMPOSITION AND PHYSICOCHEMICAL PARAMETERS**

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In chemical terms, we can define doce de leite (DL) as a mix of milk and sucrose, in which Maillard reactions (MR) take place during the thermal and evaporation treatment stages. MR, therefore, determines not just the main flavor and color, but also DL microstructure and texture. The combination of these two commodities (milk and sugar) plus the manufacturing process itself adds value to the product, mainly due to the strong influence of the compounds generated by MR. However, there are few studies published about correlation of MR compounds with commercial DL composition and physicochemical parameters. Thus, the objective of this study was to evaluate the correlation between total solids (TS), carbohydrates, protein and pH of 11 industrial DL products (with and without added sugar) with 5 different chemical markers: Furosine (Fur), Carboxymethyllysine (CML), Lysinoalanine (LAL), Pyrraline (Pyr) and free 5-hydroxymethyl-2-furaldehyde (HMF). Results showed that TS were positively correlated with Fur ( $r = 0.92$ ,  $p = 0.01$ ), HMF ( $r = 0.81$ ,  $p = 0.01$ ) and Pyr ( $r = 0.94$ ,  $p = 0.01$ ) for the group of all 11 samples (G1). The increase in TS content naturally increases the constituent amounts, however, this effect was not observed for CML and LAL, which did not present any correlation with TS. This indicates slow oxidation reactions (CML effect) and unfavorable "sugar-independent" protein cross-linking (LAL effect), possibly related to reducing sugar in the DL. Carbohydrates were positively correlated with Fur ( $r = 0.85$ ,  $p = 0.01$ ) and HMF ( $r = 0.85$ ,  $p = 0.01$ ) only in the DL subgroup with added sugar (G3), indicating that sugar-free (diet) products have a specific behavior, mainly due to the substitution of sucrose by other ingredients. Carbohydrates and LAL presented a negative correlation ( $r = -0.94$ ,  $p = 0.01$ ) in the subgroup of samples without DL with the sucrose pre-caramelization manufacturing step (G2). Possibly this type of technology favors the formation of LAL because of the high initial temperature, as this product had the highest LAL value among all samples ( $10.4 \pm 1.3$  mg/100 g). Carbohydrates and CML also presented a negative correlation ( $r = -0.82$ ,  $p = 0.02$ ) in the DL subgroup with added sugar but without sucrose pre-caramelization (G4), indicating the specificity of diet DL and the influence of the processing type on oxidation reactions. There was no correlation between protein and the analyzed parameters. Finally, pH was negatively correlated with Fur ( $r = -0.89$ ,  $p = 0.01$ ), HMF ( $r = -0.88$ ,  $p = 0.01$ ) and Pyr ( $r = -0.86$ ,  $p = 0.01$ ) in the group G1, positively correlated with CML ( $r = 0.82$ ,  $p = 0.03$ ) in the subgroup G4 and did not present correlation with LAL in any group. In conclusion, correlations between these factors may provide evidence for understanding MR in industrial DL products.

**Keywords:** Dulce de leche, Maillard reaction, composition, industry, milk

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**L32****MULTIRESPONSE KINETIC MODELLING OF MAILLARD REACTION DURING UHT-TREATMENT OF MILKS****Aytul Hamzalioglu<sup>1\*</sup>, Isil Aktag<sup>2</sup>, Vural Gokmen<sup>3</sup>**<sup>1 3</sup> Hacettepe University, Department of Food Engineering, Ankara, Turkey<sup>2</sup> Munzur University, Department of Culinary Arts and Gastronomy, Tunceli, Turkey

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Milk comprises a major part in daily diet for human owing to its balanced protein, carbohydrate, lipid and mineral content. However, its composition makes it a favourable media for microorganisms and thus limits the shelf-life. In order to increase the shelf-life of milks, ultra-high-temperature (UHT) processing, which is heating milks at higher temperatures (130-150°C), is commonly applied. Such high temperatures ensure the inactivation of bacteria and enzymes providing the extended shelf life more than 6 months, however, heat-treatment under these harsh conditions also leads to chemical changes decreasing the nutritional value of milk as well as formation of possible harmful compounds. Maillard reaction is responsible from the major changes in milks as a consequence of UHT-treatment. This study aims to kinetically investigate in depth the Maillard reaction induced changes in milk during UHT-treatment. For this purpose, a comprehensive kinetic model including the elementary steps for Maillard reaction was proposed. The changes in lactose, reducing sugars, free amino acids, bound lysine, dicarbonyl compounds, 5-hydroxymethylfurfural and AGEs including carboxymethyllysine (CML) and carboxyethyllysine (CEL) were monitored in milks during heating at 110, 120, 130 and 140°C. Kinetically dominant pathways of Maillard reaction and their rate constants were unravelled by means of multi-response kinetic modelling approach. The results of the model indicated that formation of dicarbonyl compounds, glucosone, 3-deoxyglucosone, 1-deoxyglucosone, glyoxal and methylglyoxal proceeded through the cleavage of lysine from the main intermediate lactulosyl-lysine. On the other hand, formation of this intermediate had also an important role on CML and CEL formation. By using this model, obtained kinetic rate constants were employed to predict the progress of Maillard reaction in UHT-treated milks. Time-temperature history of milks was used in the calculation of furosine, CML, CEL amounts as well as lysine loss during UHT-treatment. The amounts of these compounds were efficiently (80-97 % approximately to real amounts) estimated in UHT-treated milks. This study provided in depth kinetical evaluation of Maillard reaction and its application into real-time prediction of Maillard reaction products in UHT-treated milks.

**Keywords:** multi response kinetic modelling, UHT-treated milks, Maillard reaction, furosine, lysine loss

**L33****INFLUENCE OF PRESSURE ON MAILLARD-TYPE REACTIONS****Uwe Schwarzenbolz<sup>1\*</sup>, Thomas Henle<sup>2</sup>**<sup>1 2</sup> Technische Universität Dresden, Dresden, Germany

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In order to treat foodstuffs more gently than thermal processes while maintaining the same level of safety, various novel methods have been developed, among which high-pressure treatment stands out due to its greatest market maturity. Although "substantial equivalence" to traditional methods can be assumed, there are food chemical aspects that are not yet sufficiently characterized. In this presentation, by a combination of literature data as well as own research results, the influence of pressure on chemical reactions in foods, with emphasis on Maillard-type reactions in combination with a brief historical background, is outlined. First insights on the Maillard-reaction (MR) under high pressure can be found in T. Tamaoka (Agric. Biol. Chem. 1991, 55, 2071–2074). Especially condensations, which take place at the early stage and browning reactions (i.e. end stage), were regarded and found to be hindered by pressure. Further investigations with individual compounds suggested a decrease in the velocity of early stages of the MR [3], while the melanoidine formation is reduced. In a more general view, reactions of carbonyl compounds with lysine, which can be regarded as core of the MR, seem to be slower at high pressure [4]. On the other hand, reactions of carbonyls with arginine residues tend to be accelerated by pressure. For example, an enhanced formation of pentosidine (containing an arginine residue) can be observed while pyrrolidine (lysine derivative) is suppressed by increasing pressure. These observations are also supported in protein model systems. With respect to volatiles a change in the organoleptic quality of MR reaction models can be noticed and a significant decrease in the concentration of Strecker aldehydes was measured comparing heat to high pressure treatment of foods. Apart from that, there is evidence that the reactivity of carbohydrates is rather higher under pressure, as an increase in the mutarotation velocity as well as an intensified degradation of dicarbonyls can be noticed. Pressure also does have influence on cross-linking reactions. While sugar induced cross-links, may be due to an altered carbohydrate reactivity, is reduced, the formation of dehydroalanine derivatives like lysinoalanine is slightly enhanced by pressure. In the course of the MR also unwanted substances, like acrylamide and furan, can be formed. As the key influence factor on these so-called process contaminants is the heat impact, their concentration was found to be lower in reaction mixtures as well as in complex foods after pressure treatment. From the general view, it can be concluded that Maillard-like reactions under pressure follow different pathways as at atmospheric pressure and therefore a simple transfer of established reactions schemes from one set of conditions to another is not valid.

**Keywords:** Maillard reaction, pressure, amino acids



# **POSTER SESSION**

**P1****COMPREHENSIVE EVALUATION OF THE COMPOSITION OF BAKED INSECT-BASED PRODUCTS****Kateřina Šebelová<sup>1\*</sup>, Beverly Hradecká<sup>2</sup>, Jana Hajšlová<sup>3</sup>**<sup>1 2 3</sup> University of Chemistry and Technology Prague, Department of Food Analysis and Nutrition, Prague, Czech Republic

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Products containing insects are a new type of food that appears on the market. Edible insects are used in the food industry as an alternative and sustainable source of animal protein. For example, it can be added, in the form of powder, to baked goods as a partial substitute for flour. Due to the EU legislation, which defines insect species authorized for the food industry, it is essential to be able to detect and determine the content of these species in food products on the market and ensure the quality and safety of these products. In this study, metabolomic approach was used as a tool to comprehensively evaluate the composition of baked goods (biscuits) with the addition of house cricket (*Acheta domesticus*) powder. Biscuits were baked in two batches, each from a different type of wheat flour (T530 and T1700). In each batch, biscuits with different content of cricket powder (0%, 5%, 10%, and 15%) and different baking times (25, 35, and 45 minutes) were made, extracted and analyzed using ultra high-performance liquid chromatography coupled with high resolution tandem mass spectrometry (UHPLC-HRMS/MS). The acquired data were evaluated using two approaches. Principal component analysis (PCA) was performed to display the overall variability among samples and to distinguish individual tested cricket powder contents and baking conditions. The correlation analysis was carried out to find specific metabolites that would indicate the presence of house cricket powder in the tested samples and changes in the composition due to the different baking conditions. The results obtained during the study can contribute to the optimization of production procedures, ensuring the authenticity of insect-based products on the market and their quality.

**Keywords:** LC-MS, metabolomics, edible insects, novel food**Acknowledgement:** This work was supported by METROFOOD-CZ research infrastructure project (MEYS Grant No: LM2023064) including access to its facilities. This work was financial supported from a specific university research A1\_FPBT\_2023\_002, This work was supported by National Agency for Agriculture Research (Project no. NAZV-QK23020101)

**P2****EFFECT OF ECHINACEA PURPUREA EXTRACT ON NUTRITIONAL AND SENSORY QUALITY OF FRESH CHEESE****Michal Gross<sup>1\*</sup>, Eva Vitova<sup>2</sup>, Matěj Rychetsky<sup>3</sup>**<sup>1 2 3</sup> Faculty of Chemistry, Brno University of Technology, Brno, Czech Republic

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Codex Alimentarius defines fresh cheeses as „... products which are ready for consumption shortly after manufacture.” They represent a diverse group of products produced by coagulation of milk, cream or whey in three different ways: using acid, combination of acid with rennet and/or acid with elevated temperature. In this work, the model fresh acid-curd cheese samples were produced using standard technology, acetic acid was used as coagulant. Produced samples were enriched with the extract of Echinacea purpurea flowers (2% w/w) as natural flavouring ingredient. Echinacea purpurea, called purple coneflower, is a perennial herb with pink or purple flowers, belonging to the Asteraceae family. Several studies suggest that it contains bioactive substances that boost immune system, relieve pain, reduce inflammation, and have hormonal, antiviral, and antioxidant effects. It is recommended to treat e.g. urinary, vaginal or respiratory tract infections. For this reason, it is used for medicinal purposes in the form of e.g. extracts, tinctures, tablets. The main aim of this study was to investigate the effect of extract addition on antioxidant potential of produced cheese samples, simultaneously, sensory quality was evaluated in connection with aroma profile assessment. The results indicate that addition of extract significantly influenced chemical composition of cheese. Cheeses were enriched by several volatile aroma compounds, mainly Borneol, Bornyl acetate and Camphor, which were recognized as Echinacea components. They are characterized by camphor, woody, herbal aroma and are also considered as important antioxidants. Compared to control cheese (without addition of extract) enriched cheese showed increased nutritional value; total antioxidant activity (ABTS assay) was 197.1 µgTEAC/g, the total phenolics content (Folin-Ciocalteu method) 0.17 mg GAE/g of cheese; however, sensory quality was negatively influenced. Cheeses have inhomogeneous, unusual brownish colour and noticeably bitter flavour.

**Keywords:** fresh cheese, Echinacea, antioxidant activity, sensory quality**Acknowledgement:** This work was supported by a Standard project of specific research No. FCH-S-23-8330.

**P3**  
**CANCELLED**

**P4****IMPACT OF DIFFERENT EXTRACTION METHODS ON COMPOSITION AND FUNCTIONAL PROPERTIES OF FILIPENDULA ULMARIA EXTRACT****Matěj Rychetský<sup>1\*</sup>, Eva Vítová<sup>2</sup>**<sup>1 2</sup> Faculty of Chemistry, Brno University of Technology, Brno, Czech Republic

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The health benefits and ability to replace synthetic analogues have propelled natural extracts into the spotlight. As a result, there has been a substantial boom in their utilization across various industries, particularly in the food sector, which has experienced a notable increase in their adoption. One of these extracts that's being studied for its beneficial effects is that of *Filipendula ulmaria*, which has been shown to have potential antioxidant, antimicrobial and anti-inflammatory effects. In addition to the health beneficial effects mentioned above, this extract can also serve as a carrier of sensory active substances, which may make it an interesting additive in various food products. However, the problem with these extracts is that the content of the active compounds and the associated beneficial effects vary with different extraction parameters. For this reason, an optimization experiment was performed to find the extraction method that gives the best results in terms of active compounds content and antioxidant and antimicrobial activity. The experiment was designed and realized using two extraction methods - maceration and ultrasound assisted maceration with several optimization parameters - different extraction reagents (ethanol, hexane, water), solid:solvent ratio (1:1-1:10), extraction time (15 minutes-24 hours) and extraction temperature (20-90 °C). The extracts were analyzed by HS-SPME-GC-MS for their aroma active compounds content, antioxidant activity was measured by ABTS and DPPH assays, total phenolic content (TPC) was determined by Folin-Ciocalteu method and antimicrobial activity against three microorganisms - gram-negative *Escherichia coli*, gram-positive *Bacillus cereus* and yeast *Candida glabrata*, was tested by agar well diffusion method and broth dilution method. Based on the results of all the analyses, ultrasound assisted maceration under the following conditions was selected as the most suitable for obtaining an extract with an ideal composition in terms of active substances: 40% ethanol at a solid:solvent ratio of 1:5, 60 °C for 90 min.

**Keywords:** *Filipendula ulmaria*, aroma compounds, antioxidants, antimicrobial activity

## **P5 AN EVALUATION OF TARGETED AND NON-TARGETED APPROACHES TO SAUDI REFERENCE HONEY**

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Honeybee is an extensively consumed natural functional food, featuring high economic value, related to its authenticity and purity. Due to the high demand for honey, it has been wildly targeted for food adulteration with substandard honey. Emerging absorbent resin technology has affected the honey market. Honey adulteration is a serious global matter that requires cooperation among regulatory agencies, synchronization of standards. Saudi Arabia has seen an increase in its consumption of honey in recent years. For both commercial as well as health reasons, honey's authenticity is of the utmost importance.

**Keywords:** Saudi Arabia, sucrose, diastase, honey adulteration, HPLC

**P6****BIOPROSPECTING OF ARTEMISIA SCOPARIA, PANAX GINSENG AND PHELLODENDRON SP. EXTRACTS**

**Kamila Bechynska**<sup>1\*</sup>, Adela Kollarova<sup>2</sup>, Vit Kosek<sup>3</sup>, Lucie Slovakova<sup>4</sup>, Jan Polak<sup>5</sup>, Jana Hajslova<sup>6</sup>

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Many herbal extracts are known to contain biologically active molecules, some of which find uses in pharmaceuticals production. In the recent years, substances with a potential to inhibit lipolysis are being searched. One of potential applications is a treatment of obstructive sleep apnea, chronic disease which causes hypoxia and disrupts adipocyte metabolism. The aim of our study was to isolate and identify novel bioactive secondary metabolites from *Artemisia scoparia*, *Panax ginseng* and *Phellodendron sp.*, which are presumed to contain lipolysis inhibitors. To obtain the entire set of compounds with different polarities, three types of extracts were prepared from each dried plant: aqueous, methanolic and dichloromethane extracts. These extracts were then fractionated using high performance liquid chromatography Waters AutoPurification System coupled to single quadrupole mass spectrometry detector (QDa). Nine sets of fractions were collected and their antilipolytic activity was measured. An enzymatic assay based on measurement of released free glycerol concentration was employed both for spontaneous and isoprenaline-induced lipolysis. Significant fractions with the highest inhibitory activity were further analyzed using ultra high-performance liquid chromatography coupled with tandem high resolution mass spectrometry, in order to identify compounds responsible for respective bioactivity.

**Keywords:** bioprospecting, bioactivity, obstructive sleep apnea, antilipolytic activity

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## P7 PHYTOCANNABINOID PROFILES IN HEMP SEEDS AND PRODUCTS THEREOF

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*Cannabis sativa* L. (hemp) is a plant that contains unique secondary metabolites called phytocannabinoids. Currently, this plant is widely used in the food industry, with hemp seeds and derived products such as hemp oil and flour being particularly popular. The seeds themselves contain antioxidants and polyunsaturated fatty acids but they do not contain any phytocannabinoids unless they come into contact with resin found in the glandular trichomes of female flowers. While there are many phytocannabinoids, those belonging to the tetrahydrocannabinol group are regulated by law due to their psychoactive effects. In relation to the need for monitoring the content of psychoactive substances, a maximum limit has been set for the sum of delta-9-tetrahydrocannabinol ( $\Delta^9$ -THC) and its acidic precursor delta-9-tetrahydrocannabinolic acid ( $\Delta^9$ -THCA). In 2022, the European Commission approved an amendment to Commission Regulation (EU) No 1881/2006, which sets maximum limits for certain contaminants in food, including  $\Delta^9$ -THC. The limit is set at 3 mg/kg for hemp seeds and products derived from hemp seeds, and 7.5 mg/kg for hemp oil. This study focuses on the quantitative determination of 42 phytocannabinoids in hemp seeds and oils or flour. The obtained results of  $\Delta^9$ -THC content were also subsequently compared with the Acute Reference Dose (ARfD) established by EFSA. The analysis was performed using ultra-high performance liquid chromatography coupled with high resolution mass spectrometry (U-HPLC-HRMS). The results revealed diverse profiles of phytocannabinoids in hemp seeds and their products. The seeds contained low concentrations of target analytes, with non-psychoactive components and predominantly acidic forms of phytocannabinoids, such as cannabidiolic acid (CBDA) or cannabidivarinic acid (CBDVA), being predominant. Oils and flours exhibited varying ratios of phytocannabinoids. The highest finding of total phytocannabinoid content in hemp oils exceeded 1400 mg/kg. Such a high finding was observed in only 2 samples, while the remaining samples did not exceed the level of total phytocannabinoid content of 100 mg/kg. In each tested product group, there was a product that would exceed the ARfD set for  $\Delta^9$ -THC when consumed in the recommended amount. These findings can be used for the assessment and quality control of these products, as well as for further research and development in the field of hemp products with therapeutic potential.

**Keywords:** phytocannabinoids, hemp seeds, hemp flour, hemp oil, U-HPLC-HRMS

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**P8**  
**CANCELLED**

**P9****FOOD REFORMULATION: EFFECT OF REDUCING THE SALT CONTENT OF SELECTED TYPES OF CHEESE ON THE KEY AROMA COMPOUNDS**

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The goal of food reformulations is to offer consumers traditional products with a modified recipe so that they meet the modern requirements of a healthy lifestyle and diet. One way to reformulate food is to reduce the salt (sodium chloride) content, whose increased consumption contributes to high blood pressure and increased risk of cardiovascular disease. However, reducing the salt content of cheese has its challenges, as salt plays an important role in the production technology - it contributes not only to the taste and smell of the cheese, but also to its resulting texture, and, last but not least, to the durability of the product. A lower salt content in cheeses can be achieved by partially or completely replacing sodium chloride with potassium chloride, using potassium citrates or phosphates (processed cheeses), or increasing the water content while reducing the fat content. However, these replacements must not significantly change the sensory properties of the final product to make the reformulated product acceptable to the consumer. An important step before the determination of key odorants of particular samples of reformulated cheeses is training and testing of assessors who participate in the olfactometric evaluation. The set of assessors was tested on 15 compounds typical of cheese using sniffing sticks (fa Olfasense) or prepared aqueous solutions of standards. Subsequently, the selected assessors - experts were trained using aroma extract dilution analysis (AEDA) on a mixed solution of selected compounds. In 4 types of reformulated cheeses (processed cheese, white cheese, Dutch-type cheese, fresh cheese), key aroma compounds were determined using gas chromatography with a mass detector in connection with an olfactometer. The salt content of the reformulated cheese ranged from 0.2 g of NaCl / 100 g of fresh cheese (usual content 0.8 g / 100 g) to 3.1 g of NaCl / 100 g of white cheese (usual content 3.5 g / 100 g). The representation of key odorants varied depending on the type of cheese, the salting time (a richer profile of aroma compounds in cheeses with a shorter salting time, i.e., with a lower amount of salt) and the ripening time (a richer profile of aroma compounds with a longer ripening time). Acetic acid, acetoin, diacetyl, octanoic acid, 2-butanone, butane-2,3-diol, acetone and heptane with different NIF (nasal impact frequency) for different types of cheese were determined as the main aroma compounds of the tested cheeses.

**Keywords:** reformulation, sodium chloride, cheese, key aroma compounds, olfactometry

**Acknowledgement:** Supported by the National Agency for Agricultural Research, Czech Republic (Project No. QK1910100), Programme ZEMĚ.

## P10 EXPLORING THE MAILLARD REACTION AND CAMELIZATION IN HONEY AND RELATED PRODUCTS

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Honey is known not only as a natural sweetener and preservative but also for its medicinal potential. Fresh honey is primarily composed of saccharides, specifically glucose and fructose in approximately a 1:1 ratio, with small amounts of maltose, sucrose, and other oligosaccharides. Under the conditions of the Maillard reaction and caramelization, saccharides are potential precursors of key reactive intermediates,  $\alpha$  dicarbonyl compounds ( $\alpha$ -DCs). In terms of the effects of  $\alpha$ -DCs on human health, their dual nature can be observed. On one side, an excess of  $\alpha$ -DCs can lead to the formation of advanced glycation end-products (AGEs) and may be connected with various lifestyle diseases such as cardiovascular diseases, Alzheimer's disease, and type 2 diabetes mellitus. On the other hand, products of  $\alpha$ -DCs are involved in the formation of food color and flavour, and the antibacterial effects of certain  $\alpha$ -DCs, such as methylglyoxal (MGO), can be also highlighted. MGO is naturally present in significant quantities in New Zealand manuka honey and has gained increased recognition among consumers in recent years. Among the  $\alpha$ -DCs formed during non-enzymatic browning reactions in honey and honey-based products, 3-deoxyglucosulose (3-DG) is the most commonly occurring compound. It leads to the formation of more stable products, especially 5-hydroxymethylfuran-2-carbaldehyde (HMF) and other compounds such as 2,3-dihydro-3,5-dihydroxy-6-methyl-(4H)-pyran-4-one (DDMP). Monitoring the content of HMF in honey can indicate the progress of the Maillard reaction due to heat treatment and aging. According to European directives, only honey with an HMF concentration below 40 mg/kg can be marketed. Monitoring  $\alpha$ -DCs formation in honey and honey-related products offers the potential for more accurate and earlier detection of heat treatment and/or ageing compared to the HMF approach. Additionally,  $\alpha$ -DC composition can help in detecting potential adulteration, such as the presence of unauthorized surrogates like glucose/fructose syrups. In this study, the profile and content of  $\alpha$ -DCs, HMF and DDMP in various honeys and honey products available on the Czech and Slovak markets, including mead and royal jelly, were analyzed. The aim was to compare the composition between different types of honey (mixed flower, monofloral, forest, creamed) and the effect of natural or accelerated ageing and heating of honey and mead to gain further insights into the Maillard reaction and caramelisation processes. Factors such as creaming, fructose/glucose ratio, color and minor saccharides were correlated with  $\alpha$ -DC content and compared with glucose/fructose syrups. Correlations with HMF and other chemical markers were also investigated for a more detailed understanding of the mechanism of  $\alpha$ -DC reactions. HPLC-PDA and LC-MS methods were used to analyze  $\alpha$ -DCs as quinoxaline derivatives.

**Keywords:** Maillard reaction,  $\alpha$ -dicarbonyl compounds, HMF, honey, mead

**Acknowledgement:** This work was supported from the grant of Specific university research - grant No A2\_FPBT\_2023\_006.

**P11****EFFECTS OF POLYPHENOLS ON VOLATILE PROFILE AND ACRYLAMIDE FORMATION IN A MODEL OF WHEAT BREAD SYSTEM****Maria Barbara Różańska<sup>1\*</sup>, Agnieszka Makowska<sup>2</sup>, Sylwia Mildner-Szkudlarz<sup>3</sup>**<sup>1 2 3</sup> Poznan University of Life Sciences, Poznań, Poland

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Aroma is one of the most significant sensory attributes in terms of product acceptability to consumers. Although thermal processing induces reactions that have significant effects on flavor and taste. Moreover, several unfavorable Maillard reaction-derived chemical hazards—such as acrylamide, heterocyclic aromatic compounds, and advanced glycation end products (AGEs) are formed. Phenolic compounds may reduce the contents of acrylamide, they may also adversely affect the volatile profiles of the final products. The formation of toxic and potentially carcinogenic acrylamide (GC-MS) and volatile compounds (SPME/GC-MS) through the Maillard reaction, lipid oxidation, and yeast fermentation was studied in model bread samples after the addition of phenolic compounds: (+)-catechin, quercetin, gallic, ferulic, caffeic acids at the level 0.1%, 0.5%, 1.0%, and 2.0% w/w. The addition of as little as 0.1% polyphenols to bread significantly reduced acrylamide (16.2%–95.2%). Our results indicated that quercetin, (+)-catechin, and ferulic acid exhibited inhibitory effects at the lowest concentration (0.1%), while gallic acid and caffeic acid did so at the highest level (2.0%). Of all the phenolic compounds, ferulic acid showed the highest level of acrylamide inhibition regardless of concentration. Although the phenolic compounds mitigate acrylamide, this adversely affected bread volatile profile. The results showed ferulic acid was the most effective antioxidant at reducing the level of pleasant components derived from fermentation (~42%). At the highest level (2.0%), caffeic acid most significantly suppressed Maillard-type volatiles (75.9%), followed by gallic acid (74.3%), ferulic acid (65.6%), (+)-catechin (62.4%), and quercetin (59.3%). The most promising additive seems to be (+)-catechin, as it exerted the most significant inhibitory effect on acrylamide formation at the lowest treatment level, it is thus possible that lower concentrations will continue to have the reducing effect without altering the generation of flavors.

**Keywords:** phenolic compounds, bread, acrylamide, GC-MS, Maillard reaction**Acknowledgement:** This study was financially supported by the National Science Centre, Poland (Project No. 2013/09/B/NZ9/01626).

**P12****CHANGES IN CHEMICAL COMPOSITION AND OXIDATIVE STABILITY OF COLD-PRESSED OILS OBTAINED FROM BY-PRODUCT ROASTED BERRY SEEDS****Maria Barbara Róžańska<sup>1\*</sup>, Sylwia Mildner-Szkudlarz<sup>2</sup>**<sup>1 2</sup> Poznań University of Life Sciences, Poznań, Poland

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Fruit by-products have recently attracted increased attention on account of their health benefits. Fruit seeds are a major by-product of the food industry, and creating new uses for them by converting them to value-added products would prevent their disposal as waste, and would promote sustainable and competitive worldwide production. Cold-pressed oils from fruit seeds are among the richest sources of natural microconstituents such as polyunsaturated fatty acids (PUFA), carotenoids, polyphenols, phytosterols, tocopherols, and other compounds with high biological activity. However, it should be noted that the oxidation of PUFAs is the main reaction that decreases oil quality. Therefore, roasting seeds (15 min at 140 °C) prior to oil pressing may be considered a promising technique to improve the oil's oxidative stability—especially oils that contain almost 95% of PUFA. This study aimed to determine the effect of seed roasting on compositional parameters and oxidative stability of cold-pressed blackcurrant, raspberry, chokeberry, and strawberry seed oils, with rapeseed oil as a reference sample. The chemical characterization of cold-pressed berry oils included peroxide value (PV), acid value (AV), and p-anisidine value (p-AnV), fatty acid compositions, phytosterols, tocopherols, and phenolic compounds determination. The differential scanning calorimetry (DSC) technique was used to study the process of oxidation of the oil samples. Roasting seeds decreased the peroxide concentration by about 50%, while secondary oxidation products increased by 9%–91%. There were no significant differences in the fatty acid composition of the berry seed oils following roasting. In addition, no significant decrease in the compositional parameters or oxidative stability of roasted rapeseed oil was found. Results of DSC analyses showed that rapeseed oil from both roasted and unroasted seeds possessed the longest induction time, with the shortest being recorded for chokeberry (unroasted seeds). The raspberry seed oil had similar oxidative stability to rapeseed oil, probably due to the high concentration of  $\gamma$ -T,  $\delta$ -T, and ellagic acid possessing high antioxidant activity. Our results indicated that roasting seeds prior to pressing them for oil had a positive effect on the oil's stability and may contribute to the design of the cold-pressing process, helping to increase the stability of these novel oils and, as a consequence, all owing commercial use in various applications.

**Keywords:** berry by-product, fatty acids, tocopherols, roasting**Acknowledgement:** This study was financially supported by the National Centre of Science, Poland (Project No. 2017/27/N/NZ9/00905).

**P13****ASSESSMENT OF CHLORINATED PARAFFINS AS PRECURSORS OF 2- AND 3-CHLOROPROPANEDIOL ESTERS IN REFINED VEGETABLE OILS**

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Fatty acid esters of 2- and 3-chloropropanediol (2- and 3-MCPDEs) are a group of heat-induced contaminants, occurring primarily in refined oils, fats, and their products. They are formed in a high-temperature environment, such as vegetable oil deodorization. Regarding the toxicity of MCPDEs, there is a potential health concern among high-consumer groups of younger age. In the worst-case scenario, infants receiving only formula may slightly exceed the safe level (EFSA 2018). Since 2021, the maximum limits for 3-MCPDEs are in force. In recent years, considerable efforts have been made to elucidate the mechanisms of their formation, including the identification of their precursors, especially chlorine donors. Previous studies have shown that in addition to inorganic chlorine compounds, organochlorine compounds (present in crude oils mostly as exogenous contaminants), which decompose at elevated temperatures, can contribute to MCPDEs formation. However, their structure, origin, and potential role in this process are yet to be fully understood. Therefore, the aim of this study was to assess the potential of one group of lipophilic environmental contaminants, chlorinated paraffins (CPs), which are unstable at elevated temperatures (HCl dechlorination takes place) to contribute to the formation of MCPDEs. Laboratory-scale model systems representing bleached vegetable oils contaminated with technical mixture and individual CPs were designed and subjected to constant heat treatment (230 °C for 2 hours) to simulate the deodorization process. Gas chromatography coupled to tandem mass spectrometry (GC-MS/MS) was used for the targeted analysis of the total 2- and 3-MCPD content after hydrolysis. CPs were analyzed, after isolation on a silica microcolumn, by gas chromatography coupled to high-resolution mass spectrometry (GC-HRMS). A substantial increase in MCPDEs levels (up to 3 times the control samples) was observed. Their formation seems to correlate very well with CPs concentration. Furthermore, the decomposition profile and individual contribution of selected CP congeners were discussed. Based on the generated data, we can conclude that processing of plant oils contaminated by CPs, might result in elevated concentrations of MCPDEs.

**Keywords:** processing contaminants, vegetable oils, MCPD esters, chlorinated paraffins, deodorization

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**P14****COMPARISON OF VOLATILE PROFILES OF CARROT CHIPS PRODUCED BY DEEP AND VACUUM FRYING USING GAS CHROMATOGRAPHY COUPLED TO HIGH RESOLUTION MASS SPECTROMETRY (GC-HRMS)**

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Over the last decade, the consumer's interest in healthier food has been constantly growing. As a result, food products labelled as "heathy food" appear on the market. Among these products are, for example, vegetable chips. However, the vegetable chips produced by deep frying could be even less heathy than potato chips. Many studies reported high content of acrylamide and other processing contaminants in vegetable chips. For this reason, the alternative technology of frying, which allows to produce sensory satisfying product and eliminate the formation of process contaminates, is required. Among these alternative frying technologies is vacuum frying. In this study, solid-phase microextraction followed by gas chromatography coupled to high-resolution mass spectrometry (SPME-GC-HRMS) was applied for evaluation of volatile profiles of carrot chips produced by deep and vacuum frying. Samples were represented by carrot chips produced from orange, purple and yellow carrots, within the study was also investigated the influence of frying time on volatile profiles of carrot chips (1, 1.75, 2 min for deep frying and 4, 5, 6 min for vacuum frying. The data obtained using SPME-GC-HRMS analysis were handled by chemometric methods such as principal component analysis (PCA) and partial least squares discriminative analysis (PLS-DA). The chemometric evaluation of the data showed good separation of the samples into groups both according to the type of frying and according to the type of carrot. Among other things, the statistical processing of the data enabled the selection and identification of characteristic substances corresponding to the frying technology, which in the case of vacuum frying were terpenic substances and in the case of deep frying, products of the Maillard reaction.

**Keywords:** gas chromatography, mass spectrometry, carrot chips, volatile profiles, vacuum frying

**Acknowledgement:** This work was supported from the grant of Specific university research - grant No A1\_FPBT\_2023\_002

## P15 THE FATE OF GLYCOALKALOIDS DURING THE CULINARY PROCESSING OF POTATOES

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Potatoes (*Solanum tuberosum*) are an important world crop and raw material for the production of popular food products. They are associated not only with nutritionally significant components, but also the presence of toxic glycoalkaloids (GAs), specifically  $\alpha$ -solanine and  $\alpha$ -chaconine, is described in them. These toxic metabolites can have an adverse effect on the consumer's health at higher intakes. The European Commission therefore recommended (2022/561/EU) to monitor the concentrations of these naturally occurring substances in potatoes and their products in order to obtain a high-quality data set for health risk assessment. During the processing of potatoes, degradation of the original substances occurs and therefore it is necessary to study their degradation products, such as  $\beta$ -solanine/chaconine,  $\gamma$ -solanine/chaconine and their aglycone solanidine. This study aimed to monitor the spectrum of GAs in potato products (n = 20) purchased in the market network of the Czech Republic. A U-HPLC-HRMS method was developed and validated, which also enabled the monitoring of target analytes for which no analytical standard was available. Some concentrations in the analyzed sample exceeded the indicative level of the EU Commission recommendation of 100 mg/kg for the sum of  $\alpha$ -solanine and  $\alpha$ -chaconine, the sum of the concentrations in all these samples was in the range of 32 to 144 mg/kg. In addition, this study also paid attention to the influence of the culinary processing of potatoes. High levels of signals corresponding to the target analytes were detected in samples of fried crisps, degradation products were also observed after scraping, grating or slicing raw potatoes. This part of the here presented research led to the acquisition of an important complex data set summarizing the presence of hydrolysis products of the original glycoalkaloids.

**Keywords:** glycoalkaloids, liquid chromatography, mass spectrometry, degradation products, potatoes

**Acknowledgement:** This work was supported from the grant of Specific university research - grant No. A2\_FPBT\_2023\_003.



**P16****POTENTIAL APPLICATION OF BAG ON VALVE TECHNOLOGY IN THE FOOD INDUSTRY. A CASE STUDY: OLIVE OIL**

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Background: Packaging technology, has experienced a notable transformation over past decades, especially for aerosol applications. In this context, Bag on Valve (technology) stands out as a new packaging alternative, based on the principle of compartmentalized containers. BOV technology has been mainly implemented in packaging of personal care products. This new system provides improved functional and safety properties, as well as providing added value to the product, compared to conventional aerosols. Consequently, this packaging technology has been gaining much interest in the Food Industry. Objectives: The main goals of this review were: a) description of the BOV technology; b) comparative effects of conventional and BOV packaging systems on chemical, nutritional and bioactive food properties, with a special focus on the olive oil industry. Methods: This work was conducted according to the methodology of systematic review. The reviewed articles were compiled using the electronic databases Web of Science and Scopus. Combinations of the keywords packaging, aerosol, olive oil, filling processes, and bag on valve were chosen for the search. Articles were selected by either verifying the title and abstract or reading the full text. Results: A BOV aerosol package consists in two compartments, an outer container made up of an aluminium or tin can and an internal flexible pouch with several layers and welded to the valve. Once the valve is crimped into the can, the inner bag is filled with a propellant which can be an inert gas such as compressed air or nitrogen [1]. Concerning olive oil industry applications, the study conducted by Torres-Robles et al. [2] was focused on the effect of different types of containers on quality and phenolic content of extra virgin olive oil (EVOO) stored for 1 year under light or dark conditions. The results demonstrated that light exposure decreased the physicochemical and sensory properties and significantly reduced the total phenolic content, particularly in those stored in PET containers. Kishimoto [3] evaluated the storage of EVOO in glass bottles with different levels of transparency, concluding that the best packaging materials were glass bottles covered with aluminium foil. In contrast, De Leonardis et al. [4] studied the storage of olive oil in a bag-in-box system during 24 months, concluding that this package was able to preserve the quality indices within the established values for EVOO. Conclusion: The BOV system has emerged as an innovative aerosol packaging system with potential applications in Food Industry. The case study of the EVOO has shown that BOV technology could extend the shelf life within the same commercial category. Future studies are required to evaluate more potential applications of BOV in the packaging of spray food products.

[1] Niemiec K, et al. 2018

[2] De la Torre-Robles A, et al. 2019

[3] Kishimoto, N 2019; (4) De Leonardis A, et al. 2021

**Keywords:** packaging, aerosol, olive oil, filling processes, bag on valve

**Acknowledgement:** OTRI-UGR SCHOLARSHIP NUMBER P37B/22/02

**P17****APPLICATION OF NEW TECHNOLOGIES IN THE OLIVE OIL INDUSTRY FOR NUTRITIONAL AND BIOACTIVE QUALITY IMPROVEMENT**

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**Background:** In recent years the consumer interest in functional foods has increased. In this way, industry investigation has been focused on the development and application of different stabilization and encapsulation techniques to the treatment and incorporation of hydro- and lipophilic bioactive compounds in different food matrices. Among the different foods, virgin olive oil provides a target matrix for this development according to its composition, health benefits, and daily consumption. **Objectives:** A literature review was conducted on new technologies applied to incorporate bioactive components into the olive oil industry with two goals: to improve the nutritional quality and to introduce and formulate new functional ingredients.

**Methods:** Articles were selected based on a scientific literature review using the electronic databases Pubmed, Scopus, and Web of Science. Searches were conducted using combinations of the terms edible oils, new process, food processing, formulations, innovation, emulsion, and oleogel. Articles were selected by either verifying the title and abstract or reading the full text.

**Results:** on the one hand, the application of emerging technologies in the extraction of olive oil as Electric Pulse, High Pressure Processing, Ultrasound Technology, and Microwave Heating technologies, improves the content of phenols, phytosterols, and tocopherol while preserving the organoleptic characteristics [1]. However, the extent of improvement appears to be dependent on the specific technology and process conditions used, highlighting the need for further research in this area. Iqdiyam et al. [2] found that the integration of high-power ultrasound and control of oxygen concentration within the malaxation headspace could have the potential to enhance various aspects of extra virgin olive oil quality, including chemical indices, antioxidant compounds, and sensory attributes, which is an excellent way to extend its shelf life and improve its nutritional profile. On the other hand, the formulation of olive oil with extracted compounds from different parts of medicinal and aromatic plants has also been proposed to improve the nutritional profile, extend shelf life and enhance the oxidative stability of olive oil during long-term storage [3]. Pinto et al. [4], discusses the applications of oleogel in order to deliver functional molecules in foods. Single and double emulsions have been used, but W/O/W has the best potential for protecting bioactive compounds from food.

**Conclusion:** Advanced technologies and bioactive compounds have promising implications for improving the quality, and stability of virgin olive oil. According to scientific and industrial demands, these innovations offer valuable opportunities for developing healthier and more functional virgin olive oil products.

[1] Pérez M et al., 2021;10(3):417

[2] Iqdiyam BM et al., 2019;244:1-10

[3] Gharby S et al., 2022;11(20):3258

[4] Pinto TC, et al., 2021;7(3):86

**Keywords:** edible oil, new process, emulsion, oleogel, food processing

**Acknowledgement:** OTRI-UGR SCHOLARSHIP NUMBER P37/21/04.

## **P18**

### **STUDIES ON THE METABOLIZATION OF DIETARY AMADORI PRODUCTS BY PROBIOTIC BACTERIA**

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Our predominantly thermally treated food contains larger quantities of various Maillard reaction products (MRPs) which we ingest with our daily diet. The most important group in terms of quantity are the Amadori products (APs), of which we ingest up to 1 g per day [1]. APs are formed in the first stage of the Maillard reaction from reducing sugars and amino components by Amadori rearrangement. Preliminary work has already shown that only a very small proportion of the ingested amount of APs can be recovered in urine and feces [2]. Thus, Amadori products enter the colon largely unmodified, where they encounter a variety of different bacterial species - the colonic microbiome [3]. To date, there is very little information on how these substances are metabolized by the intestinal bacteria. Exactly which bacterial species are responsible for their degradation has only been investigated rudimentarily so far. One group of microorganisms that is positively discussed in the context of intestinal health are the so-called probiotic bacteria (e.g. *Lactobacillus plantarum*, *Bifidobacterium bifidum*), which are available as capsules from pharmacies or online shops. To investigate whether Amadori products can be metabolized by probiotic bacteria, various commercially available probiotic preparations, as well as single pure strains thereof, were incubated anaerobically for 72 h in a minimal medium containing free N-ε-fructosyl-lysine (FruLys). Analysis of degradation or formation of metabolites was performed by LC-MS/MS and GC/MS. Of the nine preparations tested, one was able to completely degrade the Amadori product within 72 h. Among others, free lysine could be quantified as a metabolite. Through model incubations with individual pure strains of the respective preparation, three new deglycating bacterial species were identified. Apparently, it is possible for certain probiotic bacteria to utilize alimentary Amadori products as a carbon source.

[1] T. Henle, *Kidney Int. Suppl.* 2003, 63, 145-147.

[2] C. Delgado-Andrade et al., *Amino Acids.* 2012, 43, 595-602.

[3] M. Snelson, M.T. Coughlan *Nutrients.* 2019, 11

**Keywords:** Maillard Reaction, Amadori products, probiotic bacteria, metabolization

## P19 FORMATION AND DEGRADATION OF FREE GLYCATION PRODUCTS DURING THE BREWING PROCESS

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The glycation products formed by the Maillard reaction between amino components and reducing sugars during the thermal treatment of food are precursors for flavor compounds and color-forming substances. In beer production, malt already contributes to a baseline level of glycation products [1]. An inventory of individual glycation products in beer has already been conducted in our research group [2], where dicarbonyl compounds were identified as relevant precursors for aging flavors [3]. However, little is known about the influence of individual process steps during malt production and the brewing process on the formation and degradation of individual glycation products. In this study, beer was produced using a small-scale brewing system in a 10 L laboratory scale, and the free Amadori products, dicarbonyl compounds, and late-phase glycation end products (AGEs) were monitored throughout the entire process. The quantification of Amadori products and AGEs was performed using LC-QQQ-MS with specific mass transitions. Dicarbonyl compounds were detected using HPLC-UV after derivatization with ortho-phenylenediamine. It was shown that the choice of malt had the greatest influence on the concentration of glycation products. During the brewing process, all Amadori products showed similar effects, while dicarbonyl compounds and AGEs behaved differently from each other. For example, an increase in 3-deoxyglucosone was observed during mashing, while concentration changes of other analyzed compounds (such as Amadori products, 3-deoxymaltosone, and the AGE pyrraline) were not significant. On the other hand, wort boiling led to a decrease in Amadori products, while 3-deoxyglucosone and MG-H1 showed an increase in concentration. Wort boiling and fermentation had the greatest influence on the levels of Maillard compounds, while variations in mash procedures had little noticeable effect. The choice of yeast also has an impact on the levels of Maillard compounds in the final product, as confirmed in recent studies [4].

[1] Hellwig & Henle (2020) Maillard Reaction Products in Different Types of Brewing Malts. *Journal of Agricultural and Food Chemistry*, 68: 14274-14285.

[2] Hellwig et al. (2016) Free and Protein-Bound Maillard Reaction Products in Beer: Method Development and a Survey of Different Beer Types, 38: 7234-7243.

[3] Nobis & Kwasnicki et al. (2021) The Influence of De Novo Formation of Aging Aldehydes 10: 2668

[4] Kertsch et al. (2023) *European Food Research and Technology*, 249:103-118

**Keywords:** Maillard, glycation, beer, malt, brewing

## P20 FORMATION AND MS BASED IDENTIFICATION OF PYROGLUTAMATE CONTAINING PEPTIDES IN ROASTED COCOA

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Cocoa has become the target of increased scientific research as consequence of its beneficial properties for human health. A complex mixture of various compounds identified in cocoa beans has an influence on the aroma and taste of chocolate. Identification of cocoa-related compounds is by far not complete. Recently, pyroglutamic acid has been detected as a so far overlooked novel compound in cocoa roasted samples. Pyroglutamic acid (pGlu) also known as pidolic acid or 5-oxoproline, is a bioactive glutamine derivate that can variably change the aroma of food and beverages, preserve the quality and nutritional value of food. During fermentation, aroma precursors, such as free amino acids, short-chain peptides and reducing sugars are formed. Cocoa bean roasting is a critical step in chocolate processing, where the typical aroma through Miallard reaction and texture are created<sup>1</sup>. The generation of pGlu was reported to be directly related to roasting step by cyclization of glutamic acid. Previous studies showed that pGlu has pharmacological properties and has antimicrobial and antitumoral, mitogenic, and antidiabetic activities. Since pGlu is a ubiquitous but never studied in cocoa beans natural amino acid derivative, this work is focused on identification of pGlu in roasted cocoa beans from Ecuador and Ivory Coast. As the cyclization of N-terminal glutamine is accelerated by increasing the temperatures, the roasted cocoa beans were chosen for this study. The samples were extracted, and analyzed with LC-MS/MS using UPLC-ESI-Q-q-TOF (Bruker, USA). All the raw MS/MS data were imported to PEAKS Studio 11.0 (BSI, Waterloo, Ontario, Canada) for peptide identification with pGlu modifications. The variable modifications were set to include Gln->pyro-Glu (N-terminal). The parent mass error tolerance was set to 10 ppm and the fragment mass error tolerance to 0.02 Da. MS/MS data were processed against a database of UniProtKB (<http://www.uniprot.org/>) Theobroma cacao. Peptides were identified, sequenced by MS/MS and annotated based on their characteristic fragmentation pattern in the positive-ion mode<sup>2</sup>. For the roasted samples from Ecuador, it was identified the range of peptides from 535 - 1371 with 6.5-8.3% confirmed peptides sequences containing pGlu. The roasted beans from Ivory Coast contained higher number of peptides in a range of 1285 to 1696 with higher percental amount of pGlu ranging from 8% to 13%. Another distinctive feature for the samples from Ivory coast is that the major of abundant peptides contain 4-6 amino acids while for Ecuadorian samples the peptides include 4-14 amino acids.

1. Andruszkiewicz PJ, D'Souza RN, Corno M, Kuhnert N. [doi.org/10.1016/j.foodres.2020.109164](https://doi.org/10.1016/j.foodres.2020.109164)

2. D'Souza RN, Grimbs A, Grimbs S, Behrends B, Corno M, Ullrich MS, Kuhnert N. [doi: 10.1016/j.foodres.2018.04.068](https://doi.org/10.1016/j.foodres.2018.04.068).

**Keywords:** cocoa bean, pyroglutamic acid, roasting, tandem MS

**P21****INFLUENCE OF MEDIUM QUALITY FOR LIPID OXIDATION AND ALPHA-TOCOPHEROL CONCENTRATION ON THE FORMATION OF OXIDATION PRODUCTS****Iveta Hradkova<sup>1\*</sup>, Barbora Soukupova<sup>2</sup>, Marketa Bercikova<sup>3</sup>, Vladimir Filip<sup>4</sup>**<sup>1 2 3 4</sup> University of Chemistry and Technology Prague, Prague, Czech Republic

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Lipid oxidation is the most widely spread reaction together with the Maillard reaction in food. During the antioxidant activity study, the choice medium for oxidation is problematic. The medium should contain the minimum of naturally occurring antioxidants that change overall results (antioxidant activity of naturally occurring antioxidants is determined with added antioxidant), and also the fatty acid composition of the medium must also be suitable for lipid oxidation - the presence of polyunsaturated fatty acids is required. In this study, sunflower oil was chosen as the raw material suitable for lipid oxidation. Sunflower oil was converted to fatty acid methyl esters, and then distilled at low pressure and high temperature (105 and 135°C) to maximise the removal of naturally occurring antioxidants, mainly tocopherols. The degree of lipid oxidation was monitored at storage temperature 60°C with added alpha-tocopherol at concentrations of 0,75 and 1,5 mmol/kg by conjugated diene content, p-anisidine value, induction period (Rancimat) at 120°C, 9-oxononanoic acid methyl ester concentration. The medium for lipid oxidation, which was distilled at higher temperature (135°C), contained an undetectable amount of naturally occurring tocopherols, but during distillation, secondary oxidative products (2-alkenals) formed to a great extent. The concentration of 0,75 mmol/kg of added alpha-tocopherol showed the maximum antioxidant effect, while the concentration of 1,5 mmol/kg of added alpha-tocopherol had a pro-oxidant effect.

**Keywords:** lipid oxidation, alpha-tocopherol, naturally occurring antioxidants**Acknowledgement:** This work was supported by funding from the Ministry of Agriculture of the Czech Republic (NAZV QK22010135)

**P22****COMPARISON OF DIFFERENT EXTRACTION TECHNIQUES FOR DETERMINATION OF VOLATILE COMPOUNDS OF RAPESEED OILS**

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The aim of this study is the comparison of four extraction techniques for the isolation of volatile compounds from rapeseed oils. The headspace solid-phase microextraction (HS-SPME), steam distillation (SD), distillation on a thin-film evaporator and the solvent-assisted flavour evaporation (SAFE) technique were used for extraction. The prepared extracts were analysed using gas chromatography coupled to mass spectrometry and the olfactometric detector (GC-MS-O). The extraction step is one of the most important parts of the analysis, therefore in this study, significant attention is paid to the advantages and limitations of individual extraction techniques. There were considerable differences in the volatile profile among the extracts obtained. The largest number of volatile compounds (60) and key aroma compounds (31) was identified in the extract obtained by thin-film evaporator. On the contrary, the lowest number of volatile compounds (24) and key aroma compounds (5) was detected using the HS-SPME technique. The major disadvantage of SD was thermal degradation and formation of thermal artifacts. The results of the study show that the key compounds responsible for the aroma of rapeseed oils were  $\alpha$ -pinene, oct-1-en-3-one, 4-isothiocyanato-1-butene and 2-isopropyl-3-methoxypyrazine. All the above mentioned techniques can be used for the analysis of key odorants of rapeseed oils, but it is more advantageous to prepare the extract using a thin-film evaporator or the SAFE technique. The observed differences among the results show the importance of using at least two extraction techniques simultaneously to get a complete range of volatile compounds. This study provides useful information on the volatile profile of rapeseed oils and the use of individual extraction techniques. A similar approach could be used for other types of food.

**Keywords:** volatile compounds, extraction, SPME, SAFE, steam distillation

**Acknowledgement:** Supported by the National Agency for Agricultural Research, Czech Republic (Project No. QK22010135), Programme ZEMĚ.

## **P23 FERMENTATION OF VEGETABLE AND VEGETABLE-FRUIT JUICES WITH USE OF STARTER CULTURES**

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The growing interest in plant-based food and beverages creates space in the market for the development of new vegetable and fruit products. One of these innovations is fermented juices or smoothies, which are called "Lactojuice". The main microorganisms used in the fermentation of plant-based beverages are lactic acid bacteria, which produce several substances with positive effects on human health and at the same time decompose antinutrients. This type of beverages is also an alternative to the dairy products, but presents similar health-promoting properties, is cholesterol and lactose free, and also, satisfy vegan, vegetarian, or allergic consumers. From another point of view these fermented beverages provide the solution the large wastage of vegetables and fruits issue by increasing shelf life of these commodities. The aim of this work was the development of the vegetable and vegetable-apple juices fermented by the single strain culture of bacterium *Lactiplantibacillus plantarum* CCDM 181 and the mixed starter culture VEGE 047 (Danisco®). The juices were evaluated in terms of pH value, organic acids content (lactic, acetic), microbial quality (number of lactic acid bacteria, yeasts, moulds) and sensory properties. The results show that adding the starter cultures to the vegetable and vegetable-apple juices leads to a rapid reduction in pH, which is crucial for suppressing spoilage microorganisms. The pH value is stable within 72 hours at 30 °C. The samples with *Lpb. plantarum* CCDM 181 after fermentation contain a higher amount of lactic acid and acetic acid than the samples with the mixed starter culture VEGE 047. Significantly lower levels of the acids are in vegetable-apple juice samples, which is caused by low pH of apple juice that suppressed the metabolic activity of lactic acid bacteria. On the other hand, the best sensory properties were observed in the vegetable-apple juice inoculated with the mixed starter culture. Fermentation of vegetable juice and vegetable-fruit juice using the starter cultures can lead to the products that are especially interesting for the content of health-promoting substances such as acetic and lactic acid. The addition of sweet fruit juice is desirable to improve sensory properties, but from the technological point of view, it should be added at the end of the fermentation process.

**Keywords:** lacto-fermented juices, fermentation, lactic acid bacteria, starter culture



**P24****MAILLARD REACTION PRODUCTS IN COMPLEX FOOD SAMPLES: HOME MADE VERSUS CANNED FOOD****Theresa Kaeubler<sup>1\*</sup>, Friederike Manig<sup>2</sup>, Thomas Henle<sup>3</sup>**<sup>1 2 3</sup> Chair of Food Chemistry, Technische Universität Dresden, Dresden, Germany\*Corresponding author - E-mail: [theresa.kaeubler1@tu-dresden.de](mailto:theresa.kaeubler1@tu-dresden.de), Phone: +49(0)351-463-32618

Maillard reaction products (MRPs) are formed during the non-enzymatic reaction between amino compounds and reducing sugars by thermal processes, storage and preservation of food. MRPs shape the sensory characteristics with regard to taste, scent and color, but are also discussed in the context of the pathogenesis of metabolic diseases [1], culminating in recommendations for a "MRP-free" diet or avoidance of industrial processed food [2]. It is frequently postulated that a consumption of industrially processed foods leads to a higher intake of MRPs [3]. However, little is known about the quantitative relevance of dicarbonyl compounds and glycated amino acids in home-made and/versus industrially processed food. For the present study, 13 commercially available canned food meals were recreated by home cooking with respect to main ingredients and energy intake. For both meal groups, individual MRPs were analyzed i) after cooking/processing and cooling and ii) after ready-to-eat heating. Quantitation of the dicarbonyl compounds methylglyoxal (MGO), glyoxal (GO), 3-deoxyglucosone (3-DG), 3-deoxygalactosone (3-DGal) as chinoxalines after derivatization with o-phenyldiamine and hydroxymethylfurfural (HMF) was performed by HPLC-UV [4]. The glycated amino acids (pyrraline, N $\delta$ -(5-methyl-4-oxoimidazolin-2-yl)-ornithin (MG-H1), N-(4-methyl-5-oxo-1-imidazolin-2-yl)sarcosine (MG-HCr), N $\epsilon$ -(carboxyethyl)lysine (CEL), N $\epsilon$ -(carboxymethyl)lysine (CML)) were analyzed via LC-MS/MS by using the respective isotopologues as internal standards [5]. It could be shown that no pronounced differences were observed between the MRP content of a home-made and the corresponding canned food item. The widely held thesis, both in public and in scientific communities, that industrial production leads to a higher heat load of food and thus to a higher daily intake of MRPs cannot be sustained.

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[3] Dr. Vlassara's A.G.E.-Less Diet: How Chemicals in the Foods We Eat Promote Disease, Obesity, and Aging and the Steps We Can Take to Stop it. H. Vlassara, S. Woodruff, G. E. Striker, Garden City, Square One Publishers, 2016.

[4] J. Degen, M. Hellwig, T. Henle, *J Agr Food Chem*, 60 (2012) 7071.[5] F. Manig, M. Hellwig, F. Pietz, T. Henle, *Foods*, 11 (2022) 2112.**Keywords:** Maillard reaction products, home-made and/versus industrially processed food**Acknowledgement:** This study is embedded in the EU-wide ePIDEMic project under the umbrella of JPI ERANET HDHL and was supported by the BMBF grant no. 01EA1909.

**P25****HOMOCITRULLINE - DOES IT PLAY A ROLE IN FOOD?**

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Posttranslational modifications of food proteins are often mentioned in the context of the Maillard reaction or oxidative processes. An underinvestigated aspect is the scenario of carbamylation. Free or protein-bound lysine may be modified under certain conditions to form homocitrulline, a structural analogon of citrulline prolonged by a methylene group. The majority of publications about homocitrulline deal with homocitrulline as physiological marker of a plenty of diseases (Jaisson et al. 2018). However, literature about homocitrulline in food is rare. To our knowledge, the presence of homocitrulline in food was only described for heated milk and, by adding urea, the formation of homocitrulline could be observed in a time- and temperature-dependent manner (Metwalli et al., 1998). The rationale of our project was thus to analyze homocitrulline in a broad range of food items and to test homocitrulline as a marker for heat treatment and maturation. The analysis was performed by using high pressure liquid chromatography coupled to mass spectrometry (LC-MS/MS) as stable isotope dilution analysis after enzymatic hydrolysis of food items. As food samples, e. g. dairy products, meat and meat products, vegetarian substitutes and cereals were included. Concentrations in foods ranged from <LOD to 80 mg/kg protein for vegetarian meat balls and up to 580 mg/kg of protein for condensed milk. In milk, plant drinks and meat, an impact of heat treatment could be detected. An increase in the homocitrulline concentration during maturation was observed for cheese, but not in meat products after storage.

In conclusion, our study revealed a broad incidence of homocitrulline in different food items such as milk, meat and meat products, nuts, cheese and legumes.

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[2] A. Metwalli, W. L. Lammers, M. Boekel, *Journal of Dairy Research* 65 (1998) 579.

**Keywords:** homocitrulline, urea, lysine, posttranslational modification, carbamoylation

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**P26****THE EFFECT OF CULINARY TREATMENT ON THE CONTENT AND FORMS OF VITAMIN B9 IN SPINACH**Lenka Libenská<sup>1\*</sup>, Lucie Drábová<sup>2</sup>, Jana Pulkrabová<sup>3</sup><sup>1 2 3</sup> University of Chemistry and Technology Prague, Department of Food Analysis and Nutrition, Prague, Czech Republic

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Vitamin B9, also called folates or folic acid, is a group of hydrophilic essential substances that are known for their importance in proper fetal development, especially in the early stages of pregnancy[1-3]. However, this vitamin is indispensable for all individuals across the population[3]. The name 'folic acid' comes from the Latin (folia translate as leaves) and it implies that leafy vegetables – such as spinach – are a good source of this vitamin. Unfortunately, vitamins are not stable compounds, and therefore significant losses have been observed for vitamin B9 during storage and food processing[1, 4, 5]. As food production processes evolve and are optimized, the question arises as to how the changes made will affect the content of nutritionally important components.

In this study the most known dietary folates source was studied. Baby spinach leaves (*Spinacia oleracea*) were either analysed fresh or after freezing (-18 °C, 24 h), drying (50 °C, to constant weight), blanching (2 min in hot water) or boiling (100 °C, 2 min). For determination of total folate content was used optimized and with BCR 485 (Mixed vegetables reference material) verified method using acidic hydrolysis and liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS). For observing the percentage of naturally occurring folate forms were samples measured without hydrolysis. To determine the changes in folic acid (FA) and 10-formylfolic acid (10-FFA) content were samples hydrolysed with acetic acid and alpha-amylase. Changes in 5-methyltetrahydrofolate (5-MTHF) and 5-formyltetrahydrofolate (5-FTHF) content were measured in samples hydrolysed only with acetic acid.

The average content of vitamin B9 in fresh baby spinach leaves is  $10.6 \pm 0.1$  µg/g dry matter. Sample freezing caused significant decrease in the content of 5-FTHF, while drying caused decrease in 10-FFA content accompanied by increase in both 5-MTHF and 5-FTHF. The highest folate loss was observed in boiled spinach leaves.

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2. Öncü-Kaya, E.M., Determination of Folic Acid by Ultra-High Performance Liquid Chromatography in Certain Malt-based Beverages after Solid-Phase Extraction. *Celal Bayar Üniversitesi Fen Bilimleri Dergisi*, 2017: p. 623-630.
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5. Biezanowska-Kopec, R., et al., Effect of storage conditions on folic acid content in folic acid-fortified bread - A model study. *Polish Journal of Food and Nutrition Sciences*, 2010. 60: p. 147-151.

**Keywords:** vitamin B9, vegetables, food processing, folates, LC-MS/MS

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**P27****EFFECTS OF STORAGE TEMPERATURE AND GRAIN FORM ON THE VOLATILE ORGANIC COMPOUNDS IN FRAGRANT RICE****Kanphassorn Wimonmuang<sup>1</sup>, Young-Sang Lee<sup>2\*</sup>**

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Fragrant rice is globally esteemed for its market value, and its intricate aroma is vital for sensory properties. However, long-term storage of fragrant rice poses challenges as unstable flavor-determining volatiles may degrade, and off-flavor-inducing volatiles may accumulate. To investigate storage-induced volatile changes, the 'Chunhyejinseonhyang' fragrant rice was stored in brown and paddy rice forms at 4, 10, 15, and 25°C up to 12 months, and subsequent changes in volatiles were evaluated by Headspace-Solid Phase Microextraction coupled with Gas Chromatography-Mass Spectrometry. A total of 60 volatile compounds were identified, and the temperatures, grain form, and duration of storage significantly affected their compositions. Based on their semi-quantitative time-series changing patterns during storage, volatiles could be categorized into seven groups: 12 continuously increased (e.g., 1-pentanol), 1 not significantly changed (e.g., tetradecane), 5 newly produced (e.g., (E)-2-octenal), 6 disappeared (e.g., 2,6-lutidine), 23 initially increased and then decreased (e.g., decanal), 12 inconsistently changed depending on storage conditions (e.g., 1-octen-3-ol), and 1 continuously decreased (e.g., 4,6-dimethyldodecane). The relative amount of 2-acetyl-1-pyrroline (2AP), the most important flavor-determining volatile of rice, in brown rice stored at 4°C rapidly increased by 24% during the initial one month of storage, followed by a continuous decrease up to 12 months by 45% compared to pre-storage. Under high (25 °C) temperature conditions, however, 2AP gradually decreased, resulting in a 53% loss within 30 days and over 90% loss after 12 months of storage regardless of storage grain forms. Storing rice in brown rice form at high temperature readily increased off-flavor inducing volatiles, such as aldehydes, alcohols, and furan volatiles, which may lead to deterioration in the quality of fragrant rice. All these results suggest that storing fragrant rice in rough rice form at a temperature lower than 15 °C is appropriate to minimize alterations in volatiles and subsequent market quality of fragrant rice.

**Keywords:** volatile organic compound, fragrant rice, rice storage, 2-acetyl-1-pyrroline

## P28 MODIFICATION OF CASEIN MICELLES AS POTENTIAL NANOCARRIER

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Caseins ( $\alpha$ -,  $\beta$ -,  $\kappa$ -casein), which belong to the phosphoprotein family, are an important component of proteins in mammalian milk. Individual caseins are predominantly present in colloidal aggregates called micelles. [1] Casein micelles are animal species-specific in size, structure, and composition. In addition to their natural function as transport vehicles for minerals and essential amino acids for newborns, native casein micelles can also be used as nanocarriers for bioactive substances such as lysozyme. [2,3] In relation to this, we investigate whether the functionality of casein micelles as nanocarriers can still be maintained, or improved, by enzymatic and/or non-enzymatic modification. The following study focuses first on the investigation of the (internal) structure of modified casein micelles. For this purpose, casein micelles obtained from raw milk were enzymatically cross-linked using microbial transglutaminase (mTG) [EC 2.3.2.13] or non-enzymatically cross-linked using methylglyoxal (MGO). Some of the modified casein micelles were treated with EDTA so that by disrupting the calcium phosphate bridges inside, uncross-linked individual caseins can be cleaved and washed out of the micelle. As a result, so-called 'casein micelle skeletons' remain, which are expected to continue to function as nanocarriers. Scanning electron microscopy images illustrate the differences in the structure of native and modified casein micelles. Furthermore, this allows identifying structural differences between the two modification methods. Investigations of the washed-out supernatant from the 'skeletons' by RP-HPLC-UV provide information on the possible composition of enzymatically and non-enzymatically modified casein micelles. These results can be used specifically for subsequent loading experiments with bioactive substances.

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[2] Livney, Curr. Opin. Colloid Interface Sci. 2010, 15

[3] Duerasch et al., J. Agric. Food Chem. 2020, 68

**Keywords:** casein micelles, modification, nanocarrier

**P29****SELECTED MAILLARD REACTION PRODUCTS AND THEIR YEAST METABOLITES IN COMMERCIAL WINES****Anna-Lena Kertsch<sup>1\*</sup>, Juliet Wagner<sup>2</sup>, Thomas Henle<sup>3</sup>**<sup>1 2 3</sup> TU Dresden, Professur für Lebensmittelchemie, Dresden, Germany

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During beer and wine production, Maillard reaction products (MRPs) are formed, which have a particular influence on the taste and aroma of the fermented beverages. [1,2] Compared to beer, very little is known about individual Maillard compounds and especially corresponding yeast metabolites in wine. In this context, Amarone wines are of particular interest. For their production, a grape drying step is incorporated, resulting in the concentration of the ingredients as well as possible Maillard reaction processes. [3] In this study, 36 selected wines (Amarone-, Ripasso-, Bordeaux-, and Red- and White wines from Valpolicella and Germany) were analyzed by HPLC-UV and GC-MS concerning the amounts of 3-deoxyglucosone (3-DG), 3-deoxygalactosone (3-DGal), methylglyoxal (MGO), glyoxal (GO), 5-hydroxymethylfurfural (HMF) and furfural (FF), 3-DG was found to be the dominant compound with values ranging between 3.3 mg/L and 35.1 mg/L. The contents of 3-DGal, MGO, GO, HMF, and FF were in a single digit mg/L range. In addition to the MRPs, the yeast metabolites originating from 3-DG (3-deoxyfructose (3-DF) and 3-deoxy-2-ketogluconic acid (3-DGA)), 2,5-bis(hydroxymethyl)furan (BHMF) and 5-formyl-2-furancarboxylic acid (FFCA), both formed from HMF, as well as the FF metabolites furfuryl alcohol (FFA) and furan-2-carboxylic acid (FCA) were detected and some of them quantitated in wines for the first time. The amounts were between 0.1 mg/L and 53.5 mg/L with especially high contents of the oxidation products. Differences between Red- and White wines indicate that enological parameters like grape variety, production method and ageing may have an influence on the MRP contents in wines. No significant difference was found between Amarone wines and the other Red wines studied in terms of their MRP contents.

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**P30****STABILITY OF CAROTENOIDS AND PHENOLIC COMPOUNDS DURING STORAGE AND PROCESSING OF SELECTED VEGETABLES**

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Vegetables are highly important for humans as they provide essential nutrients, vitamins, and minerals for optimal health and well-being. Those phytochemicals include carotenoids, tocopherols, phenolic compounds and much more. Both carotenoids and phenolics play a crucial role in the human diet, providing numerous health benefits and contributing to overall well-being. They act as powerful antioxidants, protecting the body against harmful free radicals and oxidative stress. Additionally, some carotenoids, such as  $\beta$ -carotene, are converted into vitamin A in the body, which is essential for maintaining healthy vision, supporting the immune system, and promoting proper growth and development. Vegetables can be consumed as fresh or processed. In order to prolong the shelf-life of vegetables, they undergo processing that inactivates enzymes or pathogens. Moreover, some vegetables can be consumed only after processing. These can lead to various changes, not only affecting sensory properties but also having significant implications for nutrition. These changes may involve the degradation or oxidation of carotenoids and phenolic compounds, resulting in their loss, while in some cases, their content may increase due to their release from bound forms or cells. The presented study aimed to monitor and analyze the stability of carotenoids and phenolic compounds during long-term storage and processing (boiling, frying, baking, freeze-drying) of carrots, tomatoes, sweet potatoes, green peas and spinach, with the goal of understanding the trends in the changes observed in these vegetables. Carotenoids and phenolic profiles were monitored by combining quantitative and qualitative determination. Quantitative determination of carotenoids included a hydrolysis step and was realized employing HPLC-DAD. Monitoring of carotenoid degradation products that may form during processing was performed employing UHPLC-MS. Phenolic profiles and stability were also monitored employing UHPLC-MS including quantification of the major presented compounds.

**Keywords:** carotenoids, phenolic compounds, monitoring

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## P31 APPROACHES TO MINIMIZE ACRYLAMIDE IN OXIDIZED CALIFORNIA STYLE OLIVES

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Acrylamide (AA) is an undesirable process contaminant classified by IARC as a probable human carcinogen (Group 2A). According to an EFSA opinion, the dietary intake of AA should therefore be reduced [1]. Hence, AA benchmark levels (BML) in food and mitigation measures were established in Commission Regulation (EU) 2017/2158 [2]. In addition to foods such as potato chips and French fries, which are commonly associated with AA, olives have also been identified by EFSA as a potential source of AA [1]. As there are insufficient data and thus no BML for olives so far, olives have been included in the Commission Recommendation (EU) 2019/1888 [3]. AA is formed during food processing primarily via the Maillard reaction. During this process, the free amino acid asparagine reacts with reducing sugars, particularly at temperatures above 120°C. In olives, an alternative formation pathway via precursors from fat degradation is probably responsible for AA formation [4]. The results of a German nationwide monitoring project in 2021 showed that significant amounts of AA were found in blackened "California style" olives. In contrast, green and naturally ripened black olives contained rather low AA amounts [5]. The widespread in AA levels in different olive varieties is attributed to differences in production methods. Raw olives have a very bitter taste, which is removed by treatment with brine or lye baths. During the production of "California style" olives, the oxidation of phenolic compounds is stimulated by an additional supply of air, which gives the olives their characteristic black colour. It is assumed that the subsequent sterilization in combination with the oxidation process is responsible for the formation of the high AA amounts [4]. Numerous olive samples from the retail sector were analysed at CVUA Stuttgart in 2019 to 2023, and their AA contents confirmed the results of the monitoring project. In addition, indications were found that there might be a relationship between the AA content in oxidized olives and the packaging and preservation that was used. In some samples of oxidized olives sold in plastic containers and/or to which preservatives had been added, significantly lower AA levels were detected. It can be assumed that these samples, in contrast to olives in jars, were not subjected to sterilization at high temperatures. Lower temperatures during heat preservation, also in combination with the use of preservatives, could thus represent a possible measure to mitigate the AA content in oxidized olive products.

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[2] Commission Regulation (EU) 2017/2158 of 20 November 2017.

[3] Commission Recommendation (EU) 2019/1888.

[4] S. Charoenprasert, A. Mitchell, J. Agric. Food Chem. 2014, 62, 8716.

[5] Berichte zur Lebensmittelsicherheit, BVL Report, 2021, 149.

**Keywords:** acrylamide, olives, mitigation measures, California style olives, oxidized olives



**P32****ASSESSMENT OF THE PATULIN CONTAMINATION LEVEL IN SELECTED APPLE-BASED PRODUCTS AVAILABLE IN RETAIL IN BELGIUM****Emmanuel K. Tangni<sup>1\*</sup>, Julien Masquelier<sup>2</sup>, Els Van Hoeck<sup>3</sup>**<sup>1 2 3</sup> Sciensano, Brussels, Belgium

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Apple and apple derivatives (e.g., juices, puree) are the most important foodstuffs contaminated with patulin (PAT) in the human diet. To routinely monitor these foodstuffs and ensure that the PAT levels are below the maximum permitted levels, a method using liquid chromatography combined with tandem mass spectrometry (LC-MS/MS) has been developed. Afterwards, the method was successfully validated, reaching quantification limits of 1.2 µg/L for apple juice, cider and 2.1 µg/kg for puree. Recovery experiments were performed with samples fortified with PAT in the range of 25-75 µg/L for juice/cider and 25-75 µg/kg for puree. Results showed overall average recovery rates of 85% (RSDr=13.1%) and 86% (RSDr = 2.6%) with maximum extended uncertainty (U<sub>max</sub>, k=2) of 34 and 35% for apple juice/cider and puree, respectively. Next, the validated method was applied to 103 juices, 42 purees and 10 ciders purchased on the Belgian market in 2021. PAT was not found in the cider samples, but it was present in 54.4% of the tested apple juices (up to 191.1 µg/L) and 7.1% of the puree samples (up to 35.9 µg/kg). When comparing the results to the maximum levels set by Regulation EC n° 1881/2006 (i.e. 50 µg/L for juices and 25 µg/kg for puree, for adults and 10 µg/kg for infants and young children), exceedances were observed in five apple juices and one puree sample, for infants and young children. Using these data, the potential risk assessment for consumers is suggested and the quality of apple juices and purees sold in Belgium needs further regular surveillance.

**Keywords:** mycotoxins; patulin; apple; juices; puree

## **P33**

# **EVALUATION OF SEMICARBAZIDE LEVELS IN SAUDI FLOUR PRODUCTS**

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Background: Azodicarbonamide (ADA) is an approved food additive in Saudi Arabia as a dough improver. During heating process, ADA undergoes chemical reactions that results in the formation of a byproduct called semicarbazide (SEM). There is a concern about the safety of SEM containment as in vitro studies report risk of endocrine disturbance as well as tumor synthesis in rats. Given that there are no studies investigating this issue in Saudi, our objective is to assess the presence of SEM in local bread products.

Methods: This is a cross sectional study for bakery products produced by factories in Saudi Arabia. SFDA food registration data base was used to determine the number of bread factories to be included. After verifying bread's country of origin, imported products as well as unavailable products in the market were excluded. The final estimate for the sample was 27. All samples were logged in the Laboratory Information Management System (LIMS) and analyzed directly after reception through Liquid Chromatography-Mass Spectrometry (LCMSMS), in order to avoid sample deterioration.

Results: A total of 27 bread samples were collected. 18 (66%) of them were from the central region, and 9 (33%) were from the western region. An equal number of white toast and flat bread were analysed (8, 29%) for each, and 2 (7%) were Sandwich rolls, and the remaining 6 (22%) were other types of bread. As a result, no value of SEM was detected in our samples.

**Keywords:** semicarbazide, azodicarbonamide, food additives, safety

**P34****CHEMICAL INFORMATICS AND AI ALGORITHMS FOR DETECTING UNKNOWN CHEMICAL CONTAMINANTS IN FOOD SAMPLES USING LC-MS/MS****Sarah Al-Subaie<sup>1\*</sup>**<sup>1</sup> Saudi FDA, Riyadh, Saudi Arabia

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Detecting and identifying unknown chemical contaminants in food samples is a critical task to ensure food safety and public health. Traditional methods of analyzing food samples are time-consuming and often require prior knowledge of contaminants to use standards. In this poster, we present the newest approach for reference laboratories in the Saudi Food and Drug Authority (SFDA). This approach combines chemoinformatics and artificial intelligence (AI) algorithms to detect unknown chemical contaminants in food samples using liquid chromatography and tandem mass spectroscopy (LC-MS/MS). The SFDA has invested its efforts in scientific research to develop such a method based on AI algorithms, i.e., "supervised learning" using PCA. In order to carry out its oversight role as one of the most important regional organizations in responding to emergency cases. The important idea is the ability of this method to rapidly detect contaminants in different types of foods using an easy and fast extraction method (we generally relied on the extraction of metabolic materials to obtain all the contents of the sample). The proposed method involves simple and straightforward preparation of the food sample, followed by injection into LC-MS/MS and processing of the raw data. We obtained the data from the instrument using the MzMine software, then identified the compounds that appeared to us using spectral libraries such as MzCloud. When there are large amounts of similar data, we use controlled learning algorithms (machine learning -AI) to classify this data and then upload it to the GNPS network to identify metabolites and predict the most important metabolic compounds we found in the sample without using standard materials. This approach was tested on several food matrixes, such as red meat, chicken, milk, honey, herbs, and water. We were able to qualitatively detect the most important pollutants, such as mycotoxins, heavy metals, antibiotics, and hormones. The SFDA is working to establish special libraries for the chemical contaminants it monitors in the Kingdom of Saudi Arabia in food and medicine to serve as a reliable database that can be used locally and globally. In conclusion, our experiment offers a promising solution for detecting unknown chemical contaminants in food samples using a combination of chemoinformatics and artificial intelligence algorithms. This approach has the potential to revolutionize food safety testing by enabling the rapid and reliable identification of unknown contaminants, thereby protecting public health, and ensuring the safety of our food supplies.

**Keywords:** AI, chemical informatics, non-targeted analysis, food contaminants

**P35****APPLICATION OF GREEN EXTRACTION TECHNOLOGIES IN FRUITS: INNOVATIONS WITHIN THE EXCEL4MED PROJECT**

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The overarching objective of EXCEL4MED is to create an Excellence Hub in Mediterranean fruit supply chains. The project aims at identifying high-impact strategies and establish lines of resilience for producers, processors, consumers and policymakers. Overall, EXCEL4MED will offer an adaptive capability in the Mediterranean supply chain preparing for novel waste valorisation strategies, production of added value fruit products following a holistic commerce, and the implementation of green innovative technological methodologies. EXCEL4MED will develop and demonstrate the solution in Mediterranean high-value perishable food supply chains: pomegranate and citrus fruits. EXCEL4MED will develop a conceptual design and pre-planning for pilots and demonstrators of electrochemical and sonochemical technologies in the food industry. For that purpose proof of concepts and scale-up validation assessments will be developed for the pomegranate and citrus food chains. This will be achieved by consolidating academic business linkages and providing evidence for strategy building and investment. Application of green processes for improved extraction of bioactive compounds from pomegranate seed oil, citrus and pomegranate pericarps will be carried out. Experimental studies are currently carried out to characterize the bioactive compounds of different pomegranate and citrus varieties. The effectiveness of Electric Field (PEF), Ultrasound (US) and Hydrodynamic Cavitation (HC) as pre-treatments are compared to standard methods e.g., using enzymes, as well as to hybrid treatments. The bioactive compounds are extracted using standard methods, following characterization and quantification. The antioxidant capacity of the bioactive substances are determined through the radical scavenging method (DPPH), the Ferric Reducing Antioxidant Power Assay (FRAP) and the ABTS radical cation reduction test. The best-performed treatment conditions will be identified and selected. EXCEL4MED will create 4 pilot projects in Malta and Greece; 3 on added value products and 1 on valorisation. At the end of the project, partners will provide the pilot regions specific recommendations for a joint policy in food processing, taking into consideration already identified innovation strategies for smart specialization.

**Keywords:** extraction, green technologies, citrus, technology, sustainability

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**P36****EFFECTS OF FERTILIZER AND HERBICIDE APPLICATIONS ON ACRYLAMIDE FORMATION POTENTIAL OF CORN GENOTYPES**

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The food products processed at high temperatures might contain high amounts of acrylamide including those made of corn, such as breakfast cereals, chips, crackers, etc. Since acrylamide is carcinogenic, reducing its production is crucial for public health. It is formed during heating from asparagine; therefore, acrylamide formation can be mitigated by lowering free asparagine. This study aims to limit the accumulation of free asparagine in corn grains in the field by applying various agricultural factors. Four different genotypes selected from the Gene Bank of Maize Research Institute in Serbia were exposed to two different sulfur fertilizers and herbicides for two years. The applications were used either alone or in combination. The asparagine levels in different genotypes varied between 330 mg/kg and 753 mg/kg. The influence of the applications was only clearly seen in the ZP735 genotype, while there was no discernible effect in the other genotypes. Chopin Evolution, a sulfur fertilizer that is applied as ammonium sulfate, reduced the amount of asparagine by 15% in the first year and by 22% in the second year. Although the usage of herbicides is crucial for plant growth, the output remained unaffected. To observe the impact of the applied agricultural factors on acrylamide formation in the finished product, chips and crackers were baked using corn flours from selected genotype with reduced levels of asparagine (ZP735). Compared to control samples, all the chips and crackers were found to contain less acrylamide. This reduction was seen more clearly in the crackers. For example, control crackers prepared from the corn flour of the first year contained 595 µg/kg acrylamide and the application of Chopin Evolution fertilizer led to a 15 % decrease in the crackers. Crackers prepared from the corn flour of the second year treated with Chopin Evolution fertilizer and Nicosulfuron herbicide decreased acrylamide content of the cracker by 45% (333 µg/kg) compared to the control cracker (605 µg/kg). These results indicate that acrylamide levels in crackers can fall below the benchmark levels set by the EU Commission regarding agricultural practices. Acrylamide content of the chips was found to be higher than the crackers (611 - 2476 µg/kg). The use of Chopin Evolution fertilizer and Nicosulfuron herbicide in combination again caused a decrease in the acrylamide content of the chips, 32.5% for the first year. These findings demonstrated that genetic or environmental factors, as opposed to agricultural approaches, had a greater impact on asparagine synthesis in corn. However, the asparagine contents can be affected by agricultural practices to a limited extent and might lead to a decrease in acrylamide formation in processed corn products.

**Keywords:** asparagine, corn products, acrylamide

**Acknowledgement:** This study was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia (project number 451-03-9/2021-14/200040) and the Scientific and Technological Research Council of Turkey (TÜBİTAK) (project number 220N414) within the bilateral project between the Republic of Serbia and the Republic of Turkey, 2021-2023.

**P37****CHILLI PEPPERS AND POWDERS ON THE CZECH MARKET: ARE THEIR AUTHENTICITY AND SAFETY OF CONCERN?**

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Chilli peppers and the products thereof are frequently used as food ingredients all over the world. However, safety issues persist, with unauthorised and excessive pesticide residue levels having been reported for fresh chilli peppers and chilli powders. High levels of multiple residues and mycotoxins were also found in chili samples within our preliminary study. Here, we determined these levels in samples obtained from the Czech market. Assessment was done on more than 400 pesticide residues and several mycotoxins. The results for the pesticides found in fresh chilli peppers were evaluated in terms of the EU maximum residue limits (Regulation (EC) No. 396/2005), whilst the processing factor (10) recommended by the European Spice Association was utilized to evaluate the equivalent MRLs for the powders. Multiple pesticide residues, including chlorpyrifos and carbofuran, both of which are prohibited in the EU, were found in both types of samples to be over MRL limits. Some chilli peppers contained excessive levels of profenofos, with the highest level being fifty times the MRL. Furthermore, azoxystrobin and fluopyram residues exceeded MRLs even in so-called BIO powders. In addition, several types of mycotoxins, including ochratoxin A that was over the maximum limit (Commission Regulation (EC) No. 1881/2006), were found in chilli powders. Furthermore, with regard to assessing the qualitative properties of chillies, the content of capsaicinoids (the compound responsible for the stimulating properties) is of interest. Capsaicin, dihydrocapsaicin, and nordihydrocapsaicin contents were measured. The Scoville Heat Unit (SHU) was related to the above-mentioned chemical analysis of capsaicin and found to be in most cases in compliance with the label which was declared by their supplier.

**Keywords:** pesticide residue, mycotoxin, capsaicinoids, Scoville heat units, UHPLC-MS/MS

**Acknowledgement:** This work was supported from the grant of Specific university research A2\_FPBT\_2023\_065; This work was supported by METROFOOD-CZ research infrastructure project (MEYS Grant No: LM2023064) including access to its facilities.

**P38**  
**CANCELLED**

**P39****ACRYLAMIDE EXPOSURE: THE POTENTIAL OF INVOLVING THE CONSUMER IN THE RESEARCH****Szternfeld Philippe<sup>1\*</sup>, Van Leeuw Virginie<sup>2</sup>, Joly Laure<sup>3</sup>, Philippe Szternfeld<sup>4</sup>**<sup>1 2 3</sup> Sciensano, Brussel, Belgium<sup>4</sup> Philippe Szternfeld, Dworp, Belgium

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Acrylamide is a processing contaminant classified as a probable carcinogen (IARC classification group 2A). Since its discovery in 2002, extensive research has been conducted to investigate acrylamide's formation mechanism and occurrence in various food categories. EU Regulation 2017/2158 proposes benchmark limits for different food products, and discussions are ongoing at the European level to extend these limits to a broader group. This regulation also provides a toolbox of mitigation measures that can be applied to reduce acrylamide formation in the final product. These measures include aspects such as raw material selection and storage conditions to reduce the levels of acrylamide precursors, namely asparagine and reducing sugars. Recommendations regarding the cooking process are also given to the food industry and retailers, such as the product's maximum temperature and final colour. However, it is crucial to consider the role of consumers in acrylamide formation, particularly when cooking at home. Therefore, involving consumers in research becomes essential. This study aimed to understand consumers' cooking practices and their influence on acrylamide generation through their behaviour. Fifteen participants were recruited and were instructed to cook various food items (buns, croquettes, sweet potato fries) according to their usual cooking habits and using their usual cooking device (toaster/oven/fryer). Following the cooking tests, participants were asked to complete a survey that included questions about their cooking habits and the criteria they used to determine when they stopped the cooking process. Samples were then analysed in laboratory to determine acrylamide content. The study revealed that acceptable levels of acrylamide content (below EU regulation 2017/2158 benchmark limits for similar food) were generated when participants respected the recommended maximum temperature and obtained a golden colour in the cooked food. Cooking time did not significantly impact acrylamide generation within these acceptable levels. No significant differences in acrylamide content were observed between different cooking modes (fryer/oven). Interestingly, participants identified the golden colour as the primary criterion for determining when to stop cooking, aligning with the colour specified in EU regulation 2017/2158. These results may be used to determine better the consumer exposure to acrylamide. These results show the potential of such studies. The involvement of consumers in scientific research allows for a complete knowledge of acrylamide, from raw materials to the final consumer. Repeating this study on a larger scale with a more representative sample of the population may provide valuable insights to assess better the risk associated with acrylamide consumption.

**Keywords:** acrylamide, food, exposure, consumer**Acknowledgement:** Federal Public Service Health, Food Chain Safety and Environment for the financial support provided



**P40****MIGRATION OF NON-INTENTIONALLY ADDED SUBSTANCES FROM MULTI-LAYER LAMINATES USED FOR STAND-UP POUCHES**Lukas Vapenka<sup>1\*</sup>, Eliska Vaclavikova<sup>2</sup>, Lenka Votavova<sup>3</sup>, Vojtech Kruzik<sup>4</sup><sup>1 2 3 4</sup> University of Chemistry and Technology Prague, Prague, Czech Republic

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One of the problems encountered in the food industry is the migration of low molecular weight substances from food contact materials (FCM). Some substances may be included in the group of substances referred to as non-intentionally added substances (NIAS). The substances are mainly impurities in intentionally used materials and additives, intermediates and products of reactions occurring during the FCM life cycle, or decomposition products. NIAS may have the potential to negatively affect the organoleptic properties of packaged foods and may also pose a health risk in some cases. This work dealt with the identification and semi-quantitative determination of the migration level of NIAS contained in multilayer laminated materials currently widely used for stand-up pouches. Migration tests were performed on packaging materials made of layers of polyethylene, polypropylene, polyethylene terephthalate, polyamide and aluminium, with appropriate adhesives and printing into a food simulant (95% ethanol) at a temperature of 60 °C for 10 days. Identification and semi-quantification were performed by gas chromatography with mass spectrometry. A total of 10 samples of infant food packaging materials were analysed and 13 different NIAS were identified. The migration levels of these substances ranged from 0.01 to 2.51 mg/kg of food simulant. The highest level of migration was determined for tris(2,4-di-tert-butylphenyl)-phosphate, which was also the most frequently migrating NIAS. This compound is formed by oxidation from the additive tris(2,4-di-tert-butylphenyl)-phosphite commonly used for antioxidant stabilization of materials based on polyolefin polymers. Alternatively, 2,4-di-tert-butylphenol itself, which was also one of the identified NIAS, is released from the phosphate form by hydrolysis. Other NIAS identified were hydrocarbons (alkanes C16 to C25) and a cyclic dimer of caprolactam (1,8-Diazacyclotetradecane-2,9-dione), which is formed by cyclization of the monomer used for production of polyamide. The last group of NIAS were various esters (linear and cyclic) of glycols with adipic and phthalic acid, which are formed by esterification of these substances originating from additives, adhesives and the polymer itself. The established migration levels do not exceed the selected limits, except for the last group of esters, for which the risk assessment is problematic and questionable.

**Keywords:** non-intentionally added substances, food contact material, migration, risk assessment**Acknowledgement:** This work was supported from the grant of Specific university research grant No A1\_FPBT\_2023\_004.

**P41****IMPACT OF PULSED ELECTRIC FIELD ON FUSARIUM PATHOGENS AND THEIR MYCOTOXINS IN BARLEY AND DURING MALTING****Nela Prusova<sup>1\*</sup>, Adam Behner<sup>2</sup>, Marcel Karabin<sup>3</sup>, Jana Chrpova<sup>4</sup>, Milena Stranska<sup>5</sup>**<sup>1 2 3 5</sup> University of Chemistry and Technology Prague, Department of Food Analysis and Nutrition, Prague, Czech Republic<sup>4</sup> The Crop Research Institute, Prague, Czech Republic

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Contamination of cereals and cereal-based products by micromycetes and their secondary toxic metabolites, mycotoxins, is a global concern, emphasizing the need for strategies to minimize their occurrence throughout the food production chain. Various physical, chemical, and biological approaches can be employed pre- or post-harvest to address this issue. Pulsed electric field (PEF) technology offers a novel approach and therefore in our study, we explored its application for both direct reduction of mycotoxin content and also indirect mycotoxin reduction during malting by targeting their fungal producers. Our objective was to investigate the impact of PEF under various conditions (pre-steeping time of barley before PEF, electrolyte medium, input voltage, current, wavelength, and pulse number) on four *Fusarium* species (*F. culmorum*, *F. graminearum*, *F. sporotrichioides* and *F. poae*) and 16 *Fusarium* and *Alternaria* mycotoxins (deoxynivalenol, deoxynivalenol-3-glucoside, 3-acetyldeoxynivalenol, 15-acetyldeoxynivalenol, nivalenol, zearalenon, neosolaniol, diacetoxyscirpenol, HT-2 toxin, T-2 toxin, enniatins A, A1, B, B1, beauvericin and tentoxin) in barley and also during malting. Determination of mycotoxins was performed using ultra-high performance liquid chromatography and high-resolution tandem mass spectrometry (U-HPLC-HRMS/MS) and *Fusarium* species analysis by real-time polymerase chain reaction (RT-PCR). Our results demonstrated significant PEF-induced reductions in trichothecenes, zearalenone, enniatins, beauvericin, and tentoxin, up to 31%, 48%, 84%, 36% and 46%, respectively, in the dry matter of barley. Additionally, the presence of reaction products resulting from hydrolysis or oxidation were observed for enniatin A1, enniatin B1, zearalenone, and tentoxin. Regarding experiments on the effect of PEF on *Fusarium* species and mycotoxins during malting, significant reductions were observed across all species under the optimized PEF conditions. The most notable decrease was observed in the case of *F. sporotrichioides* and *F. poae*, with their content in PEF-treated malt approximately four- and six-fold lower, respectively, compared to the control malt. The reduction in these micromycetes corresponded to a significant decrease of relevant mycotoxins, in particular, the content of type A trichothecenes and enniatins. Furthermore, the influence of PEF treatment on mycotoxin biosynthesis and enzymatic activity during malting was observed, depending on the intensity of PEF and the degree of pre-steeping of the barley before treatment.

**Keywords:** Pulsed Electric Field, mycotoxins, *Fusarium* species, high resolution mass spectrometry, malting

**Acknowledgement:** This work was financially supported by the Czech Science Foundation (project No 20-14649S). This work was also supported by METROFOOD-CZ research infrastructure project (MEYS Grant No: LM2023064) including access to its facilities, and a grant for specific University research - A1\_FPBT\_2023\_002.

**P42****EVALUATION OF THE SYNTHETIC SWEETENERS USAGE IN BEVERAGES AND TABLE SWEETENERS SOLD IN CZECH REPUBLIC**

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The usage of synthetic sweeteners in beverages has become increasingly popular as a way to provide sweetness without the added calories of traditional sugar. These sweeteners, such as aspartame, sucralose, and saccharin, offer a low-calorie alternative for individuals seeking to reduce their sugar intake or manage weight. However, there are risks associated with their consumption. For example, food adulteration with sweeteners may be done by adding synthetic sweeteners to the food in order to lower the energetic value without scaring the consumer with a food additive. Another example can be the substitution of natural steviol glycosides (E960) with synthetic ones to lower the price of the product because synthetic sweeteners can be up to 5 times cheaper than steviol glycosides. In addition, undeclared synthetic sweetener aspartame (E951) in food products can lead to potential health problems for people suffering from phenylketonuria. The usage of sweeteners in the European Union is strictly regulated in Regulation (EC) No 1333/2008 by setting limits for each approved sweetener. Therefore, in order to understand the real situation on the market, sets of energy drinks and soft drinks as well as table sweeteners were analysed for the presence of 7 synthetic sweeteners employing UHPLC-MS. The obtained results were later compared with the established limits as well as consumption of individual sweeteners was assessed.

**Keywords:** sweeteners, beverages, table sweeteners

**Acknowledgement:** This work was supported by METROFOOD-CZ research infrastructure project (MEYS Grant No: LM2023064) including access to its facilities. Also, this work was financial supported from a specific university research A1\_FPBT\_2023\_002.

**P43****CHARACTERIZATION AND DISCRIMINATION OF X-RAY IRRADIATED CAMEMBERT CHEESE BY UHPLC-Q-ORBITRAP-MS LIPIDOMICS AND CHEMOMETRIC ANALYSIS**

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Camembert is a bloomy rind cheeses of French origin, obtained through the activity of *Penicillium camemberti* [1]. This type of cheese is included in the ready-to-eat (RTE) food category, with potential growth risk of *Listeria monocytogenes* and thus it has a short shelf-life depending on the production process, packaging, storage and distribution conditions [2]. In this context, among non-thermal technologies, food irradiation represents a clean and safe valid alternative to preserve the hygienic quality of food and to extend the shelf-life of several foodstuffs including soft cheeses. During this treatment, foods are exposed to ionizing radiations, such as X-rays, able to destroy or inactivate pathogenic and spoilage microorganisms [3]. In this study, X-rays irradiation at a dose of 3.0 kGy was applied to Camembert cheese and possible modifications on lipid composition induced by this treatment were evaluated. Analysis of the lipidome was performed by Ultra-High Performance Liquid Chromatography coupled with Quadrupole Orbitrap Mass Spectrometry (UHPLC-Q-Orbitrap-MS) and data were processed by multivariate statistics. The results highlighted that the qualitative lipid fingerprint of Camembert, characterized by 479 compounds, categorized into 16 different subclasses, did not change after irradiation, confirming that the X-ray dose employed in this investigation did not lead to the formation of new lipid molecules due to this treatment. On the other hand, differences in the abundance of specific lipids were observed and considered for chemometric analysis. Partial Least Squares-Discriminant Analysis (PLS-DA) and Linear Discriminant Analysis (LDA) were applied showing high discriminating ability with excellent values of accuracy, specificity and sensitivity. Through the PLS-DA and LDA models, it was possible to select 40 and 24 lipids, respectively, including 3 ceramides (Cer), 1 hexosyl ceramide (HexCer), 1 lysophosphatidylcholine (LPC), 1 lysophosphatidylethanolamine (LPE), 3 phosphatidic acids (PA), 4 phosphatidylcholines (PC), 10 phosphatidylethanolamines (PE), 5 phosphatidylinositols (PI), 2 phosphatidylserines (PS), 3 diacylglycerols (DG) and 9 oxidized triacylglycerols (OxTG) as potential markers of treatment useful in food safety control plans. The results confirm that the lipidomic approach adopted is effective in implementing the knowledge of the effects of X-ray irradiation on food, also evaluating all safety aspects.

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**Keywords:** food irradiation, UHPLC-Q-Orbitrap-MS, lipidomics, chemometric analysis, Camembert cheese

**Acknowledgement:** This work is supported by funding from the Ministero della Salute (Rome, Italy) - Project code GR-2018-12367064.

## **P44 DETERMINATION OF POLYCYCLIC AROMATIC HYDROCARBONS IN TRADITIONALLY SMOKED MEAT PRODUCTS AND CHARCOAL GRILLED MEAT IN CYPRUS**

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The study highlights the investigation of the presence of polycyclic aromatic hydrocarbons (PAHs) in traditionally smoked meat products and charcoal grilled meat. Specifically, the levels of benzo(a)anthracene (BaA), chrysene (Chr), benzo(b)fluoranthene (BbF) and benzo(a)pyrene (BaP) were determined for 363 samples of smoked sausage, bacon, pork (lountza) and ham (chiomeri), grilled pork and poultry produced in Cyprus. The method is based on a saponification and liquid extraction step, followed by cleanup of the extract and finally, the determination of PAHs using high performance liquid chromatography with fluorescence detector. Most of the samples analysed (96%) were contaminated with at least one PAH. In these contaminated samples, 12% of the smoked products and 15% of the grilled meat samples exceeded the maximum levels of the EU legislation. The highest PAH concentrations were found in samples with higher fat content and longer smoking or cooking time.

**Keywords:** polycyclic aromatic hydrocarbons, charcoal grilled meat, traditionally smoked meat, HPLC-FLD

**Acknowledgement:** DiTECT project (European Union's Horizon 2020 research and innovation programme under grant agreement No 861915)

**P45****SORBATE ADDITION IN DOCE DE LEITE: EFFECT OF PH AND THERMAL PROCESSING**

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Doce de leite (DL) is a dairy product extensively produced and consumed in South America. Upon good manufacturing practice, contamination can be prevented due to the high amount of carbohydrate and solids levels. In that way, no preservatives should be needed in DL. However, due to the plurality of manufacturing sites and scales, air-borne-, packaging- and utensils contamination with yeasts and molds cannot be excluded (mainly post-processing contamination by consumer manipulation). Therefore, the addition of preservatives as sorbic acid (H-Sorb) and its salts, such as potassium sorbate (K-Sorb), are common chemical substances widely used in the DL industry and are also permitted by the Southern Common Market (Mercosul) regulation. Nevertheless, no previous study has determined the influence of preservative addition in the product during the manufacturing processes, nor if the thermal effect could be impacting in the final concentration (loss due to steam entrainment or thermal degradation). Thus, the present study determined and quantified H-Sorb, added as K-Sorb, in DL production, considering the addition of preservative before or after thermal treatment. The analyzes were carried out in a reverse phase high pressure liquid chromatography (RP-HPLC), in accordance with the ISO 9231| IDF 139 method. The effect of pH matrix (2.5, 4.8 and 7.0) on H-Sorb content was also investigated by evaporation model solutions. The analytical method fulfilled the acceptance criteria: selectivity (single peak), linearity (3.1 - 50.0 µg/mL; R<sup>2</sup> = 0.9977), precision (CV = 0.66% to 2.04%), accuracy (recoveries = 93.6% to 101.9%), detection limit (0.72 µg/mL) and quantification limit (2.17 µg/mL). In DL samples, the accuracy of H-Sorb determined by the addition of K-Sorb before/after heat treatment varied from 88.8% to 96.5%. Furthermore, the results showed that the addition of preservative, either at the beginning of the process (before the concentration step) or at the end of the latter step (evaporation), does not interfere quantitatively in its final concentration. Hence, H-Sorb seems not to evaporate at the normal conditions of pH, temperature and time applied during DL manufacture (pH ~6.9, 100 min and ~105°C). Experiments on model aqueous solutions, with different pH values, showed that at low pH values (≤ pK<sub>a</sub> of H-Sorb), the prevailing acid form seems to be partially lost during the evaporation process. In DL production, the pH of the ingredients' blend is close to neutrality (hence K-Sorb is the prevailing moiety) and the processing conditions applied are not able to reduce the pH to values in which reactions could occur in a way that would convert K-Sorbate into H-Sorb. In addition, H-Sorb analysis from commercial samples (n=10) showed that their concentrations (265 - 572 mg/kg) were within DL regulation; there was also one sample that claimed to have no added preservative, but showed 265.8 ± 12.1 mg of H-Sorb per kg DL.

**Keywords:** dulce de leche, sorbic acid, evaporation, industry, milk

**Acknowledgement:** This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior -Brasil (CAPES) - Finance Code 001. CNPq (403602/2020-3) and Alexander von Humboldt Foundation.

**P46**  
**CANCELLED**

**P47**  
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**P48****A JOINED-UP APPROACH TO THE IDENTIFICATION, ASSESSMENT AND MANAGEMENT OF EMERGING FOOD SAFETY HAZARDS AND ASSOCIATED RISKS (FOODSAFER)****Rudolf Krska<sup>1</sup>, [Alexandra Schamann](#)<sup>2</sup>, Alexandra Malachova<sup>3</sup>, Martin Wagner<sup>4\*</sup>**

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In Europe, foodborne hazards, including bacteria, parasites, toxins (chemical hazards), and allergens, cause approximately 23 million cases of illness and 5,000 deaths each year. Food safety management systems established over the past decades in European farmers and food businesses, as well as European food safety governance, need to be adapted to make them more robust in the face of changes that affect our global food systems. To achieve such an adaptation, the EU-funded project FoodSafeR was recently established, which brings together academic working groups, industry, SMEs, and policymakers. In total, the project unites a consortium of 18 organisations from 14 European countries. FoodSafeR has the goal to design, develop and test the building blocks of an innovative, pro-active, and holistic food safety management system with a focus on emerging risks at its core. The project embodies integrated approaches to food safety risk identification assessment and management in a comprehensive suite of future-oriented frameworks, tools, methods, strategies, models, guidance, and training materials. An open and accessible digital hub will be set up as a 'One-Stop-Shop' vehicle targeted at risk managers and assessors, food safety authorities and the relevant actors and stakeholders operating in the European food system. FoodSafeR will contribute to sustainable, healthy, and inclusive food systems, delivering co-benefits for climate mitigation and adaptation, environmental sustainability and circularity, sustainable and healthy nutrition, safe food consumption, reduction of food poverty, the inclusion of marginalised people, empowerment of communities, and flourishing businesses. In addition, the project will contribute to preventing food safety incidences which are caused by biological and chemical hazards in the European food system.

**Keywords:** food safety, emerging risks, future food systems

**P49****CHEMICAL CHARACTERIZATION OF WHITE ACQUALAGNA TRUFFLE (TUBER MAGNATUM PICO) BY UHPLC-QTOF-MS, GC-MS, ICP-MS AND SENSORY ANALYSIS**

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White Truffle is the most expensive edible hypogeous fungi that grow in symbiosis with the roots of the host plants, and it is appreciated for its extraordinary taste and smell. Global demand for truffles has increased significantly in recent years, and the requirement for traceability of truffle origin has become an important aspect of the truffle trade. Therefore consumers are interested in regional and authentic food products and the specification of the geographical origin of the European truffles is particularly relevant [1]. Considering the scarce scientific literature, this work represents the first study with the characterization of the Acqualagna white truffle. A new untargeted metabolomics approach, using UHPLC-Q-TOF allowed us to identify 456 potential metabolites. Analysis were performed both in positive and in negative polarity acquisition modes and mass spectra were processed using MS-DIAL, which provides information about mass accuracy, isotopic pattern, and spectral matching with the library of each compound. More than 60 compounds reported a total identification score match greater than 90%. Moreover, the elemental composition was investigated through ICP-MS and revealed a higher content of K, P, S, Ca and Mg, representing 97% of the elements investigated. The correspondence between the elements present in the water released from the soil and in the truffle was also taken into consideration to detect which elements of the soil have been assimilated. The aroma profile of fresh truffles was performed using the gas chromatography-mass spectrometry system (GC-MS) and was compared with the sensorial analysis. The results of the volatile profile consist mainly of Bis(methylthio)methane (78.72%) and other 50 minor constituents. This study identified for the first time the natural mixture of key odorants and showed the correspondence between odorants and molecules of the white Acqualagana truffle. In conclusion, this is the first step to characterize and determine the origin traceability of the Acqualagna white truffle, a local food defined by its unique composition.

[1] Segelke, T. et al. (2020). Food authentication: Species and origin determination of truffles (*Tuber* spp.) by inductively coupled plasma mass spectrometry and chemometrics. *Journal of Agricultural and Food Chemistry*, 68(49), 14374-14385.

**Keywords:** white truffle, UHPLC-QTOF-MS, GC-MS, ICP-MS, sensory analysis  
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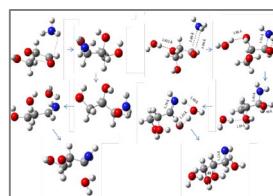
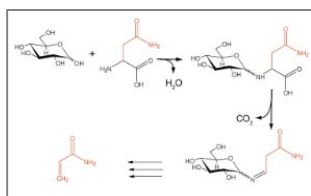
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